HIV Drug Resistance Assessment Among Women Who Seroconverted During the MTN-025/HOPE Open-Label Extension Dapivirine Vaginal Ring Trial

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Background: Clinical trials of dapivirine (DPV) vaginal ring have shown it is safe, effective, and desired by women as an HIV prevention option. The risk of drug resistance is a potential concern for DPV ring users who acquire HIV. We conducted a comprehensive resistance evaluation of plasma samples from the women who seroconverted during the Microbicide Trials Network-025/HIV Open-label Prevention Extension (HOPE) study of DPV ring.

Methods: Plasma collected on the visit at which seroconversion was detected was tested by next-generation sequencing with unique molecular identifiers for non-nucleoside reverse transcriptase inhibitor (NNRTI) drug resistance mutations (DRM) present at ≥1% frequency. Bulk-cloned plasma-derived recombinant HIV was phenotyped in a TZM-bl–based assay for susceptibility to DPV and other NNRTI. HIV-1 RNA was retrospectively quantified in plasma samples collected before HIV seroconversion.

Results: Among 38 participants who seroconverted in HOPE, 7 (18%) had NNRTI DRM detected by next-generation sequencing with unique molecular identifiers including A98G, K103N, V106M, E138A, and V179D. Six of 7 samples with NNRTI DRM had <3-fold reduction in susceptibility to DPV. Only 1 sample with K103N and V179I polymorphism had 9-fold reduction in susceptibility to DPV, but this genotype occurred in an individual who did not use DPV ring, likely indicating transmitted resistance. Detection of NNRTI resistance was not higher in individuals who remained on DPV ring >3 months after acquiring HIV infection.

Conclusions: NNRTI resistance among women who seroconverted during HOPE was infrequent and selection of DPV-specific mutations was not detected. DPV ring is considered a safe and effective option for HIV prevention in women.

Key Words: HIV-1, HIV prevention, pre-exposure prophylaxis, dapivirine ring, HIV drug resistance

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INTRODUCTION

The number of incident HIV infections in African women and girls continues to exceed Joint United Nations Programme on HIV/AIDS targets for new HIV infections globally, with major setbacks in HIV response in the past 2 years due to disruptions from COVID-19, emphasizing the continued need for diverse HIV prevention options.[1,2] A silicone elastomer intravaginal ring containing 25 mg of the non-nucleoside reverse transcriptase inhibitor (NNRTI) dapivirine (DPV) is the first topical product approved for HIV prevention in women and was recommended for use by the World Health Organization in January 2021 after a positive scientific opinion from the European Medicines Agency in July 2020.[3] DPV vaginal ring has received regulatory approval in several African countries with reviews in progress in others. DPV ring has broad desirability, acceptance, and safety in women throughout their reproductive life. A recent demonstration study in Zimbabwe showed that most of the participants aged 18–25 years accepted DPV ring (81%) compared with oral PrEP (19%), especially from rural areas.[4] Development of multipurpose rings that include levonorgestrel with DPV is underway to provide both HIV prevention and contraception from a single device.[5–10]

Two Phase III trials (the Microbicide Trials Network [MTN]-020/ASPIRE study and the International Partnership for Microbicides [IPM] 027/Ring Study) and 2 open-label

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follow-on studies (MTN-025/HIV Open-label Prevention Extension (HOPE) and IPM 032/DREAM) demonstrated the efficacy of DPV ring in reducing the risk of HIV infection among African women. NNRTI resistance frequency among HIV seroconverters across the 4 studies ranged from 10% to 28%. The frequency of individual NNRTI drug resistance mutations (DRM) did not differ in the placebo and DPV ring arms indicating that NNRTI resistance was likely transmitted rather than selected by DPV ring. The only exception was in the Ring Study, where E138A was detected more frequently in the DPV arm (10.7% DPV vs 3.4% placebo); however, the difference in proportion was not statistically significant (P = 0.2).[11–15]

DPV has never been used clinically for HIV treatment; thus, its resistance profile has predominantly been derived from in vitro selection experiments and cell-based drug susceptibility assays. These studies have identified common NNRTI mutations including V90I, L100I, K103N, V106I, E138K, Y181C, and Y188L to be associated with DPV resistance or cross-resistance.[16,17] Because DPV is solely used for HIV prevention, mutation scores or resistance interpretations for DPV are not available in routinely used HIV drug resistance databases.[18,19]

