Birth Defects and Adverse Pregnancy Outcomes in Hospital-based Birth Surveillance in Eswatini

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Background: The Botswana Tsepamo study identified an initial neural tube defect (NTD) safety signal with dolutegravir antiretroviral therapy (ART) exposure at conception. We conducted similar surveillance in 5 hospitals in Eswatini from September 2021 to September 2023 to evaluate the prevalence of birth defects and adverse pregnancy outcomes by maternal HIV status and ART regimen/timing.

Methods: Routine pregnancy history and HIV/ART status were collected from clinic records. Women of live or stillborn infants with birth defects consented for interviews and photographs of defects. A medical geneticist reviewed blinded interview data and photographs.

Results: Of 45,836 women with live-born or stillborn infants, 13,577 (29.6%) were living with HIV; 11,581 (86.0%) were receiving ART at conception (84.1% dolutegravir). Overall, birth defects were confirmed in 387 (0.8%) women. Comparing women with and without HIV, there were no significant differences in major defects (0.48% vs. 0.38%) or NTD (0.10% vs. 0.08%). In women with HIV, there were no significant differences between those on dolutegravir versus non-dolutegravir at conception for major defects (0.53% vs. 0.49%) or NTD (0.08% vs. 0.22%). Stillbirths were significantly higher in women with HIV than those without (2.6% vs. 1.9%, $P < 0.001$), as was low birthweight and preterm delivery (11.8% vs. 10.4%, $P < 0.001$; 12.5% vs. 10.7%, $P < 0.001$, respectively). There were no significant differences in outcomes by ART regimen.

Conclusions: While these data from sub-Saharan Africa further strengthen the lack of a NTD safety signal in women with HIV on ART, there remained elevated adverse birth outcomes despite treatment compared to women without HIV.

Key Words: HIV, birth defect, dolutegravir, antiretroviral

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A neural tube defect (NTD) safety signal with dolutegravir exposure at conception was unexpectedly identified in pregnant women living with HIV (WLH) in the Botswana Tsepamo birth outcomes study. This signal brought attention to the need for reliable data on the safety of new drugs in pregnancy and improved pharmacovigilance systems in resource-limited settings.

An unscheduled, preliminary analysis from the Tsepamo study in May 2018 reported 4 NTDs among 426 dolutegravir (Tivicay, manufactured by Viiv Healthcare, London, United Kingdom) exposures at conception (0.94%) compared to 14 in 11,300 non-dolutegravir exposures at conception (0.12%) in WLH.[1] These preliminary data led the World Health Organization (WHO), US Food and Drug Administration and European Medicines Agency to issue a caution about using this highly potent and effective antiretroviral (ARV) drug in women of reproductive potential and pregnant women.[2–4] At that time, only limited and fragmented data were available related to birth defects with dolutegravir exposure at conception.[5] There was no single database containing sufficient prospective exposures to dolutegravir at conception to assess an uncommon (~1/1000 births) outcome such as NTD. Additional data collected from the Botswana study over time increased the number of dolutegravir exposures at conception from 426 to 9460. Re-analysis suggested that the prevalence of NTD with dolutegravir antiretroviral therapy (ART) at conception (0.11%) was actually not significantly different from exposure to dolutegravir started during pregnancy (0.06%), non-dolutegravir ART at conception in women with (0.11%) or without HIV (0.07%).[6]

To provide further evidence from an additional African country, we conducted a similar birth defect surveillance study in Eswatini. Like Botswana, Eswatini is a sub-Saharan African (SSA) country without folate food fortification, which has a protective effect against NTD.[7] The country has one of the highest adult HIV prevalences in the world (24%), with a prevalence of 36% among pregnant women, and 93% of deliveries are with skilled personnel (a proxy for facility delivery).[8,9] Surveillance was conducted in 5 of 12 public hospitals with maternity facilities in Eswatini, representing approximately 77% of all births nationally (based on countrywide 2020 estimates). The transition to dolutegravir-based ART in Eswatini started in 2019, with scale-up nationally in early 2021.[10] In our initial report of the first year of surveillance (September 2021 to September 2022), including over 24,000 women giving birth, of the 7586 WLH, 65% were receiving dolutegravir ART at conception, and 99% of those starting ART for the first time during pregnancy initiated dolutegravir-based therapy.[11] We demonstrated that NTD prevalence was not significantly different among WLH on dolutegravir-based ART at conception, non-dolutegravir-based ART at conception and women without HIV (0.08%, 0.15% and 0.08%, respectively).[11] These data, combined with the updated Botswana data, demonstrated that the initial increase in NTD prevalence detected in the Tsepamo study in 2018 was likely due to small numbers of exposures to the drug of concern, resulting in inadequate data to inform NTD prevalence.

