Acceptability and Detection of High-Risk Human Papilloma Virus using Self-Collected Sampling for Cervical Cancer Screening among HIV-Positive and HIV-Negative Women in Tanzania

Research Article

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Abstract

Objective: Cervical cancer is responsible for the greatest number of cancer-related cases and deaths among Tanzanian women. Although a national cancer screening program using visual inspection with acetic acid (VIA) was established in Tanzania in 2010, participation remains low. Self-sampled human papillomavirus (HPV) tests have recently been recommended as a method to increase screening participation. We evaluated the acceptability and detection of high-risk HPV (Hr-HPV) using self-collected sampling for cervical cancer screening among Tanzanian HIV-negative and HIV-positive women.

Methods: A cross-sectional study design was used to recruit women from five clinics offering cervical cancer screening in Dar es Salaam and the surrounding Pwani Region. Eighteen women (9 HIV+ and 9 HIV-) were recruited from each clinic using convenience quota sampling. A trained nurse instructed women who provided informed consent on the self-collection of vaginal samples. A survey to assess self-sampling acceptability was administered after self-collection was completed. All women were then screened with VIA. The self-collected samples were genotyped for Hr-HPV using the AmpFire® Multiplex High Risk HPV Assay.

Results: Among 90 women who participated in the study, the median age was 34 years (IQR: 28 - 37). Thirty-four percent of women indicated they preferred self-collection, 30% indicated that either method was fine and 36% indicated that they would prefer clinician-collected sampling. A high proportion of all women (≥90%) found self-sampling acceptable based on six different indices including convenience and ease. Thirty-eight (42.2%) women were Hr-HPV positive. Hr-HPV prevalence was 28.9% (13 women) in HIV-negative women and 55.6% (25 women) in HIV-positive women.

Conclusion: Self-collected samples were well accepted in this sample of HIV-positive and negative women. Although self-collection may address the low cervical cancer screening participation in Tanzania, if confirmed in larger studies, cost-effective strategies to triage the potentially high proportion of HPV positive women are needed.

Keywords: Human Papilloma Virus; Cervical Cancer; Self-Collected Test; Human Immunodeficiency Virus; Screening.

Introduction

Cervical cancer is the leading cause of cancer-related morbidity and mortality among Tanzanian womenwith approximately 10,241 diagnosed with and 6,525 dying from cervical cancer in the last year.[1] Current estimates indicate an age-standardized incidence rate of 62.5 cases per 100,000 women and an age-standardized mortality rate of 42.7 deaths per 100,000 women.[2]

In comparison, Europe has an age-standardized cervical cancer incidence of 10.7 per 100,000 women with a mortality rate of 3.76 per 100,000 women and globally the incidence rate is approximately 13.3 cases per 100,000 women and the mortality rate is 7.25 deaths per 100,000 women.[2] Additionally, approximately 5% of Tanzania's adult population lives with HIV,[3] which is an independent risk factor for cervical cancer.[4-6]

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Citation: Kandali Samwel, Julius Mwaiselage, Redempta Mbatia, Madalyn Nones, Pendo Bukori, et al., Acceptability and Detection of High-Risk Human Papilloma Virus using Self-Collected Sampling for Cervical Cancer Screening among HIV-Positive and HIV-Negative Women in Tanzania. *Int J Chronic Dis Ther*. 2024;9(2):148-153.

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Cervical cancer screening with visual inspection with acetic acid (VIA) has been available as part of a national program since 2010.[7] In 2021, approximately 1.5 million women were screened for cervical cancer by VIA, which is about 40% of the eligible population for screening in Tanzania.[8] A number of factors are associated with low uptake of cervical cancer screening among women in Tanzania, including a low clinician-to-patient ratio, a lack of knowledge about screening, as well as fear about screening procedures and results. [9-11] There are also identifiable subgroups in Tanzania, such as women with lower education levels or high parity as well as those who reside in rural areas, for whom screening rates are lower than for women as a whole.[10] Given these concerns and the relatively high prevalence of HIV, increasing screening uptake is an important public health priority.[12]

