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High Risk Human Papillomavirus (Hr-HPV) Prevalence and Genotypes Detected in Women ≥ 30 Years Old who have Never been Screened for Cervical Cancer in Harare, Zimbabwe

Raymond Chibvongodze[1*], Tafadzwa Dupwa[2], Elson Mberi[3], Chantal Nyirakanani[4] and Lucy Wangari Muchiri[5]

[1]Department of Medical Laboratory Sciences, University of Zimbabwe, P.O.Box A178, Avondale, Harare, Zimbabwe.
[2]Clinical Trial Research Centre, University of Zimbabwe, P.O.Box A178, Avondale, Harare, Zimbabwe.
[3]Department of Hematology, University of Zimbabwe, P.O. Box A178, Avondale, Harare, Zimbabwe.
[4]Faculty of Applied Sciences, University of Liege, Place du 20 Aout, Liege, Belgium.
[5]Department of Human Pathology, University of Nairobi, P.O.Box 19676-00202, Nairobi, Kenya.

Authors' contributions

This work was carried out in collaboration among all authors. Author RC designed the study and wrote the first draft of the manuscript. Author TD performed the statistical analysis. Authors EM and LWM reviewed the first draft. Author CN performed literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Background: Almost 50% of all cervical cancer cases are diagnosed in patients who have never been screened for cervical cancer before. There is an established cause - effect relationship between Hr-HPV infection and cervical cancer. Therefore, knowing the prevalence and high risk HPV genotype distribution in this group of women helps to formulate vaccination policies in a country. There is paucity of such information in Zimbabwean women.
Aim: To determine high risk HPV prevalence and genotypes in women who have never been

**Corresponding author: E-mail: rayscientist@gmail.com;*

screened for cervical cancer. To compare HPV positivity rates between women of ages' <30 and ≥30 years.
Study Design: Cross sectional descriptive study.
Place and Duration of Study: Cimas Medical Laboratories. Between January 2017 to December 2019.
Methodology: High risk HPV DNA screening was done using the Cepheid Xpert HPV qualitative screening test. Specimens positive for the pooled HPV subtypes were further typed using the HPV Genotypes 14 Real-TM Quant test kit to characterize specific subtypes. A t –test was used to compare the HPV positivity rates between women of ages' <30 and ≥30 years. A p-value <0.05 was regarded as statistically significant.
Results: The HPV positivity rates were 20% and 31% in women of ages' ≥30 and <30 years respectively. There was a statistically significant difference in HPV positivity between these two groups (p=0.03). HPV 52 was the most common subtype (15.8%) followed by HPV 18 (14.3%), HPV 16 (11.9%), HPV 35 (9.5%) and HPV33 & 58 which both contributed 8.3% of all subtypes detected.
Conclusions: Approximately 20% of women 30 years old and above have an HPV infection. HPV 52, 18 and 16 are the most common HPV subtypes in Zimbabwe.
Recommendation: HPV 52 should be included in the vaccines currently being used in Zimbabwe which are predominantly composed of HPV 16 and 18.

Keywords: Human Papillomavirus; cervical cancer; liquid based cytology; deoxyribonucleic acid.

1. INTRODUCTION

Squamous cell carcinoma of the cervix is the most common infection associated malignancy in women [1]. It ranks as the 3rd most prevalent cancer in women [1]. Approximately one third of a billion cervical cancer related deaths are recorded worldwide annually [2]. However, the figures are believed to be higher as most cervical cancer deaths go unreported in developing countries [2]. Developed countries witnessed an enormous decrease in cervical cancer due to organized and well executed Pap smear-based mass cervical cancer screening programs [3]. Most (>80%) of the global cervical cancer cases are reported in developing countries like Zimbabwe [2]. Cervical cancers recorded in Africa alone contribute one quarter of the global cervical cancer burden [3].

Human Papillomavirus (HPV) infection is necessary for cervical neoplasia [3]. HPV is the most common sexually transmitted virus with >75% of women having acquired the infection at some point in their lives [4]. Most of the infections are transient and are cleared by the host cell mediated immunity with 24 months [4]. HPV persist in only a minority of women with the ultimate result being, the abnormal proliferation of the cervical epithelium [4].

Based on epidemiological association with genital cancers HPVs are classified as high risk or low risk genotypes [3]. High risk genotypes have a higher oncogenic potential, and these include 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 68, 73 and 82 genotypes [3]. Low risk genotypes are rarely implicated in genital cancers and usually cause benign warts. These include 6, 11, 44, 54, 61, 70, 72, and 81 [3].