The high rate of NNRTI resistance in individuals who seroconverted in HOPE (20%) in the context of increasing rates of pretreatment NNRTI resistance in countries with planned DPV ring introduction remains a threat to the success of DPV ring rollout.[13,20] To better understand resistance risk with seroconversion on DPV ring, we conducted an in-depth evaluation of HIV infections in HOPE for NNRTI resistance including high-sensitivity, next-generation genotype analyses and phenotypic susceptibility of plasma-derived recombinant viruses against a panel of NNRTIs.

METHODS

Study Samples

MTN-025/HOPE was a Phase IIIB study of DPV ring conducted between August 2016 and October 2018 at 14 clinical research sites in Malawi, South Africa, Uganda, and Zimbabwe, as previously described.[13] This study enrolled 1456 women who had remained HIV negative after participating in the Phase III double-blinded placebo-controlled MTN-020/ASPIRE study and offered them access to DPV ring for 12 months, with the option to participate in the study while declining ring use.[12] DPV ring is not approved by the US Food and Drug Administration. HOPE (NCT02858037) was approved by the institutional review board at each site, and all participants provided written informed consent. Participants were tested for HIV monthly for the first 3 months, then quarterly for the remainder of the study using 2 concurrent HIV rapid tests, with confirmation of positive results by the Geenius HIV-1/2 Supplemental Assay (Bio-Rad). HIV-1 RNA PCR (Roche TaqMan or Abbott M2000) was retrospectively performed on all samples from the first positive rapid through quarterly collected preseroconversion stored samples until an undetectable HIV RNA result was obtained. The minimum length of time that a participant was potentially on product during acute HIV-1 infection was estimated using the time frame between last undetectable HIV RNA result and first positive rapid test result. This study included 38 participants who seroconverted on HOPE, including 35 participants who became HIV positive after enrollment and 3 who enrolled into HOPE with undetected acute HIV infection. All testing described in this study was conducted on archived plasma samples collected on the visit at which seroconversion was detected.

Assessment of DPV Ring Adherence

Residual DPV in returned rings was measured as a proxy for DPV ring adherence using acetone extraction and high-pressure liquid chromatography (Parexel, Bloemfontein, South Africa), as previously described.[21]

Evaluation of HIV Drug Resistance by Next-Generation Sequencing and NNRTI Susceptibility Testing

Plasma collected at the study visit at which HIV seroconversion was confirmed was analyzed for HIV-1 mutations in reverse transcriptase targeting amino acids 81–149 and 152–212 with a laboratory-developed Illumina Miseq next-generation sequencing (NGS) assay using unique molecular identifier (UMI)–tagged HIV cDNA to remove PCR bias using bioinformatics tools and reduce sequencing error while providing precise quantification of HIV templates sampled during library preparation, as previously reported.[22–24] HIV drug resistance mutations and HIV-1 subtypes were identified using the Stanford Genotypic Resistance Interpretation Algorithm v9.0.[25] Plasma-derived full-length reverse transcriptase from participants who became seropositive in HOPE was bulk-cloned into HIV-1_{xxLAI} and the recombinant virus was assayed for NNRTI susceptibility in TZM-bl cells to calculate fold-change in 50% inhibitory concentration (IC_{50}), as previously described.[24,26] DPV was kindly provided by International Partnership for Microbicides (Silver Spring, MD). Nevirapine (NVP), efavirenz (EFV), etravirine (ETR), and rilpivirine (RPV) were obtained through the AIDS Research and Reference Reagent Program, Division of AIDS, NIAID, and NIH.

Statistical Analysis

Four-parameter, nonlinear regression for curve fitting was used to generate NNRTI IC_{50} values using GraphPad Prism 9 software (GraphPad Software, Inc; Boston, MA). Fold-change was calculated as IC_{50} of mutant HIV-1/wild-type HIV-1.