The current report provides an additional year of birth surveillance data, through September 2023, including over 45,000 pregnancies and nearly 6000 additional women living with HIV. We aimed to determine the prevalence of birth defects, including NTD,

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and adverse pregnancy outcomes among live and stillborn infants by maternal HIV status and ART regimens and timing.

MATERIALS AND METHODS

Detailed study methods have been reported previously.[11] In summary, we conducted a cross-sectional, observational study to evaluate birth outcomes of women delivering September 2021 to September 2023 in 5 government hospitals across all 4 regions of Eswatini. Routine data on pregnancy history and HIV/ART status were collected from paper and electronic clinic records from maternity wards. Limited data on miscarriages were also abstracted from hospital registers to the extent that they were documented. Women with live or stillborn infants with birth defects identified through routine surface examination provided consent for interviews capturing detailed history and exposure data and photographs of newborns' birth defects. Blinded interview data and photographs were reviewed by a medical geneticist for confirmatory defect diagnosis and classified by organ group based on the Metropolitan Atlanta Congenital Defects Program.[12] Data were collected by trained research assistants based at each study site.

Stillbirth was defined as fetal death at ≥28 weeks gestation, reflecting fetal viability locally with preterm birth in Eswatini,[13] or a fetal death at <28 weeks gestation with a weight of ≥1000 g per the WHO definition of late fetal death.[14] Miscarriages were recorded in non-maternity wards as fetal deaths at <28 weeks gestational age. Rarely, weight and/or gestational age were not recorded, and the outcome was defined based on clinical notes in registers. Stillbirth rates were calculated using all pregnancies as the denominator (live-born, stillborn or miscarriage). Medical abortions are uncommon in Eswatini and were only included if birth defects were identified via ultrasound, which is infrequently performed at public health facilities. Low birthweight (LBW) rates were defined as infants weighing <2500 g (WHO definition) among livebirths only. Preterm delivery (PTD) rates were defined as infants delivered at <37 weeks gestation among livebirths only. Examinations were not routinely conducted for miscarriages; data from miscarriages were only used in this paper as part of the denominator to calculate the stillbirth rate. ART at conception was determined using the date of drug initiation and the last menstrual period and defined as maternal ART received up to 8 weeks after the last menstrual period date (≤6 weeks after the estimated conception date).

We describe rates of birth defects by HIV status and ART regimen at conception. Infants who experienced one or more major defects, irrespective of whether they also had any minor defects, were classified as having a major birth defect. Infants who experienced at least 1 minor defect and no major defects were classified as having a minor birth defect. Major defects were categorized as NTD or non-NTD. Defect prevalence was calculated as the number of events divided by the total number of live and stillbirths; the numerator included medical abortions with confirmed defects. Chromosomal trisomies identifiable by physical appearance were excluded from the analysis.

Study ethical approval was provided by the Eswatini Health and Human Research Review Board (EHHRRB028/2021) and the Advarra Institutional Review Board (Pro00055975) in the United States.

Statistical Analysis

Descriptive statistics were calculated for women and infant characteristics among live and stillbirths overall and grouped by ART regimen at conception. Medians and interquartile ranges were calculated for continuous variables, and frequencies and percentages were provided for categorical variables. Descriptive statistics were also reported for characteristics/risk factors among women with infants with major birth defects. Rates of birth defects, LBW and PTD were estimated overall and by HIV and ART status with corresponding 95% confidence intervals (CIs). A composite adverse birth outcome variable was then derived that included major birth defects, LBW, PTD, stillbirth and miscarriages. Unadjusted and adjusted log-binomial regression models were used to examine the association between this outcome and factors with available routine data from all women: maternal HIV status, age at delivery, gravidity and delivery/miscarriage year. Relative risks (RRs) were reported with corresponding 95% CIs. All tests were 2-sided, and the level of statistical significance was set at 0.05. Data were analyzed using SAS (v9.4; Cary, NC).

