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Cardiac Effects of Antiretroviral Therapy in HIV-Negative Infants Born to HIV-Positive Mothers: The NHLBI CHAART-1 Cohort Study

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Abstract

Objective—To investigate the possible effects of antiretroviral therapy (ART) *in utero* on cardiac development and function in HIV-negative children.

Background—ART reduces vertical HIV transmission. Long-term cardiotoxicity after *in utero* exposure to ART is unknown in children but has occurred in young animals.

Methods—Using a prospective multi-site cohort study design, we compared echocardiograms taken between birth and 24 months in two groups of HIV-negative infants of HIV-positive mothers: 136 infants exposed to ART (ART+) and 216 unexposed infants (ART−).

Results—Mean LV mass Z-scores were consistently lower in ART+ girls than in ART− girls: differences in mean Z-scores were −0.46 at birth (P=0.005), −1.02 at 6 months (P<0.001), −0.74 at 12 months (P<0.001), and −0.79 at 24 months (P<0.001). Corresponding differences in Z-scores for boys were smaller: 0.13 at 1 month (P=0.42), −0.44 at 6 months (P=0.01), −0.15 at 12 months (P=0.37), and −0.21 at 24 months (P=0.21). Septal wall thickness and LV dimension were smaller than expected in ART+ infants, but LV contractility was consistently about 1 SD higher at all ages (P<0.001). In ART+ infants, LV fractional shortening was higher than in ART− infants; girls showed a greater difference.

Conclusion—Fetal exposure to ART is associated with reduced LV mass, LV dimension, and septal wall thickness Z-scores and increased LV fractional shortening and contractility up to age 2 years. These effects are more pronounced in girls than in boys. Fetal ART exposure may impair myocardial growth while improving depressed LV function.

Keywords

Pediatric; HIV; Antiretroviral Therapy; Cardiomyopathy

INTRODUCTION

Abnormalities of left ventricular (LV) structure and function are associated with human immunodeficiency virus (HIV) infections,[1–6] possibly with antiretroviral therapy (ART),[7–10] and even mild abnormalities independently predict mortality in HIV-infected children.[3–5] HIV-exposed but negative newborns show depressed LV contractility that persists at 5 years of age, whereas HIV-positive newborns experience chronic and progressive abnormalities in LV structure and function.[6]

Mitochondrial abnormalities have been reported in animals and children exposed to ART with inconsistent results.[7–12] Such abnormalities could cause cardiomyopathy in exposed children although the clinical implications of mitochondrial abnormalities in HIV-negative infants exposed to ART are uncertain.[7–12]

Nearly 10,000 ART− and HIV-exposed but negative infants are born annually in the U.S. but are not routinely followed for suspicion of heart disease.[8–10, 13]

To study the cardiac effects of perinatal exposure to ART, we compared two cohorts of HIV-negative children born to HIV-positive mothers.

METHODS

In the CHAART-1 cohort study,[14] 166 HIV-negative infants exposed to ART *in utero* were prospectively identified between June 2003 and January 2006 from the WITS study of 2842 infants born to HIV-positive women.[15] CHAART-1 participants came from 5 of the 7 WITS sites; 4 also participated in the P^2C^2-HIV study. CHAART-1 participants were enrolled in WITS and were 2 years old or less so that primary endpoints could be measured at age 2. CHAART-1 recruited children were followed until loss to follow-up, the child withdrew, December 2006, or whichever came first.

Maternal exclusion criteria for CHAART-1 children included diabetes, phenylketonuria, a Mendelian or chromosomal defect, a heart defect requiring medication or surgery, or pregnancy exposures to chemotherapy, radiation, or drugs associated with heart disease in offspring.

The P^2C^2-HIV study prospectively described cardiovascular disorders in children born to HIV-positive mothers enrolled between 1990 and 1994 at 6 centers.[16] Of the 463 HIV-negative infants enrolled during gestation or within 28 days after birth, 262 had no *in utero* or postnatal ART exposure and were the non-ART-treated controls. The P^2C^2-HIV study showed that independently of ART, HIV-negative children born to HIV-positive women have abnormal LV structure and function, making them an essential control group when assessing the impact of ART therapy in the CHAART-1 study.[6]

CHAART-1 sites received IRB approval to collect serial echocardiograms. Written informed consent was obtained from parents or guardians.

Prospective data were collected from the CHAART-1 and P^2C^2-HIV datasets. Cardiac function was evaluated through serial echocardiograms starting at birth or the visit closest to CHAART-1 enrollment and ending on December 31, 2006. Echocardiograms were scheduled at 6 intervals from 1 to 48 months of age. The corresponding P^2C^2-HIV collection was scheduled at 14 intervals from birth to 54 months. This report analyzes echocardiographic data for participants up to age 2 years.

The P^2C^2-HIV echocardiogram acquisition protocol was followed.[2,14] Echocardiographic data were digitized by the same independent, blinded cardiologist who had measured the previous P^2C^2-HIV and Boston control echocardiographic data.[17]

Statistical Methods

CHAART-1 compared two primary echocardiographic endpoints at age 2 years with respect to *in utero* ART exposure: LV mass and fractional shortening. Alpha was set at 0.025 to correct for multiple comparisons. All other analyses were carried out at the 0.05 alpha level and should be considered as suggestive findings in need of future confirmation.

General linear models were used to determine whether ART independently predicted changes in these 2 measures and to identify any ART-covariable interactions. Serial echocardiographic measurements from birth to 2 years were analyzed using generalized estimating equations.[18] Twins and infants whose maternal ethnicity was other than African-American, Caucasian, or Hispanic were excluded.

The echocardiographic measures were standardized to Z-scored deviations using Boston infants and children as the normal reference population.[2] Z-scores were used as an

adjustment mechanism to correct echocardiographic measurements for heart growth and development. Z-scores of zero correspond to an average heart parameter measurement, negative below average and positive above average. Numeric values correspond to changes in standard deviation units. The methodology is identical to the growth charts versus age for height and weight in children that are provided by the Centers for Disease Control and Prevention. As long as the comparison is made with populations who are not markedly different from the normal control reference population with regard to factors such as body-mass index, level of habitual exercise (i.e., exclusion of elite athletes), and blood pressure, no differences in Z-scores are anticipated.