RESULTS

NNRTI Resistance Mutation Detection by Next-Generation Sequencing

All 38 samples (100%) from women who seroconverted during the HOPE study (including 3 who enrolled with acute HIV infection) were successfully sequenced by UMI-NGS

TABLE 1. Plasma HIV-1 RNA Levels, HIV-1 Genotype, Phenotype, and Adherence Levels of Individuals Who Seroconverted in the MTN-025/HOPE DPV Vaginal Ring Open-Label Extension Study

PID	Plasma HIV-1 RNA Copies/mL at Seroconversion	Range (min, max) in days of RNA Positive Preseroconversion Ring Exposure*	HIV-1 Reverse Transcriptase Genotype by Next-Generation Sequencing with % Mutant Frequency	# Of UMI (% Frequency Detection Limit)†	Mean DPV IC_{50} ± SD (Fold-Change)‡	Average DPV (mg) Released in Past 1–3 Months from Used Rings (Adherence Interpretation)
1	280,868	1, 84	Wild type	16,458 (1)	0.42 ± 0.02 (0.5)	3.3 (Inconsistent to Near-Consistent Use)
2	18,280	82, 160	Wild type	894 (1)	0.95 ± 0.09 (1.1)	No Ring Collected
3	1170	27, 122	Wild type	135 (5)	0.37 ± 0.01 (0.4)	5.3 (28 Days Consistent Use)
4	47,362	1, 83	Wild type	1006 (1)	0.64 ± 0.07 (0.8)	3.4 (Inconsistent to Near-Consistent Use)
5	6294	1, 89	Wild type	1373 (1)	0.56 ± 0.07 (0.7)	3.4 (Inconsistent to Near-Consistent Use)
6	11,120	31, 101	Wild type	743 (1)	0.59 ± 0.07 (0.7)	No Ring Collected
7	869,098	1, 35	Wild type	7672 (1)	0.44 ± 0.005 (0.5)	3.0 (Inconsistent to Near-Consistent Use)
8	21,122	1, 83	Wild type	2513 (1)	0.62 ± 0.08 (0.7)	No Ring Collected
9	1538	73, 157	Wild type	201 (5)	0.34 ± 0.02 (0.4)	1.1 (Inconsistent to Near-Consistent Use)
10	93,325	29, 121	Wild type	4884 (1)	0.54 ± 0.13 (0.6)	No Ring Collected
11	7993	91, 120	Wild type	393 (1)	1.24 ± 0.32 (1.5)	5.0 (28 Days Consistent Use)
12	95,169	1, 88	Wild type	7587 (1)	0.96 ± 0.12 (1.1)	5.2 (28 Days Consistent Use)
13	6110	1, 79	Wild type	636 (1)	0.74 ± 0.08 (0.9)	3.3 (Inconsistent to Near-Consistent Use)
14	21,616,060	No data	Wild type	11,683 (1)	0.50 ± 0.08 (0.6)	4.6 (28 Days Consistent Use)
15	3,225,929	1, 28	Wild type	15,000 (1)	0.78 ± 0.10 (0.9)	5.0 (28 Days Consistent Use)
16	4,011,745	1, 91	Wild type	29 (10)	0.58 ± 0.10 (0.7)	3.6 (Inconsistent to Near-Consistent Use)
17	1,774,522	1, 64	Wild type	47 (10)	0.41 ± 0.02 (0.5)	5.4 (28 Days Consistent Use)
18	47,700	1, 85	Wild type	1596 (1)	0.39 ± 0.03 (0.5)	3.6 (Inconsistent to Near-Consistent Use)
19	12,500	166, 250	Wild type	110 (5)	0.69 ± 0.03 (0.8)	2.1 (Inconsistent to Near-Consistent Use)
20	808,000	27, 110	Wild type	10,280 (1)	0.68 ± 0.06 (0.8)	No Ring Collected
21	317	1, 84	Wild type	36 (10)	0.32 ± 0.02 (0.4)	No Ring Collected
22	2407	89, 167	Wild type	729 (1)	0.53 ± 0.15 (0.6)	1.3 (Inconsistent to Near-Consistent Use)
23	2845	1, 78	Wild type	486 (1)	No result	No Ring Collected
24	1,620,792	1, 28	Wild type	17,500 (1)	0.55 ± 0.09 (0.7)	0.3 (Inconsistent to Near-Consistent Use)
25	250,360	83, 167	Wild type	18,239 (1)	0.27 ± 0.04 (0.3)	1.8 (Inconsistent to Near-Consistent Use)
26	10,897	27, 61	Wild type	2502 (1)	1.04 ± 0.17 (1.2)	No Ring Collected
27	1,630,000	1, 84	Wild type	14,686 (1)	0.47 ± 0.09 (0.6)	1.9 (Inconsistent to Near-Consistent Use)