RESULTS

A total of 45,836 women with live-born or stillborn infants (median age 26 years, interquartile range: 21–32) were enrolled in the study; 32,259 (70.4%) had an HIV-negative status and 13,577 (29.6%) were living with HIV (Table 1); 4 women had an unknown HIV status and 3 women had medical abortions due to a birth defect identified via ultrasound (not shown in Table 1). Of the 13,577 WLH, 13,465 (99.2%) had data on their ART regimen; 11,581 (86.0%) received ART at conception. Of those receiving ART at conception, 84.1% received a dolutegravir-based regimen; most women on non-dolutegravir ART at conception received an efavirenz-based regimen (95.7%, 1759). Of the 1884 women newly initiated on ART during pregnancy, 99.5% were initiated on dolutegravir-based ART and 0.5% were initiated on efavirenz-based ART.

In total, 46,569 infants were born to women enrolled in the study (Table 1); 2.3% (1066) were stillborn and 12.0% were LBW (<2500 g). Eighty infants (0.2%) died within 24 hours of life. The most common causes of death were birth asphyxia (n = 36) and prematurity with associated complications (n = 22).

In total, 387 women delivered 388 infants (one twinset) with ≥1 birth defects (Fig. 1): 215 (0.5%) infants had ≥1 major birth defects with or without minor defects and 173 (0.4%) infants had ≥1 minor defects only. Of infants with major defects, 141 (65.6%) were born to women without HIV and 74 were born to WLH. Among women with infants who had major birth defects, 57 (26.5%) were on dolutegravir-based ART at conception, 12 (5.6%) on non-dolutegravir-based ART at conception (all efavirenz-based) and 5 (2.3%) were diagnosed with HIV during pregnancy and not on ART at conception (Table 2).

Of the 215 infants with major defects, 40 (18.6%) infants had NTDs, 26 of whom had an NTD as the sole major defect (1 child had 2 NTDs) and 14 of whom had both NTDs plus ≥1 additional major defects. There was no significant difference in NTD prevalence between WLH and women without HIV (0.08% vs. 0.10%, respectively, $P = 0.67$). Among WLH, there was no significant difference in NTD prevalence between those receiving dolutegravir-based ART at conception compared to non-dolutegravir-based ART at conception (0.08% vs. 0.22%, $P = 0.11$). Excluding infants with NTD as a sole defect (but including the 14 children who had both a major defect and NTD), major defect prevalence was not significantly different between women with and without HIV (0.48% vs. 0.38%, $P = 0.14$). Similarly, among WLH, there was no significant difference in major defects between those receiving dolutegravir-based ART at conception compared to non-dolutegravir-based ART at conception (0.53% vs. 0.49%, $P = 0.81$). Infants diagnosed with minor defects only were excluded from Table 2; most minor defects (92.5%, 160) were polydactyly, postaxial hand or unspecified.

The overall rates of stillbirth, LBW and PTD were 2.1%, 10.8% and 11.2%, respectively (Table 3). Stillbirths were significantly greater ($P < 0.001$) among WLH compared to women

TABLE 1. Maternal Pregnancy and Antiretroviral History and Pregnancy Outcomes and Infant Birth Information among Live and Stillbirths

