Performance of two-stage cervical cancer screening with primary high-risk HPV testing in women living with HIV

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Precis

Colposcopy following positive high-risk human papillomavirus testing maintained sensitivity and improved positive predictive value of high-grade cervical dysplasia among women living with human immunodeficiency virus.

Abstract

*Objective***:** To evaluate the performance of cervical cancer screening algorithms for women living with human immunodeficiency virus (HIV), utilizing primary high-risk human papillomavirus testing (hrHPV) testing followed by cytology, visual inspection with acetic acid (VIA), or colposcopy.

Methods: Prospective cohort study of women living with HIV in Botswana. All participants underwent hrHPV testing. Participants with positive hrHPV results underwent cytology, VIA, colposcopy, and biopsy. Participants with negative hrHPV results also underwent cytology. Histopathology was the reference standard for determination of pre-invasive cervical disease and cervical cancer. Sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and likelihood ratios (LR) of hrHPV-based two-stage screening algorithms were calculated.

Results: Among 300 women screened, 88 (29%) had a positive hrHPV test, and 29 of the 88 (35%) hrHPV-positive women had CIN2+ on histopathology. hrHPV followed by colposcopy resulted in a sensitivity of 83%, specificity of 49%, PPV of 47%, LR+ of +1.6 and LR- of -0.4. hrHPV followed by VIA resulted in a reduced sensitivity of 59%, specificity of 49%, PPV of 39%, LR+ of +1.2 and LR- of -0.8. hrHPV testing followed by cytology also resulted in a reduced sensitivity of 62%, specificity of 77%, PPV of 60%, LR+ of +2.7 and LR- of -0.5. Stratification by HPV 16/18/45 did not improve performance of the algorithms.

Conclusion: In a high-risk HIV population, hrHPV testing followed by colposcopy demonstrated the highest sensitivity and PPV in detecting high-grade cervical dysplasia. Allocating resources to colposcopy in resource-limited settings may be more effective than other screening strategies.

Clinical Trial Registration: 2-stage Cervical Cancer Screening in Botswana, https://clinicaltrials.gov/ct2/show/NCT03324009, NCT03324009

Introduction

Cervical cancer is the fourth leading cause of cancer death in women worldwide and the leading cause of cancer death in women in Botswana.[1,2,3] The disease burden in Botswana is impacted by the high prevalence of human immunodeficiency virus (HIV), which is 22% among people aged 15-49 years and is a well-established risk factor for cervical cancer.[4,5,6] Most cervical cancers are associated with infection with high-risk human papillomavirus (hrHPV) types.[7,8,9] Globally, HPV prevalence is variable, ranging from 15-45%, with higher prevalence in women living with HIV.[10,11,12] HPV 16, 18, and 45 are the high-risk types most commonly associated with cervical cancer in Africa.[13,14,15] Among women living with HIV, persistent hrHPV positivity and infection with multiple types are strong risk factors for cervical cancer.[16]

Cervical cancer is largely preventable and treatable where screening and treatment programs are available.[17,18,19,20] Cervical cancer screening strategies are most effective when based on local evidence and tailored to the population and resource infrastructure.[21] Current programming in Botswana utilizes a combination of cytology (Pap smear) and visual inspection with acetic acid (VIA). However, there is mounting evidence that primary hrHPV testing is the most effective screening strategy because of its high sensitivity (95%).[22] hrHPV testing is increasingly included in some national guidelines.[23,24,25] hrHPV testing is planned for future national programming in Botswana, but the guidelines for managing positive hrHPV results remain unclear, particularly among women living with HIV.[26,27,28] Appropriate triage of a positive hrHPV result is necessary to prevent overtreatment of hrHPV when it is associated with no or low-grade cervical dysplasia. The best two-stage screening strategy is unknown for women living with HIV in resource-limited settings[29,30, 34]

In this study, we investigated the performance of primary hrHPV testing followed by cytology, VIA and colposcopy impression to predict pre-invasive cervical disease in women living with HIV in Botswana. We hypothesized that VIA, cytology and colposcopy would perform similarly as a triage test in women living with HIV who test positive for hrHPV. Evaluating cervical cancer screening algorithms with primary

hrHPV testing in women living with HIV is essential for establishing an evidence-based screening strategy in this high-risk population.