Interaction terms of ART exposure with gender, ethnicity and age that were significant at 0.05 were included in the final models. Alpha at 0.05 determined statistical significance in all analyses, except for the primary endpoints (see above), because these were hypothesis-generating analyses. The effect of ART exposure on primary endpoints was analyzed without adjustments and with adjustment for sex, ethnicity, significant maternal characteristics and significant sex-ART interaction.

All analyses were performed using the SAS software package, version 8.2. (SAS Institute, Inc., Cary, N.C.)

RESULTS

Demographic and Clinical Characteristics

Data were collected from 136 ART-exposed HIV-negative infants from the CHAART-1 centers and from 216 non-ART-exposed HIV-negative infants from the P^2C^2-HIV study (Table 1).[19] Infant sex and maternal ethnicities were similar. However, the maternal CHAART-1 cohort used tobacco (22% versus 37%) and illicit drugs (15% versus 29%) less, and their infants had higher birth weights. CHAART-1 mothers experienced vaginal bleeding during pregnancy (25% versus 3%) and Cesarean-section delivery (40% versus 18%) more frequently. Immunologic studies were similar, except for a higher mean CD8 T-lymphocyte count in the P^2C^2-HIV mothers (P=0.04; Table 1). *In utero* ART exposure for the 136 CHAART-1 infants was: 6 (4%) for mono-ART, 11 (8%) for combination ART (2 or more ART drugs without a non-nucleoside reverse transcriptase inhibitor or protease inhibitor), and 119 (88%) for highly active-ART (3 or more ART drugs including a non-nucleoside reverse transcriptase inhibitor or protease inhibitor).

Data included 234 serial echocardiograms from CHAART-1 infants (mean, 1.7 per child) and 516 serial echocardiograms from P^2C^2-HIV infants (mean: 2.4 per child). Mean systolic and diastolic blood pressure Z-scores were higher in ART-exposed males compared to unexposed males (Table 1). Adjustment variables for each analysis in a Table are presented in the footnote of that Table.

Primary Endpoint Evaluations

The unadjusted LV mass Z-score for all CHAART-1 infants at age 2 years was 0.53 SD less than the P^2C^2-HIV cohort (P=0.006). Left ventricular mass and ART remained associated after adjusting for sex, sex-ART interaction (P=0.009), ethnicity, and maternal vaginal bleeding (P=0.01). Both the unadjusted and adjusted LV mass Z-scores were smaller in the CHAART-1 group when pooling across sex; most of the difference was among girls, who showed a greater reduction (Table 2).

The unadjusted LV fractional shortening Z-score for CHAART-1 infants was 0.45 SD greater than that of the P^2C^2-HIV infants (P=0.01) at age-2. For this outcome, there was no sex-ART interaction. Left ventricular fractional shortening and ART remained significantly

associated after adjusting for sex and ethnicity. Both the unadjusted and adjusted LV fractional shortening Z-scores were higher in the CHAART-1 group, with girls showing a greater but non-significant difference (Table 2).

Longitudinal Analyses of Cardiac Outcomes

Left ventricular fractional shortening and LV mass endpoints were similar to the age 2 years outcomes, except that the sex-ART interaction for LV fractional shortening was more prominent and statistically significant. The sex-ART differences for these 2 endpoints were present by age 6 months and remained at age 2 years.

Adjusted LV end-systolic dimension Z-scores were lowest in the ART-exposed group, with the greatest differences in girls (0.52 SD lower in the ART-exposed than the unexposed girls). Adjusted LV end-diastolic dimension Z-scores were lower in the ART-exposed girls at 6 months and age-2 (0.44 SD and 0.51 SD, respectively). Left ventricular contractility was 0.72 SD and 1.11 SD higher in ART-exposed boys and girls, respectively. End-diastolic septal wall thickness was 0.82 SD and 1.42 SD lower in ART-exposed boys and girls, respectively.

For the cardiac endpoints without sex-by-ART interaction, results were combined across sexes (Table 3).

Other Treatment Interactions for Cardiac Endpoints

Cardiac outcomes had statistically significant sex- or time-by-treatment interactions (Figures 1 and 2). Among CHAART-1 boys, mean LV mass was higher at 1 month and decreased over 6 months; in P^2C^2-HIV boys it increased over 6 months (Figure 1A). The CHAART-1 girls were consistently about a half of a standard deviation below the average of the P^2C^2-HIV girls (Figure 1B).

Left ventricular fractional shortening measurements for CHAART-1 boys changed from being above (but still below normal) at 1 month to approximately the same (and normal) at age 2 years when compared to P^2C^2-HIV boys (Figure 1C). For girls, the difference between the adjusted means of the two cohorts was greatest at 1 month and remained statistically significant at age-2 (0.87 SD, $P<0.001$, Table 2, Figure 1D). Left ventricular end-diastolic dimension (Figures 1E and 1F), end-systolic dimension (Figure 1G and 1H), contractility (Figures 1I and 1J) and end-diastolic septal wall thickness (Figures 1K and 1L) differed by sex and ART treatment (Table 2).

Left ventricular end-diastolic posterior wall thickness (Figure 2A), heart rate (Figure 2B), and weight-for-height Z-scores (Figure 2C) differed by cohort (Table 3). CHAART-1 infants consistently had higher LV end-diastolic posterior wall thickness, but their Z-scores remained approximately a half SD below zero ($P<0.01$). Heart rate measures were 0.18 SD higher in the CHAART-1 infants at all ages ($P=0.002$). The heart rate Z-scores in the CHAART-1 infants increased from 0.36 SD at birth to 1.06 SD at 2 years ($P<0.01$). Weight-for-height Z-scores were 0.79 SD higher in the CHAART-1 infants than in the P^2C^2-HIV infants at 1 month only ($P<0.001$) with the CHAART-1 measurements above normal and the P^2C^2-HIV measurements below normal ($P<0.01$). Both cohorts improved to normal after 1 month.

Left ventricular end-systolic posterior wall thickness (Figure 2D), end-systolic septal wall thickness (Figure 2E), and LV afterload (Figure 2F) show a qualitative (crossing) interaction between age and exposure to ART (Table 3), resulting in significant associations with ART only at certain ages. Left ventricular end-systolic posterior wall thickness measurements of both cohorts were below normal at 1 month, with higher values for the CHAART-1 cohort.