(*continued on next page*)

TABLE 1. (*Continued*) Plasma HIV-1 RNA Levels, HIV-1 Genotype, Phenotype, and Adherence Levels of Individuals Who Seroconverted in the MTN-025/HOPE DPV Vaginal Ring Open-Label Extension Study

PID	Plasma HIV-1 RNA Copies/mL at Seroconversion	Range (min, max) in days of RNA Positive Preseroconversion Ring Exposure*	HIV-1 Reverse Transcriptase Genotype by Next-Generation Sequencing with % Mutant Frequency	# Of UMI (% Frequency Detection Limit)†	Mean DPV IC_{50} ± SD (Fold-Change)‡	Average DPV (mg) Released in Past 1–3 Months from Used Rings (Adherence Interpretation)
28	1,010,000	1, 25	Wild type	15,557 (1)	0.77 ± 0.18 (0.9)	4.5 (28 Days Consistent Use)
29	4200	1, 83	100% K103N	216 (5)	2.22 ± 0.65 (2.6)	5.0 (28 Days Consistent Use)
30	2061	1, 62	100% E138A, 100% V179D	11 (>20)	1.31 ± 0.04 (1.6)	2.8 (Inconsistent to Near-Consistent Use)
31	407	126, 153	100% A98G	131 (5)	0.74 ± 0.03 (0.9)	4.3 (Inconsistent to Near-Consistent Use)
32	2,751,104	1, 85	100% K103N	4105 (1)	2.36 ± 0.17 (2.8)	2.6 (Inconsistent to Near-Consistent Use)
33	75,993	1, 94	87% K103N§	2940 (1)	7.94 ± 0.60 (9.4)	Did Not Receive Ring
34	36,976	27, 111	100% V106M, 100% V179D	1435 (1)	1.10 ± 0.26 (1.3)	No Ring Collected
35	84,167	1, 83	100% K103N	18,492 (1)	0.66 ± 0.11 (0.8)	4.1 (Inconsistent to Near-Consistent Use)
36	15,232	AHI at enrollment	Wild type	3218 (1)	0.41 ± 0.04 (0.5)	No Ring Use
37	9,903,949	AHI at enrollment	Wild type	15,000 (1)	1.04 ± 0.16 (1.2)	1.1 (Inconsistent to Near-Consistent Use)
38	1421	AHI at enrollment	Wild type	730 (1)	0.38 ± 0.03 (0.4)	No Ring Collected

*The minimum length of time a participant was on product before seroconversion was estimated for each participant by proximity of the last positive viral load result to the visit in which seroconversion was detected. Because HIV-1 infection could occur at any time before the visit in which HIV-1 RNA was detected, the maximum amount of time a participant may have been on product before seroconversion includes all days up to the last negative viral load result.

†% Frequency detection limit was determined based on the number of unique UMI barcode present in the sample. For 95% confidence, 298, 54, and 29 UMI consensus sequences were required to call 1%, 5%, and 10% minor variants, respectively.

‡FC was calculated as IC_{50} of plasma-derived recombinant HIV-$_{1xxLAI}$/wild-type batch control HIV-$_{1xxLAI}$ (IC_{50} 0.84 nM; n = 29).

§Participant 33 also had V179I, a polymorphism that is listed as a major nucleoside reverse transcriptase inhibitor mutation by Stanford Genotypic Resistance Interpretation Algorithm v9.0.[25]

AHI, acute HIV infection; FC, fold-change; PID, participant identifier; UMI, unique molecular identifier.