Methods

We conducted a prospective cohort study of women seeking care at the infectious disease care clinic at Princess Marina Hospital in Gaborone, Botswana. The infectious disease care clinic provides care to people living with HIV at Princess Marina Hospital, the regional tertiary referral hospital. Women included in the study were HIV-positive, greater than 24 years of age, and competent to understand study procedures and give informed consent. Women were excluded if they were currently pregnant, currently menstruating heavily or with persistent vaginal discharge, had a previous hysterectomy, or had a previous diagnosis of cervical cancer.

Eligible women were provided study information by a research assistant or study nurse while waiting for their scheduled clinical visit at infectious disease care clinic and offered voluntary participation. After obtaining informed consent, we administered a questionnaire including demographic data, HIV treatment history, history of cervical cancer screening, and knowledge about cervical cancer. In addition to patient report, the electronic medical record was searched for results of prior cervical cancer screening. The institutional review boards of the Botswana Ministry of Health and Wellness, the University of Botswana, and the Beth Israel Deaconess Medical Center approved this study. The ethics committee of Princess Marina Hospital also approved this study.

All participants underwent a speculum examination of the cervix by a trained study nurse, at which time samples were collected from the cervix for hrHPV testing and for cervical cytology using a Cervex-brush®. HPV specimens were placed in a PreservCyt® transport medium and testing was performed using the Xpert® HPV Assay (Cepheid, Sunnyvale, CA) at the Botswana Harvard AIDS Initiative Partnership Laboratory.

The Xpert® HPV assay tests for 14 hrHPV types, including 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68. Cytology was prepared by spreading collected cervical cells from a Cervex-brush® onto a glass slide and fixing with a spray fixative at the collection site. Cytology was sent to the National Health Laboratory for processing and pathologist evaluation and reported using the revised Bethesda classification.[31] Abnormal lower genital tract cytology was evaluated at two thresholds: abnormal squamous cells of undetermined significance (ASC-US) or worse, and high-grade squamous intraepithelial lesion (HSIL) or worse.

Because there are no clinical guidelines for management of positive hrHPV results in Botswana, we also collected cytology at the time of hrHPV sample collection to ensure that all participants were screened according to current cervical cancer screening guidelines in Botswana. We referred participants who tested negative for hrHPV to colposcopy if they had a study cytology of HSIL or had a prior abnormal cytology result and study cytology result of ASC-US or worse (≥ASC-US) in accordance with current Botswana National Cervical Cancer Prevention Programme algorithms. We referred all participants who tested positive for any hrHPV type to VIA and colposcopy, regardless of their cytology result. At the time of the colposcopy visit, participants underwent a speculum examination of the cervix with both VIA and colposcopy performed by providers who were blinded to the HPV test results and cytology results. VIA was performed by a trained nurse midwife who had participated in the Botswana Ministry of Health and Wellness national VIA training program and was experienced in performing VIA in the clinical setting. Visual assessment was performed after applying 5% acetic acid to the cervix using a cotton swab and findings were categorized as normal, abnormal with recommendation for cryotherapy, or abnormal with recommendation for loop electrosurgical excision procedure (LEEP). In the analysis, we considered lesions recommended for cryotherapy as "low-grade" and lesions recommended for LEEP as "high-grade". Subsequently, a gynecologist blinded to the VIA assessment performed colposcopy and normal, low-grade or high-grade impression was recorded. All participants had a biopsy collected at the time of colposcopy. If there was a visible lesion, a punch biopsy or LEEP was performed according to current best practice in

Botswana. If no lesion was visible, a small endocervical excision or an endocervical curettage was performed. All women with cervical intraepithelial neoplasia ≥ CIN2 (CIN2+) on biopsy or endocervical curettage were referred for an excisional procedure. Women with histopathology showing CIN3 with microinvasion or invasive cervical cancer were referred to gynecologic providers for further assessment and treatment.