Both cohorts improved to normal and became approximately the same at age-2. End-systolic septal thickness Z-scores from the CHAART-1 cohort were 0.40 SD lower than the P^2C^2-HIV measurements during the first year ($P<0.05$). Measurements from both cohorts became approximately the same at age-2. Left ventricular afterload showed the CHAART-1 cohort starting at 1 month with an average Z-score 0.47 SD lower than the P^2C^2-HIV measurements and ending approximately the same as the P^2C^2-HIV measurements at 12 months and age 2 years.

DISCUSSION

Infants of HIV-positive mothers exposed to ART show improved LV contractility and fractional shortening during the first 2 years of life compared to a non-ART exposed cohort of HIV-negative infants born to HIV-positive mothers. However, ART exposure was also associated with reduced LV mass, septal thickness, and dimension, that were also below normal. In other settings, these 3 changes can lead to progressive LV dysfunction.[20–22] The long-term cardiac effects of fetal exposure to ART are unclear. Our findings are consistent with the hypothesis that control of myocardial growth differs from control of myocardial function and that ART exposure is associated with less myocardial growth but better myocardial function.

Higher LV contractility leads to a smaller LV end-systolic volume, a higher wall thickness and hence lowers LV wall stress at end-systole. A fall in LV wall stress reduces the stimulus to LV hypertrophy, and hence, lower LV mass is the anticipated response to higher LV contractility.

Our ART-exposed cohort had better maternal health than did the P^2C^2-HIV cohort and better infant LV contractility and fractional shortening. Left ventricular fractional shortening normalized in both groups by age 2 years, except for CHAART-1 females, and LV contractility was better in the CHAART-1 group and remained normal or slightly above normal across all visits. Adding the significant maternal variables to the final multivariate models did not change the ART exposure effects on echocardiographic parameters, suggesting that ART itself, and not improved maternal health, affects LV function either directly or indirectly. We incorrectly speculated that improved maternal health resulted in improved LV contractility and fractional shortening.[6]

As in rodents,[23,24] ART exposure inhibited myocardial growth, being associated with decreased septal thickness, LV dimension and mass. These changes suggest either an overall loss of cardiac tissue with ART exposure, an inability of the septum to grow in response to increasing body-surface area, or both. Weight-for-height was not significantly different by ART exposure with follow-up, suggesting that BSA-normalized Z-scores for these variables are appropriate.

Although our results suggest that ART is associated with early, improved LV function, longitudinal changes in other factors may lead to increasing mechanical stress, such as lower LV mass, that may result in increased LV afterload, that could compromise long-term LV function, a finding found in anthracycline-treated childhood cancer survivors.[20–22] Mechanistically, these changes could be induced by apoptosis and by decreased hypertrophic-signaling mechanisms related to ART-growth inhibiting effects, ART-associated DNA mitochondrial mutations or gene polymorphism shifts,[23–25] mechanisms similar to that for pediatric cancer survivors with doxorubicin-associated cardiomyopathy, a well-characterized example of late cardiotoxicity following an early childhood drug exposure.[26]

Perinatal myocardiocyte injury and death are significantly related to inflammation.[27] The Fetal Inflammatory Response Syndrome is associated with increased neonatal morbidity.[28] Immune Reconstitution Inflammatory Syndrome in ART-exposed HIV-infected mothers may heighten inflammation that contributes to myocardiocyte injury and reduced neonatal LV mass.[29]

Exposure to ART was also associated with higher heart rates, which have been associated with rapid progression of childhood HIV-associated disease.[30] Sustained tachycardia, as observed with ART exposure, may be cause or effect, but with secondary LV hypoplasia, may explain our LV growth findings. Mild LV restrictive disease, with a reduced capacity for LV dilation and with a secondary tachycardia, is also possible. Reduced end-diastolic volume, as the initial limiting factor to cardiac output with secondary compensatory tachycardia is certainly the anticipated response to detraining and to a restrictive cardiomyopathy, so this sequence is also plausible. Alternatively, the decreased LV growth may be secondary to a direct ART effect, such as mitochondrial DNA damage from ART-associated oxidative stress resulting in LV hypoplasia that contributes to the observed increased LV contractility and heart rate, especially in girls.[23–25]

The effects of ART exposure on LV mass, dimension, fractional shortening, and septal thickness were more pronounced in girls. These differences might predict early and advanced cardiovascular disease in girls as they age. Our findings are similar to zidovudine's effects in mice, where females were more sensitive to ART than were males.[31] Similarly, newborn girls have greater myocardial injury marker elevations than do newborn boys.[27] Doxorubicin cardiotoxicity suggests an increased vulnerability of the developing female myocardium.[20–22]

The magnitude of the Z-score differences in this study may appear to be small relative to those that guide the daily clinical decisions by cardiologists. However, their magnitude is consistent with findings in doxorubicin-treated pediatric cancer survivors,[20–22] who 20 to 30 years later have a 15-fold increase in the rate of heart failure compared with siblings and an 8.2-fold increase in cardiac mortality compared with U.S. population mortality data.[32,33]

Zidovudine and its metabolites cross the placenta and are found in the fetal heart.[34] The hypothesis that zidovudine may be associated with cardiac mitochondrial dysfunction is supported by animal studies and by limited clinical data.[35–39] Yet, zidovudine monotherapy showed no effect on the same LV structure and function measurements in the current study in either HIV-negative or positive children.[11,12] Follow-up studies of HIV-negative infants born to HIV-positive women have found neither detrimental effects of perinatal zidovudine exposure nor clinically important cardiovascular disease.[8–10,12,40–42]

Combination ART (96% of CHAART-1 infants had combination therapy) has early and marked effects on LV structure and function. In rodents, combination ART produces more mitochondrial DNA mutations than monotherapy.[23–25,31] In contrast to nucleoside reverse transcriptase inhibitors, protease inhibitors in general do not cross the placenta suggesting the effects seen likely result from nucleoside reverse transcriptase inhibitor combinations rather than protease inhibitors. Combination ART compared with a nucleoside reverse transcriptase inhibitor alone in HIV-negative children was associated with more lymphocyte abnormalities.[43] Other abnormalities in ART-exposed HIV-negative children include anemia, neutropenia, thrombocytopenia, lower CD4 and CD8-cell counts, and increased micronucleated erythrocytes.[43–45]

Study Limitations

The P^2C^2-HIV study (1991–1996) and the CHAART-1 study (2003–2006) were performed in different eras (pre-HAART and HAART, respectively). This non-concurrent design was used for ethical reasons because ART therapy could not be withheld from pregnant women. However, the cardiac protocol, clinical sites, study personnel, and the person measuring echocardiograms were largely common to both studies.

