IMPORTANCE Most studies examining the association of prenatal antiretroviral (ARV) exposures with congenital anomalies (CAs) in children born to human immunodeficiency virus (HIV)–infected women have been reassuring, but some evidence suggests an increased risk with specific ARV agents.

OBJECTIVE To evaluate the association of in utero ARV exposures with CAs in HIV-exposed uninfected children.

DESIGN, SETTING, AND PARTICIPANTS Prospective cohort study design. The Pediatric HIV/AIDS Cohort Study’s Surveillance Monitoring of ART Toxicities (SMARTT) Study was performed at 22 US medical centers among 2580 HIV-exposed uninfected children enrolled in the SMARTT Study between March 23, 2007, and June 18, 2012.

EXPOSURES First-trimester exposure to any ARV and to specific ARV medications.

MAIN OUTCOMES AND MEASURES The primary endpoint was a CA based on physician review of infant physical examinations according to the Antiretroviral Pregnancy Registry modification of the Metropolitan Atlanta Congenital Defects Program. Rates of CAs were estimated overall and by birth year. Logistic regression models were used to evaluate the association of CAs with first-trimester ARV exposures, adjusting for demographic and maternal characteristics.

RESULTS Congenital anomalies occurred in 175 of 2580 children, yielding a prevalence of 6.78% (95% CI, 5.85%-7.82%); 242 major CAs were confirmed, including 72 musculoskeletal and 55 cardiovascular CAs. The prevalence of CAs increased significantly among successive birth cohorts (3.8% for children born before 2002 and up to 8.3% for those born 2008-2010). In adjusted models, no association of first-trimester exposures with CAs was found for any ARV, for combination ARV regimens, or for any drug class. No individual ARV in the reverse transcriptase inhibitor drug classes was associated with an increased risk of CAs. Among protease inhibitors, higher odds of CAs were observed for atazanavir sulfate (adjusted odds ratio [aOR], 1.95; 95% CI, 1.24-3.05) and for ritonavir used as a booster (aOR, 1.56; 95% CI, 1.11-2.20). With first-trimester atazanavir exposure, risks were highest for skin (aOR, 5.23) and musculoskeletal (aOR, 2.55) CAs.

CONCLUSIONS AND RELEVANCE Few individual ARVs and no drug classes were associated with an increased risk of CAs in HIV-exposed infants after adjustment for calendar year and maternal characteristics. While the overall risk remained low, a relative increase was observed in successive years and with atazanavir exposure. Given the low absolute CA risk, the benefits of recommended ARV therapy use during pregnancy still outweigh such risks, although further studies are warranted.
The use of combination antiretroviral (ARV) regimens for prevention of mother-to-child transmission of human immunodeficiency virus (HIV) and for treatment of HIV-infected pregnant women has contributed to a substantial reduction in HIV-infected infants. However, the safety of in utero exposure to such combination ARV regimens remains a concern, particularly as newer agents are approved and an increasing percentage of women enter pregnancy already receiving ARV therapy.

Most prior studies examining the risk of congenital anomalies (CAs) according to in utero ARV exposure have been reassuring, but some evidence has suggested an increased risk of CAs overall or for certain CAs with specific ARVs. In the international Antiretroviral Pregnancy Registry, the estimated prevalence of CAs was 2.9% among more than 6900 children with first-trimester ARV exposures, similar to the rate among children exposed in later trimesters. The Women and Infants Transmission Study found no increase in the overall rate of defects (3.56 per 100 live births) compared with the general population estimate of 2.76 from the Metropolitan Atlanta Congenital Defects Program (MACDP) but reported an increased risk of hypospadias after exposure to zidovudine (zidovudine or azidothymidine) during the first trimester. Two studies from US-based cohorts have shown an increased overall risk of CAs among infants with first-trimester efavirenz exposure. A single animal study and case reports have also reported CAs associated with efavirenz exposure, leading to recommendations against its use in pregnancy, although specific risks have not been confirmed.

Previous studies predominantly included children born before 2007, preventing evaluation of newer ARVs and combinations with increasing use. In the United States, prenatal use of tenofovir disoproxil fumarate, emtricitabine, and lopinavir has increased dramatically since approval in 2000 to 2003 to 40% to 50% use by 2010, while nelfinavir mesylate use has declined substantially following safety warnings. Atazanavir use increased by 2010 to approximately 20%. Through 2011, an Italian cohort showed similar trends. In addition to changes in specific ARVs, most infants in previously studied cohorts were not exposed to ARVs in the first trimester, a critical window for teratogenicity. We used an ongoing US-based pregnancy cohort, the Surveillance Monitoring of ART Toxicities (SMARTT) Study of the Pediatric HIV/AIDS Cohort Study network, to examine the association of in utero ARV exposures and infant CAs during the past 15 years. Our objectives were (1) to evaluate changes in the rate of CAs over time as new ARVs and regimens were used and (2) to evaluate the association of in utero ARV exposure with CAs.

