Antiretroviral Drug Use and HIV Drug Resistance Among Young Women in Rural South Africa: HPTN 068

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Background: Antiretroviral (ARV) drugs are used for HIV treatment and prevention. We analyzed ARV drug use and HIV drug resistance in a cohort of young women in rural South Africa enrolled in the HIV Prevention Trials Network (HPTN) 068 study, which evaluated the use of a cash transfer conditional on school attendance to reduce HIV incidence.

Methods: ARV drug testing was performed using plasma samples from 2526 young women. This included 2526 enrollment samples (80 HIV-infected and 2446 HIV-uninfected) and 162 seroconversion samples (first HIV-positive study visit). Testing was performed using a qualitative assay that detects 20 ARV drugs from 5 drug classes. HIV drug resistance testing was performed with the ViroSeq HIV-1 Genotyping System for samples that had HIV viral loads ≥400 copies per milliliter.

Results: At enrollment, ARV drugs were detected in 10 (12.5%) of 80 HIV-infected young women. None of 2446 HIV-uninfected young women had ARV drugs detected at enrollment. ARV drugs were also detected in 16 (9.9%) of 162 seroconverters. At enrollment, 9 (13.4%) of 67 young women with genotyping results had HIV drug resistance; resistance was also detected in 9 (6.9%) of 131 seroconverters with genotyping results.

Conclusions: Most of the HIV-infected young women in this cohort from rural South Africa were not taking ARV drugs, suggesting they were unaware of their HIV status or were not in care. HIV drug resistance was detected in young women with both prevalent and new HIV infection.

Key Words: ARV, drug resistance, HPTN 068, young women, South Africa

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INTRODUCTION

Recent research in HIV treatment and prevention supports an expanding role for use of antiretroviral (ARV) drugs.[1] In HIV-infected individuals, antiretroviral therapy (ART) reduces HIV-associated morbidity and mortality,[2,3] increases life expectancy,[4] and reduces the risk of HIV transmission to sexual partners and infants.[5–7] Early initiation of ART, regardless of CD4 cell count, is now recommended for HIV treatment, prevention of HIV transmission in serodiscordant couples, and prevention of mother-to-child transmission worldwide.[8,9] ARV drug regimens are also

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Y.Z. and M.V.S. contributed equally to this work.

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recommended for HIV-uninfected individuals for pre-exposure prophylaxis (PrEP), post-exposure prophylaxis, and other reasons (eg, treatment of hepatitis).[1,8,10–12] Some ARV drugs are also used off-label for recreational purposes.[13,14] The availability of ARV drugs, recommendations for their use in both HIV-infected and HIV-uninfected individuals, and prevalence of ARV drug use for different indications vary geographically and among different risk groups. This highlights the need for surveillance of ARV drug use and HIV drug resistance in different populations and settings.

ARV drug testing provides an objective method for assessing ARV drug use. Methods based on liquid chromatography–tandem mass spectroscopy are often used to quantify the levels of ARV drugs for research studies.[15–18] These assays typically measure only one or a few ARV drugs. The cost and complexity of this type of testing limit its use for large surveys of ARV drug use. Our group developed a lower-cost, high-throughput method based on high-resolution mass spectrometry that detects 20 ARV drugs from 5 drug classes.[19] Compared with a gold standard method based on liquid chromatography–tandem mass spectroscopy, the multidrug assay achieved 89.1%–100% concordance for detecting ARV drugs in clinical plasma samples.[20] In a second report, the multidrug assay had 100% sensitivity and specificity for detecting tenofovir (TFV) and emtricitabine (FTC) when those drugs were present at concentrations consistent with daily PrEP use.[19] We have used the multidrug assay to evaluate ARV drug use in clinical trials and cohort studies,[19,21–24] including a population-level survey of >7000 HIV-infected adults in sub-Saharan Africa.[24] We also used the assay to evaluate ARV drug use in a cohort of 1806 HIV-uninfected women in the United States; in that study, ARV drugs were detected in women at 2 of the 10 study sites (7% in Bronx, NY; 15% in Baltimore, MD).[23]