with most of the participants (76%) having a limit of detection for the frequency of DRM in the virus population of ≥1%. The limit of detection for the other 24% ranged from 5 to >20% (Table 1). Of the 38 samples, 31 (82%) had HIV-1 with no detectable NNRTI DRM (ie, <1% mutant frequency) and 7 (18%) had NNRTI DRM detected by NGS-UMI, including A98G, K103N, V106M, E138A, and V179D. All DRM detected were present at >20% frequency. Major NNRTI DRM selected by DPV in vitro, including L100I, E138K, V179F, and Y181C/I were not detected. No correlation between drug resistance and ring use was noted. Of the 7 participants with NNRTI resistance, one did not use DPV ring, one did not return rings for the 3 months before seroconversion, 2 had DPV levels indicating inconsistent use of DPV ring, and 3 had DPV ring levels indicating 28 days of continuous use during at least 1 of the last 3 months. Of the 31 participants with wild-type HIV-1, 11 (35%) reported no ring use, 13 (42%) had inconsistent ring use as estimated by residual DPV ring levels, and 7 (23%) had continuous ring use as estimated by residual DPV ring levels. No NNRTI DRM (≥1% frequency) was detected in the 3 participants who enrolled into HOPE with acute HIV infection.

Risk of Resistance With Delayed Detection of Seroconversion

Preseroconversion HIV RNA was used to approximate the duration of ring exposure during acute HIV infection and before seroconversion was detected by rapid antibody testing for the 35 of 38 participants who were HIV RNA undetectable at HOPE enrollment. At seroconversion, all participants had detectable HIV RNA ranging from 317 to 21.6 million copies (cp)/mL (median 42,169 cp/mL). For most of the HOPE participants (21 of 35, 60%), seroconversion was detected within 3 months of first detectable HIV RNA and 5 of these 21 participants (24%) had NNRTI resistance. Only 13 of 35 participants (40%) had detectable HIV-1 RNA more than 3 months before seroconversion, and 2 of these 13 (15%) had NNRTI resistance. The risk of resistance was not greater in those with delayed detection of seroconversion ($P > 0.01$, χ^2).

Susceptibility of Plasma-Derived HIV-1 to NNRTIs

Susceptibility testing of participants' plasma samples was successfully completed (N = 37) except 1 sample from

a participant who never received DPV ring. For the 30 samples including the 3 from individuals with acute infection at enrollment without known NNRTI DRM, susceptibility against DPV remained similar to a wild-type control HIV-1 (0.3–1.5 fold-change [FC]), suggesting that no novel mutations causing reduced susceptibility to DPV were present. Similarly, participant-derived HIV-1 without known NNRTI DRM remained susceptible to ETR (mean FC 0.6), RPV (mean FC 0.7), NVP (mean FC 1.3), and EFV (mean FC 1.1).

Of the 7 samples with NNRTI DRM, only 1 (87% K103N, 75% V179I polymorphism) had reduced susceptibility to DPV (9.4-FC) compared with a wild-type control. This participant never received DPV ring. Two samples with 100% K103N had only low-level decreases in susceptibility to DPV (2.6 and 2.8-FC) while 1 sample with only 100% K103N had no change in DPV susceptibility (0.8-FC). The remaining 3 samples (with genotypes E138A/V179D, A98G, and V106M/V179D all at 100% frequency) had no change in DPV susceptibility relative to wild-type HIV-1 (0.9-FC to 1.6-FC). Samples with known NNRTI DRM had expected reductions in susceptibility to currently used NNRTIs, including K103N to NVP (24–210-FC) and EFV (5–38-FC). The V106M/V179D genotype conferred resistance to both NVP and EFV (Fig. 1).

DISCUSSION

We undertook a comprehensive assessment of the 38 seroconversions in the HOPE open-label study of DPV ring and found infrequent NNRTI resistance. However, the overall rate of resistance observed among participants becoming HIV seropositive (20%) was higher than that observed in the DPV ring arm of the Phase III ASPIRE trial (10.4%),[12,24] but 20% is consistent with the contemporaneous national NNRTI pretreatment drug resistance rates of 10%–30% in Uganda, South Africa, and Zimbabwe as reported in the 2019 World Health Organization HIV Drug Resistance Report.[27] That the frequency of NNRTI resistance in HOPE participants who became HIV seropositive is similar to that of circulating NNRTI resistance in the community argues for transmitted drug resistance in HOPE participants and against selection of resistance by DPV. This thesis is further supported by the Phase III ASPIRE trial in which the frequency of NNRTI resistance was not higher in the DPV arm than the placebo arm. The findings from the current HOPE study are also consistent with those of the parallel, open-label IPM 027 study, which also found infrequent NNRTI resistance with ring use (15.5% NNRTI resistance in the DPV arm; 13.8% in the placebo arm).[15]