Methods

The protocol was approved by appropriate institutional review boards, with written informed consent by mothers or guardians for study participation for themselves and their children. We analyzed data from HIV-infected pregnant women and their children enrolled in the SMARTT Study. This study includes 2 cohorts, static and dynamic. Between 2007 and 2009, the static cohort enrolled mothers or caregivers and their children younger than 12 years who had detailed information on ARV use during pregnancy and pregnancy outcomes. In 2007, the dynamic cohort began enrolling pregnant women and their infants between 22 weeks of gestation and 1 week after delivery into prospective surveillance.

Information on ARV use during pregnancy and medical conditions, including pregnancy outcomes, was collected by medical record abstraction. Congenital anomalies were identified at study-specified newborn and 1-year-old physical examinations for those in the dynamic cohort and from physical examinations performed in prior studies for those in the static cohort. Participants were considered evaluable for this analysis if they were enrolled and had a study visit by July 1, 2012.

Outcome Measure

The outcome of interest was the presence of a CA, defined as an abnormality in the structure of a body part that was documented within the first year of life. Congenital anomalies were recorded on study-specific anomaly and diagnosis forms. All study authors (blinded to ARV exposures) reviewed the reported CAs and classified them according to the Antiretroviral Pregnancy Registry modification of the MACDP classification scheme, a well-documented system for categorizing CAs. According to this system, an infant with at least 1 major anomaly or at least 2 conditional anomalies in the absence of a major anomaly is considered a CA case. Additional information was requested from sites if needed to classify potential anomalies. Each CA was reviewed by at least 2 physicians (M.J.C., R.H., R.B.V.D., K.R., J.S.R., M.R., H.A.M., and D.H.W.), and discrepancies were discussed to obtain consensus.

Prenatal ARV Exposures

The primary exposure of interest was reported maternal use of ARVs during the first trimester (<14 weeks of gestation). Highly active ARV therapy (HAART) regimens were defined as those containing 3 or more ARVs from 2 or more drug classes (nucleoside reverse transcriptase inhibitors, nonnucleoside reverse transcriptase inhibitors, protease inhibitors [PIs], and integrase inhibitors). Children were classified according to first-trimester exposure to any ARV, to individual ARVs, to ARV drug classes, and to HAART. We also evaluated these exposures at any time during pregnancy and by timing of the first ARV exposure.

Potential Confounders

Confounding was evaluated using prior knowledge (based on biological mechanisms and previous literature) and descriptive statistics from our cohort through the use of directed acyclic graphs. Potential confounders evaluated included self-reported race/ethnicity, prepregnancy body mass index, advanced maternal age at delivery (>35 years), health conditions such as pregestational diabetes mellitus, maternal sexually transmitted infections during pregnancy, self-reported substance use (tobacco and alcohol or other drug use), plasma HIV RNA concentration (viral load) and CD4 counts (earliest available measures in pregnancy), and first-trimester use of other medications previously reported to be associated with CA risk.
(eg, folate antagonists and antidepressants, including selective serotonin reuptake inhibitors). Socioeconomic status was also considered, as reflected by annual household income and caregiver education levels. Low birth weight (<2500 g), preterm birth (<37 weeks of gestation), and delivery by cesarean section were described but not included as potential confounders because these measures could be on the causal pathway between ARV exposure and CA status and cesarean section might be preferentially performed when a CA was suspected.

Statistical Analysis
Rates of CAs (95% CIs) were estimated overall and by calendar year and compared descriptively with the MACDP rates for the US population. The characteristics of children with and without CAs were compared using χ² test, Fisher exact test, and Wilcoxon rank sum tests as appropriate. Logistic regression analysis was used to evaluate the association between the in utero ARV exposures described above and confirmed CAs. Adjusted models included birth cohort and other noted confounders with P < .10 in multivariable models. Separate analyses were conducted for certain CA categories (eg, cardiovascular, musculoskeletal, skin, and male genital), although these had limited power.