In 2021, the World Health Organization recommended on the use of human papillomavirus (HPV) testing for cervical cancer screening in low and middle-income countries, including Tanzania.[13] Reasons for this include the fact that the test can be conducted from a self-collected sample or a clinician-collected sample and importantly has a high sensitivity for the detection of high-grade dysplasia and cancer compared to VIA. In a study conducted in Tanzania, HPV testing was shown to be significantly more sensitive than VIA using detection of HSIL on cytology as the outcome.[4] Studies conducted inTanzania have shown that self-collection is acceptable.[14, 15] In a study by Katanga et al. most women (79.8%) preferred self-collection to clinician-collected samples (16.5%).[14] Previous studies, including a systematic review of HPV self-collection studies conducted in Africa, have also demonstrated moderate to strong agreement between clinician-collected samples and self-collected samples, suggesting the feasibility of self-administered tests to adequately detect HPV. [14, 16] Based in part, on these findings and the WHO recommendation, the Tanzanian Ministry of Health recently developed a plan to integrate HPV-based testing into the national cervical cancer screening program.[17] The success of this plan will depend, however, on understanding how best to integrate HPV self-collection based testing with a screening program that's currently structured based on VIA and also how HPV self-collection may perform in a mixed population of HIV-positive and HIV negative women.

To address this, we conducted a pilot study to assess self-sampling acceptability and compare HPV test results to VIA test results stratified by HIV status in 90 women attending healthcare facilities across Tanzania.

Patients and Methods

Sample Selection

IRB approval for this study was provided by the Tanzanian National Institute for Medical Research and the University of Minnesota Institutional Review Board. This cross-sectional study consisted of women selected from five sites: the Ocean Road Cancer Institute (ORCI) in Dar es Salaam, Bagamoyo District Hospital (located in a rural coastal area north of Dar es Salaam), Mkuranga District Hospital (located in a rural area south of Dar es Salaam), Chalinze District Hospital (in a rural region west of Dar es Salaam) and Kisarawe District Hospital (located in a semi-urban area west of Dar es Salaam). The ORCI is a designated

cancer hospital while the remaining study sites are all district-level hospitals. The 5 study sites were selected based on their geographic range to ensure inclusion of women from a variety of urban and rural areas from Dar es Salaam and the surrounding Pwani region. All sites are government funded hospitals that provide regular cervical cancer screening services and HIV care. For this pilot study, eighteen women (nine HIV-positive women and nine HIV-negative women) were selected from each study site. Convenience quota sampling was used for study recruitment to ensure equal numbers of HIV-positive and HIV-negative women. Enrollment took place over nine months from February 2019 to October 2019. To participate in the study, women had to be 25 years of age or older, provide informed consent, not have a current diagnosis of cervical cancer, provide a medical history to confirm HIV status, and have the ability to self-collect a sample for HPV testing.

Following an informed consent process, each participant was administered a demographic and health-related questionnaire. Detailed instructions were then provided by a trained study nurse for HPV self-collection; an instructional poster was placed in the self-collection room for additional guidance. Women were provided with all equipment necessary to obtain the specimen, including a collection brush and a private setting for sample collection. After self-collection, participants underwent VIA performed by a trained nurse; women who were VIA-positive were treated by cryotherapy or LEEP based on their VIA results.[13]

The self-collected samples were stored and shipped at room temperature to the Ocean Road Cancer Institute laboratory, in Dar-es-salaam, Tanzania for processing. This site has trained molecular technicians who perform HPV testing for research studies conducted in Tanzania.[14] Samples were extracted with the Qiagen DNeasy Blood and tissue kit (Qiagen Inc, Valencia, California USA; cat number 69506) according to the manufacturer's instructions. The concentrations of the extracted DNA were determined using a nanodrop spectrophotometer. DNA samples were then stored at -20°C until further analysis. Genotyping was done using the Ampifire® Hr- HPV assay BIO-RAD Real-time PCR according to the manufacturer's instructions.[18, 19] Only samples that tested positive for Hr-HPV DNA were considered HPV-positive for this study. The assay identifies 15 high-risk Hr-HPV genotypes (16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, and 68) as has been previously described.[18, 19] Results for Hr-HPV were shared with the study nurse who then contacted each participant. HPV positive women were asked to follow up with their clinic for a VIA screening.

Post sampling survey

Each participant completed a survey immediately following self-collection. The survey was adapted from a survey administered to women in Malaysia.[20] For our study, the survey was translated into Swahili, focused on self-collection only, and administered after collection was completed (post-collection); a copy is available from the authors. Participants were asked if they preferred self-collection of HPV samples, clinician-collected sampling, or if either was fine. Acceptability of self-collection was measured according to six separate indices: experience with self-collection, ease of collection, convenience, embarrassment associated with self-collection, discomfort associated with the collection procedure, and confidence in the ability to correctly collect a sample.