Young women in sub-Saharan Africa have significantly higher rates of HIV infection and acquire HIV infection at a younger age than their male peers.[25] In this study, we evaluated ARV drug use and HIV drug resistance in a large cohort of young women in rural South Africa who participated in the HIV Prevention Trials Network (HPTN) 068 study.[26,27] This study enrolled both HIV-infected and HIV-uninfected young women and evaluated the effect of a conditional cash transfer on HIV incidence.[26,27] The overall annual incidence in the HPTN 068 cohort was 1.8%.[27] The study was conducted in the Bushbuckridge subdistrict in Mpumalanga Province, South Africa. Poverty, unemployment, and circular labor migration are common in this region. Resources for HIV care may be more limited in rural areas of South Africa compared with urban areas.[28] Those in rural populations may also be less likely to know their HIV status and may have lower adherence to ART, compared with their urban counterparts.[29–31] These issues highlight the need to evaluate ARV drug use and HIV drug resistance in young women in rural African settings.

METHODS

Study Cohort

Plasma samples were obtained from HPTN 068, a phase III randomized controlled trial (NCT01233531, enrollment period: March, 2011 to December, 2012; the study ended in 2015).[26,27] The HPTN 068 study enrolled 2537 young women aged 13–20 years who were attending high school. Participants had annual study visits through their expected high school graduation date, as previously described (main study).[26,27] A subset of participants had 1 additional follow-up visit after exiting from the main study (follow-up study). South Africa is currently implementing the largest ART program in the world,[32] and many clinics are designated as ART clinics. In HPTN 068, access to care was provided by government referral to the health care system. Clinics were within walking distance in all communities, and treatment was free of charge. At the start of the trial, ART was available for individuals with CD4 cell counts under 350 cells/mm^3; the threshold for ART initiation was changed to 500 cells/mm^3 near the end of the trial. Active linkage to care was provided in the main HPTN 068 study, using a process similar to peer navigation, where study counselors offered to provide transportation to the clinic and to accompany young women to their first HIV care visit.

Laboratory Methods

HIV testing was performed at enrollment (all participants) and at annual follow-up visits for participants who were HIV-uninfected at enrollment. CD4 cell count testing was performed at study site for HIV-infected participants at their first HIV-positive study visit and follow-up visits.[26] All other laboratory testing procedures described in this report were performed retrospectively at the HPTN Laboratory Center (Johns Hopkins University School of Medicine, Baltimore, MD, USA). This included HIV testing to confirm results from site testing and to confirm HIV seroconversion events,[27] HIV viral load testing using the RealTime HIV-1 Viral Load assay (Abbott Molecular, Des Plaines, IL; limit of quantification: 40 copies/mL), ARV drug testing, and HIV drug resistance testing.

ARV drug testing was performed using a qualitative assay based on high-performance liquid chromatography coupled with high-resolution accurate-mass mass spectrometry (QExactive-Orbitrap; Thermo Scientific, San Jose, CA).[19,20] This assay detects 20 ARV drugs from 5 drug classes; this includes 9 protease inhibitors (PIs) (amprenavir, atazanavir, darunavir, indinavir, lopinavir, nelfinavir, ritonavir, saquinavir, and tipranavir); 6 nucleoside/nucleotide reverse transcriptase inhibitors (NRTIs) [abacavir, FTC, lamivudine (3TC), stavudine (d4T), TFV, and zidovudine (ZDV)]; 3 non-nucleoside reverse transcriptase inhibitors (NNRTIs) [efavirenz (EFV), nevirapine (NVP), and rilpivirine (RPV)]; 1 CCR5 receptor antagonist (maraviroc); and 1 integrase strand transfer inhibitor (raltegravir). The lower limit of detection was 2 ng/mL for 6 drugs (nelfinavir, NVP, RPV, abacavir, maraviroc, and ritonavir) and 20 ng/mL for the remaining 14 drugs.

HIV genotyping was performed using the ViroSeq HIV-1 Genotyping System v2.0 (Abbott Molecular, Des Plaines, IL) for samples with HIV viral loads ≥400 copies per milliliter. GenBank Accession numbers: KY883695-KY883762, KY888784-KY888875, and KY921717-KY921757.