To confirm the robustness of results, several sensitivity analyses were conducted. Analyses were repeated with restriction to cases having at least 1 major CA (eg, excluding children with only conditional CAs) and exclusion of those with a chromosomal anomaly. We repeated all analyses with restriction to the dynamic cohort because its prospective follow-up from birth reduces the risk of recall bias and misclassification and this restriction eliminates overlap with previous cohorts (Women and Infants Transmission Study, Pediatric AIDS Clinical Trials P219c protocol, and International Maternal Pediatric Adolescent AIDS Clinical Trials P1025 protocol). Last, sensitivity analyses that included random effects were conducted to control for multiple children born to the same mother and to adjust for the clustering of children within research sites. Because of observed time trends, analyses were repeated with stratification by, rather than adjustment for, birth cohort but yielded similar results and are not presented. Analyses were conducted using statistical software (SAS, version 9.2; SAS Institute), and 2-sided P ≤ .05 was considered statistically significant. Because the SMARTT study is a safety investigation, no correction for multiple comparisons was used to minimize the probability of not detecting true associations (type II error); however, the large number of tests increases the risk of spurious associations, and findings warrant confirmation in future studies.

Results

Study Population and CA Status by Demographic and Maternal Characteristics
The demographic and maternal characteristics of 2580 participants at 22 US medical centers (1380 in the dynamic cohort born between April 2, 2007, and June 29, 2012, and 1200 in the static cohort born between March 27, 1995, and December 27, 2008) enrolled by July 1, 2012, are summarized in Table 1. After team review, 175 infants met the modified MACDP criteria for a confirmed CA case, yielding a prevalence of 6.78% (95% CI, 5.85%-7.82%). One hundred sixty-two unique children had at least 1 major CA (prevalence, 6.27%; 95% CI, 5.37%-7.29%), and 13 children had 2 or more conditional CAs but no major anomalies. These 162 children had a total of 242 confirmed major CAs; musculoskeletal (n = 72) and cardiovascular (n = 55) anomalies were most common (eTable 1 in the Supplement). The prevalences of CAs were 3.8%, 5.2%, 8.0%, 8.3%, and 5.7% for children born before 2002, 2002 to 2004, 2005 to 2007, 2008 to 2010, and after 2010, respectively, with a significantly increasing trend (P = .03) in successive years. However, no significant overall difference was observed in prevalences between the static cohort vs the dynamic cohort (6.4% vs 7.1%, P = .53).

No significant difference was found in the distribution of CA cases by demographic or socioeconomic characteristics other than birth cohort (Table 1). Cases were more often delivered by cesarean section and more often preterm births than noncases, but no association was observed with higher maternal viral load (HIV RNA concentration >1000 copies/mL) or with tobacco and alcohol or other drug use. The use of selective serotonin reuptake inhibitors was rare during the first trimester, occurring in 30 of 2580 (1.2%), and only one of these infants had a CA. The use of folate antagonists (cotrimoxazole or pyrimethamine) was reported by 107 mothers, 6 (5.6%) of whom had CAs.

Multivariable logistic models for CA case status adjusted for low maternal CD4 count (<250 cells/mm³) early in pregnancy and birth cohort. For musculoskeletal anomalies, adjusted models also included maternal alcohol consumption during the first trimester (adjusted odds ratio [AOR], 2.09; 95% CI, 0.92-4.72). Of 2580 children, 63 (6 cases and 57 noncases) lacked detailed information regarding maternal ARV use needed to identify trimesters of exposure, yielding 2517 children for evaluation of ARV exposures.

Association of In Utero ARV Exposures With CAs
A significantly higher prevalence of CAs was observed for children exposed to HAART (8.1% for exposed vs 5.8% for unexposed) or to PIs (8.5% for exposed vs 5.8% for unexposed) in the first trimester, but these associations did not persist in adjusted models (Table 2). No individual nucleoside reverse transcriptase inhibitors were associated with an increased risk of CAs, but the combination of didanosine plus stavudine, while rare (<1% exposed), was associated with an 8-fold higher odds of CAs. For nonnucleoside reverse transcriptase inhibitors, neither efavirenz nor nevirapine was associated with CAs.

For PIs, a significantly higher prevalence of cases was observed among children exposed to atazanavir (11.7% vs 6.2%), lopinavir (9.4% vs 6.3%), and ritonavir when used as a booster (>99% of use) (9.3% vs 5.8%). Associations persisted in adjusted models for atazanavir and ritonavir (Table 2). Atazanavir was usually used in combination with ritonavir (88.5%) and often with certain nucleoside reverse transcriptase inhibitors. The combinations of atazanavir with ritonavir, tenofovir, or emtricitabine were each associated with an increased
Table 1. Demographic and Maternal Characteristics of 2580 SMARTT Study Infants Overall and by Congenital Anomaly Case Status