The primary outcome was performance of two-stage cervical cancer screening algorithms in detecting high grade cervical dysplasia. We defined high-grade cervical dysplasia as a colposcopy result of cervical intraepithelial neoplasia grade 2 or higher (CIN2+). Using histopathology collected at time of colposcopy as the gold standard, we calculated the sensitivity, specificity, positive predictive value (PPV) negative predictive value (NPV), and likelihood ratios (LR) to detect high-grade cervical dysplasia for 1) cytology following a positive hrHPV test, 2) VIA impression following a positive hrHPV test and 3) colposcopy impression following a positive hrHPV test. For each two-stage screening strategy, we evaluated test performance at two cutoffs. For cytology, we evaluated cut-offs of ASC-US and HSIL. For VIA and colposcopy, we evaluated cut-offs of low-grade and high-grade impressions. In addition, we repeated this analysis stratified by hrHPV type (16/18/45 and other hrHPV).

Data were entered into a REDCap electronic database by a designated research assistant and accuracy of data entry were verified by the study nurse and principal investigator. Descriptive statistics are presented as median with interquartile range or proportion. We compared categorical variables with the chi-square or Fisher's exact test and continuous variables with the Wilcoxon rank sum test. We considered two-sided p values <0.05 statistically significant and used SAS 9.4 (SAS Institute, Cary, North Carolina) for analyses.

The goal of a two-stage algorithm to detect high-grade cervical dysplasia is to increase PPV while maintaining sensitivity and specificity. In a prior cervical cancer screening study among a population of women with a relatively high HIV prevalence, the PPV of a hrHPV positive test for high-grade cervical

dysplasia was 24% (Denny, 2000). Our sample size calculation was targeted to detect an improvement in PPV from 24% for hrHPV testing alone to 49% for the two-stage algorithms. Assuming a two-sided alpha of 0.05, a sample size of 81 participants with hrHPV was needed to yield 80% power to detect the specified difference. Based on preliminary data from a recent study of women living with HIV in Botswana, we assumed hrHPV-positivity would be 30% (unpublished data). Thus, we needed to enroll 270 participants with HIV to yield 81 who would be hrHPV positive. To allow for 10% loss to follow-up between the primary hrHPV testing and colposcopy we aimed to enroll at least 300 participants.

Authors' Data Sharing Statement

Will individual participant data be available (including data dictionaries)? Yes

What data in particular will be shared? All of the individual participant data collected during the trial, after deidentification.

What other documents will be available? Study protocol

When will data be available (start and end dates)? Beginning 3 months and ending 5 years following article publication.

With whom? Investigators whose proposed use of the data has been approved by an independent review committee identified for this purpose.

For what types of analyses? To achieve aims in the approved proposal

By what mechanism will data be made available? Proposals should be directed to rluckett@bidmc.harvard.edu. To gain access, data requestors will need to sign a data access agreement.

Results

We recruited participants from April to July 2018, and all follow-up colposcopy visits were completed by August 2018. Of the 312 women living with HIV enrolled, 12 were lost to follow-up, deemed ineligible or withdrawn before cervical samples were collected at the first study visit, leaving 300 (96%) who underwent hrHPV testing and cytology collection. Of those participants, 88 (29%) had a positive hrHPV result. Among those 88 who were hrHPV positive, we did not have colposcopy results for 6 (3 were lost to follow-up, 1 withdrew, 1 became ineligible due to pregnancy, and 1 biopsy specimen was lost in the laboratory) and had histopathology results from colposcopy for 82 women for this analysis. Additionally, two participants who were hrHPV-negative underwent colposcopy for cytology of HSIL (Figure 1).

Baseline characteristics were similar among women who tested positive and negative for hrHPV (Table 1). The majority of women reported having undergone prior cervical cancer screening (95%). There were no differences between groups in prior abnormal screening results or cervical excisional procedures. Only 5 women had a recent CD4 count of < 200/μL, and all of the participants were taking antiretroviral therapy. Only two women reported a history of smoking, and both tested negative for all hrHPV types.