Statistical Methods

Associations between ARV drug detection and individual characteristics and associations between HIV drug resistance and individual characteristics were examined using logistic regression (SAS, version 9.4; SAS Institute, Cary, NC).

Ethical Considerations

All participants provided written informed consent for participation in the HPTN 068 study. The study was approved by institutional review boards at the University of North Carolina at Chapel Hill and the University of the Witwatersrand Human Research Ethics Committee.

RESULTS

Study Cohort

HPTN 068 enrolled 2537 young women; 8 were excluded from this analysis (4 did not meet enrollment criteria; 4 had inconclusive HIV status at enrollment, Fig. 1A). At enrollment, 81 (3.2%) of the remaining 2529 were HIV-infected, and 2448 (96.8%) were HIV-uninfected. Overall, 164 women acquired HIV infection after study enrollment (Fig. 1B); 107 acquired HIV infection in the main HPTN 068 study (before their expected graduation date), and 57 acquired HIV infection in the follow-up study (after their expected graduation date).[27] We evaluated ARV drug use in 3 groups of women in this cohort: (1) those who were HIV-infected at study enrollment, (2) those who were HIV-uninfected at study enrollment, and (3) those who acquired HIV after study enrollment (seroconverters). HIV drug resistance was evaluated in women with HIV infection, at the first HIV-positive study visit.

Detection of ARV Drugs in Women at Study Enrollment

ARV drug testing was performed using enrollment samples from 2526 of the 2529 women (80 from HIV-infected women; 2446 from HIV-uninfected women, Fig. 1A). ARV drugs were detected in 10 (12.5%) of 80 samples from women who were HIV-infected at enrollment; 6 samples had 1 NNRTI with 1 or 2 NRTIs, 3 samples had 1 NNRTI alone, and 1 sample had 1 NRTI alone (Table 1). ARV drugs from other ARV drug classes were not detected. The most common NNRTI detected was EFV (n = 5), and the most common NRTI detected was 3TC (n = 7). Five of the 10 samples with ARV drugs detected had a viral load <400 copies per milliliter (Table 1). No ARV drugs were detected in samples from the 2446 HIV-uninfected women at enrollment.

Detection of ARV Drugs in Women Who Acquired HIV Infection After Study Enrollment

Overall, 164 of the women who were uninfected at enrollment acquired HIV infection during the main study or follow-up study (Fig. 1B). None of the 164 women had drugs detected at enrollment (before HIV infection). Samples were available from the first HIV-positive study visit for 162 of the 164 women. ARV drugs were detected in 16 (9.9%) of 162 samples [7 (6.5%) of 107 women during the main study; 9 (16.4%) of 55 women infected during the follow-up study, Table 1]. All 16 samples had EFV detected; 14 samples also had 1 or 2 NRTIs detected (8 had TFV+FTC, 4 had TFV+3TC, and 2 had FTC alone, Table 1). Thirteen (81.3%) of the 16 samples had viral loads <400 copies per milliliter (Table 1).

HIV Drug Resistance

HIV genotyping results were obtained for 198 (93.8%) of the 211 women who had a viral load ≥400 copies per milliliter. This included 67 women who were HIV infected at enrollment (enrollment samples were tested, Fig. 1A) and 131 women who acquired HIV infection after enrollment (the first HIV-positive sample was tested, Fig. 1B). Eight (4.0%) of the 198 samples had one or more ARV drugs detected. Major resistance mutations were identified in 18 (9.1%) of the 198 samples (Table 1). This included samples from 9 (13.4%) of the 67 women who were HIV-infected at enrollment and 9 (6.9%) of the 131 seroconverters (Table 1). At least 1 NNRTI resistance mutation was detected in all 18 cases. The most common NNRTI resistance mutations detected were K103N (n = 11), V106M (n = 3), and Y181C (n = 4). These mutations confer resistance to NVP and EFV, and Y181C also confers resistance to RPV. Major PI resistance mutations were not detected in any of the samples.

Five of the 18 women with HIV drug resistance had multiclass resistance (NNRTI + NRTI resistance); all 5 were HIV-infected at study enrollment. The following combinations of mutations were detected: K103N+M184V (n = 2), K103N +Y181C+M184V, V106M+M184I/V, and Y181C+G190A +M184V. The mutation, M184V, which was detected in all 5 cases, confers resistance to 3TC, FTC, and other NRTIs. ARV drugs were detected in all 5 cases. The viral loads in these cases ranged from 7425 to 388773 copies/mL).