These indices were measured on a five-point Likert scale with '1' indicating the lowest level of satisfaction and '5' indicating the highest level of satisfaction. For all Likert scales, a score ≥4 was considered a 'high rating/positive score' and a score <4 was considered a 'low rating/negative score.'

Statistical Analysis

All survey data and test results were entered in R (version 4.2.1 for Windows) for descriptive analysis and hypothesis testing. Participant age was categorized as <30 or ≥ 30 years; the number of sexual partners in the last 5 years was categorized as ≤2 or >2 partners; age at first intercourse was categorized as <18 or ≥18 years. Cut-points were based on a previous study evaluating HPV prevalence by HIV status as well as the median value for variables such as age at first intercourse and past sexual partner number. [21] Sampling preference was categorized into three levels: preference for self-collection, preference for clinician-collection, or either.

The proportion of women who were positive based on their self-collected HPV tests was compared to the proportion positive by VIA, since the latter is currently recommended for screening in Tanzania. Concordance between a positive result for any HPV type from self-collected specimens and VIA results was evaluated using kappa statistics. Data on demographic and sexual history were used to identify potential characteristics associated with an HPV-positive test result and/or an abnormal VIA test result. Crude prevalence ratios and 95% confidence intervals were used to estimate the association between participant characteristics and preferred sampling method (self vs clinician) as well as the association between HIV positivity and HPV infection. Acceptability of HPV self-collection was also assessed, stratified by HIV status.

Results

Demographics

The final sample consisted of 90 women (half (n=45) of whom were HIV-positive) with a median age of 34 years (IQR: 28 - 37). No difference was observed in participant age distribution across the study sites. When considering marital status, 51 women (56.7%) were married, 24 women (26.7%) were widowed, divorced or separated, and 15 women (16.7%) were single. Seventy-two women (80%) reported a formal education (primary school, secondary school, college, or university) while 20% of women reported no formal education. The median age of first sexual intercourse was 18 years (IQR: 17 - 18.6). Seven (10.8%) of the 65 women who responded to the question regarding contraceptive use indicated the recent use of condoms. All HIV-infected women were using ART and self-reported adherence to their current regimen. The median duration of usage was 36 months (IQR: 12 - 84).

Acceptability and Preference of HPV Sampling

In terms of preferences for HPV self-collection sampling, 31 women (34.4%) indicated that they would prefer self-collection while 32 women (35.6%) indicated that they would prefer clinician- collected sampling and 27 women (30%) indicated that either method was fine. Of the women who preferred clinician collection, 26 (81.3%) indicated that they felt more confident with clinician-collection. There were no significant differences between preferences for self-collected versus clinician-collected samples for any of the assessed demographic variables, including HIV-status (Table 1). The demographic distribution for women who preferred self-collection was similar to that of the entire sample. Of those that preferred self-collection, 18 women (58.1%) were HIV-positive (Table 1). Each of the acceptability indices had a high level of positive scores: ninety percent of women scored the experience highly. Eight-five women (94.4%) scored the indices of easy-to-do, convenient, not embarrassing, and no discomfort or pain as high. Eighty-seven women (96.7%) scored a four or above for confidence in self-sampling ability (Table 2).

HPV Prevalence in HIV-positive and HIV-negative Women

Table 1. Demographic characteristics of study participants in study on HPV self-collection, Tanzania, by preference for sampling type (Prevalence ratios are provided between preferences of clinician-collection versus self-collection).

Participant Characteristic	N (%) Or Median (IQR)	Preferred self-sampling, N(%)	Prefer either, N(%)	Preferred clinician-collection, N(%)	PR	95% CI
Total (N)	90 (100)	31 (34.4)	27 (30.0)	32 (35.6)		
Age*	34 (28-37)	34 (27-40)	33 (28-36)	35 (27-41)		
Education						
No Primary Education	18 (20)	7 (38.9)	2 (11.1)	9 (50.0)	Referent	
Primary School	37 (41.1)	13 (35.1)	9 (24.3)	15 (40.5)	0.94	0.48 - 1.87
Secondary School or Higher	35 (38.9)	11 (31.4)	16 (45.7)	8 (23.5)	0.76	0.38 - 1.48
Marital Status						
Single	15 (16.7)	5 (33.3)	5 (33.3)	5 (33.3)	Referent	
Widowed/Divorced/Separated	24 (26.7)	9 (37.5)	5 (20.8)	10 (41.7)	1.06	0.48 - 2.30
Married/Living with Partner	51 (56.7)	17 (33.3)	17 (33.3)	17 (33.3)	1	0.49 - 2.02
Past Sexual Partners						
<2 Partners	49 (54.4)	15 (30.6)	18 (36.7)	16 (32.7)	Referent	
>2 Partners	41 (45.6)	16 (39.0)	9 (30.0)	16 (39.0)	0.97	0.59- 1.60
Age at First Sex						
>18 Years of Age	54 (60.0)	19 (35.2)	15 (27.8)	20 (37.0)	Referent	
<18 Years of Age	36 (40.0)	12 (33.3)	12 (33.3)	12 (33.3)	0.97	0.58 - 1.63
HIV status						
HIV-negative	45 (50.0)	13 (28.9)	17 (37.8)	15 (33.3)	Referent	
HIV-positive	45 (50.0)	18 (40.0)	10 (22.2)	17 (37.8)	0.9	0.54 - 1.51

