



# Prevalence of HPV among HIV-negative women of child-bearing age in Lomé, Togo

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**Aim:** This study aimed to assess the prevalence and the distribution of HPV genotypes among HIV-negative women of child-bearing age in Lomé, Togo. **Materials & methods:** From April 2014 to September 2015, a cross-sectional study was conducted among HIV-negative women attending gynecological consultation in six health centers in Lomé. Cervical swabs were obtained from 324 women. HPV test was performed using HPV Direct Flow Chip. **Results:** The prevalence of any type and oncogenic HPV was 9.3 and 8.3%, respectively. A total of 13 different genotypes HPV, high risk (16, 18, 35, 45, 52, 53, 68, 82) and low risk (6, 40, 43, 44/65, 62/81), were found. **Conclusion:** Findings from this study provide essential insights for planning future public health strategies, including HPV vaccination programs.

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**Keywords:** HIV-negative women • HPV genotyping • human papillomavirus • Lomé, Togo • oncogenic HPV • prevalence

The association between cervical cancer and HPV is well known. HPV is the most common cause of cervical cancer, which is the second most frequent female cancer worldwide [1]. HPV is mainly transmitted through sexual contact and most people are infected with HPV shortly after the onset of sexual activity [2]. Cervical cancer is caused by sexually acquired infection with certain types of HPV [2,3]. In 2018, cervical cancer was the fourth most frequent cancer in women with an estimated 570,000 new cases worldwide and represented 7.5% of all female cancer deaths. Every year, more than 85% of cervical cancer-related deaths occur in less developed regions [2].

More than 200 genotypes of HPV have been described. Some HPV are classified as low-risk oncogenic HPV (LR-HPV), which cause papilloma and condyloma acuminata. High-risk oncogenic HPV (HR-HPV) are cancer causing [2]. HPV types 16 and 18 account for nearly 70% of all cases of invasive cervical cancer worldwide. This percentage rises to 90% for HPV6/11/16/18/31/33/45/52/58 [3]. Main factors associated with HPV are: multiple sexual partners, early sexual activity, prolonged oral contraception use, smoking, poor socio-economic conditions, associated sexually transmitted infections and HIV infection [3–5].

The burden of cervical cancer and HPV infection can be reduced and control strategies rely on HPV vaccination and early detection of benign or precancerous cervical lesions [5]. To date, three HPV vaccines are widely marketed internationally: Cervarix<sup>®</sup> (GlaxoSmithKline), Gardasil<sup>®</sup> (Merck), and Gardasil-9<sup>®</sup> (Merck), which protect against HPV 16/18, HPV 6/11/16/18 and HPV-6/11/16/18/31/33/45/52/58, respectively [6,7]. These genotypes, generally identified in more than 70% of cervical cancer cases worldwide, are still relevant in most African countries, while others not included in the vaccines have been found to be circulating and might be contributing to clinical conditions [8,9]. Thus, describing HPV type circulation in Togo is needed to inform vaccination program.

In Togo, cervical cancer is a public health problem and it is the second most common cancer in women [10], with an estimated mortality rate of 12.5% in 2018 in Togo [2]. Few data are available on circulating genotypes of HPV among key populations and women infected with HIV in the country [8,11,12]. However, no data are available on HPV prevalence in the general population, while Togo had received funding from the Global Alliance for Vaccines and Immunization to carry out a demonstration project for the administration of HPV vaccine, in two districts (Tchamba and Golfe) for 2 school years (2015–2016, 2017–2018). It is important to have essential data for planning future public health strategies, including HPV vaccination programs. The present study aimed to estimate the prevalence of HPV and to describe the distribution of HPV genotypes among HIV-negative women of child-bearing age in Lomé, Togo.

## Materials & methods

### Study design & setting

A cross-sectional study was conducted over a period of 18 months (from April 2014 to September 2015) among women attending gynecological consultation in six health centers in Lomé: three public health clinics (the department of Histology–Embryology of the teaching hospital ‘Centre Hospitalier Universitaire Sylvanus Olympio’, the ‘Centre Médical Social Agoe’ and the ‘Centre Médical Social Adidogomé’) and three private clinics (Alpia, Biasa and Wossinu-Gbogbo).

### Sample size & participants

We included all HIV-negative women, aged 18 years and older, who came for a gynecological visit during the study period. Women who gave their approval for the HPV cervical sampling and gave their informed written consent were recruited.