Women Characteristics	Women With HIV on Dolutegravir at Conception (n = 9743)	Women With HIV on Non-dolutegravir ART at Conception (n = 1838)	Women Diagnosed With HIV During Pregnancy, Not on ART at Conception (n = 1884)	Women With HIV on Unknown Regimen (n = 112)	Women Without HIV (n = 32,259)	Total (n = 45,836)
Women's age (yr), median (IQR)	31 (27–36)	31 (27–36)	26 (22–31)	28.5 (23–35)	25 (20–30)	26 (21–32)
No. (%)						
Women's age (yr)						
<18	108 (1.1)	6 (0.0)	72 (3.0)	1 (0.0)	2714 (8.0)	2901 (6.3)
18–34	6629 (68.0)	1271 (69.0)	1563 (83.0)	82 (73.0)	26,145 (81.0)	35,694 (77.9)
≥35	3006 (30.0)	561 (30.0)	249 (13.0)	29 (25.0)	3400 (10.0)	7245 (15.8)
Gravida						
1	932 (10.0)	140 (7.6)	642 (34.1)	27 (24.1)	12,849 (39.8)	14,591 (31.8)
2–5	7830 (80.0)	1528 (83.1)	1206 (64.0)	75 (67.0)	18,485 (57.3)	29,127 (63.6)
≥6	980 (10.1)	169 (9.2)	35 (1.9)	10 (9.0)	919 (2.9)	2113 (4.6)
Missing	1	1	1	0	6	9
Gestational age at delivery (wk)						
<32	214 (2.2)	53 (2.9)	46 (2.4)	3 (2.7)	634 (2.0)	950 (2.1)
≥32 to <37	1052 (10.8)	190 (10.3)	226 (12.0)	9 (8.0)	3008 (9.3)	4485 (9.8)
≥37	8476 (87.0)	1594 (86.8)	1611 (85.6)	100 (89.3)	28,612 (88.7)	40,397 (88.1)
Missing	1	1	1	0	5	8
Type of pregnancy						
Singleton	9555 (98.1)	1807 (98.4)	1852 (98.4)	109 (98.2)	31,772 (98.5)	45,099 (98.4)
Twin	184 (1.9)	30 (1.6)	30 (1.6)	2 (1.8)	467 (1.5)	713 (1.6)
Triplet	1 (0.0)	0	0	0	9 (0.0)	10 (0.0)
Missing	3	1	2	1	11	18
Infant/delivery characteristics	**Women with HIV on Dolutegravir at Conception (n = 9929)**	**Women with HIV on Non-dolutegravir ART at Conception (n = 1868)**	**Women Diagnosed with HIV during Pregnancy, Not on ART at Conception (n = 1914)**	**Women with HIV on Unknown Regimen (n = 114)**	**Women without HIV (n = 32,744)**	**Total (n = 46,569)**
Birth outcomes						
Live birth	9661 (97.3)	1809 (96.8)	1852 (96.8)	111 (97.4)	32,072 (97.9)	45,507 (97.7)
Stillbirth	268 (2.7)	59 (3.2)	62 (3.2)	3 (2.6)	672 (2.1)	1066 (2.3)
Sex						
Male	5040 (50.8)	976 (52.3)	945 (49.4)	58 (51.3)	16,880 (51.6)	23,902 (51.3)
Female	4885 (49.2)	890 (47.7)	967 (50.6)	55 (48.7)	15,842 (48.4)	22,640 (48.6)
Could not be determined	1 (0.0)	1 (0.1)	0	0	10 (0.0)	12 (0.1)
Missing	3	1	2	1	12	19
Weight (g)						
<1500 (VLBW)	200 (2.0)	50 (2.7)	38 (2.0)	1 (0.9)	543 (1.7)	832 (1.8)
1500–2499 (LBW)	1106 (11.1)	197 (10.5)	235 (12.3)	16 (14.1)	3209 (9.8)	4763 (10.2)
>2500	8619 (86.9)	1619 (86.8)	1639 (85.7)	96 (85.0)	28,961 (88.5)	40,938 (88.0)
Missing	4	2	2	1	31	40
Early infant death						
Within 24 h	17 (0.2)	0	5 (0.3)	0	58 (0.2)	80 (0.2)
>24h, but before discharge	5 (0.1)	1 (0.1)	0	0	9 (<0.1)	15 (<0.1)
Died, but missing timing	4	3	0	0	37	44
Mode of delivery						
Vaginal	8415 (84.8)	1633 (87.5)	1634 (85.5)	91 (80.5)	27,792 (84.9)	39,567 (85.0)
C-section	1511 (15.2)	234 (12.5)	278 (14.5)	22 (19.5)	4939 (15.1)	6986 (15.0)
Missing	3	1	2	1	13	20

VLBW indicates very low birthweight.