Nine of the 18 women with HIV drug resistance acquired HIV infection during the HPTN 068 study; all 9 of these women had resistance to NNRTIs (7 had K103N, 1 had V106M, and 1 had Y181C). ARV drugs were only detected in 1 of the 9 samples; the remaining 8 seroconverters who did not have drugs detected may have been infected with resistant HIV strains. ARV drug testing also revealed that some of the newly infected women were at risk of acquiring additional drug resistance. One woman who had a viral load of 4188 copies per milliliter and did not have any resistance mutations was taking EFV, tenofovir disoproxil fumarate (TDF), and 3TC; a second woman who had a viral load of 766 copies per milliliter and did not have NRTI resistance mutations was taking NRTIs (TDF and FTC, along with EFV).

Factors Associated With Detection of ARV Drugs and HIV Drug Resistance

We examined the association of demographic, laboratory, and clinical factors with detection of ARV drugs and HIV drug resistance (Table 2). Not surprisingly, a significantly higher prevalence of ARV drugs was observed among women

who had a lower viral load at their first HIV-positive visit (odds ratio: 0.21, 95% confidence interval: 0.11 to 0.4; $P < 0.0001$). ARV drugs were also detected more frequently in women who had 2 deceased parents (double orphans) compared with those had 2 living parents (5/18 = 27.8% vs. 16/153 = 10.5%, $P = 0.04$). There was no significant association of ARV drug detection with age, CD4 cell count, infection group, pregnancy history, food insecurity, school attendance, depression, or alcohol use. The only factor associated with HIV drug resistance was having 1 parent deceased, compared with having 2 living parents ($P = 0.04$). The percentage of women who had viral loads <400 copies per milliliter was similar for those with 2 living parents vs. 1 or no living parents [1.7 (0.5, 6.3); $P = 0.45$].

DISCUSSION

In this study, we characterized ARV drug use and HIV drug resistance in a large cohort of young women in rural South Africa. Previous reports have described significant differences between urban and rural areas of South Africa in ART coverage, HIV prevalence among women, and the rate of acquired drug resistance among adults failing first-line ART.[33–35] This report adds to previous studies by examining these issues in school-aged girls in a rural area, by evaluating ARV drug use as well as drug resistance, and by analyzing both acquired and transmitted drug resistance.

The HPTN 068 trial enrolled young women aged 13–20 years in 2011 and 2012. Previous studies have estimated the level of ART coverage in South Africa to be 66%–81%