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Total (N = 2580)</th>
<th>No. (%)</th>
<th>Noncases (n = 2405)</th>
<th>Cases (n = 175)</th>
<th>P Value&lt;sup&gt;b&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cohort</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>.53</td>
</tr>
<tr>
<td>Dynamic</td>
<td>1380 (53.5)</td>
<td>1282 (53.3)</td>
<td>98 (56.0)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Static</td>
<td>1200 (46.5)</td>
<td>1123 (46.7)</td>
<td>77 (44.0)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Birth cohort</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>.02</td>
</tr>
<tr>
<td>&lt;2002</td>
<td>391 (15.2)</td>
<td>376 (15.6)</td>
<td>15 (8.6)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2002-2004</td>
<td>343 (13.3)</td>
<td>325 (13.5)</td>
<td>18 (10.3)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2005-2007</td>
<td>525 (20.3)</td>
<td>483 (20.1)</td>
<td>42 (24.0)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2008-2010</td>
<td>940 (36.4)</td>
<td>862 (35.8)</td>
<td>78 (44.6)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2011-2012</td>
<td>381 (14.8)</td>
<td>359 (14.9)</td>
<td>22 (12.6)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female sex</td>
<td>1254 (48.6)</td>
<td>1177 (48.9)</td>
<td>77 (44.0)</td>
<td>.21</td>
<td></td>
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<tr>
<td>Race</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>.87</td>
</tr>
<tr>
<td>White</td>
<td>701 (27.2)</td>
<td>651 (27.1)</td>
<td>50 (28.6)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Black/African American</td>
<td>1703 (66.0)</td>
<td>1592 (66.2)</td>
<td>111 (63.4)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>14 (0.5)</td>
<td>13 (0.5)</td>
<td>1 (0.6)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Latino/Hispanic</td>
<td>845 (32.8)</td>
<td>781 (32.5)</td>
<td>64 (36.6)</td>
<td>.28</td>
<td></td>
</tr>
<tr>
<td>Maternal age &gt;35 y at delivery</td>
<td>336 (13.0)</td>
<td>307 (12.8)</td>
<td>29 (16.6)</td>
<td>.20</td>
<td></td>
</tr>
<tr>
<td>Annual household income &lt;$20 000</td>
<td>1683 (65.2)</td>
<td>1570 (65.3)</td>
<td>113 (64.6)</td>
<td>.72</td>
<td></td>
</tr>
<tr>
<td>Caregiver not a high school graduate</td>
<td>894 (34.7)</td>
<td>831 (34.6)</td>
<td>63 (36.0)</td>
<td>.68</td>
<td></td>
</tr>
<tr>
<td>Delivery by cesarean section</td>
<td>1402 (54.3)</td>
<td>1293 (53.8)</td>
<td>109 (62.3)</td>
<td>.03</td>
<td></td>
</tr>
<tr>
<td>Low birth weight &lt;2500 g</td>
<td>483 (18.7)</td>
<td>440 (18.3)</td>
<td>43 (24.6)</td>
<td>.045</td>
<td></td>
</tr>
<tr>
<td>Preterm birth &lt;37 wk of gestation</td>
<td>527 (20.4)</td>
<td>477 (19.8)</td>
<td>50 (28.6)</td>
<td>.008</td>
<td></td>
</tr>
<tr>
<td>Pregnancy complication</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Toxemia or preeclampsia</td>
<td>145 (5.6)</td>
<td>129 (5.4)</td>
<td>16 (9.1)</td>
<td>.04</td>
<td></td>
</tr>
<tr>
<td>Gestational diabetes mellitus</td>
<td>116 (4.5)</td>
<td>108 (4.5)</td>
<td>8 (4.6)</td>
<td>.85</td>
<td></td>
</tr>
<tr>
<td>Pregestational diabetes mellitus</td>
<td>51 (2.0)</td>
<td>45 (1.9)</td>
<td>6 (3.4)</td>
<td>.15</td>
<td></td>
</tr>
<tr>
<td>Either of the above diabetes mellitus</td>
<td>161 (6.2)</td>
<td>148 (6.2)</td>
<td>13 (7.4)</td>
<td>.51</td>
<td></td>
</tr>
<tr>
<td>Maternal immunologic and virologic status</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HIV RNA concentration &lt;1000 copies/mL at delivery</td>
<td>390 (15.1)</td>
<td>368 (15.3)</td>
<td>22 (12.6)</td>
<td>.38</td>
<td></td>
</tr>
<tr>
<td>HIV RNA concentration &gt;1000 copies/mL early in pregnancy</td>
<td>1316 (51.0)</td>
<td>1227 (51.0)</td>
<td>89 (50.9)</td>
<td>.99</td>
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<tr>
<td>CD4 count &gt;250 cells/mm&lt;sup&gt;3&lt;/sup&gt; at delivery</td>
<td>368 (14.3)</td>
<td>350 (14.6)</td>
<td>18 (10.3)</td>
<td>.17</td>
<td></td>
</tr>
<tr>
<td>CD4 count &gt;250 cells/mm&lt;sup&gt;3&lt;/sup&gt; early in pregnancy</td>
<td>470 (18.2)</td>
<td>447 (18.6)</td>
<td>23 (13.1)</td>
<td>.10</td>
<td></td>
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<tr>
<td>Maternal substance use during pregnancy</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hard drug use&lt;sup&gt;c&lt;/sup&gt;</td>
<td>68 (2.6)</td>
<td>63 (2.6)</td>
<td>5 (2.9)</td>
<td>.81</td>
<td></td>
</tr>
<tr>
<td>Illicit drug use, including hard drugs&lt;sup&gt;c&lt;/sup&gt;</td>
<td>206 (8.0)</td>
<td>191 (7.9)</td>
<td>15 (8.6)</td>
<td>.77</td>
<td></td>
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<tr>
<td>Alcohol consumption</td>
<td>196 (7.6)</td>
<td>180 (7.5)</td>
<td>16 (9.1)</td>
<td>.46</td>
<td></td>
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<tr>
<td>Tobacco use</td>
<td>446 (17.3)</td>
<td>414 (17.2)</td>
<td>32 (18.3)</td>
<td>.76</td>
<td></td>
</tr>
<tr>
<td>Maternal medication use during pregnancy</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Methadone hydrochloride treatment</td>
<td>22 (0.9)</td>
<td>22 (0.9)</td>
<td>0</td>
<td>.40</td>
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<tr>
<td>Pain medication</td>
<td>99 (3.8)</td>
<td>91 (3.8)</td>
<td>8 (4.6)</td>
<td>.55</td>
<td></td>
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<tr>
<td>First-trimester SSRI</td>
<td>30 (1.2)</td>
<td>29 (1.2)</td>
<td>1 (0.6)</td>
<td>.72</td>
<td></td>
</tr>
<tr>
<td>First-trimester folate antagonist</td>
<td>107 (4.1)</td>
<td>101 (4.2)</td>
<td>6 (3.4)</td>
<td>.84</td>
<td></td>
</tr>
<tr>
<td>Maternal STI during pregnancy</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gonorrhea</td>
<td>72 (2.8)</td>
<td>64 (2.7)</td>
<td>8 (4.6)</td>
<td>.15</td>
<td></td>
</tr>
<tr>
<td>Chlamydia</td>
<td>215 (8.3)</td>
<td>199 (8.3)</td>
<td>16 (9.1)</td>
<td>.67</td>
<td></td>
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<tr>
<td>Trichomonas</td>
<td>282 (10.9)</td>
<td>268 (11.1)</td>
<td>14 (8.0)</td>
<td>.17</td>
<td></td>
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<tr>
<td>Syphilis</td>
<td>76 (2.9)</td>
<td>72 (3.0)</td>
<td>4 (2.3)</td>
<td>.82</td>
<td></td>
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<tr>
<td>Any of the above</td>
<td>511 (19.8)</td>
<td>478 (19.9)</td>
<td>33 (18.9)</td>
<td>.84</td>
<td></td>
</tr>
</tbody>
</table>