*value is provided as median (IQR)

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All 90 samples were suitable for testing for the presence of Hr-HPV. Overall, thirty-eight (42.2%) women were Hr-HPV positive (Table 3). Hr-HPV prevalence was 28.9% for HIV-negative women and 55.6% in HIV-positive women. Fifteen (39.5%) of the 38 Hr-HPV positive samples were positive for multiple HPV types. The prevalence of being HPV-positive for HIV-positive women was 1.92 times (95% CI 1.13 - 3.26) that of HIV-negative women (Table 3). Among HPV-positive women, the prevalence of testing positive for multiple Hr-HPV types in HIV-positive women was 1.74 times (95% CI 0.36 – 9.90) that of HIV-negative women. The most commonly detected Hr-HPV types were HPV 16 (18.4%), HPV 39 (26.3%), HPV 56 (13.2%), HPV 59 (15.8%) and HPV 68 (15.8%). Among women co-infected with HIV and HPV, HPV 39 was the most commonly detected Hr-HPV type (32.0%). Among HIV-negative women with HPV, HPV 16 (15.0%) and HPV 68 (15.0%) were the most commonly detected Hr-HPV types.

Comparison of HPV testing and VIA

Two women were VIA-positive. Of these, one was HPV-positive and HIV-negative while the other woman was HPV-negative and HIV-positive. Cohen's ϰ was 0.008 (95% CI -0.06–0.08) for the concordance between HPV testing and the VIA results indicating very poor agreement.

Discussion

In this study of self-collected HPV tests, a slightly lower proportion of participants preferred self-collection compared to clinician-collected sampling. However, the post-collection survey on acceptability showed a high proportion of both HIV-positive and HIV-negative women found self-collection acceptable based on different indices including experience with self-collection as well as ease and convenience. Previous studies have assessed women's acceptability of self-collected HPV samples in similar low-resource settings, including Tanzania and have found a high acceptance of HPV self-sampling.[14,16] However, in contrast to our study, some of these studies also demonstrated an overall preference for self-collection over clinician collection. The main reason provided was greater confidence in a clinician-collected sample, which is consistent with the preference for clinician collection provided in other studies.[14,16] Other reasons for the slight difference in preference noted may be due to our small sample size, differences in the number of choices provided for the response for this survey compared to others and/or clarity of the question since the Swahili version of the survey was not validated or evaluated in terms of reliability.[22] These results suggest the need for education and ways to reassure women about HPV self-collection.

This study found that both HIV-positive and HIV-negative women were infected with high-risk HPV types. The prevalence of Hr-HPV in HIV-negative (28.9%) and positive (55.6%) women in our study is similar (38.1% and 50.9%, respectively) to another study that included women from Tanzania but used clinician-collected HPV tests.[21] However, in contrast, our estimates of Hr-HPV are higher than those from a large study also conducted in Tanzania, which reported 17.2% and 46.7% for HIV-negative and HIV positive women, respectively.[23] Differences may be due to our smaller sample size and the fact our sample was younger and more women were from rural areas, both of which are associated with a higher prevalence of HPV. Other reasons could be the assay used, which can affect estimates of prevalence, as well as mode of collection (clinician versus self-collection).[23, 24]

Although no statistically significant differences were found when we compared sampling preference by HIV status, HIV-positive women had a significantly higher prevalence of positive Hr-HPV results and were more likely to test positive for multiple HPV types, which is consistent with findings from other studies. [23, 25] Taken together, these results suggest that self-collected samples can be successfully used for HPV testing to determine the presence of Hr-HPV types for both HIV positive and negative women. However, a limitation of our study is that we did not

Table 2. Number of study participants, stratified by HIV status, who indicated a 4 or higher on each of the acceptability indices for HPV self-sampling.