As previously noted, a potential limitation of the use of Z-scores is if the study population differs significantly from the reference population in terms of anthropomorphic measures of exercise. We found no evidence of such differences between our study population and the reference group.

HIV virulence might have increased in the decade between the studies, causing CHAART-1 measurements to deviate more from normal than P^2C^2-HIV measurements. However, the CHAART-1 mothers had no evidence of more advanced disease.

Conclusions

Fetal exposure to ART is associated with reduced LV mass, dimension, and septal wall thickness and higher LV fractional shortening and contractility during the first 2 years of life. We speculate that *in utero* exposure to ART may impair myocardial growth while initially improving LV function, although LV function was less than normal. These effects are more pronounced in girls. In the U.S., with more than 100,000 HIV-negative infants exposed to ART, these findings clearly indicate a need for long-term monitoring of these infants to better define the mechanism of these effects and to evaluate their long-term clinical importance.

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REFERENCES

1. Luginbuhl LM, Orav EJ, McIntosh K, Lipshultz SE. Cardiac morbidity and related mortality in children with HIV infection. JAMA. 1993; 269:2869–2875. [PubMed: 8388521]
2. Lipshultz SE, Easley KA, Orav EJ, et al. Left ventricular structure and function in children infected with Human Immunodeficiency Virus: The prospective P^2C^2 HIV multicenter study. Circulation. 1998; 97:1246–1256. [PubMed: 9570194]
3. Al-Attar I, Orav EJ, Exil V, Vlach SA, Lipshultz SE. Predictors of cardiac morbidity and related mortality in children with acquired immunodeficiency syndrome. J Am Coll Cardiol. 2003; 41:1598–1605. [PubMed: 12742303]
4. Lipshultz SE, Easley KA, Orav EJ, et al. Cardiac dysfunction and mortality in HIV-infected children: The Prospective P^2C^2 HIV Multicenter Study. Circulation. 2000; 102:1542–1548. [PubMed: 11182983]
5. Fisher SD, Easley KA, Orav EJ, et al. Mild dilated cardiomyopathy and increased left ventricular mass predict mortality: The prospective P^2C^2 HIV Multicenter study. Am Heart J. 2005; 150:439–447. [PubMed: 16169321]
6. Lipshultz SE, Easley KA, Orav EJ, et al. Cardiovascular status of infants and children of women infected with HIV-1 (P^2C^2 HIV): A cohort study. Lancet. 2002; 360:368–373. [PubMed: 12241776]
7. Blanche S, Tardieu M, Rustin P, et al. Persistent mitochondrial dysfunction and perinatal exposure to antiretroviral nucleoside analogues. Lancet. 1999; 354:1084–1089. [PubMed: 10509500]

8. Foster C, Lyall H. HIV and mitochondrial toxicity in children. J Antimicrob Chemother. 2008; 61:8–12. [PubMed: 17999978]
9. Thorne C, Newell ML. Safety of agents used to prevent mother-to-child transmission of HIV: Is there any cause for concern? Drug Saf. 2007; 30:203–213. [PubMed: 17343429]
10. Suksomboon N, Poolsup N, Ket-Aim S. Systematic review of the efficacy of antiretroviral therapies for reducing the risk of mother-to-child transmission of HIV infection. J Clin Pharm Ther. 2007; 32:293–311. [PubMed: 17489882]
11. Lipshultz SE, Orav EJ, Sanders SP, Hale AR, McIntosh K, Colan SD. Cardiac structure and function in children with human immunodeficiency virus infection treated with zidovudine. N Engl J Med. 1992; 327:1260–1265. [PubMed: 1406818]
12. Lipshultz SE, Easley KA, Orav EJ, et al. Absence of cardiac toxicity of zidovudine in infants. N Engl J Med. 2000; 343:759–766. [PubMed: 10984563]
13. Cooper ER, Charurat M, Mofenson L, et al. Combination antiretroviral strategies for the treatment of pregnant HIV-1-infected women and prevention of perinatal HIV-1 transmission. J Acquir Immune Defic Syndr. 2002; 29:484–494. [PubMed: 11981365]
14. Lavigne JE, Shearer WT, Thompson B, et al. for the CHAART Study Group. Cardiovascular outcomes of pediatric seroreverters perinatally exposed to HAART: Design of a longitudinal clinical study. Cardiovasc Toxicol. 2004; 4:187–198. [PubMed: 15371634]
15. Shearer WT, Quinn TC, LaRussa P, et al. for the Women and Infants Transmission Study Group. Viral load and disease progression in infants infected with Human Immunodeficiency Virus Type 1. N Engl J Med. 1997; 335:1337–1342. [PubMed: 9134873]
16. Kattan M, Kaplan S, Lipshultz S, et al. The pediatric pulmonary and cardiovascular complications of vertically transmitted human immunodeficiency virus (P^2C^2 HIV) infection study: Design and methods. The P^2C^2 HIV Study Group. J Clin Epidemiol. 1996; 49:1285–1294. [PubMed: 8892497]
17. Lipshultz SE, Easley KA, Orav EJ, et al. Reliability of multicenter pediatric echocardiographic measurements of left ventricular structure and function: The prospective P^2C^2 HIV study. Circulation. 2001; 104:310–316. [PubMed: 11457750]
18. Liang KY, Zeger SL. Longitudinal data analysis using generalized linear models. Biometrika. 1986; 73:13–22.
19. Pitt J, Schluchter M, Jenson H, et al. Maternal and perinatal factors related to maternal-infant transmission of HIV-1 in the P^2C^2 HIV study: The role of EBV shedding. J Acquir Immune Defic Syndr Hum Retrovirol. 1998; 19:462–470. [PubMed: 9859959]
20. Lipshultz SE, Colan SD, Gelber RD, Perez-Atayde AR, Sallan SE, Sanders SP. Late cardiac effects of doxorubicin therapy for acute lymphoblastic leukemia in childhood. N Engl J Med. 1991; 324:808–815. [PubMed: 1997853]
21. Lipshultz SE, Lipsitz SR, Mone SM, et al. Female sex and drug dose as risk factors for late cardiotoxic effects of doxorubicin therapy for childhood cancer. N Engl J Med. 1995; 332:1738–1743. [PubMed: 7760889]
22. Lipshultz SE, Lipsitz SR, Sallan SE, et al. Chronic progressive cardiac dysfunction years after doxorubicin therapy for childhood acute lymphoblastic leukemia. J Clin Oncol. 2005; 23:2629–2636. [PubMed: 15837978]
23. Torres SM, March TH, Carter MM, et al. In utero exposure of female CD-1 mice to AZT and/or 3TC: I. Persistence of microscopic lesions in cardiac tissue. Cardiovasc Toxicol. 2010; 10:37–50. [PubMed: 20101476]
24. Torres SM, Divi RL, Walker DM, et al. In utero exposure of female CD-1 mice to AZT and/or 3TC: II. Persistence of functional alterations in cardiac tissue. Cardiovasc Toxicol. Feb 13.2010 Epub ahead of print.
25. Torres SM, Walker DM, McCash CL, et al. Mutational analysis of the mitochondrial tRNA genes and flanking regions in umbilical cord tissue from uninfected infants receiving AZT-based therapies for prophylaxis of HIV-1. Environ Mol Mutagen. 2009; 50:10–26. [PubMed: 19031409]
26. Lipshultz SE, Walker VE, Torres SM, et al. Frequent mitochondrial DNA mutations and polymorphisms in long-term survivors of childhood acute lymphoblastic leukemia. Blood. 2007; 118:824A. abstract.