HPV is a DNA virus that belongs to the Papovaviral family [5]. It infects basal keratinocytes of the stratified squamous epithelium during sexual contact [5]. Infection sets into motion the expression of early genes (*E1, E2, E4, E5, E6 and E7)* that facilitate establishment and replication of the virus [6]. Replication proceeds via the latent and productive phases. The latent phase occurs when the HPV genome exists as an episome in the cellular cytoplasm and replicates in tandem with the host genome and thus, no new virions are produced [6]. In the productive phase, the HPV genome replicates independently resulting in the formation of numerous new virions which fill up the cytoplasm causing a cytopathic effect known as koilocytosis [6].

The integration of the HPV genome into the host genome results in cellular growth dysregulation (malfunction of the tumour suppressor gene p53 and elaboration of the cellular transcription factor E2F-1) which leads to increased proliferation, defective apoptosis and accumulation of cells with damaged DNA [5]. The increased proliferation of mutated cells without the decelerating effect of tumour suppressor genes results in a neoplastic transformation of the cells [6].

The most frequently used cervical cancer screening methods in Zimbabwe are visual inspection with acetic acid (VIA) and Pap smear cytology. HPV DNA testing is sparingly used as a primary cervical cancer screening test in Zimbabwe because of its prohibitive cost [1]. Currently HR-HPV DNA testing is mainly being used to triage patients with ASCUS/LSIL Pap smear results [7]. HPV DNA testing can be done using Polymerase Chain Reaction (PCR), In situ hybridization (ISH) and Hybrid Capture 2 (HC2) methods [6]. HR-HPV DNA testing has the major advantage of high reproducibility and high sensitivity [1]. However, it has a low positive predictive value (PPV) [1]. This is because most HPV infections are transient and thus there is a poor correlation with clinical disease [6]. Therefore, a positive HR-HPV result should be correlated with either the clinical picture or other tests such as Pap smear [6].

A few centres are performing HPV DNA testing in Zimbabwe because it is affordable to only a few. This has led to paucity of reliable information on the prevalence and frequency of genotypes of high-risk HPV in unscreened reproductive women in Zimbabwe. In response to this, this study aimed at investigating the prevalence and frequencies of HR-HPV genotypes in women ≥30 years in Zimbabwe. This information will be important in guiding cervical cancer screening and vaccination policies in Zimbabwe.

2. MATERIALS AND METHODS

2.1 Study Design

Cross-sectional descriptive study from January 2020 to March 2021.

2.2 Study Sites

Cimas Medical Laboratories**,** Harare, Zimbabwe.

2.3 Study Population

Women who came for cervical cancer screening at Cimas Healthcare Clinics.

2.4 Study Entry Criteria

2.4.1 Test group

Only women ≥30 years old that had no previous screening for cervical cancer were enrolled in this group.

2.4.2 Comparison group

Only women <30 years old that had no previous screening for cervical cancer were enrolled in this group.

2.5 Sampling Method

Consecutive sampling method

2.6 Sample Size

A total of 1446 women who fulfilled selection criteria were recruited in this study

2.7 Study Objectives

1. To determine high risk HPV prevalence and genotypes in women who have never been screened for cervical cancer.
2. To compare HPV positivity rates between women of ages' <30 and ≥30 years.

2.7.1 Hr-HPV DNA testing

Hr-HPV DNA screening was done using the Cepheid Xpert HPV qualitative test (CE IVD-Sunnyvale, CA 94089 USA). The Xpert qualitative test detects 14 high risk HPV types which were reported as HPV 16, HPV 18/45 and other HR HPV (31,33,35,39,51,52,56,58,59,66 and 68). Specimens positive for the pooled HPV subtypes were further typed using the HPV Genotypes 14 Real-TM Quant test kit to characterize specific subtypes.

2.8 Data Management

Patients eligible for the study were assigned a unique study number and the following data was captured: age, date of last menstrual period and any clinical symptoms noted during clinical examination. HR-HPV results and all prior data were stored in an IBM SPSS software version 21. Information stored in soft copies was protected from access from unauthorized persons by a password which was changed periodically. The data was analyzed used the IBM SPSS software version 21. Descriptive statistics were presented as proportions, tables and charts.