Abbreviations: HIV, human immunodeficiency virus; SMARTT, Surveillance Monitoring of ART Toxicities; SSRI, selective serotonin reuptake inhibitor; STI, sexually transmitted infection.

* Characteristics were unavailable for some participants, including race (n = 162), ethnicity (n = 3), maternal age (n = 53), annual household income (n = 185), caregiver education level (n = 25), delivery mode (n = 52), preterm birth (n = 45), low birth weight (n = 23), diabetes mellitus (n = 91), maternal viral load (n = 194), maternal CD4 count (n = 159), maternal substance use during pregnancy (n = 196), and maternal STI during pregnancy (146 gonorrhea, 147 Chlamydia, 149 syphilis, and 355 Trichomonas).

<sup>b</sup> P values were calculated by χ² test for birth cohort and by Fisher exact test for all other characteristics.

<sup>c</sup> Hard drugs include cocaine, heroin, and opium. Illicit drugs include these hard drugs, as well as marijuana, ecstasy, methamphetamine, and hallucinogens.
risk of CAs, with similar aORs (2.01, 2.00, and 1.85, respectively), while combinations of atazanavir with zidovudine (aOR, 0.89) or lamivudine (aOR, 1.48) showed no significant association. Of the 2 primary regimens that included ritonavir with another PI, atazanavir with ritonavir showed increased odds, while ritonavir-boosted lopinavir did not.

Specific anomalies for children exposed to first-trimester atazanavir are listed in eTable 2 in the Supplement.

Associations for ARV exposures at any time during pregnancy indicated a significantly higher risk of CAs for those exposed to the combinations of didanosine plus stavudine or to zidovudine plus lamivudine. When the rate of CAs by timing of the first exposure was examined (eTable 3 in the Supplement), the results were generally consistent with the comparisons of first-trimester exposure. However, for some ARVs the highest prevalence of CAs occurred with the first exposure during the second or third trimester (10.6% for abacavir and 17.1% for stavudine).