27. Lipshultz SE, Simbre VC, Hart S, et al. Frequency of elevations in markers of cardiomyocyte damage in otherwise healthy newborns. Am J Cardiol. 2008; 102:761–766. [PubMed: 18774003]
28. Gotsch F, Romero R, Kusanovic JP, et al. The fetal inflammatory response syndrome. Clin Obstet Gynecol. 2007; 50:652–683. [PubMed: 17762416]
29. Dhasmana DJ, Dheda K, Ravn P, Wilkinson RJ, Meintjes G. Immune reconstitution inflammatory syndrome in HIV-infected patients receiving antiretroviral therapy: pathogenesis, clinical manifestations and management. Drugs. 2008; 68:191–208. [PubMed: 18197725]
30. Shearer WT, Lipshultz SE, Easley K, et al. Alterations in cardiac and pulmonary function in pediatric rapid human immunodeficiency virus type 1 disease progressors. Pediatrics. 2000; 105:1–8. URL: http://www.pediatrics.org/cgi/contents/full/105/1/e9. [PubMed: 10617696]
31. Chan SS, Santos JH, Meyer JN, et al. Mitochondrial toxicity in hearts of CD-1 mice following perinatal exposure to AZT, 3TC, or AZT/3TC in combination. Environ Mol Mutagen. 2007; 48:190–200. [PubMed: 16395692]
32. Mertens AC, Yasui Y, Neglia JP, et al. Late mortality experience in five-year survivors of childhood and adolescent cancer: the Childhood Cancer Survivor Study. J Clin Oncol. 2001; 19:3163–72. [PubMed: 11432882]
33. Oeffinger KC, Mertens AC, Sklar CA, et al. Chronic health conditions in adult survivors of childhood cancer. N Engl J Med. 2006; 355:1572–1582. [PubMed: 17035650]
34. Chow HH, Li P, Brookshier G, Tang Y. In vivo tissue disposition of 3'-azido-3'-deoxythymidine and its anabolites in control and retrovirus-infected mice. Drug Metab Dispos. 1997; 25:412–422. [PubMed: 9107539]
35. Brogley SB, Ylitalo N, Mofenson LM, et al. In utero nucleoside reverse transcriptase inhibitor exposure and signs of possible mitochondrial dysfunction in HIV-uninfected children. AIDS. 2007; 21:929–938. [PubMed: 17457086]
36. European Collaborative Study. Exposure to antiretroviral therapy in utero or early life: the health of uninfected children born to HIV-infected women. J Acquir Immune Defic Syndr. 2003; 32:380–387. [PubMed: 12640195]
37. Barret B, Tardieu M, Rustin P, et al. Persistent mitochondrial dysfunction in HIV-1-exposed but uninfected infants: clinical screening in a large prospective cohort. AIDS. 2003; 17:1769–1785. [PubMed: 12891063]
38. Godfried MH, Boer K, Beuger S, Scherpbier HJ, Juijpers TW. A neonate with macrosomia, cardiomyopathy, and hepatomegaly born to an HIV-infected mother. Eur J Pediatr. 2005; 164:190–192. [PubMed: 15645286]
39. Walker DM, Poirier MC, Campen MJ, et al. Persistence of mitochondrial toxicity in hearts of female B6C3F1 mice exposed in utero to 3'-azido-3'-deoxythymidine. Cardiovasc Toxicol. 2004; 4:133–153. [PubMed: 15371630]
40. Culnane M, Fowler M, Lee SS, et al. Lack of long-term effects of in utero exposure to zidovudine among uninfected children born to HIV-infected women. Pediatric AIDS Clinical Trials Group Protocol 219/076 Teams. JAMA. 1999; 281:151–157. [PubMed: 9917118]
41. The Perinatal Safety Review Working Group. Nucleoside exposure in the children of HIV-infected women receiving antiretroviral drugs: absence of clear evidence for mitochondrial disease in children who died before 5 years of age in five United States cohorts. J Acquir Immune Defic Syndr. 2000; 25:261–268. [PubMed: 11115957]
42. Mofenson L, Munderi P. Safety of antiretroviral prophylaxis of perinatal transmission for HIV-infected pregnant women and their infants. J Acquir Immune Defic Syndr. 2002; 30:200–215. [PubMed: 12045684]
43. Pacheco SE, McIntosh K, Lu M, et al. for the Women and Infants Transmission Study. Effect of perinatal antiretroviral drug exposure on hematologic values in HIV-uninfected children: an analysis of the women and infants transmission study. J Infect Dis. 2006; 194:1089–1097. [PubMed: 16991083]
44. Witt KL, Cunningham CK, Patterson KB, et al. Elevated frequencies of micronucleated erythrocytes in infants exposed to zidovudine in utero and postpartum to prevent mother-to-child transmission of HIV. Environ Mol Mutagen. 2007; 48:322–329. [PubMed: 17358032]

45. Feiterna-Sperling C, Weizsaecker K, Buhrer C, et al. Hematologic effects of maternal antiretroviral therapy and transmission prophylaxis in HIV-1-exposed uninfected newborn infants. J Acquir Immune Defic Syndr. 2007; 45:42–51.