3. RESULTS AND DISCUSSION

3.1 Age Characteristics

A total of 1446 women of ages' ≥ 30 years were recruited in the study. A separate comparison

group of 205 women of ages <30 years was analyzed for comparison. Their age characteristics are summarized in Table 1.

3.2 Comparison of HPV Positivity between Women of ages' ≥ 30 years and < 30 years

Of the 1446 woman of ages' ≥ 30 years, 289 (20%) were positive for HPV DNA and 64 (31%) of the 205 women of ages' <30 years were positive for HPV DNA. A t –test was used to compare the HPV positivity rates between women of ages' <30 and ≥30 years. There was a statistically significant difference in HPV positivity between women of ages' ≥ 30 years and <30 years (p-value = 0.03). This data is illustrated in Table 2.

3.3 Number of HPV Subtypes Detected per Sample

Forty four (15.2 %) of the 289 HPV positive patients had multiple HPV subtypes detected from their samples. A total 336 HPV subtypes were detected from the 289 samples. The number of Hr-HPV subtypes detected per sample are summarized the Table 3.

3.4 Detected High Risk HPV Subtypes

All the high risk HPV subtypes on the HPV Genotypes 14 Real-TM Quant test kit (Sacace Biotechnologies - 44- 22100, Como, Italy) were detected in this study. Their frequencies are summarized in the Table 4.

3.5 Discussion

HPV infection is a prerequisite for cervical neoplasia [1]. Numerous high risk HPV subtypes are capable of initiating neoplasia after infecting the basal keratinocytes of the stratified squamous epithelium of the cervix [6]. This study aimed at investigating the prevalence of HPV as well the frequencies of the various HPV subtypes detectable from residual liquid based cytology samples from women above 30 years who have never been screened for cervical cancer. HPV is reported to have high rates of clearance in women below 30 years old [1]. Therefore, an HPV infection in women above 30 years is significant as it has a higher likelihood of causing cervical cancer [6]. This information is important in informing the formulation of HPV vaccines tailored to our population.

In this study which was done in Harare, 289 (20%) of the 1446 women were positive for HPV. This was comparable to a study done by Fitzpatrick et al in a rural population in Zimbabwe which reported an HPV positivity rate of 17% [8]. This confirms that our study findings are generalizable to any part of the country. However, the HPV positivity in this study were lower than that recorded by Mandishora and Sharita et al which reported HPV positivity rates of 72% and 43% respectively [9-10]. This difference can be explained by the enrollment of a higher proportion of HIV positive patients. Literature shows that HIV positive women are more likely to have an HPV infection, multiple HPV subtypes, more high-risk HPV subtypes and higher HPV viral load than HIV negative patients [1].

In this study, most HPV infections were in the age group 31-40 years. This differed from findings from a study by Sharita et al. which reported that most infections in the age group 18-25 years [10]. The difference can be due to the strict inclusion criteria in this study which strictly enrolled women of ages' ≥ 30

Table 1. Summary of the age characteristics of the study participants

Population	≥ 30 years	< 30 years
Number	1446	205
Mean age (years)	38.7	26.6
Age SD (years)	8.3	3.2
Age range (years)	30-79	18-29

Table 2. Independent t test analysis results for comparing HR-HPV positivity

Population	HR-HPV positivity rate	t-value	d.f	p-value
Age: ≥30 years	20%	634	204	0.03
Age: <30 years	31%			

Table 3. Number of HPV subtypes detected per sample

Number of subtypes	Number of samples	Relative frequency (%), n=289
1	245	84.8
2	41	14.2
3	3	1.0
Total	289	100

Table 4. Frequencies of high risk HPV subtypes

High risk HPV subtype	Number of subtypes	Relative frequency (%), n=336
16	40	11.9
18	48	14.3
31	5	1.5
33	28	8.3
35	32	9.5
39	5	1.5
45	10	3.0
51	21	6.3
52	53	15.8
56	17	5.1
58	28	8.3
59	6	1.8
66	16	4.8
68	27	8.0

years. A comparison of HPV positivity in women <30 years and women ≥ 30 years in this study showed that HPV positivity in women less than 30 years is statistically higher than HPV positivity in women 30 years and above. The difference is because most HPV infections in younger women are transient and are cleared by the immune system with two years before they reach 30 years old [11-12].