Since no data on HPV infection were available in the general population in Togo, the sample size calculation was based on the following assumptions: an expected prevalence of HPV infection of 30% based on estimates in a population of HIV-negative women of child-bearing age of 33% in Côte d’Ivoire [5], country of West Africa, with a precision of 6%, a significance level set at 5% and a non-response rate of 10%; the minimum sample size was estimated at 246 participants.

### Data collection

A standardized questionnaire was used to collect relevant sociodemographic, clinical and biological information during a face-to-face interview.

A cytobrush (Hospitex Diagnostics Srl) was used to collect the cells at the junction between the endocervix and the ectocervix and these cells were stored in a preservative solution provided by the manufacturer (Cytofast solution 42010600, Hospitex Diagnostics Srl, Sesto Fiorentino, Firenze, Italy). Cervical cells were transported and stored at ambient temperature (10–30°C) for a maximum time period of 5 days until manipulation at the ‘Laboratoire de Biologie Moléculaire et Immunologie’ of the ‘Faculté des Sciences de la Santé’ of the Université de Lomé.

For the HPV testing, specimens were analyzed by the HPV Direct Flow Chip system, which is intended for simultaneous detection and genotyping of 36 HPV types (high-risk HPV [16, 18, 26, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 68, 73 and 82] and low-risk HPV [6, 11, 40, 42, 43, 44, 54, 55, 61, 62, 67, 69, 70, 71, 72, 81, 84 and 89] [=CP6108]), by direct PCR, followed by reverse dot blot automatic hybridization, based on DNA-flow technology and colorimetric detection with the automatic e-BRID® System [13]. The amplification was carried out by PCR from crude-cell extracts, by using the PCR Mix and Phire Hot Start II DNA Polymerase (MAD-003930MU-P-E-30, Master Diagnostica, Granada, Spain). Amplicons were hybridized, using the optimized HPV Direct Flow Chip kit (MAD-003930M-H, Master Diagnostica) [14]. HPV chip membranes contained immobilized probes for hybridization control, beta-globin gene (for quality control of amplification), HPV-consensus sequences and genotype-specific HPV detection. The hybridization was performed automatically in sets of 15 samples using the e-BRID System (Master Diagnostica) [13,14]. HPV Direct Flow Chip had a sensibility of 100% and a specificity of 100% [15]. HPV Direct Flow Chip results were comparable with those of the other HPV tests like Linear Array HPV Genotyping Test, CLART® HPV2 and Digene® Hybrid Capture 2 HPV DNA Test [14].

### Statistical analyses

Data were entered into a Microsoft Excel database. The Chi-squared test or the exact Fisher test and the Mann–Whitney U test or the Wilcoxon test, were used for comparison of categorical and quantitative variables, respectively.

**Table 1. Socio-demographic, clinical and biological characteristics.**

Characteristics	N (n = 324)	%
<b>Age (years)</b>		
20–29	89	27.4
30–39	80	24.6
40–49	102	31.4
≥50	53	16.3
<b>Marital status</b>		
Lives alone	164	50.6
Married/cohabiting	160	49.4
<b>Education level</b>		
None or primary school	81	25.0
Secondary school	154	47.5
University	89	27.5
<b>Economic situation</b>		
IGA	206	63.5
No IGA	118	36.5
<b>Number of pregnancies</b>		
<2	197	60.8
≥2	127	39.2

IGA: Income generating activity; n: Number; %: The frequency for each data.

Proportions and odds ratio estimates were reported with their 95% CIs. Sample sizes and type, HPV prevalence and genotypes, and single- and multiple-infection profiles were described. STATA software version 14.1 (StataCorp, TX, USA) was used for the statistical analyses, which were performed at a 5% significance level. A logistic regression model was used for univariate and multivariate analyses.

### Ethical considerations

Ethical approval for this study was obtained from the ‘Comité de Bioéthique pour la Recherche en Santé (CBRS)’ (Bioethics Committee for Health Research) of the Ministry of Health of Togo (n°751/2014/MS/CAB/DGS/DPLET/CBRS). Participants provided written consent prior to participation and authorizations from the directors of selected medical centers were obtained before conducting this study.