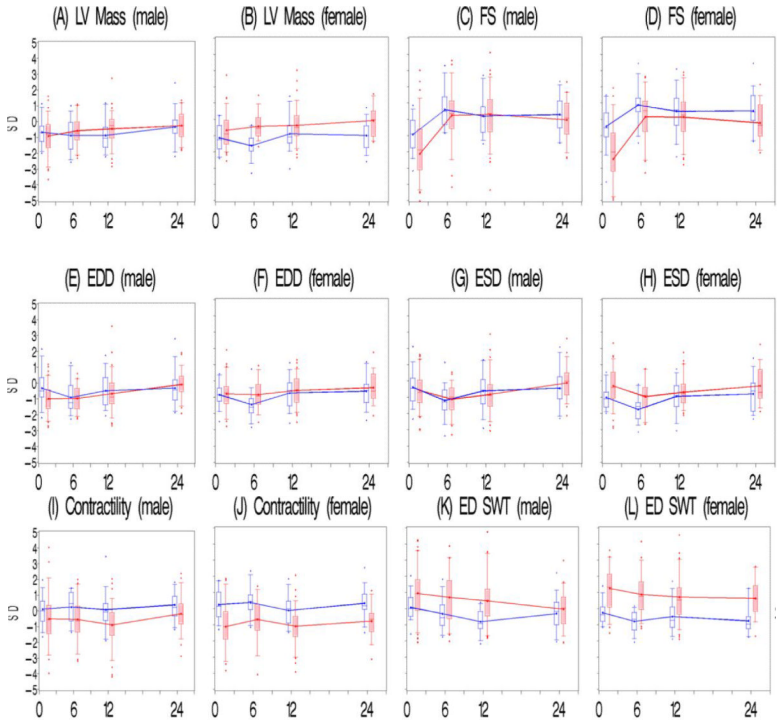

Figure 1. Cardiac Measurements of 136 CHAART-1 Infants and 216 P^2C^2-HIV Infants

CHAART-1 ART-positive infants' data are shown by the blue line with hollow boxes. P^2C^2-HIV ART-negative infants' data are shown by the red line with solid boxes. Rectangles show interquartile ranges, and vertical lines show the 5^{th} percentile to the 95^{th} percentile. Dots represent outliers. (LV=left ventricular; FS=fractional shortening; EDD=end-diastolic dimension; ESD=end-systolic dimension; ED SWT=end-diastolic septal wall thickness)

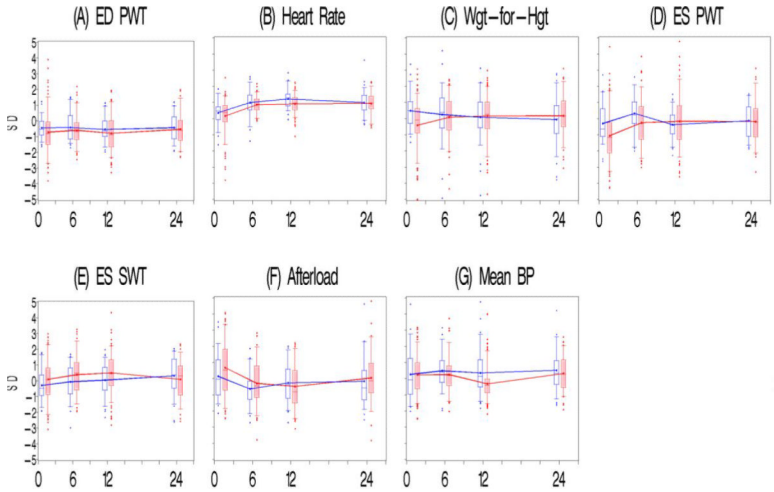

Figure 2. Cardiac and Anthropomorphic Measurements of 136 CHAART-1 Infants and 216 P^2C^2-HIV Infants

CHAART-1 ART-positive infant data are shown by the blue line with hollow boxes. P^2C^2-HIV ART-negative infant data are shown by the red line with solid boxes. Rectangles show interquartile ranges, and vertical lines show the 5th percentile to the 95th percentile. Dots represent outliers. (LV=left ventricular; ED PWT=end-diastolic posterior wall thickness; ES PWT=end-systolic posterior wall thickness; ES SWT=end-systolic septal wall thickness; BP=blood pressure)

Table 1

Demographic Characteristics, Maternal CD4 and CD8 Measurements, and Infant Blood Pressure Measurements of 352 HTV-Negative Infants and Their HlV-Positive Mothers According to Perinatal Exposure to Antiretroviral Therapy.

Variable Characteristics of mothers during pregnancy — no. (%)	CHAART-1 Infants Exposed to Antiretroviral Therapy (N=136)	P^2C^2-HTV Infants Not Exposed to Antiretroviral Therapy (N=216)	P Value*
Ethnic ity			0.20
Black	83 (61)	120 (56)	
White	10 (7)	29 (13)	
Hispanic	43 (32)	67 (31)	
Age at delivery, year			0.08
< 30	84 (62)	153 (71)	
≥ 30	52 (38)	63 (29)	
Tobacco use			0.003
Yes	30 (22)	79 (37)	
No	106 (78)	133 (62)	
Unknown	0 (0)	4 (2)	
Alcohol use			0.72
Yes	32 (24)	46 (21)	
No	104 (76)	164 (76)	
Unknown	0 (0)	6 (3)	
Illicit drug use			0.002
Yes	21 (15)	63 (29)	
No	115 (85)	149 (69)	
Unknown	0 (0)	4 (2)	
Vaginal Bleeding			0.001
Yes	34 (25)	6 (3)	
No	102 (75)	209 (97)	
Unknown	0 (0)	1 (<1)	
Gestational diabetes			0.56
Yes	3 (2)	7 (3)	
No	133 (98)	207 (96)	
Unknown	0 (0)	2 (1)	
Preeclampsia			0.26
Yes	1 (1)	5 (2)	
No	135 (99)	210 (97)	
Unknown	0 (0)	1 (<1)	
Cesarean section			<0.001
Yes	55 (40)	38 (18)	
No	81 (60)	178 (82)	