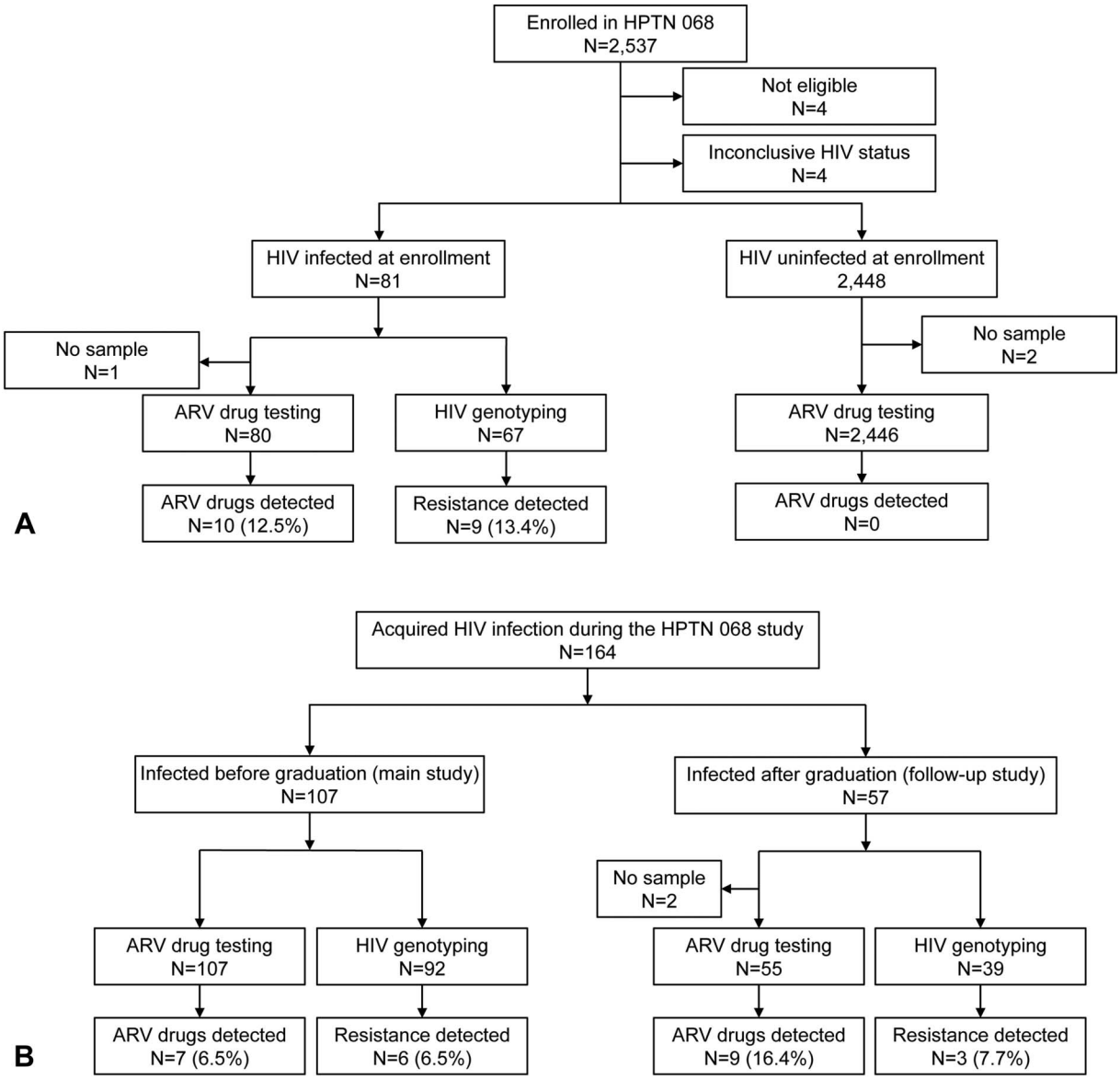

FIGURE 1. Summary of ARV drug testing and HIV resistance testing. The figure shows a summary of results for ARV drug testing and HIV resistance testing. Panel A shows results from young women who enrolled in HPTN 068. Panel B shows results for participants who acquired HIV infection after study enrollment.

TABLE 1. ARV Drugs Detected, HIV Viral Load, and HIV Drug Resistance in the Subset of HIV-Infected Participants Who Had ARV Drugs or Resistance Mutations Detected*