This study detected fourteen different HPV subtypes. HPV 52 was the most common subtype (15.8%) followed by HPV 18 (14.3%), HPV 16 (11.9%), HPV 35 (9.5%) and HPV33&58 which both contributed 8.3% of all subtypes detected. The most common subtypes detected in this study were consistent with findings by Mandishora et al and Fitzpatrick et al who reported HPV52, HPV 18 and HPV 16 as the most common subtypes [8-9]. These finding were significantly different from another study we did in Kenya which reported HPV 56 (40.7%), HPV 51 (22.2%) and HPV 68 (11.1%) as the most common subtypes [13]. These findings show that no single vaccine is capable of meeting the needs of all populations worldwide.

4. CONCLUSIONS

Approximately 20% of women of ages' ≥30 years have an Hr-HPV infection. HPV 52, 18 and 16 are the most common HPV genotypes in Zimbabwe.

5. RECOMMENDATION

HPV 52 should be included in the vaccines currently being used in Zimbabwe which are predominantly composed of HPV 16 and 18.

CONSENT

As per international standard or university standard, patients' written consent has been collected and preserved by the author(s).

ETHICAL APPROVAL

Ethical approval was obtained from the Joint Research Ethical Committee of University of Zimbabwe and Parirenyatwa Hospital (JREC), certificate number: JREC 4/2020. Permission was also granted by Cimas Medical Laboratories. During the study, strict patient confidentiality was observed. Cervical sample collection is a safe procedure. However, minor complications such as mild bleeding may be encountered in patients with cervicitis. Such spotting is usually self limiting and usually ends on its own in a few hours. Patients who received positive results were referred to gynecologists with the group for colposcopy and treatment.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Anderson J, Lu E, Sanghvi H, Kibwana S, Lu A SH. Cervical Cancer Screening and Prevention for HIV-Infected Women in the Developing World. Cancer prevention-from mechanisms to transitional benefits. 2012;231–4.
   Available:www.intechopen.com.
   Accessed on 12.03.19
2. Anorlu RI. Cervical cancer: the sub-Saharan African perspective. Reprod Heal Matters. 2008;16(32):41–9.
3. Global report: UNIAIDS report on the global AIDS pandemic. UNIAIDS; 2019.
   Available:https:// www.unaids.org.
   Accessed on 12.03.20
4. Lonky N, Mahdavi A, Wolde-Tsadik G, Bajamundi K, Felix J, et al. Evaluation of the clinical performance of high-risk human papillomavirus testing for primary screening. J Low Gen Tract Dis. 2010; 14(3):1089–2591.
5. Edmund S. Cibas. Cytology Diagnostic principles and Clinical Correlates. 3rd edition.Saunders Elsevier. Philadelphia. 2009;255-356
6. Willbur DA, Henry MR. College of American Pathologists Practical Guide to Gynecologic Cytopathology- Morphology, Management and Molecular methods. 1st edition. CAP Press. Illinois. 2008;1-274.
7. Wilbur DC, Nayar R. The Bethesda System for Reporting Cervical Cytology Definations, Criteriaand Explanatory notes. 3rd ed. Springer. 2015;89-109.
8. Fitzpatrick M, Mandishora RSD, Kartzenstein DA, McCarty K, Weber J, Chirenje ZM. HrHPV prevalence and type distrubution in rural Zimbabwe. A community based self collection study using near point of care GeneXpert HPV testing. Int J Infec Dis. 2018;82: 21-29.
9. Mandishora RSD, Christiansen I, Chin'ombe N, Duri K, Ngara B, Chirenje ZM. Genotypic diversity of anogenital HPV in women attending cervical cancer screening in Harare, Zimbabwe. J Med Vir. 2017;89:9.
10. Sharita D, Chirenje ZM, Blumenthal PD. Evaluation of a human papillomavirus assay in cervical cancer screening in Zimbabwe. BJOG.2000;107:33-38.
11. Carter JR, Ding Z, Rose BR. HPV infection and cervical disease: A review. Aust N Z J Obs Gy-naeco. 2011; 51(2):103–8.
12. Gomez DT SJ. Human Papilloma infection and cervical cancer: Pathogenesis and epidermiology. VA M. Guadalajara ; 2007: Formatex.
    Available:Citeseerx.ist.psu.edu/viewdoc/versons;jsessionid=F193788F. Accessed on 18.04.19
13. Chibvongodze R, Nyirakanani C, Ndung'u J, Ojwang J, Kyama CM. Human Papillomavirus DNA testing and Pap smear cytology co-testing as a 'test of cure' in patients previously treated for cervical lesions by LEEP at Kenyatta National Hospital. 2017;94:3.

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