## Results

### Socio-demographic & clinical characteristics of the participants

A total of 341 HIV-negative women attending ordinary gynecological consultations were invited to participate in the study. A total of 15 (4.4%) women refused to be sampled for the HPV test and were not included in the study. Two samples were damaged and were not analyzed. Finally, a total of 324 women were enrolled in this study. The median age of the study participants was 38 years old (interquartile range [IQR]: 29–47). Three-quarters (75%) of the study participants had at least secondary education and 49.3% were married or cohabiting. More than half of the women (60.8%) had fewer than two pregnancies (Table 1).

### Prevalence of HPV infection

HPV was found in 30 women, corresponding to a prevalence of HPV infection of 9.3% (95% CI: 6.3–12.39). HPV infection was more common in women 40–49 years of age (14/102; 13.7%), 20–29 years of age (9/89; 10.1%), with a secondary (16/154; 10.3%) or university (9/89; 10.1%) education level, with no income generating activities (15/118; 12.7%) and among those living alone (18/164; 10.9%). However, these findings were not statistically significant ( $p > 0.05$ ; Table 2).

The prevalence of HR-HPV was 8.3% (27/324) (95% CI: 5.6–11.9). HR-HPV were more common among women aged 40–49 years (12/102; 11.7%), 20–29 years (8/89; 8.9%), those living alone (16/164; 9.7%) and those with university degrees (9/89; 10.1%), but the differences were not statistically significant ( $p > 0.05$ ; Table 2).

**Table 2. Socio-demographic, clinical and biological characteristics according to HPV type.**

Characteristics	Total (n = 324)	‡HPV+ (n = 30)			§HR-HPV+ (n = 27)			¶LR-HPV+ (n = 7)		
	n	n	%	p-value	n	%	p-value	n	%	p-value
<b>Age (years)</b>				0.126			0.273			0.433
20–29	89	9	10.1		8	8.9		3	3.3	
30–39	80	3	3.7		3	3.7		0	0.0	
40–49	102	14	13.7		12	11.7		3	2.9	
≥50	53	4	7.5		4	7.5		1	1.8	
<b>Marital status</b>				0.374			0.461			0.513
Lives alone	164	18	10.9		16	9.7		4	2.4	
Married/cohabiting	160	12	7.5		11	6.8		3	1.8	
<b>Education level</b>				0.540			0.648			0.324
None or primary	76	5	6.1		5	6.1		0	0.9	
Secondary	138	16	10.3		13	8.4		4	3.5	
University	80	9	10.1		9	10.1		3	3.7	
<b>Economic situation</b>				0.154			0.125			0.500
IGA†	206	15	7.2		13	6.3		4	1.9	
No IGA	118	15	12.7		14	11.8		3	2.5	
<b>Number of pregnancies</b>				0.200			0.204			0.436
<2	197	22	11.1		20	10.1		5	2.5	
≥2	127	8	6.2		7	5.5		2	1.5	

p >5% indicates no statistical difference.  
 HPV +: Women infected with HPV; HR-HPV+: Women infected with high-risk HPV; LR-HPV: Women infected with low-risk HPV; n: Number; %: The prevalence in each specified category.

Among HPV-infected women, HR-HPV was observed in 90.0% (27/30) and LR-HPV in 23.3% (7/30) (Table 2).

### HPV genotypes distribution

Among the 30 women infected with HPV, HPV18 (15/30; 50%) was the most common genotype identified, followed by HPV82 and HPV45, 62/81 (3/30; 10%). Concomitant infection with both HPV16 and HPV18 was observed in one woman (1/30; 3.3%). However, infection with HPV11 or concomitant infection with HPV6 and HPV16 or HPV18 was not observed.

A total of 13 different HPV genotypes HR-HPV (16, 18, 35, 45, 52, 53, 68, 82) and LR-HPV (6, 40, 43, 44/65, 62/81) were detected (Figure 1).