Separate analyses conducted by type of anomaly indicated that first-trimester atazanavir exposure was significantly associated with musculoskeletal and skin anomalies (Table 3). Significantly higher odds of musculoskeletal anomalies were observed among infants exposed to didanosine plus stavudine in the first trimester. Ritonavir as a booster was associated with an increased risk of musculoskeletal CAs. We observed a significantly higher odds of male genital anomalies (eg, hypospadias and cryptorchidism) with first-trimester zidovudine exposure and lamivudine exposure.

For some less commonly used ARVs, including raltegravir (1.5% exposed), enfuvirtide (0.3% exposed), maraviroc (0.1% exposed), and etravirine (0.4% exposed), no first-trimester exposures were found. Raltegravir was the only one of these ARVs associated with any CAs, with a rate of 4.2% (3 of 71 exposed at any time during pregnancy) compared with 6.8% for raltegravir unexposed.

**Table 2. Association of First-Trimester Antiretroviral Exposure With Congenital Anomalies**

<table>
<thead>
<tr>
<th>Variable</th>
<th>% Exposed</th>
<th>Defect Rate, No./Total No. (%)</th>
<th>Odds Ratio (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Antiretroviral drug class</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Any antiretroviral</td>
<td>48.4</td>
<td>93/1219 (7.6)</td>
<td>1.33 (0.97-1.82)</td>
</tr>
<tr>
<td>Highly active antiretroviral treatment</td>
<td>40.7</td>
<td>83/1025 (8.1)</td>
<td>1.44 (1.05-1.97)</td>
</tr>
<tr>
<td>NNRTI</td>
<td>8.5</td>
<td>13/214 (6.1)</td>
<td>0.89 (0.50-1.60)</td>
</tr>
<tr>
<td>NRTI</td>
<td>48.1</td>
<td>92/1211 (7.6)</td>
<td>1.31 (0.96-1.80)</td>
</tr>
<tr>
<td>Protease inhibitor</td>
<td>35.2</td>
<td>75/887 (8.5)</td>
<td>1.51 (1.10-2.07)</td>
</tr>
<tr>
<td>NNRTI</td>
<td>Abacavir</td>
<td>8.8</td>
<td>15/222 (6.8)</td>
</tr>
<tr>
<td>Didanosine</td>
<td>2.1</td>
<td>5/52 (9.6)</td>
<td>1.49 (0.59-3.80)</td>
</tr>
<tr>
<td>Emtricitabine</td>
<td>14.9</td>
<td>28/374 (7.5)</td>
<td>1.15 (0.75-1.75)</td>
</tr>
<tr>
<td>Lamivudine</td>
<td>31.7</td>
<td>63/197 (7.9)</td>
<td>1.31 (0.95-1.81)</td>
</tr>
<tr>
<td>Stavudine</td>
<td>2.7</td>
<td>4/68 (5.9)</td>
<td>0.87 (0.31-2.41)</td>
</tr>
<tr>
<td>Tenofovir disoproxil fumarate</td>
<td>17.1</td>
<td>32/431 (7.4)</td>
<td>1.14 (0.77-1.70)</td>
</tr>
<tr>
<td>Zidovudine</td>
<td>28.8</td>
<td>57/726 (7.9)</td>
<td>1.28 (0.92-1.78)</td>
</tr>
<tr>
<td>Didanosine plus stavudine</td>
<td>0.3</td>
<td>2/7 (28.6)</td>
<td>5.62 (1.08-29.16)</td>
</tr>
<tr>
<td>Zidovudine plus lamivudine</td>
<td>27.2</td>
<td>57/213 (8.3)</td>
<td>1.40 (1.00-1.95)</td>
</tr>
<tr>
<td>NNRTI</td>
<td>Efavirenz</td>
<td>3.7</td>
<td>7/94 (7.4)</td>
</tr>
<tr>
<td>Nevirapine</td>
<td>4.6</td>
<td>7/115 (6.1)</td>
<td>0.90 (0.41-1.96)</td>
</tr>
<tr>
<td>Protease inhibitor</td>
<td>Atazanavir sulfate</td>
<td>8.8</td>
<td>26/222 (11.7)</td>
</tr>
<tr>
<td>Darunavir</td>
<td>2.1</td>
<td>1/54 (1.9)</td>
<td>0.26 (0.04-1.88)</td>
</tr>
<tr>
<td>Fosamprenavir calcium</td>
<td>1.7</td>
<td>4/42 (9.5)</td>
<td>1.47 (0.52-4.18)</td>
</tr>
<tr>
<td>Lopinavir</td>
<td>13.5</td>
<td>23/176 (13.9)</td>
<td>1.54 (1.03-2.31)</td>
</tr>
<tr>
<td>Nelfinavir mesylate</td>
<td>8.7</td>
<td>15/220 (6.8)</td>
<td>1.02 (0.59-1.76)</td>
</tr>
<tr>
<td>Ritonavir when used as a booster</td>
<td>25.2</td>
<td>59/635 (9.3)</td>
<td>1.65 (1.19-2.30)</td>
</tr>
<tr>
<td>Saquinavir mesylate</td>
<td>1.3</td>
<td>2/33 (6.1)</td>
<td>0.90 (0.21-3.77)</td>
</tr>
</tbody>
</table>