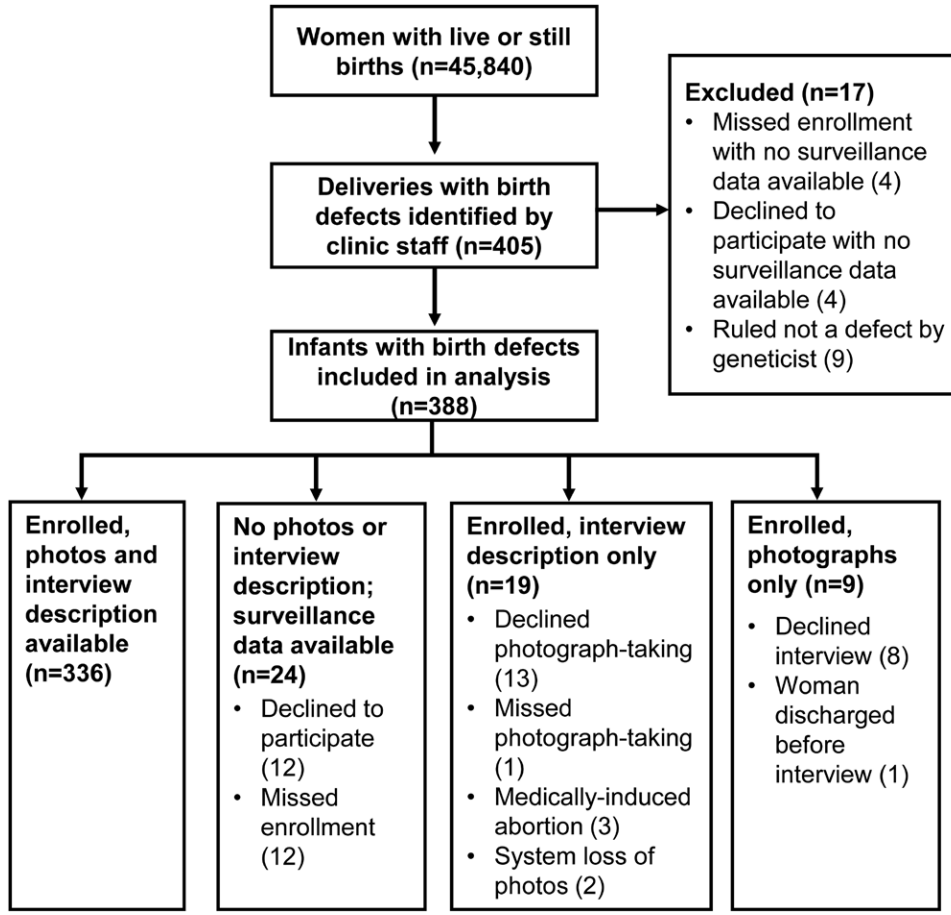

FIGURE 1. Screening and enrollment of women with infants with identified birth defects

TABLE 2. Prevalence of Birth Defects by Maternal HIV and ART Status

HIV and ART Status at Conception	Live/ Stillbirths*	All Major Defects† (n)	Prevalence (95% CI)	NTD (n)	Prevalence (95% CI)	*P* Value	Major Defects Excluding NTD as Sole Defect‡ (n)	Prevalence (95% CI)	*P* Value
Living without HIV	32,259	141	0.44 (0.37–0.52)	27	0.08 (0.06–0.12)	0.67	124	0.38 (0.32–0.46)	0.14
Living with HIV	13,465	74	0.55 (0.44–0.69)	13	0.10 (0.06–0.17)		65	0.48 (0.38–0.61)	
ART at conception									
Dolutegravir	9743	57	0.59 (0.45–0.76)	8	0.08 (0.04–0.16)	0.11§	52	0.53 (0.41–0.70)	0.81§
Non-dolutegravir	1838	12	0.65 (0.37–1.14)	4	0.22 (0.08–0.56)		9	0.49 (0.26–0.93)	
No ART	1884	5	0.27 (0.11–0.62)	1	0.05 (0.01–0.30)		4	0.21 (0.08–0.54)	
Total¶	45,724	215	0.47 (0.41–0.54)	40	0.09 (0.06–0.12)		189	0.41 (0.36–0.48)	

*Determined based on number of women with live and stillbirths; none of the major birth defects occurred in multiple births.

†Includes 3 medically induced abortions (2 women living with HIV, 1 on dolutegravir at conception and 1 on EFV at conception and 1 woman without HIV).

‡Excludes 26 infants with NTD as the sole major defect; includes 14 infants with both major defect(s) plus NTD.

§Test of association only performed between ARV regimens (dolutegravir vs. non-dolutegravir).

¶Total excludes 116 women with live/still births on an unknown ART regimen at conception (n = 112) or unknown HIV status (n = 4) as no birth defects were identified in these groups.

without HIV (2.6% vs. 1.9%). LBW and PTD were also significantly greater ($P < 0.001$) among live infants born to WLH compared with those without HIV (LBW 11.8% vs. 10.4%; PTD 12.5% vs. 10.7%, respectively). There were not any significant differences in stillbirth, LBW or PTD among infants born to WLH and receiving dolutegravir-based ART at conception compared to non-dolutegravir-based ART at conception or those receiving no ART at conception who initiated ART during pregnancy.