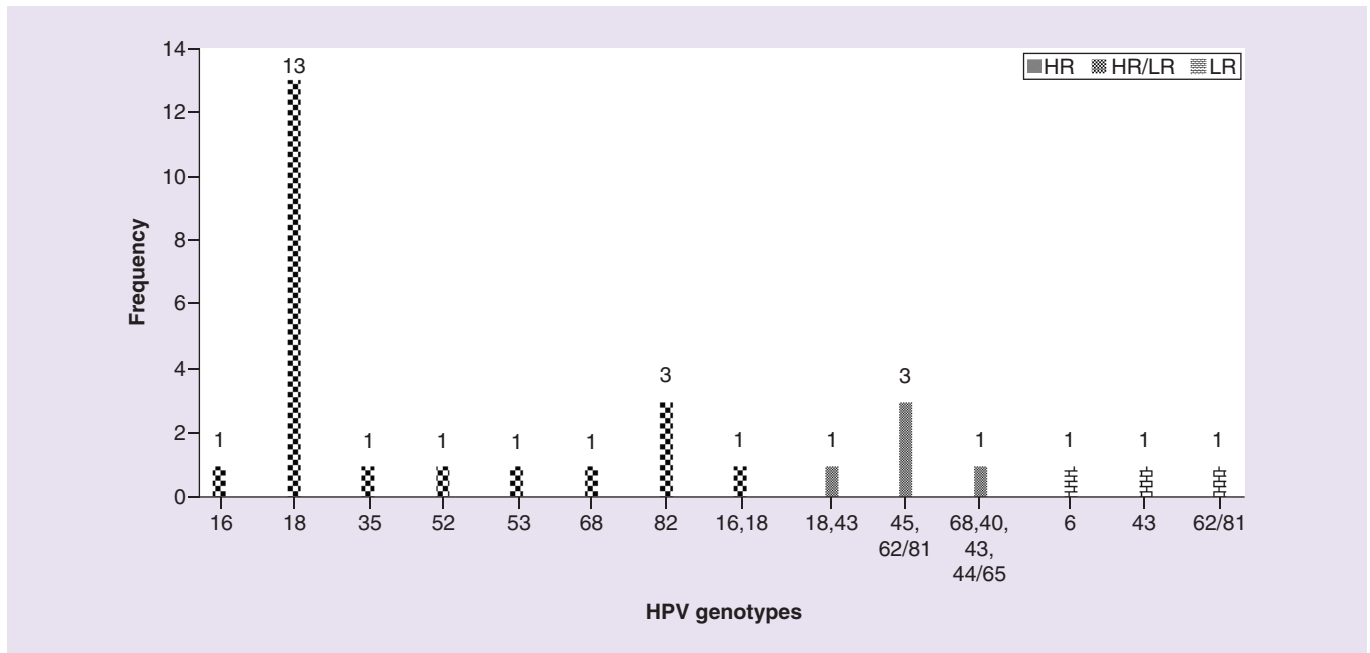
Single and multiple infections were observed in 80% (24/30) and 20% (6/30) of participants, respectively. Among women infected with HR-HPV, 77.7% (21/27) had a single infection (16, 18, 35, 52, 53, 68 and 82), 3.7% (1/27) had an infection with two HR-HPV (16, 18) and 23.8% (5/21) had one HR-HPV and at least one LR-HPV (40, 43, 44/65, 62/81). Of oncogenic strains identified, the Cervarix® and Gardasil-9® vaccines covered two strains for 53.3% and four strains for 70% of the participants, respectively.

### Factors associated with HR-HPV infection

In univariate analyses, none of the explanatory variables were associated with HR-HPV infection (Table 3).

### Discussion

In the present study, the prevalence of any-type HPV was relatively low (9.3%) and this is comparable to the overall prevalence reported in Ghana (10.7%), in Egypt (10.4%, 10.3%) and Northern Africa (10.9%) [16,17]. It is one of the lowest prevalence reported in sub-Saharan Africa. Indeed, in a meta-analysis of 1 million women with normal cytological findings published in 2010, Bruni *et al.* reported a prevalence of 24% in sub-Saharan Africa [17]. This prevalence is also low compared with the prevalence found in other West African countries, with 33.2% in Benin, 24.3% in Burkina Faso and 50.3% in Côte d'Ivoire [5,9,18]. This difference could be explained by many factors