Status	Study Visit	ARV Drugs Detected		Viral Load	Mutations Detected	
		NNRTI	NRTI		NNRTI	NRTI
HIV-infected at enrollment (ARV drugs detected)	Enrollment	EFV	3TC, TFV	68	—	—
	Enrollment	NVP	3TC, TFV	<40	—	—
	Enrollment	NVP	3TC, ZDV	<40	—	—
	Enrollment	EFV	3TC	<40	—	—
	Enrollment	EFV	3TC	<40	—	—
	Enrollment	NVP	3TC	9968	*A98G*, *K101E*, **Y181C, G190A**	**M184V**
	Enrollment	EFV	None	164,089	**K103N,** *P225H*	**M184V**
	Enrollment	EFV	None	388,773	*K101E*, **K103N,** *P225H*	**M184V**
	Enrollment	NVP	None	7425	*A98G*, **K103N, Y181C**	**M184V**
	Enrollment	None	3TC	36,062	**V106M**	*A62V*, **M184I/V**
HIV-infected at enrollment (no drugs detected)	Enrollment	None	None	119,267	**K103N**	—
	Enrollment	None	None	73,392	**V106M**	—
	Enrollment	None	None	1263	**Y181C**	—
	Enrollment	None	None	15,225	**G190A**	—
HIV-infected after enrollment (ARV drugs detected)	Year 1	EFV	TFV, 3TC	<40	—	—
	Year 1	EFV	TFV, 3TC	475	No sample	No sample
	Year 1	EFV	TFV, 3TC	56	—	—
	Year 1	EFV	TFV, FTC	4188	—†	—†
	Year 2	EFV	None	137	—	—
	Year 3	EFV	TFV, FTC	207	—	—
	Graduation	EFV	TFV, 3TC	<40	—	—
	Post-grad	EFV	TFV, FTC	766	**K103N**	—†
	Post-grad	EFV	TFV, FTC	181	—	—
	Post-grad	EFV	TFV, FTC	71	—	—
	Post-grad	EFV	TFV, FTC	<40	—	—
	Post-grad	EFV	TFV, FTC	<40	—	—
	Post-grad	EFV	TFV, FTC	<40	—	—
	Post-grad	EFV	FTC	<40	—	—
	Post-grad	EFV	FTC	<40	—	—
	Post-grad	EFV	None	85	—	—
HIV-infected after enrollment (no drugs detected)	Year 1	None	None	22,247	**K103N**	—
	Year 2	None	None	3664	**K103N**	—
	Year 2	None	None	8860	*V179E*, **Y181C**	*D67N*, *K70E*
	Year 3	None	None	10,159	**K103N**	—
	Year 3	None	None	94,359	**K103N**	—
	Graduation	None	None	91,235	**V106M**	—
	Post-grad	None	None	11,872	**K103N**	—
	Post-grad	None	None	50,037	**K103N**	—

*The table shows laboratory data from the subset of 38 HIV-infected participants who had one or more ARV drugs detected or had one or more HIV drug resistance mutations detected. This included 14 participants who were HIV-infected at study enrollment (10 with ARV drugs detected; 4 with no ARV drugs detected) and 24 participants who acquired HIV infection after enrollment (16 with ARV drugs detected; 8 with no ARV drugs detected). Samples were collected at enrollment or at the first HIV-positive visit (study visit). Years 1, 2, and 3 indicate the visit type (years after enrollment). Graduation indicates a visit that occurred at the expected graduation date. Post-grad indicates a visit that occurred after 1–2 years after the expected graduation date. HIV genotyping was performed for the 19 samples that had HIV viral loads ≥400 copies per milliliter; 1 sample was not available for testing. Major resistance mutations (shown in bold) were detected in 18 (94.7%) of the 19 samples with genotyping results. NNRTI and NRTI resistance mutations are shown; PI resistance mutations were not detected. Accessory mutations are shown in italics. Viral load data are shown as copies per milliliter.

†These women were at risk of having additional resistance mutations.

(Joint United Nations Programme survey; 2011, 2013),[36,37] 57.8% (women aged 15–59 years, population survey in KwaZulu-Natal that included ARV drug testing; 2013)[38] and 31.7% (aged 15–32 years, population survey in Kwazulu-Natal; 2009–2011).[24] In HPTN 068, ARV drugs were only detected in 12.5% of the participants who were infected at enrollment and 9.9% of the seroconverters. The lower frequency of ARV drug use in the HPTN 068 cohort may reflect lack of knowledge of HIV status, lower adherence to ART among adolescents and young adults, less access to ART in the study communities, or other factors.[27]

At the time HPTN 068 was conducted, the recommended first-line ART regimen included 2 NRTIs (TDF+3TC or FTC) and a NNRTI (EFV or NVP).[39] In HPTN 068, a NNRTI

TABLE 2. Association of Demographic and Other Factors With ARV Drug Use and HIV Drug Resistance Among HIV-Infected Women*