Response	HIV-Positive, n (%) (N=45)	HIV-Negative, n (%) (N=45)	Total, n (%) (N=90)
Good experience	40 (89.9)	41 (91.1)	81 (90.0)
Easy to do	43 (95.6)	42 (93.3)	85 (94.4)
Very convenient	42 (93.3)	43 (95.6)	85 (94.4)
Not embarrassing	43 (95.6)	42 (93.3)	85 (94.4)
No discomfort	41 (91.1)	44 (97.8)	85 (94.4)
Very confident	43 (95.6)	44 (97.8)	87 (96.7)

Table 3. Counts, Prevalence ratio and 95% confidence interval* for a positive HPV result based on HIV status.

		HPV status		
		HPV-positive	HPV-negative	Total
HIV status	HIV-positive	25	20	45
	HIV-negative	13	32	45
	Total	38	52	90

*Prevalence Ratio: 1.92 (95% CI: 1.13 - 3.26)

Figure 1. Flowchart for HPV testing that leverages VIA, which is currently widely available and used for cervical cancer screening in Tanzania, as well as genotyping (adapted from current WHO guidelines).[13]

HPV – human papillomavirus
VIA – visual inspection with acetic acid
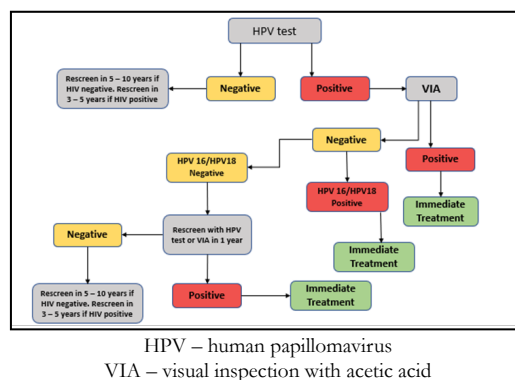
collect biopsies on women. Thus, while the higher prevalence of HPV in our sample suggests the ability of HPV testing to better detect high-grade disease and cancer, we were not able to determine test accuracy in relationship to biopsy-confirmed CIN 3 or cancer. This is an important consideration, especially for HIV positive women, given the higher prevalence of Hr-HPV and potential for overtreatment. Other limitations, as noted previously, are our relatively small sample size which limits our ability to detect significant differences based on demographic characteristics of our sample. In addition, our sample was recruited using convenience sampling, and included relatively young women from Dar es Salaam and hospitals in the surrounding Pawani region which limits comparability of our estimates of HPV prevalence to those expected from older populations and/or sampled from other regions across Tanzania.

Although self-collection was well accepted by both HIV positive and negative women and provided samples that were adequate for HPV testing, it is important to consider the effectiveness of self-collection as a cervical cancer screening tool in comparison to VIA, which has, until recently been recommended and used for screening in Tanzania. In our study, one participant was discordant with VIA-positive and HPV-negative results. This could be due to a false positive VIA test result, as found in Katanga et al. (2019) or the presence of non-HPV lesions.[24] The other participant who was VIA-positive also tested positive for Hr-HPV. Of note, this study found the majority of those who were HPV-positive were VIA-negative. This notable difference in test performance may be due to our small sample size, so should be interpreted with caution. However, in a large study of 3,767 women in Tanzania by Dartell et al. (2014), 4.5% of women were VIA positive compared to 20.1% who were Hr-HPV positive.[4] These results suggest that follow-up from a positive HPV result will not only add additional costs to the healthcare system but may also overburden clinics that have until now, provided VIA-based screening to a limited number of women. Use of VIA, which is widely available in Tanzania, as well as genotyping may help identify women who need immediate treatment and those who can undergo rescreening at a later date. An example algorithm, which is a slight variation on that proposed by the WHO is presented in Figure 1.[13] The effectiveness and cost-effectiveness of different algorithms that combine these options, especially for HIV-positive women and that can be implemented across Tanzania remains to be determined.

In conclusion this pilot study highlights the potential for HPV self-collection as a cervical cancer screening option in Tanzania- that could increase screening participation. In this study, all the self-collected samples were adequate for testing, and we were able to detect Hr-HPV types in both HIV-positive and HIV-negative women. Additionally, self-collection was well-accepted in women irrespective of HIV status. However, given the high Hr-HPV positivity compared to VIA positivity in this study, especially among HIV positive women, if confirmed in larger studies, countries such as Tanzania will need to consider how best to incorporate this approach to screening into their existing infrastructure to avoid over burdening healthcare facilities and providers.

Acknowledgment

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