Of the 88 (29%) women who were positive for any hrHPV type, 15 of the 300 screened had HPV 16 (prevalence 5%); 21 of the 300 screened had HPV 18/45 (prevalence 7%); and 66 of the 300 screened had other hrHPV types (prevalence 22%). Among the 82 women with a positive hrHPV test who had histopathology results, 29 (35%) had CIN2+ (Table 2). The prevalence of CIN2+ by hrHPV type was 31%, 21%, and 43% for HPV 16, HPV 18/45, and other hrHPV types, respectively. Among the 11 participants co-infected with multiple hrHPV types, the prevalence of CIN2+ was 45%.

We compared the performance of the two-stage cervical cancer screening algorithms. hrHPV followed by colposcopy impression had a sensitivity of 83%, specificity of 49%, PPV of 47%, LR+ of +1.6 and LR- of -0.4. hrHPV testing followed by VIA resulted in a reduced sensitivity of 59%, specificity of 49%, PPV

of 39%, LR+ of +1.2 and LR- of -0.8 at the low cut-off point of "low-grade impression". hrHPV testing followed by cytology also resulted in a reduced sensitivity of 62%, specificity of 77%, PPV of 60%, LR+ of +2.7 and LR- of -0.5 at the ASC-US threshold (Table 3). Triaging hrHPV positive women with colposcopy impression, VIA and cytology missed CIN2+ diagnoses in 5, 12, and 11 women in our cohort, respectively. Evaluation of the two-stage algorithms stratified by HPV 16/18/45 versus other hrHPV types did not improve the performance of any algorithm (Table 4).

Four women had histopathology results of cancer or microinvasive CIN 3. One of these women had HPV18/45 and the other three had other hrHPV types. All four had a cytology result of HSIL. Three had low-grade impressions on both VIA and colposcopy, while one had a high-grade impression on both VIA and colposcopy.

Discussion:

Primary hrHPV testing followed by colposcopy was the most sensitive two-stage algorithm for cervical cancer screening among women living with HIV in Botswana. Both VIA and cytology as second-stage screening methods had unacceptably low sensitivity, missing approximately one-third of women with high-grade cervical lesions. Triaging hrHPV positive results with VIA or cytology eliminated the benefit of the high sensitivity that primary hrHPV testing provides. Further, triaging of hrHPV positive results based on type did not improve the performance of any two-stage algorithm.

One third of women in our study with positive hrHPV primary screening had high-grade cervical disease, which is a higher proportion than found in other populations living with HIV.[32] Our population also had a higher prevalence of high-grade dysplasia among women with other hrHPV compared to women with HPV 16 or 18/45. This is consistent with prior studies in Botswana that showed heterogeneous HPV types associated with high-grade precancerous cervical lesions among women living with HIV (16, 18, 35, 58, and 61) and a lower prevalence of HPV 16 and 18 positivity in cervical cancer specimens.[33,34,35] This

cross-sectional data does not support triaging strategies based on hrHPV type, as may be considered in other African settings.[36]

Primary hrHPV testing followed by colposcopy results in a high number of referrals for colposcopy, presenting challenges in resource-limited settings.[37] Guidelines for low- and middle-income countries have presumed that scaling up colposcopy is not feasible.[38,39] Recent trends in cervical cancer screening in the region have focused on visual inspection strategies as opposed to colposcopy training.[40] However, consideration of available data to plan effective screening programs is vital. Our findings support concerns raised in prior studies that VIA and cytology triaging of women with hrHPV may have variable or low sensitivity, particularly in women living with HIV, and that referral to colposcopy may be a better alternative.[41,42,43,44] Building on the infrastructure that visual inspection has developed may facilitate roll-out of colposcopy, if coupled with the training of nurses and general practice providers in the region. In Botswana, for instance, the VIA programming has equipped a number of facilities with capability to perform LEEP, and many LEEP sites have colposcopes not currently in use. If rapid hrHPV testing were available in the future, same-day triage with colposcopy and treatment at these sites would be feasible.