Delayed seroconversion and selection of resistance can be a risk for individuals who become infected with HIV while using pre-exposure prophylaxis (PrEP) for HIV prevention. HIV drug resistance selected from the use of PrEP could affect success of future treatment or could be transmitted to a partner and contribute to rising community resistance prevalence.[28] In the HPTN-084 trial of long-acting injectable cabotegravir (CAB-LA) as PrEP, 44% of infections in the CAB-LA arm developed mutations in the integrase gene, likely due to delayed detection of infection with ongoing low-level replication that enabled selection of resistance.[29–31] By contrast, only 2 of 13 individuals (15%) who continued ring use for at least 3 months after HIV infection had NNRTI resistance mutations and neither had decreases in viral susceptibility to DPV. These observations again suggest that the NNRTI mutations were likely transmitted by a partner at the time of infection and not selected for specific resistance to DPV.

The level of ring adherence among participants, estimated by the amount of residual DPV in the 3 most recently returned rings before seroconversion, was associated with NNRTI resistance, although the number of participants with resistance was small (n = 7). Higher adherence is associated with greater protection from DPV ring, and this may be due to high levels of DPV locally in the vagina with limited systemic DPV exposure, thus also limiting the potential for drug-selective pressure.[21,32] Individuals who

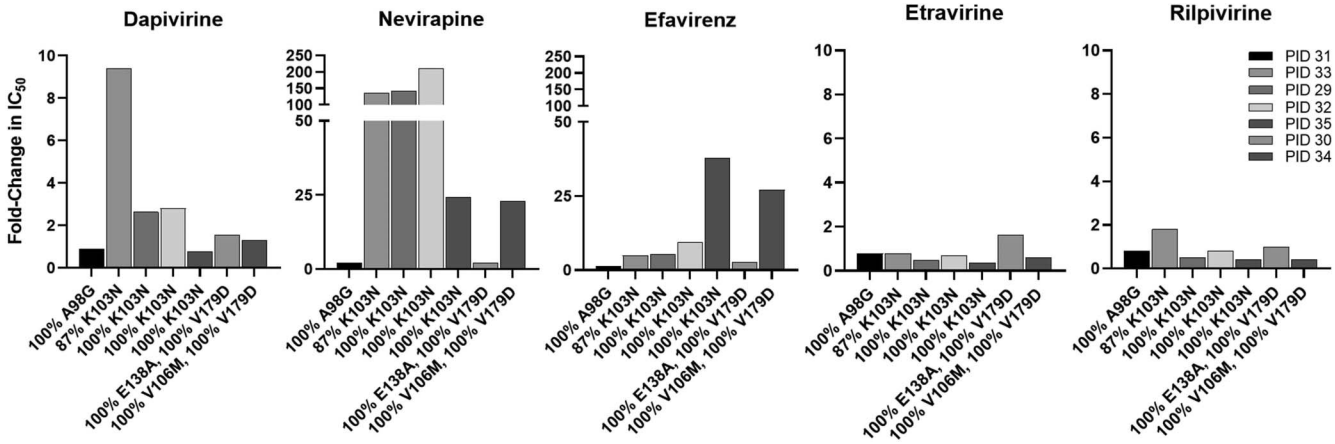

FIGURE 1. Fold-change in IC_{50} of plasma-derived recombinant HIV-1 containing non-nucleoside drug resistance mutations from individual participants who seroconverted in the HOPE trial in comparison with wild-type controls. Determined in TZM-bl cells against dapivirine, nevirapine, efavirenz, etravirine, and rilpivirine. Each bar represents a different participant with the genotype listed below the bar.

used the ring for greater than 3 months before seroconversion did not have a greater risk of resistance compared with individuals who had seroconversion detected within 3 months of first positive HIV RNA. Thus, the risk of NNRTI resistance selection in plasma may be minimal despite infection and prolonged use of ring before detection of seroconversion.