	Detection of ARV Drugs				HIV Drug Resistance			
Characteristics	**Yes (N = 26)**	**No (N = 216)**	**OR (95% CI)**	***P***	**Yes (N = 18)**	**No (N = 180)**	**OR (95% CI)**	***P***
Age (year)†‡	18.5 (16, 21)	19 (17, 20)	0.99 (0.79 to 1.24)	0.93	17.5 (16, 19)	19 (17, 20)	0.79 (0.61 to 1.01)	0.06
Viral load (log_{10})†‡	1.8 (1.6, 2.9)	4.2 (3.7, 4.7)	0.21 (0.11 to 0.4)	**<0.0001**	4.3 (4.0, 4.9)	4.3 (3.9, 4.8)	1.09 (0.54 to 2.21)	0.80
CD4 cell count ($cell/mm^3$)†‡	484.5 (353, 754)	559 (400, 573)	0.98 (0.82 to 1.18)	0.86	414.5 (313.5, 665.5)	534 (383.5, 731)	0.88 (0.7 to 1.11)	0.29
First HIV-positive visit								
Study enrollment	10 (12.5%)	70 (87.5%)	Ref		9 (13.4%)	58 (86.6%)	Ref	
Main study	7 (6.5%)	100 (93.5%)	0.49 (0.18 to 1.35)	0.17	6 (6.5%)	86 (93.5%)	0.45 (0.15 to 1.33)	0.15
Follow-up study	9 (16.4%)	46 (83.6%)	1.37 (0.52 to 3.63)	0.53	3 (7.7%)	36 (92.3%)	0.54 (0.14 to 2.12)	0.37
Ever pregnant§								
Yes	6 (12.8%)	41 (87.2%)	1.27 (0.48 to 3.35)	0.64	2 (5.1%)	37 (94.9%)	0.48 (0.1 to 2.16)	0.34
No	20 (10.4%)	173 (89.6%)	Ref		16 (10.2%)	141 (89.8%)	Ref	
Missing	—	2 (100.0%)			—	2 (100.0%)		
Orphan§								
Both parents alive	16 (10.5%)	137 (89.5%)	Ref		7 (5.8%)	113 (94.2%)	Ref	
Single orphan	2 (3.6%)	54 (96.4%)	0.32 (0.07 to 1.43)	0.13	8 (15.7%)	43 (84.3%)	3.00 (1.03 to 8.79)	**0.04**
Double orphan	5 (27.8%)	13 (72.2%)	3.29 (1.04 to 10.44)	**0.04**	3 (18.8%)	13 (81.2%)	3.73 (0.86 to 16.19)	0.08
Missing data	3 (20.0%)	12 (80.0%)			—	11 (100.0%)		
Food insecurity§								
Yes	13 (12.6%)	90 (87.4%)	1.37 (0.6 to 3.09)	0.45	8 (9.6%)	75 (90.4%)	1.23 (0.45 to 3.34)	0.68
No	13 (9.6%)	123 (90.4%)	Ref		9 (8.0%)	104 (92.0%)	Ref	
Missing data	—	3 (100.0%)			1 (50.0%)	1 (50.0%)		
School days missed per month§								
0–2 days	20 (11.5%)	154 (88.5%)	Ref		13 (9.4%)	125 (90.6%)	Ref	
≥3 days	6 (10.2%)	53 (89.8%)	0.87 (0.33 to 2.29)	0.78	5 (9.8%)	46 (90.2%)	1.05 (0.35 to 3.09)	0.94
Missing data	—	9 (100.0%)			—	9 (100.0%)		
Depression§‖								
Yes	9 (13.2%)	59 (86.8%)	1.30 (0.55 to 3.08)	0.55	5 (9.1%)	50 (90.9%)	1.10 (0.36 to 3.33)	0.87
No	17 (10.5%)	145 (89.5%)	Ref		11 (8.3%)	121 (91.7%)	Ref	
Missing data	—	12 (100.0%)			2 (18.2%)	9 (81.8%)		
Alcohol use¶								
Never or less than once a month	23 (10.4%)	198 (89.6%)	Ref		17 (9.4%)	163 (90.6%)	Ref	
Once a month or more	3 (14.3%)	18 (85.7%)	1.43 (0.39 to 5.25)	0.59	1 (5.6%)	17 (94.4%)	0.56 (0.07 to 4.5)	0.59

*The table shows the association of demographic and other factors associated with ARV drug detection and HIV drug resistance. $P < 0.05$ is bolded.
†Median (interquartile range).
‡These variables were assessed at the first HIV-positive visit.
§These variables were assessed at the enrollment visit.
‖Depression was measured using the Short Form Children's Depression Index (CDI, 10 items). A cut-off of 7 was used, where an index ≥7 or above is depressed, and an index <7 is not depressed.
¶This variable was assessed during follow-up (from enrollment through the expected graduation date).
CI, confidence interval; N, number; OR, odds ratio; ref, reference.