This study highlights the acute need to improve screening for cervical cancer and raises concern about the frequency of screening in women living with HIV in low- and middle-income countries. Current national strategy in Botswana recommends screening with cytology or VIA in women living with HIV every three years. While many of the participants had been screened before (over 90%), only 11% of women reported a prior abnormal result and 2-3% reported a prior excisional procedure. Our high prevalence of high-grade pre-invasive cervical disease supports the need for frequent screening to ensure diagnosis of disease prior to progression to malignancy. In addition to high rates of pre-invasive cervical disease, the rate of detection of cervical cancer in our screening cohort was relatively high at 2%. This included 3 women enrolled but immediately referred for suspicion for clinical stage IB cervical cancer on examination and 4 women with histopathology concerning for Stage IA cervical cancer (cervical cancer or microinvasive

CIN3). This rate was similar to another screening cohort in Zambia where 6 of 200 (3%) women living with HIV had invasive cervical cancers detected at the time of screening, but higher than other settings.[45] In a large cervical cancer screening cohort of 79,506 women in India, 238 (0.3%) invasive cervical cancers were detected (Sankaranarayanan, 2009). In a cervical cancer screening cohort of 1128 women living with HIV in India, 5 (0.4%) invasive cervical cancers were detected. [46]

We found lower rates of hrHPV prevalence among women living with HIV than reported in the literature, which may highlight the improvement in HIV management over time with higher antiretroviral therapy utilization and viral suppression.[47,48] Botswana has had continuous access to antiretroviral therapy in the public sector since 2002, with initiation of antiretroviral therapy at graduated CD4 counts over time (initially 200 then 350) until a test-and-treat policy was initiated in 2016. Demographic differences in study populations may also contribute to this difference. Our study had a higher median age than in studies conducted in the United States, Kenya and Brazil. Additionally, the population in New York had higher risk behaviors, as indicated by high rates of smoking and on-going intravenous drug use.[49] The study population in Brazil was pregnant which may have resulted in increased immunosuppression and higher hrHPV detection rates.[50] Rates of hrHPV prevalence among women living with HIV in the region generally range from 47-57%, however, the prevalence is lower in women aged 40-49.[51,52] In a similarly-aged cohort of women in Zambia, where 90% of participants were on antiretrovirals and only 77% virally suppressed, hrHPV positivity was 47% (Chibwesha, 2016). On-going evaluation of hrHPV rates in women living with HIV in the modern antiretroviral therapy are necessary to understand if our findings are generalizable.

Our study has limitations. Our confidence intervals are wide around sensitivity, specificity, PPV and NPV as a result of our relatively small sample size. Further, research in larger populations will help to clarify if the difference in performance detected in this study is significant. The cohort was recruited from an HIV treatment center, which may represent a unique population of health-seeking individuals and may not be

representative of a broader population. Ease of communication and follow-up of abnormal results may not therefore be replicated in a larger population. However, we found many women were not only reachable, but proactively followed-up their results, indicating that improved education about cervical cancer may reduce loss to follow-up and maximize dissemination of results. While the goal of this study was to evaluate screening algorithms that would be possible with pathology services currently available, external validation of cytology and histopathology specimens was not performed and thus accuracy compared to an expert gynecologic cytopathologist and pathologist was not evaluated. History of cervical cancer screening is primarily self-reported with limited ability to confirm results in the electronic medical record. In regards to study design, the effect of co-infection with multiple hrHPV types could not be assessed because the study sample was not sufficiently powered for this subgroup. Finally, one VIA nurse and colposcopist conducted the evaluations; therefore, performance of these tests may not be generalizable.