**Figure 1. Human papillomavirus genotypes.**  
HPV: Human papillomavirus; HR: High risk; LR: Low risk.

**Table 3. Factors associated with HR-HPV infection.**

Characteristics	‡HR-HPV infection				Univariate analysis	
	No		Yes		OR	p-value
	N	%	N	%		
<b>Age (years)</b>						0.683
<35	122	41.1	10	37.0	1	
≥35	175	58.9	17	63.0	1.19	0.53–2.77
<b>Marital status</b>						0.350
Lives alone	148	49.8	16	59.3	1	
Married/cohabiting	149	50.2	11	40.7	0.68	0.30–1.51
<b>University level</b>						0.477
Yes	80	26.9	9	33.3	1	
No	217	73.1	18	66.7	0.74	0.33–1.78
<b>Economic situation</b>						0.086
IGA	193	65.0	13	48.1	1	
No IGA	104	35.0	14	51.9	2.00	0.90–4.46
<b>Number of pregnancies</b>						0.146
<2	177	59.6	20	74.1	1	
≥2	120	40.4	7	25.9	0.52	0.20–1.21

p >5% indicates no statistical difference.  
%: The prevalence in each specified category; HR-HPV: High-risk HPV; IGA: Income generating activities; n: Number; OR: Odds ratio.

such as the laboratory technique used, the age and HIV status of the population. In our population, all women were HIV-negative.

In our study, two population groups aged 20–29 years (10.1%) and 40–49 years (13.7%) had higher prevalence of HPV. Similar findings, with both peaks of age, have been documented in other studies [11,17,19].

The prevalence of HR-HPV was 8.3%. HR-HPV were more common among women aged 40–49 years (11.7%), living alone (9.7%) and with a university degree (10.1%). In Togo, high prevalence of HR-HPV has been previously

reported in key populations, with 44% among men having sex with men, 32.9% among female sex workers and 16.7% in HIV-infected women [8,11,12].

A total of 13 different HPV genotypes HR-HPV (16, 18, 35, 45, 52, 53, 68, 82) and LR-HPV (6, 40, 43, 44/65, 62/81) were found in the study's population. The identification of different genotypes, proves that there is a cocirculation of multiple HPV types in Lomé, Togo. Among the 30 HPV infected women, the most common HPV types were HPV18 (50%), HPV82 (10%) and HPV45 (10%). This distribution of HPV in Togo is consistent with literature data, in the general population for early diagnosis. HPV16 is the most common reported genotype in cancer or precancerous lesions [17–19]. Our results align with findings reported in Burkina Faso and in Ghana, neighboring countries in Togo, which found more HPV18 than HPV16 [9,20,21].

HPV35 found in our study was also found among men having sex with men and female sex workers in Togo, and in women of child-bearing age in Abidjan, Côte d'Ivoire [5,8,12]. Therefore, it is important to identify circulating genotypes for better management and protection of populations against HPV infections.

Some factors that could be responsible for these variations include the type of assay used, differences in population, multiple HPV infections and varying exposures of individuals to different risk factors [5]. Thus, the comparison of the prevalence between studies should be made with caution.

There are more single HPV infections than multiple HPV infections in this study. Infection with multiple HPV genotypes, which was found to be associated with an increased risk of HPV persistence [22], was relatively low in the present study. Furthermore, these findings are comparable to those of Ouedraogo and Traore *et al.* in Burkina Faso, Piras *et al.* in Benin and Krings *et al.* in Ghana [9,18,19,21]. Low detection of multiple HPV infections could be influenced by the specificity of the tests used for screening or diagnosis, especially if the test does not cover most genotypes circulating in the geographical area. In our study, we used a PCR and hybridization to detect the HPV DNA (18 HR and 18 LR-HPV) [14].

In this study, neither HR-HPV nor any type HPV infection was associated with age, marital status, education, economic situation and the number of pregnancies. This could be explained by the low prevalence of HPV reported in our study. However, the prevalence of genital HR-HPV is also directly related to some risk factors [2,22]. These risk factors could be responsible for HPV acquisition, persistence and development of cervical cancer. These risk factors can be early onset of sexual activity, low socioeconomic status, illiteracy, multiple sexual partners, marital status, alcoholism, smoking and use of oral contraceptives [5,9,19,22]. People who are immunocompromised such as those living with HIV or who have coinfection with other sexually transmitted agents are also more likely to have persistent HPV infections and a more rapid progression to precancer and cancer [5,8,12]. In a previous study among HIV infected women, a prevalence of 22.2% was reported in Togo [11].