Variable Characteristics of infants — no. (%)	CHAART-1 Infants Exposed to Antiretroviral Therapy (N=136)	P2C2-HTV Infants Not Exposed to Antiretroviral Therapy (N=216)	P Value*
Sex			0.90
Male	74 (54)	116 (54)	
Female	62 (46)	100 (46)	
Premature birth (<37 wk)			0.24
Yes	15 (11)	33 (15)	
No	121 (89)	181 (85)	
Birth weight			0.09
< 2500 g	12 (9)	32 (15)	
≥ 2500 g	124 (91)	182 (84)	
Unknown	0 (0)	2 (1)	
Mean (95% CI) maternal CD4 and CD8 measurements during pregnancy			
CD4 count (cells/mm^3)	532.2 (491.7 − 572.7)	586.2 (517.1 − 655.2)	0.18
CD4 (%)	30.7 (28.9 − 32.5)	30.3 (28.7 − 31.8)	0.73
CD8 count (cells/mm^3)	796.1 (739.3 − 852.9)	904.4 (820.4 − 988.4)	0.04
CD8 (%)	45.7 (43.8 − 47.6)	48.0 (46.1 − 50.0)	0.09
Mean (95% CI) infant blood pressure Z-scores at the time of echocardiography†			
Diastolic			
Overall	0.43 (0.26 − 0.59)	0.23 (0.14 − 0.33)	0.04
Boys	0.50 (0.26 − 0.73)	0.22 (0.10 − 0.34)	0.04
Girls	0.35 (0.12 − 0.57)	0.25(0.10 − 0.39)	0.45
Systolic			
Overall	0.31 (0.12 − 0.50)	0.005 (−0.1 − 0.1)	0.006
Boys	0.50 (0.20 − 0.78)	−0.004 (−0.15 − 0.14)	0.003
Girls	0.10 (−0.14 − 0.34)	0.02 (−0.14 − 0.17)	0.56
Echocardiography age groups (%)†			
Birth visit (0 – 2 months)	54 (23)	173 (34)	-
6-month visit (5 – 8 months)	50 (21)	96(19)	-
1-year visit (11 – 22 months)	64 (27)	155 (30)	-
2-year visit (23 – 34 months)	66 (28)	92 (18)	-

*Chi-square analysis: Observations with unknown data were excluded from P-value calculation.

†The values are based on 234 measurements of infants exposed to antiretroviral therapy and 516 measurements of unexposed infants.

Table 2

Sex-Specific Cardiac Outcomes among 352 HIV-Negative Infants Born to HIV-Positive Mothers, by Exposure to Antiretroviral Therapy[1]

Cardiac Measure	Sex	Study	Population means				Adjusted estimates‡			
			Birth	6 months	1 year	2 years	Birth	6 months	1 year	2 years
Left ventricular mass Z-scores[2]										
	Male	CHAART	−0.67	−0.88	−0.87	−0.34	−0.72†	−1.01†	−0.64†	−0.43†
		P2C2	−0.92	−0.58	−0.46	−0.26	−0.85†	−0.58†	−0.49†	−0.23*
		Δ	0.25	−0.30	−0.41	−0.08	0.13	−0.44	−0.15	−0.21
							P=0.42	P=0.01	P=0.37	P=0.21
	Female	CHAART	−1.14	−1.63	−0.89	−0.99	−1.17†	−1.46†	−1.09†	−0.88†
		P2C2	−0.67	−0.43	−0.37	−0.08	−0.71†	−0.44†	−0.35†	−0.09
		Δ	−0.47	−1.20	−0.52	−0.91	−0.46	−1.02	−0.74	−0.79
							P=0.005	P<0.001	P<0.001	P<0.001
Left ventricular fractional shortening Z-scores[2]										
	Male	CHAART	−0.93	0.59	0.20	0.30	−0.89†	0.58†	0.21	0.28
		P2C2	−2.12	0.24	0.30	−0.04	−2.12†	0.36*	0.29*	−0.05
		†	1.19	0.35	−0.10	0.33	1.22	0.22	−0.08	0.33
							P<0.001	P<0.36	P<0.69	P<0.12
	Female	CHAART	−0.43	0.89	0.50	0.52	−0.59†	0.89†	0.52†	0.59†
		P2C2	−2.44	0.15	0.14	−0.22	−2.35†	0.13	0.06	−0.28
		Δ	2.01	0.74	0.36	0.74	1.76	0.76	0.46	0.87
							P<0.001	P=0.003	P=0.04	P<0.001
Left ventricular end-diastolic dimension Z-scores[2]										
	Male	CHAART	−0.32	−0.90	−0.48	−0.31	−0.49†	−1.04†	−0.45†	−0.35†
		P2C2	−0.99	−0.96	−0.66	−0.08	−0.91†	−0.96†	−0.66†	−0.20*
		Δ	0.67	−0.06	0.19	−0.23	0.42	−0.08	0.20	−0.15
							P=0.009	P=0.61	P=0.22	P=0.36
	Female	CHAART	−0.85	−1.46	−0.74	−0.63	−0.79†	−1.34†	−0.75†	−0.64†
		P2C2	−0.79	−0.84	−0.58	−0.41	−0.86†	−0.90†	−0.60†	−0.14