Of the 7 seroconversions with NNRTI resistance, all mutations detected by UMI-NGS were present at high frequency and consistent with mutations detected by standard virus population sequencing.[13] Reduced DPV susceptibility was only observed in 1 participant with the NNRTI mutation K103N present in combination with a V179I polymorphism. V179I in combination with the E138A HIV-1 subtype C polymorphism also confers higher levels of resistance in vitro.[24]

Taken together, the results of the HOPE study provide strong evidence that DPV ring is a safe HIV prevention option that offers several unique benefits. With limited systemic exposure and low risk of resistance with seroconversion, implementation of DPV ring could occur with current HIV testing strategies including self-testing to enable demedicalized implementation of an efficacious HIV prevention product. The consequences of transmitted NNRTI resistance, if it occurs, can be lessened with the expansion of tenofovir/lamivudine/dolutegravir (DTG) first-line antiretroviral therapy, which is highly active against NNRTI-resistant HIV-1. Nevertheless, ongoing and updated surveillance of pretreatment HIV drug resistance will be essential in regions with high circulating rates of NNRTI resistance to assess any potential impact on efficacy of DPV ring.

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REFERENCES

1. The Joint United Nations Programme on HIV/AIDS (UNAIDS). *UNAIDS Data 2021. [JC3032E]*. 29 November 2021 ed; 2021. Accessed June 01, 2022.
2. UNAIDS. *IN DANGER: UNAIDS Global AIDS Update 2022. Licence: CC BY-NC-SA 3.0 IGO*; 2022. Accessed July 19, 2023.
3. World Health Organization (WHO). *Guidelines: Updated Recommendations on HIV Prevention, Infant Diagnosis, Antiretroviral Initiation and Monitoring*; 2021. Published March 2021. Available at: https://apps.who.int/iris/bitstream/handle/10665/341052/9789240023581-eng.pdf?sequence=1&isAllowed=y. Accessed June 01, 2022.
4. Mavudze J, Moga T, Moyo I, et al. *HIV Sero-Conversions Among Adolescent Girl and Young Female Dapivirine Vaginal Ring (DPV-R) Users: Early Learnings from a Demonstration Project in Zimbabwe*. Abstract 2. 2023.
5. Shapley-Quinn MK, Laborde N, Luecke E, et al. Acceptability of the dapivirine vaginal ring in postmenopausal US women. *AIDS Patient Care STDs*. 2022;36:97–105.
6. Reddy K, Mathebula F, Katz A, et al. Ring-ing in the future: participant and male partner perspectives regarding future use of the dapivirine vaginal ring for HIV prevention. *AIDS Behav*. 2022;26:1923–1932.
7. Mayo AJ, Browne EN, Montgomery ET, et al. Acceptability of the dapivirine vaginal ring for HIV-1 prevention and association with adherence in a Phase III trial. *AIDS Behav*. 2021;25:2430–2440.
8. Roberts ST, Hawley I, Luecke E, et al. Acceptability and preference for 3-month versus 1-month vaginal rings for HIV-1 risk reduction among participants in a Phase 1 trial. *J Womens Health*. 2022;31:1029–1039.
9. Dallal Bashi YH, Murphy DJ, McCoy CF, et al. Silicone elastomer formulations for improved performance of a multipurpose vaginal ring releasing dapivirine and levonorgestrel. *Int J Pharm: X*. 2021;3:100091.
10. Murphy DJ, Dallal Bashi YH, McCoy CF, et al. In vitro drug release, mechanical performance and stability testing of a custom silicone elastomer vaginal ring releasing dapivirine and levonorgestrel. *Int J Pharm: X*. 2022;4:100112.
11. Nel A, van Niekerk N, Kapiga S, et al. Safety and efficacy of a dapivirine vaginal ring for HIV prevention in women. *N Engl J Med*. 2016;375:2133–2143.
12. Baeten JM, Palanee-Phillips T, Brown ER, et al. Use of a vaginal ring containing dapivirine for HIV-1 prevention in women. *N Engl J Med*. 2016;375:2121–2132.
13. Baeten JM, Palanee-Phillips T, Mgodi NM, et al. Safety, uptake, and use of a dapivirine vaginal ring for HIV-1 prevention in African women (HOPE): an open-label, extension study. *Lancet HIV*. 2021;8:e87–e95.