Table 4 presents the major birth defects by maternal HIV/ART status, with the specific types of NTD and other major non-NTD organized by organ group, respectively. Multiple defects in the same infant in the same organ group were combined, but some infants had multiple defects in more than 1 organ category, so the numbers do not sum to the totals. Sixteen of the NTDs (40.0%) were myelomeningocele/meningocele. Of 189 infants with major defects excluding NTD, the most common was varus foot malformation (23.3%, 44). See Table, Supplemental Digital Content 1,

TABLE 3. Prevalence of Other Birth Outcomes by Maternal HIV and ART Status

HIV Status	Deliveries and Miscarriages*	Stillbirths† (n)	Stillbirth Prevalence (95% CI)	*P* Value	Live births†	LBW (n)	LBW Prevalence (95% CI)	*P* Value	PTD (n)	PTD Prevalence (95% CI)	*P* Value
Living without HIV	35,784	673	1.9 (1.8–2.0)	<0.001	32,071	3339	10.4 (10.1–10.8)	<0.001	3439	10.7 (10.4–11.1)	<0.001
Living with HIV	14,979	392	2.6 (2.4–2.9)		13,433	1581	11.8 (11.2–12.3)		1677	12.5 (11.9–13.1)	
Unknown HIV status	42	2	4.8 (1.3–15.8)		2	0	--		0	--	
ART at conception											
Dolutegravir	10,523	268	2.6 (2.3–2.9)	0.23	9661	1124	11.6 (11.0–12.3)	0.82‡	1197	12.4 (11.8–13.1)	0.69‡
Non-dolutegravir	1957	59	3.0 (2.3–3.9)		1809	207	11.4 (10.1–13.0)		218	12.1 (10.6–13.6)	
No ART	2095	62	3.0 (2.3–3.8)		1852	234	12.6 (11.2–14.2)		249	13.4 (12.0–15.1)	
Unknown ART	404	3	0.7 (0.3–2.2)		111	16	14.4 (9.1–22.1)		13	11.7 (7.0–19.0)	
Total	50,805	1067	2.1 (2.0–2.2)		45,506	4920	10.8 (10.5–11.1)		5116	11.2 (11.0–11.5)	

*Data available for 4232 (86.9%) of 4870 miscarriages. Of 4232 miscarriages, 1154 mothers were living with HIV (7.7% miscarriages in 14,979 deliveries and miscarriages) and 3040 were living without HIV (8.5% in 35,784 deliveries and miscarriages), and 38 mothers had unknown HIV status (90.5% miscarriages in 42 deliveries and miscarriages). There were 3 medically induced abortions (2 women living with HIV, 1 on dolutegravir at conception and 1 on EFV at conception, and 1 woman without HIV).

†Determined based on number of live and stillbirths, including multiple births, and not the number of women as in Table 2.

‡Test of association only performed between ARV regimens (dolutegravir vs. non-dolutegravir).

TABLE 4. Type of Neural Tube Defect by Maternal HIV and ART Status at Conception Among Live and Stillbirths

Birth Defects	Dolutegravir (n = 9743)	Non-Dolutegravir ART (n = 1838)	No ART (n = 1884)	HIV-Negative Status (n = 32,259)	Total (n = 45,724)
No. of infants with major defects*	57	12	5	141	215
Neural tube defects†	8	4	1	27	40
Myelomeningocele or meningocele with (5) without (11) hydrocephalus	4		0	12	16
Anencephaly/Acrania	3	2	0	4	9
Anencephaly	0	0	0	1	1
Encephalocele	0	0	0	4	4
Craniorachischisis	0	0	0	2	2
Spina bifida NOS	1	2	1	3	7
Occipital encephalocele; microcephaly	0	0	0	1	1
No. of infants with major defects (excluding NTD)	52	9	4	124	189
Musculoskeletal defects	26	6	2	79	113
Defects of face, eye, ear and neck	14	5	9	23	51
Limb defects	12	1	0	21	34
Other (non-NTD) nervous system defects	7	1	0	16	24
Genitourinary defects	3	0	0	10	13
Syndromes	1	0	1	3	5
Gastrointestinal defects (upper, lower)	0	0	0	5	5
Skin and skin derivatives	0	0	1	3	4
Other	2	0	0	6	8

*Numbers do not sum to group totals because some infants had more than one major defect.

†Fourteen infants were diagnosed with an NTD plus other major non-NTD defect(s).

http://links.lww.com/INF/G75, for a description of other potential risk factors and exposures among women of infants with identified birth defects.

Unadjusted and adjusted RRs of a composite adverse birth outcome are presented in Table 5. In the adjusted model, only an HIV-positive status (RR: 1.22, 1.16–1.29) and maternal age <18 years (RR: 1.20, 1.04–1.38) were associated with an increased risk of any adverse outcome.