Follow-up of this cohort is currently underway to evaluate the best interval and modality for longitudinal screening. Further research on the performance of technology-based cervical cancer screening methods compared to current available methods in low- and middle-income countries is also being planned in a larger population. Balancing the cost of these strategies with clinical effectiveness is essential and a cost-effectiveness evaluation of these strategies in Botswana is being explored. Finally, regional adoption of a test-and-treat policy for HIV may continue to impact cervical cancer rates in the long-term as long-standing antiretroviral therapy use and initiation of treatment at higher CD4 levels may reduce incidence of cervical dysplasia, progression of dysplasia, and increase the likelihood of CIN regression.[53] On-going research in our population living with HIV is essential to understand this impact.

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Table 1: Baseline characteristics of the study population

Characteristic	All n = 300*	hrHPV positive n = 88	hrHPV negative n = 212	p
Age, years [interquartile range]	46 [42-52]	44 [40-51]	47 [42-52]	0.05
Education				0.40
≤Primary	94 (31)	24 (27)	70 (33)	

≥Secondary	206 (69)	64 (73)	142 (67)	
Employed	197 (66)	63 (72)	134 (63)	0.21
Marital status				0.85
Single	215 (72)	61 (69)	154 (72)	
Married	55 (18)	18 (20)	37 (17)	
Divorced	12 (4)	3 (3)	9 (4)	
Widowed	18 (6)	6 (7)	12 (6)	
Parity[$]				0.15
0	11 (4)	5 (6)	6 (3)	
1-3	199 (66)	58 (66)	141 (67)	
≥4	75 (25)	24 (27)	51 (24)	
Sexual partners				0.83
1-5	186 (62)	55 (63)	131 (62)	
≥6	100 (33)	28 (32)	72 (34)	
Missing	14 (5)	5 (6)	9 (4)	
Postmenopausal	106 (35)	27 (31)	79 (38)	0.38
CD4 Count (per µL)				0.63
<200	5 (2)	2 (2)	3 (1)	
200-500	83 (28)	27 (31)	56 (26)	
>500	212 (71)	59 (67)	153 (72)	
Detectable viral load	11 (4)	6 (7)	5 (2)	0.12
Currently on a~~A~~ntiretroviral therapy~~RT~~	300 (100)	88 (100)	213 (100)	--
Length of time on antiretroviral therapy~~ART~~, years [interquartile range]	14 [11 – 15]	14 [9 – 15]	14 [12 – 15]	0.09
History of cervical cancer screening				
Yes	285 (95)	79 (90)	206 (97)	0.02
Pap ≥ASC-US	27 (9)	11 (14)	16 (8)	0.44
VIA positive	3 (1)	1 (1)	2 (1)	1.0
History of cervical excisional procedure	6 (2)	3 (3)	3 (1)	0.18

***All table entries are number of study subjects (%) unless otherwise noted**

[$]Data available for 285 participants

ART: antiretroviral therapy; ASC-US: abnormal squamous cells of undetermined significance; VIA: visual section with acetic acid

Table 2: Prevalence of CIN2+ (per 100 women living with HIV) who tested positive for high-risk HPV and underwent colposcopy

HPV type	Number undergoing colposcopy (n)	Number with CIN2+ (n)	Prevalence of CIN2+ (%) [95% CI]
Any high-risk HPV type	82	29	35% [25 – 47]

HPV 16*	13	4	31% [9 – 61]
HPV 18/45*	19	4	21% [6 – 46]
Other high-risk HPV type*	61	26	43% [30 – 56]
>1 high-risk HPV type	11	5	45% [17 – 77]
*Infection with these sub-types is not mutually exclusive CIN2+: cervical intraepithelial neoplasia grade 2 or higher			

Table 3: Performance of two-stage screening in detecting CIN2+ among women living with HIV who tested positive for high-risk HPV and underwent colposcopy