Abbreviations: NNRTI, nonnucleoside reverse transcriptase inhibitor; NRTI, nucleoside reverse transcriptase inhibitor.

*Each row reflects a separate logistic regression model, both unadjusted and adjusted, for low maternal CD4 count (<250 cells/mm³) early in pregnancy and birth cohort.*
Sensitivity Analyses
When restricting cases to children with major anomalies and when excluding 11 children with chromosomal anomalies (eTable 1 in the Supplement), the significant associations with first-trimester atazanavir, ritonavir (as a booster), and the combination of didanosine plus stavudine persisted, with similar estimated effects. Similarly, sensitivity analyses accounting for multiple children per mother and for clustering within research site provided results almost identical to those in Table 2.

In the dynamic cohort, a higher percentage of infants were exposed during the first trimester to HAART (47.2%) and to PIs (41.9%), but none were exposed to didanosine plus stavudine. In adjusted models, no significant associations for first-trimester exposures were observed for the dynamic cohort, and the association for atazanavir was attenuated (aOR, 1.55; 95% CI, 0.91-2.63). However, when ARV exposures at any time during pregnancy were evaluated, significantly higher odds of CAs were observed among infants in the dynamic cohort exposed to lamivudine (aOR, 2.13; 95% CI, 1.26-3.60), zidovudine (aOR, 2.06; 95% CI, 1.23-3.44), zidovudine plus lamivudine (aOR, 2.43; 95% CI, 1.45-4.06), and abacavir (aOR, 1.58; 95% CI, 1.00-2.49). In contrast, a protective association was found with darunavir exposure (aOR, 0.21; 95% CI, 0.05-0.84).

Examination of ARV exposures within the dynamic cohort by trimester of the first exposure indicated that the increased risk for zidovudine, lamivudine, and their combination was observed for both the first and later trimesters compared with those never exposed to these specific ARVs or combinations. The increased risk for abacavir in the dynamic cohort was only observed for those first exposed later in pregnancy compared with abacavir unexposed (aOR, 2.20; 95% CI, 1.31-3.71). For musculoskeletal anomalies, those exposed to first-trimester atazanavir had significantly increased odds of CAs (aOR, 2.49; 95% CI, 1.25-4.95).

Discussion
We observed an overall prevalence of 6.78 CAs among every 100 live births, which is considerably higher than that in prior studies of HIV-exposed infants in the United States and the United Kingdom, with reported prevalences ranging from 2.8% to 5.5%,4-7,12,13 and higher than the rate of 3.2% in a recent Italian cohort,10 but it is similar to the 6.2% rate reported by a Latin American study.27 We observed an increasing trend in the rate of CAs from before 2002 through 2010, followed by a slight decline through 2012. The higher rates of CAs may reflect a real increase consistent with temporal trends demonstrated in population studies,28,29 increased ascertainment given the study-required evaluation for anomalies, and longer follow-up than some studies. They may also be partially attributable to the increasing percentage of mothers receiving ARVs early in pregnancy, which was less than 30% in earlier studies3,6,7,12,13 but is approximately 50% of the present cohort.

The association of first-trimester atazanavir exposure with CAs, particularly musculoskeletal and skin anomalies, has not previously been reported to our knowledge and warrants further investigation. The International Maternal Pediatric Adolescent AIDS Clinical Trials P1025 protocol13 also reported higher rates of CAs with first-trimester atazanavir exposure (9.2% vs 5.3% for atazanavir unexposed; aOR, 1.83), although not attaining significance. Most prior studies included births before 2007 and did not reflect the increasing use of this particular ARV up to 20% by 2010 since its 2003 approval.16 Furthermore, exposures to particular ARV combinations may be associated with higher risks. We observed higher odds of CAs for first-trimester atazanavir exposure when combined with ritonavir, tenofovir, or emtricitabine, all with increased