DISCUSSION

Our study adds nearly 50,000 births and 10,000 women on dolutegravir at conception to further strengthen the data from an SSA setting without national folate fortification on the lack of an NTD safety signal. NTD prevalence was 0.08%, both among WLH on dolutegravir at conception and women without HIV. These rates are similar to the updated data from the Tsepamo study from 2022, in which the NTD prevalence was reported as 0.11% (95% CI 0.06%–0.19%) and 0.07% (95% CI 0.05%–0.08%) in women on dolutegravir at conception and without HIV, respectively.[6] By the time our study had commenced in Eswatini, dolutegravir transition was well underway and there were fewer exposures to efavirenz-based ART than in the Tsepamo study. In our study, NTD prevalence in women on non-dolutegravir-based ART at conception (96% of which were births among women on efavirenz at conception) was 0.22% with 1838 exposures with wide confidence intervals; this contrasts with the NTD prevalence in women on efavirenz-based ART at conception in the Tsepamo study of 0.08%, which included 14,432 births among women on efavirenz at conception.[6] We found no significant differences in NTD or major birth defects overall by maternal HIV status or by maternal ART regimen at conception in WLH.

Other birth surveillance studies in Kenya, South Africa and Uganda also did not find associations between major birth defects overall and HIV status[15,16] or by dolutegravir versus non-dolutegravir use at conception in WLH.[16] Overall major birth defect prevalence in Eswatini (0.5% and 0.4% among women with and without HIV, respectively) was similar to those reported in other birth surveillance studies in SSA.[17–19] The major defect prevalence rates in Eswatini and other African countries are lower than overall birth defect prevalence

TABLE 5. Adverse Pregnancy Outcomes (Composite) Estimated from Unadjusted and Adjusted Models Among All Women

Variable	Unadjusted			Adjusted		
	N	Relative Risk	95% CI	N	Relative Risk	95% CI
Maternal HIV status	50,033			45,847		
Positive		1.07	1.03–1.11		**1.22**	1.16–1.29
Negative (R)		1			1	
Maternal age at delivery (yr)	50,031			45,847		
<18		0.81	0.73–0.90		**1.20**	1.04–1.38
18–35		0.91	0.86–0.96		1.05	0.96–1.14
>35 (R)		1			1	
Gravida	48,611			45,847		
1		0.93	0.83–1.04		0.98	0.84–1.14
2-5		1.01	0.91–1.13		0.93	0.81–1.06
≥6 (R)		1			1	
Delivery/miscarriage (yr)	45,854			45,847		
2021		0.999	0.92–1.09		0.99	0.91–1.08
2022		0.98	0.92–1.04		0.98	0.92–1.04
2023 (R)		1			1	

R indicates reference group.
Bold values indicate significance at the 0.05 level.

rates in the United States and Europe because they are based on the detection of visually obvious defects identified by surface examination of the infant at birth in countries where ultrasound evaluations during pregnancy are rare,[20] whereas birth registries in resource-rich settings often include internal defects identified following birth through one year of age or identified by ultrasound examinations.[21,22] An evaluation of healthcare claims data between 2008 and 2020 in the United States compared the incidence rates of NTDs, stillbirth and pregnancy loss by HIV status and periconceptional ART exposure. They did not find significant differences in NTD risk ratios between pregnant women with periconceptional dolutegravir or non-dolutegravir exposure and those without HIV; however, periconception ART exposure groups had higher risk ratios for pregnancy loss compared to those without HIV.[23]

Our results also showed that a prevalence gap of adverse birth outcomes persists between women without HIV and WLH regardless of ART regimen. Our overall rates of adverse birth outcomes of 2.1% stillbirth, 10.8% LBW and 11.2% PTD were similar to or slightly lower than published rates from other low-income and middle-income country settings: 2.2% to 2.5%,[24–26] 11.7% to 15.5%[26–28] and 12.6% to 12.7%,[26,27] respectively. However, WLH compared to those without HIV experienced significantly greater non-birth defect adverse birth outcomes. Moreover, when adverse outcomes were combined into one composite variable, HIV-positive status was associated with a 1.2 times risk of adverse outcomes when accounting for other factors, including gravidity and delivery year, which have been used in similar models of birth defect surveillance.[29,30] Women younger than 18 years also had 1.2 times the risk of adverse outcomes compared to women older than 35 years after adjusting for confounders.