This study presents some limitations. First, we recruited participants in clinical setting and findings from our study may not be generalized to all HIV-negative women of child bearing age in Togo. Second, cytological analyses of cervical swabs were not available to assess the association between HPV infection and clinical lesions. Third, as this study focuses more on the prevalence of HPV, sexual factors were not documented, such as the age at first intercourse, sexual partners and use of hormonal contraceptives. Hence, we did not have a broad set of factors for the analysis of risk factors.

Despite these limitations, to our knowledge, this was the first study reporting the prevalence of HPV infection and its genotype distribution among HIV-negative women of child-bearing age in Lomé, Togo. The previous studies were conducted among key populations (men who have sex with men and female sex workers). Further studies should be carried out among adolescents in order to have a complete distribution of HPV in Togo.

## Conclusion

In our study, in HIV-negative women of child-bearing age in Togo, we found that one in ten women has an HPV infection. It is advisable to look for possible cross-reactions of the vaccine strains used in order to better protect the population against strains circulating in the country. Therefore, we strongly call for a program to screen for HPV in Togo. It is also urgent to introduce vaccines against HPV and propose a program against cervical cancer in Togo.

## Author contributions

All authors gave input or revised the final manuscript critically. All authors read and approved the final manuscript and take public responsibility for its content. Additional contribution by authors: conceptualization was performed by YT Nyasenu, FA Gbeasor-Komlanvi, A Ehlan, DK Ekouevi and A Dagnra. Data curation was performed by YT Nyasenu, FA Gbeasor-Komlanvi, SA-R Issa, MK Tchankoni and BM Yambiyo. Formal analysis was performed by YT Nyasenu, FA Gbeasor-Komlanvi, MK Tchankoni, BM Yambiyo



and DK Ekouevi. Funding acquisition was performed by M Prince-David and A Dagnra. Investigation was performed by YT Nyasenu, FA Gbeasor-Komlanvi, DK Ekouevi and A Dagnra. Methodology was performed by YT Nyasenu, A Ehlan, FA Gbeasor-Komlanvi, MK Tchankoni, BM Yambiyo, M Salou and DK Ekouevi. Project administration was performed by YT Nyasenu, A Ehlan, M Prince-David and A Dagnra. Supervision was performed by A Dagnra, M Salou, A Ehlan and DK Ekouevi. Validation was performed by YT Nyasenu, FA Gbeasor-Komlanvi, A Ehlan, M Prince-David, M Salou, DK Ekouevi and A Dagnra. Visualization was performed by A Ehlan, FA Gbeasor-Komlanvi, DK Ekouevi and A Dagnra. YT Nyasenu, FA Gbeasor-Komlanvi and DK Ekouevi wrote the original draft. Writing, reviewing and editing was performed by YT Nyasenu, FA Gbeasor-Komlanvi, DK Ekouevi, A Ehlan, M Salou, M Prince-David and A Dagnra.

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### Financial & competing interests disclosure

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No writing assistance was utilized in the production of this manuscript.

### Ethical conduct of research

The authors state that they have obtained appropriate institutional review board approval or have followed the principles outlined in the Declaration of Helsinki for all human or animal experimental investigations. In addition, for investigations involving human subjects, informed consent has been obtained from the participants involved.

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### Summary points

- HPV has been recognized as being involved in 99.7% of cervical cancer. Cervical cancer is the second most frequent female cancers worldwide and in Togo, with an estimated mortality rate of 12.5% in Togo in 2018. Limited data on HPV are available among HIV-negative women in Togo.
- A cross-sectional study was conducted from April 2014 to September 2015, among HIV-negative women attending gynecological consultation in six health centers in Lomé. Cervical swabs were collected among women who gave their informed consent to participate in the study. HPV test was performed using HPV Direct Flow Chip.
- A total of 324 women were included.
- The prevalence of any type and oncogenic HPV was 9.3 and 8.3%, respectively.
- Among the 30 women infected with HPV, HPV18 (15/30; 50%) was the most common genotype identified, followed by HPV82 (3/30; 10%) and HPV45 (3/30; 10%).
- A total of 13 different genotypes HPV, high risk (16, 18, 35, 45, 52, 53, 68, 82) and low risk (6, 40, 43, 44/65, 62/81), were detected.
- Single and multiples infections were observed in 80% (24/30) and 20% (6/30) of participants, respectively.
- In univariate analyses, there are no socio-demographic and clinical factors associated with high-risk HPV.
- Findings from this study provide essential insights for planning future public health strategies and for the HPV vaccination program.

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