Two-stage screen using different cut-offs		Biopsy result		Two-stage screen characteristics				
		CIN2+ (n)	≤ CIN1 (n)	Sensitivity (%) [95% CI]	Specificity (%) [95% CI]	PPV (%) [95% CI]	NPV (%) [95% CI]	LR +/- [95% CI]
Cytology	NILM	11	41	--	--	--	--	
	≥ ASC-US	18	12	62% [42–79]	77% [64–88]	60% [41–77]	79% [65–89]	+ 2.7 [1.1 4.3] -0.5 [0.2-0.7]
	≥ HSIL	9	4	31% [15–51]	92% [82–98]	69% [39 – 91]	71% [59–81]	
Visual inspection with acetic acid (VIA)	normal	12	26	--	--	--	--	
	≥ low-grade impression	17	27	59% [39–76]	49% [35–63]	39% [24–55]	68% [51–83]	+ 1.2 [0.7–1.6] - 0.8 [0.4–1.3]
	≥ high-grade impression	4	5	14% [3–32]	91% [79–97]	44% [14–79]	66% [54–76]	
Colposcopy impression	normal	5	26	--	--	--	--	
	≥ low-grade impression	24	27	83% [64–94]	49% [35–63]	47% [33–62]	84% [66–95]	+ 1.6 [1.1–2.1] -0.4 [0.1–0.7]
	≥ high-grade impression	4	5	14% [4–32]	91% [79–97]	44% [14–79]	66% [54–76]	

CIN2+: cervical intraepithelial neoplasia grade 2 or higher; CI: confidence interval; PPV: positive predictive value; NPV: negative predictive value; NILM: negative for intraepithelial lesion or malignancy; ASC-US: abnormal squamous cells of undetermined significance; HSIL: high-grade squamous intraepithelial lesion

Table 4: Performance of two-stage screening in detecting CIN2+ among women living with HIV who tested positive for high-risk HPV and underwent colposcopy stratified by HPV type

Study Arm	CIN 2+ (n)	≤ CIN 1 (n)	Sensitivity (%) [95% CI]	Specificity (%) [95% CI]	PPV (%) [95% CI]	NPV (%) [95% CI]

hrHPV + Cytology	**HPV 16/18/45**						
	NILM	3	17	--	--	--	--
	≥ ASC-US	5	7	63 (24 – 91)	71 (49 – 87)	42 (15 – 72)	85 (62 – 97)
	≥ HSIL	3	2	38 (9 – 76)	92 (73 – 99)	60 (15 – 95)	81 (62 – 94)
	Other hrHPV						
	NILM	9	26	--	--	--	--
	≥ ASC-US	17	9	65 (44 – 83)	74 (57 – 88)	65 (44 – 83)	74 (57 – 88)
	≥ HSIL	8	3	31 (14 – 52)	91 (77 – 98)	73 (39 – 94)	64 (49 – 77)
hrHPV + VIA	**HPV 16/18/45**						
	Normal	4	15	--	--	--	--
	≥ low-grade impression	4	9	50 (16 – 84)	63 (41 – 81)	31 (9 – 61)	79 (54 – 64)
	≥ high-grade impression	1	1	13 (0 – 53)	96 (79 – 100)	50 (1 – 99)	77 (58 – 90)
	Other hrHPV						
	Normal	10	14	--	--	--	--
	≥ low-grade impression	16	21	62 (41 – 80)	40 (24 – 58)	43 (27 – 61)	58 (37 – 78)
	≥ high-grade impression	4	5	16 (5 – 36)	86 (70 – 95)	44 (14 – 79)	59 (44 – 72)
hrHPV + Colposcopy	**HPV 16/18/45**						
	Normal	1	10	--	--	--	--
	≥ low-grade impression	7	14	88~~100~~ (47 – 100)	42 (22 – 63)	33 (15 – 57)	91 (59 – 100)
	≥ high-grade impression	2	3	25 (3 – 65)	88 (68 – 97)	78 (58 – 91)	81 (62 – 94)
	Other hrHPV						
	Normal	4	18	--	--	--	--
	≥ low-grade impression	22	17	85 (65 – 96)	51 (34 – 69)	56 (40 – 72)	82 (60 – 95)
	≥ high-grade impression	4	3	15 (4 – 35)	91 (77 – 98)	57 (18 – 90)	59 (45 – 72)