Similar differential PTD and LBW rates by maternal HIV or ART status have been reported previously.[31–33] Aligned with our study definition of ≥28 weeks gestation, stillbirth rates in 4 SSA countries were 2.9% for WLH and 1.9% in women without HIV.[26] In a provincial birth surveillance study in South Africa, compared to women without HIV, stillbirths were higher among WLH only if they were not on ART; however, LBW and late pregnancy-related deaths were higher among all WLH.[34] Among WLH, those initiating ART in early pregnancy had better birth outcomes than both those initiating preconception and in late pregnancy. In another study in Zimbabwe that looked at PTD, LBW and small for gestational age (SGA), only SGA was associated with maternal HIV infection in multivariable analysis, though the authors did highlight the lack of accurate gestational age determinations as a limitation in the absence of diagnostics, like fetal ultrasound.[25]

Potential biological mechanisms could explain some of these differences in adverse birth outcomes by HIV status. Elevated inflammatory markers have been associated with adverse outcomes in pregnant women without HIV infection.[35,36] In non-pregnant persons with HIV receiving ART, evidence of persistent inflammation despite long-term suppressive therapy has been observed.[37,38] Several studies have shown higher levels of inflammatory markers in pregnant WLH on ART compared to pregnant women without HIV, although the studies differed in the specific cytokines evaluated.[39–44] The authors have hypothesized that residual inflammation despite ART could affect pregnancy outcomes. A study in India found that elevated levels of interleukin 17A and interleukin 1β were associated with adverse pregnancy outcomes (including PTD and LBW) in both women with and without HIV.[42] Pregnant WLH on ART had higher levels of several inflammatory biomarkers compared to pregnant women without HIV in a US study; immune activation was more pronounced in those with perinatal HIV and those on protease inhibitor ART.[40] In Uganda, cytokine profiles in pregnant WLH receiving ART were compared to pregnant persons without HIV, finding a distinct maternal plasma cytokine profile in those with HIV associated with ongoing maternal inflammation, particularly in those with detectable viral load despite ART.[43] The proinflammatory, Th1 and pleiotropic cytokines were more often detected in pregnant WLH on ART compared to women without HIV, with PTD associated with increased detection of Th17 cytokines and SGA with Th2 and Th17 cytokines in South Africa.[44] Another study found lower placental weight and smaller placental area in WLH receiving ART from conception than those without HIV.[45]

Numerous studies have shown that adverse birth outcomes are highest among WLH who are not receiving ART.[46] ART has critical benefits for maternal health and reduces perinatal HIV transmission as well as adverse perinatal outcomes. However, while ART significantly reduces the risk of adverse birth outcomes in pregnant WLH, the risk remains higher than in women without HIV.

Our study has some limitations. Surveillance data were collected based on existing clinical records, resulting in some missing data, though we had rigorous quality controls in place for data abstraction and procedures to help ensure the triangulation of data across different source documents. Because primary data on risk

factors were only collected from women with infants with birth defects, there were few variables collected among the entire study population, which limited our examination of the HIV status and birth outcome association in the multivariable model, and thus our ability to minimize potential effects of confounding. Addressing this would have required significant expansion of routinely collected data under a surveillance approach or greater primary data collection among women who delivered infants who were not diagnosed with birth defects, neither of which would have been practical options for this study.

It should be acknowledged that it can be resource-intensive from a human and financial perspective to establish and maintain birth surveillance activities in busy maternity wards and other departments to capture miscarriages.[47] However, there are now several models of birth surveillance in a number of countries across the African continent. Similar to Eswatini, these have included components like training on physical examinations, engaging a medical geneticist and implementing other quality control procedures that contribute to a successful approach and generate critical outcome data in a routinized way.[17,48] Use of mobile applications to provide real-time support to clinical staff and help to ensure standardization can further strengthen this model.[17]

These data underscore the lack of an NTD safety signal with dolutegravir conception, finding no statistical differences by HIV or ART regimen and using a surveillance approach that covers three-quarters of the facility deliveries in a country with a high burden of HIV and no national folate supplementation. The continued elevation in adverse birth outcomes despite ART in pregnant WLH points toward the need to further examine potential mechanisms for this increase, which appears more related to HIV itself rather than ART regimens. This surveillance approach should continue to be used to monitor safety in pregnancy to inform policy and optimize HIV prevention and treatment, particularly as new ARV drugs and formulations are introduced (eg, long-acting ARVs for pre-exposure prophylaxis) and could be used to assess the safety of drugs in pregnancy for other conditions, such as tuberculosis.

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L.M. conceived the original idea. M.M.G., P.N.K., C.C., B.N., W.M., N.D.M., N.M. and L.M. designed the study. P.N.K. and M.K. were responsible for data collection and data management with study oversight from M.M.G., C.C., V.T. and B.N. H.J.H. analyzed the data. M.M.G., H.J.H. and P.N.K. had full access to all the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. As provided the final outcome data used for the results. M.M.G. wrote the manuscript with significant contributions from L.M. All authors reviewed and approved the manuscript.

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