Table 3. Analyses Conducted by Type of Anomaly

<table>
<thead>
<tr>
<th>Type of Anomaly</th>
<th>Defect Rate, No./Total No. (%)</th>
<th>Odds Ratio (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Exposed</td>
<td>Unexposed</td>
</tr>
<tr>
<td>Atazanavir sulfate</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cardiovascular</td>
<td>7/222 (3.2)</td>
<td>33/2295 (1.4)</td>
</tr>
<tr>
<td>Musculoskeletal</td>
<td>11/222 (5.0)</td>
<td>46/2295 (2.0)</td>
</tr>
<tr>
<td>Skin</td>
<td>3/222 (1.4)</td>
<td>6/2296 (0.3)</td>
</tr>
<tr>
<td>Ritonavir when used as a booster</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cardiovascular</td>
<td>16/635 (2.5)</td>
<td>24/1882 (1.3)</td>
</tr>
<tr>
<td>Musculoskeletal</td>
<td>22/635 (3.5)</td>
<td>35/1882 (1.9)</td>
</tr>
<tr>
<td>Zidovudine</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male genital</td>
<td>8/726 (1.1)</td>
<td>6/1791 (0.3)</td>
</tr>
<tr>
<td>Lamivudine</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male genital</td>
<td>8/797 (1.0)</td>
<td>6/1720 (0.3)</td>
</tr>
<tr>
<td>Didanosine plus stavudine</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cardiovascular</td>
<td>0/7</td>
<td>40/2510 (1.6)</td>
</tr>
<tr>
<td>Musculoskeletal</td>
<td>1/7 (14.3)</td>
<td>56/2510 (2.2)</td>
</tr>
</tbody>
</table>

Abbreviation: NA, not applicable.
* Models adjusted for any anomaly (low mater nal CD4 count <250 cells/mm3) early in pregnancy and birth cohort, cardiovascular anomaly (birth cohort), musculoskeletal anomaly (low maternal CD4 count early in pregnancy and first-trimester maternal alcohol consumption), skin anomaly (low maternal CD4 count early in pregnancy), and male genital anomaly (maternal age >35 years at delivery).
use during the past decade,\textsuperscript{16-18} than with older ARVs (zido-
vedine or lamivudine). In contrast, when ritonavir was used to
boost PIs other than atazanavir (primarily lopinavir), it was
not associated with higher odds. Finally, while various spe-
cific anomalies were reported for atazanavir-exposed chil-
dren, the increased risk was highest for musculoskeletal and
skin anomalies of generally milder severity.

In contrast to some prior studies,\textsuperscript{12,17} we observed no asso-
ciation of CAs with first-trimester efavirenz exposure. A
meta-analysis\textsuperscript{19} also found no increased risk of overall CAs with
efavirenz exposure. We confirmed an increased risk of male
genital anomalies with first-trimester zidovudine exposure\textsuperscript{1-3};
this association remained marginally significant in the dy-
namic cohort and was thus not entirely attributable to over-
lap with prior studies.

Our study has several strengths, including its large size, the
use of the well-validated MACDP classification system, and
complete information on maternal health, substance use, and
pregnancy complications. We also considered other medica-
tions used during pregnancy such as selective serotonin reup-
take inhibitors and folate antagonists. However, a limitation
of our study is the possibility of selection bias: mothers of static
cohorts with CAs may have been more willing to par-
ticipate, which could have artificially increased the preva-
ience of CAs and may have accounted for the higher rate during
2005 to 2010 compared with more recent years. Conversely,
allowing enrollment up to 1 week after birth may exclude in-
fants with severe CAs incompatible with life. In addition, the
MACDP classification system, while providing specific objec-
tive criteria for identifying anomalies, may not allow discrimi-
nation by defect severity. Misclassification and lack of specifi-
city of CA outcomes, as well as potential exposure misclassifica-
tion, could have resulted in attenuation of findings;
therefore, we evaluated specific CAs and both individu-
als and combinations of ARVs in increasing use.

Conclusions

In conclusion, our study was reassuring in confirming the lack
of an increased risk of CAs among children exposed to ARVs
during the first trimester of pregnancy. We observed a higher
prevalence of CAs than has been reported in the general popu-
lation, but after adjustment for calendar year and maternal
characteristics, no relative increase in risk was observed for
those exposed vs unexposed to HAART or to PI-based regi-
ments early in pregnancy. However, while the absolute risk of
CAs was low, some individual drugs, particularly atazanavir,
showed relative increases in the risk of overall CAs and spe-
cific anomalies, which warrant further study. As World Health
Organization 2013 ARV guidelines are implemented globally,
an increasing percentage of women with HIV will be ex-
pected to enter pregnancy already receiving ARVs.\textsuperscript{15} There-
fore, risks associated with in utero ARV exposures must be con-
sidered to identify optimal regimens based on their safety
profiles.

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REFERENCES