Cardiac Measure	Sex	Study	Population means				Adjusted estimates‡			
			Birth	6 months	1 year	2 years	Birth	6 months	1 year	2 years
		Δ	−0.05	−0.62	−0.16	−0.22	0.07	−0.44	−0.15	−0.51
							P=0.66	P=0.006	P=0.34	P=0.003
Left ventricular end-systolic dimension Z-scores										
	Male	CHAART	−0.39	−1.20	−0.61	−0.44	−0.57†	−1.25†	−0.78†	−0.35†
		P^2C^2	−0.58	−1.12	−0.82	−0.12	−0.53†	−1.21†	−0.74†	−0.31†
		Δ	0.19	−0.08	0.22	−0.33	−0.04	−0.04	−0.04	−0.04
							P=0.78	P=0.78	P=0.78	P=0.78
	Female	CHAART	−1.01	−1.75	−0.95	−0.80	−0.91†	−1.59†	−1.12†	−0.69†
		P^2C^2	−0.30	−0.96	−0.69	−0.30	−0.39†	−1.07†	−0.60†	−0.17
		Δ	−0.72	−0.79	−0.26	−0.50	−0.52	−0.52	−0.52	−0.52
							P<0.001	P<0.001	P<0.001	P<0.001
Left ventricular contractility Z-scores										
	Male	CHAART	0.10	0.24	0.08	0.38	0.14	0.31*	−0.09	0.39†
		P^2C^2	−0.51	−0.55	−0.90	−0.18	−0.58†	−0.41†	−0.81†	−0.33†
		Δ	0.61	0.79	0.98	0.56	0.72	0.72	−0.72	0.72
							P<0.001	P<0.001	P<0.001	P<0.001
	Female	CHAART	0.26	0.38	−0.11	0.33	0.15	0.33†	−0.07	0.41†
		P^2C^2	−1.11	−0.67	−1.09	−0.77	−0.95†	−0.78†	−1.17†	−0.70
		Δ	1.37	1.05	0.98	1.10	1.11	1.11	1.11	1.11
							P<0.001	P<0.001	P<0.001	P<0.001
End-diastolic septal wall thickness Z-scores										
	Male	CHAART	0.08	−0.32	−0.81	−0.31	0.12	−0.28*	−0.41†	−0.55†
		P^2C^2	0.95	0.70	0.48	−0.03	0.94†	0.54†	0.41†	0.27
		Δ	−0.87	−1.01	−1.29	−0.27	−0.82	−0.82	−0.82	−0.82
							P<0.001	P<0.001	P<0.001	P<0.001
	Female	CHAART	−0.25	−0.79	−0.49	−0.76	−0.15	−0.55†	−0.68†	−0.82†
		P^2C^2	1.26	0.88	0.72	0.64	1.27†	0.87†	0.74†	0.60†
		Δ	−1.51	−1.67	−1.21	−1.40	−1.42	−1.42	−1.42	−1.42

Cardiac Measure	Sex	Study	Population means				Adjusted estimates‡			
			Birth	6 months	1 year	2 years	Birth	6 months	1 year	2 years
							P<0.001	P<0.001	P<0.001	P<0.001

[1] Crude measurements and estimates from regression models are adjusted for age, sex, ethnicity, and interaction between sex and exposure to antiretroviral therapy.

[2] Model is also adjusted for interactions between age and exposure to antiretroviral therapy.

* P<0.05.

† P<0.01.

‡ Adjusted estimates from GEE models only.

Table 3

Cardiac and Anthropomorphic Outcomes among 352 HTV-Negative Infants Born to HIV-Positive Mothers, by Exposure to Antiretroviral Therapy[12]

Cardiac or Anthropom orphic Measure		Population means				Adjusted estimates‡			
	Study	Birth	6 month	1 year	2 years	Birth	6 month	1 year	2 years
Left ventricular end-diastolic posterior wall thickness Z-scores									
	CHAART	−0.44	−0.42	−0.54	−0.43	−0.52†	−0.38†	−0.60†	−0.40†
	P2C2	−0.72	−0.60	−0.80	−0.54	−0.70†	−0.57†	−0.78†	−0.58†
	Δ	0.28	0.18	0.26	0.10	0.18	0.18	0.18	0.18
						P=0.02	P=0.02	P=0.02	P=0.02
Heart rate Z-scores									
	CHAART	0.36	0.99	1.22	1.00	0.36†	1.02†	1.13†	1.06†
	P2C2	0.17	0.86	0.92	0.94	0.17†	0.83†	0.95†	0.88†
	Δ	0.19	0.13	0.30	0.06	0.18	0.18	0.18	0.18
						P=0.002	P=0.002	P=0.002	P=0.002
Weight-for-height Z-scores									
	CHAART	0.49	0.26	0.09	−0.05	0.39†	0.28	0.14	0.01
	P2C2	−0.41	0.09	0.16	0.17	−0.40†	0.08	0.17	0.14
	Δ	0.90	0.17	−0.07	−0.23	0.79	0.20	−0.03	−0.13
						P<0.001	P=0.39	P=0.88	P=0.52
Left ventricular end-systolic posterior wall thickness Z-scores[3]									
	CHAART	−0.30	0.32	−0.37	−0.13	−0.33*	0.30*	−0.34†	−0.13
	P2C2	−1.06	−0.25	−0.16	−0.18	−1.03†	−0.19	−0.15	−0.22
	Δ	0.76	0.58	−0.21	−0.05	0.70	0.49	−0.19	0.09
						P<0.001	P=0.01	P=0.25	P=0.64
End-systolic septal wall thickness Z-scores[3]									
	CHAART	−0.29	−0.06	0.05	0.33	−0.33*	−0.04	0.08	0.35*
	P2C2	0.10	0.39	0.50	0.10	0.12	0.40†	0.48†	0.09
	Δ	−0.39	−0.45	−0.45	0.23	−0.46	−0.44	−0.40	0.26
						P=0.01	P=0.03	P=0.009	P=0.19

Cardiac or Anthropom orphic Measure		Population means				Adjusted estimates‡			
	Study	Birth	6 month	1 year	2 years	Birth	6 month	1 year	2 years
Left ventricular afterload (end-systolic wall stress) Z-scores[3]									
	CHAART	0.15	−0.63	−0.26	−0.16	0.20	−0.64†	−0.24	−0.20
	P^2C^2	0.68	−0.29	−0.48	0.05	0.68†	−0.34*	−0.47†	0.09
	Δ	−0.53	−0.34	0.23	−0.20	−0.47	−0.30	0.23	−0.29
						P=0.04	P=0.15	P=0.23	P=0.26

[1] Estimates from regression models are adjusted for age, sex, and race or ethnicity.

[2] Sex-specific effects of exposure to antiretroviral therapy are not shown because they were non-significant; therefore, data from boys and girls are pooled.

[3] Model is also adjusted for interactions between age and exposure to antiretroviral therapy.

* P<0.05.

† P<0.01.

‡ Adjusted estimates from GEE models only.