14. Nel A, van Niekerk N, Van Baelen B, et al. Safety, adherence, and HIV-1 seroconversion among women using the dapivirine vaginal ring (DREAM): an open-label, extension study. *Lancet HIV*. 2021;8:e77–e86.
15. Steytler J, Craig C, van der Ryst E, et al. Characterization of viruses in Phase 3 and Phase 3b trials (the ring study and the dapivirine ring extended access and monitoring trial) of the dapivirine vaginal ring for human immunodeficiency virus type 1 infection risk reduction. *Clin Infect Dis*. 2022;76:996–1002.
16. Schader SM, Oliveira M, Ibanescu RI, et al. In vitro resistance profile of the candidate HIV-1 microbicide drug dapivirine. *Antimicrob Agents Chemother*. 2012;56:751–756.
17. Fletcher P, Harman S, Azijn H, et al. Inhibition of human immunodeficiency virus type 1 infection by the candidate microbicide dapivirine, a nonnucleoside reverse transcriptase inhibitor. *Antimicrob Agents Chemother*. 20092009;53:487–495.
18. Parkin N, Chappey C, Maroldo L, et al. Phenotypic and genotypic HIV-1 drug resistance assays provide complementary information. *JAIDS J Acquir Immune Defic Syndr*. 2002;31:128–136.
19. Rhee SY, Gonzales MJ, Kantor R, et al. Human immunodeficiency virus reverse transcriptase and protease sequence database. *Nucleic Acids Res*. 2003;31:298–303.
20. World Health Organization. *HIV Drug Resistance Report 2021*. Geneva, Switzerland: World Health Organization; 2021.
21. Brown ER, Hendrix CW, van der Straten A, et al. Greater dapivirine release from the dapivirine vaginal ring is correlated with lower risk of HIV-1 acquisition: a secondary analysis from a randomized, placebo-controlled trial. *J Int AIDS Soc*. 2020;23:e25634.
22. McCormick K, Penrose KJ, Sethi R, et al. *Comparison of HIV Drug-Resistant Mutant Detection by NGS with and without Unique Molecular Identifiers (UMI)*. Presented at: 27th International Workshop on HIV Drug Resistance and Treatment Strategies Oral Abstract; 2018; Johannesburg, South Africa.
23. Boltz VF, Rausch J, Shao W, et al. Ultrasensitive single-genome sequencing: accurate, targeted, next generation sequencing of HIV-1 RNA. *Retrovirology*. 2016;13:87.
24. Parikh UM, Penrose KJ, Heaps AL, et al. HIV-1 drug resistance among individuals who seroconverted in the ASPIRE dapivirine ring trial. *J Int AIDS Soc*. 2021;24:e25833.
25. Liu TF, Shafer RW. Web resources for HIV type 1 genotypic-resistance test interpretation. *Clin Infect Dis*. 2006;42:1608–1618.
26. Brehm JH, Koontz DL, Wallis CL, et al. Frequent emergence of N348I in HIV-1 subtype C reverse transcriptase with failure of initial therapy reduces susceptibility to reverse-transcriptase inhibitors. *Clin Infect Dis*. 2012;55:737–745.
27. WHO. *World Health Organization HIV Drug Resistance Report 2019*. Document WHO/CDS/HIV/19.21. Geneva, Switzerland: WHO; 2019.

28. Parikh UM, Mellors JW. How could HIV-1 drug resistance impact preexposure prophylaxis for HIV prevention? *Curr Opin HIV AIDS*. 2022;17:213–221.
29. Landovitz RJ, Donnell D, Clement ME, et al. Cabotegravir for HIV prevention in cisgender men and transgender women. *N Engl J Med*. 2021;385:595–608.
30. Marzinke MA, Grinsztejn B, Fogel JM, et al. Characterization of human immunodeficiency virus (HIV) infection in cisgender men and transgender women who have sex with men receiving injectable cabotegravir for HIV prevention: HPTN 083. *J Infect Dis*. 2021;224:1581–1592.
31. Eshleman S, Fogel JM, Halvas EK, et al. *CAB-LA PrEP: Early Detection of HIV Infection May Reduce InSTI Resistance Risk. Abstract 95*. Presented at: Conference on Retroviruses Opportunistic Infections; 2022; Virtual, February 12-16, 2022.
32. Nel A, Haazen W, Nuttall J, et al. A safety and pharmacokinetic trial assessing delivery of dapivirine from a vaginal ring in healthy women. *AIDS*. 2014;28:1479–1487.