(EFV or NVP) alone or in combination with 1 or 2 NRTIs (TFV, 3TC, d4T, and ZDV) was detected in 9 of 10 participants who had ARV drugs detected at enrollment and in all seroconverters who had ARV drugs detected at their first HIV-positive study visit. Because NRTIs have shorter half-lives compared with NNRTIs, detection of NRTIs may depend on the time between drug dosing and sample collection; we considered detection of an NNRTI alone to be consistent with ART. In the 2 cases where EFV was detected alone, it is also possible that the drug was being used for recreational purposes.[13,14] The detection of 3TC alone in 1 woman suggests that she was not adherent to an ART regimen or that she was using the drug for another reason (eg, treatment of hepatitis B virus infection).

We also assessed ARV use among >2400 young women in HPTN 068 who were HIV-uninfected. In a previous report, we found unusual patterns of ARV drug use among HIV-uninfected women in the United States (EFV and/or a PI).[23] ARV drugs were not detected in any of the HIV-uninfected young women in the HPTN 068 cohort; of note, the study was performed before the approval of PrEP in South Africa.[8]

In HPTN 068, HIV drug resistance was detected in samples from 13.4% of the participants infected at enrollment and 6.9% of the seroconverters. A subset of the participants with drug resistance did not have ARV drugs detected (6.0% infected at enrollment; 6.1% of the seroconverters). Some of these participants may have taken ARV drugs in the past, or between the annual study visits. Therefore, these numbers represent a maximal estimate of transmitted drug resistance. The estimated level of transmitted drug resistance among seroconverters is categorized as moderate based on the World Health Organization (WHO) guidelines.[40] In a national survey conducted in South Africa, the prevalence of pretreatment drug resistance was 9% among adults aged 18 years and older (7.4% in Mpumalanga Province).[41] In a study conducted in KwaZulu-Natal where most of the participants were women, the prevalence of pretreatment resistance was 8.7%.[42] In HPTN 068, 5 young women had multiclass drug resistance at study enrollment. ARV drugs were detected in samples from all 5 young women, suggesting that the multiclass resistance was acquired because of ARV drug exposure.

In the HPTN 068 cohort, 30% of the young women who were taking ARV drugs were not virally suppressed. In a separate study, we estimated that 12.6% of the young women in HPTN 068 were viremic controllers; those young women maintained viral loads <2000 copies per milliliter for at least 12 months in the absence of ARV drug use. In this study, 13 (41.9%) of the 31 young women who had viral loads <400 copies per milliliter at their first HIV-positive visit did not have ARV drugs detected; this indicates that low viral load is a poor surrogate marker for ARV drug use in this cohort.[43]

In the HPTN 068 study, double orphans were at significantly higher risk of HIV infection than those with 2 living parents.[26] In this study, we also found that double orphans were more likely to be taking ARV drugs than women with 2 living parents; the reason for this difference is not clear. We also found that single orphans were more likely to have HIV drug resistance than women with 2 living parents; a similar trend was seen for double orphans, but this difference was not statistically significant. Previous studies have found that better relationships between caregivers and orphaned children improved the adherence to ART.[44,45]

The findings from this study indicate a need for improvements in HIV care. In this cohort of young women in rural South Africa, ARV drug use was relatively infrequent among young women with both prevalent and new HIV infection. Because HPTN 068 did not collect information about the availability and uptake of HIV testing in these communities, it was not possible to determine whether the low rate of ARV drug use in this cohort reflected lack of knowledge of HIV status, limited access to ART, lack of interest in HIV treatment, or other factors. Among the young women who were using ARV drugs, many were not virally suppressed, and many had HIV drug resistance. This indicates a need for expanded programs for HIV/AIDS education and counseling to improve ART adherence. The relatively high frequency of drug resistance among young women with new HIV infection who were not using ARV drugs at their first HIV-positive study visit also suggests a need for broader HIV/AIDS education and ART counseling in the study communities, to reduce the rate of transmitted drug resistance.

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