Original Article

Prevalence of precancerous cervical lesions and high-risk human papillomavirus types in Yaounde, Cameroon

Richard Tagne Simo[1], Arsène Godlove Djoko Nono[1], Hervet Paulin Fogang Dongmo[2], Paul Faustin Seke Etet[2], Bertrand Kiafon Fonyuy[3], Armel Hervé Nwabo Kamdje[2], Nicolas Njintang Yanou[4], Phélix Bruno Telefo[5]

[1] *Department of Biomedical Sciences, Faculty of Science, University of Ngaoundere, Cameroon*
[2] *Department of Physiological Sciences and Biochemistry, FMBS, University of Ngaoundere, Garoua, Cameroon*
[3] *Anato-Cytopathology Laboratory, Centre Hospitalier Dominicain Saint Martin de Porres, Yaounde, Cameroun*
[4] *Department of Biological Sciences, Faculty of Sciences, University of Ngaoundere, Ngaoundere, Cameroon*
[5] *Department of Biochemistry, Faculty of Science, University of Dschang, Cameroon*

Abstract

Introduction: Various Human papillomavirus (HPV) types cause cervical cancer, and represent the primary cause of cancer death in Africa and the second cause of most common cancers in Cameroon. Herein, we determined the prevalence of high-risk HPV types in women and associated cervical cytologic abnormalities in Yaounde, Cameroon.

Methodology: A cross-sectional study targeting HPV-positive women aged 20 and over was conducted between March and June 2020 at the Saint Martin de Porres' Health Centre in Yaounde. HPV tests were performed by PCR for detection of HPVs 16, 18, 33, and 45. The test was performed on 616 women using exfoliated cell specimens; then, we processed on cytological diagnosis with Pap smears on HPV positive specimens.

Results: The HPV types tested were detected in 137 participants, of which 38.7% with multiple HPV infections, and the remaining part with single HPV infections of type HPV 16 (28.5%), HPV 18 (17.5%), HPV 33 (10.2%), and HPV 45 (5.1%). Cervical cytologic abnormalities were found in 69.34% of participants including: LSIL (49.63%), HSIL (15.32%), ASC-US (3.66%) and AGC (0.73%). Co-infections with HPV 16 and HPV 18 were significantly associated with HSIL ($p = 0.001$) lesions, while HPV 45 was more common in participants with normal cytology ($p = 0.001$). Cervical lesion occurrence was significantly associated with the number of sexual partners ($p = 0.02$) and history of oral contraceptive pill use ($p = 0.001$).

Conclusions: Our results suggest that HPV 16 and 18 are predominant in Yaounde, and are associated with more severe precancerous lesions.

Key words: Cervical cancer; HPV; genotyping; co-infection.

Introduction

Persistent infections with high-risk human papillomavirus (HR-HPV) induce the development of cervical precancerous and cancerous lesions [1]. HR-HPV infections are the most common cause of cervical cancer, the third most frequent gynecological cancer worldwide [2]. Nearly 85% of the disease burden lies in low/middle-income countries, where cases are detected at an advanced stage due to inadequate screening, lack of awareness and screening campaigns and prophylactic HPV vaccination programs [3,4]. Cervical cancer is the leading cause of cancer death in Africa and the second cause of most common cancers in Cameroon regardless of gender, due to poor access to standard treatment [3]. Papillomaviruses generate productive infections within the stratified epithelia of the skin, oral cavity and anogenital tract. Infections of basal epithelial cells initiate the viral life cycle. Five HPV types (HPVs 16, 18, 31, 33 and 45) would mainly account for squamous cell carcinoma (SCC) (80% of cases) and adenocarcinoma (ADC) (94% of cases) of the cervix [5]. HPV 16 and 18 would be the most active types in cervical cancer development worldwide and will account for more than 70% of cervical cancer cases, partly thanks to their special abilities to escape immune surveillance [4]. Fortunately, HPV vaccines were developed, such as: (*i*) Cervarix, a bivalent recombinant vaccine targeting HPV 16 and 18; (*ii*) Gardasil, which is used in the prevention of HPVs 6, 11, 16, and 18; and (*iii*) the nanovalent vaccine Gardasil 9 used to prevent

infections with HPVs 6, 11, 16, 18, 31, 33, 45, 52, and 58. Vaccination against HR-HPV is the most recommended preventive measure. However, immune protections induced by HPV vaccines are strain-specific [6], and the distribution of HPV varies with population and geographical areas [7]. Thus, it would be advisable to assess the distribution of HR-HPV types in the female population, particularly in women with cervical precancerous or cancerous lesions, before introducing an HPV vaccine. Since Cameroon health authorities are planning to introduce an HPV vaccine to the national immunization program, the aim of this study was to characterize the high-risk HPV types present in women and associated cervical cytologic abnormalities in Yaounde, the political capital and second largest city of Cameroon.

Methodology

Participants and Study procedure

The study was performed between March and June 2020 at the Saint Martin de Porres' Health Centre (SMPHC) Yaoundé. A total of 616 women were recruited to the study. Women aged 20 and over, sexually active, HPV-positive with or without cancerous lesions were included. Women previously diagnosed with cervical cancer, bleeding on the sampling day, inflammatory or hemorrhagic smears and hysterectomized women were excluded from the study. Qualified women for the study were informed on the reason, detailed information and procedures associated to the study. Only women willing to join the study and to sign the information consent form were included. Sociodemographic information was obtained, and cervical samples collected. The study protocol and procedures were approved by the National Committee on Research Ethics for Human Health, Yaounde, Cameroon (2020/12/81/CE/CNERSH/SP), and the study was performed in accordance with international ethical standards (Declaration of Helsinki and amendments). All participants were informed of their right to withdraw from the study at any time. Data were anonymized and kept confidential.

Cervical Specimen Collection and cytology

Cervical specimens were collected by a gynecologist using standard clinical procedures. Briefly, two cervical exfoliated cell specimens were collected for HPV DNA genotyping assays and liquid-based cytological diagnosis, with a cytobrush and an Ayres spatula. Samples Slides were prepared using a liquid-based cytology (BD SurePath™ liquid-based Pap test) method. Cytological classifications of precancerous lesions were performed on slides and classified according to the Bethesda 2014 system [8]: (*i*) squamous cells abnormalities (Atypical squamous cells (ASC-US and ASC-H); low-grade squamous intraepithelial lesions (LSIL); high-grade squamous intraepithelial lesions (HSIL) and squamous cell carcinoma) and (*ii*) glandular squamous cell abnormalities (atypical glandular cells (AGC) and adenocarcinoma).

DNA extraction and amplification

1 mL of cervical specimen was added to 200 μL of proteinase K solution and the mixture was transferred to a mixing Block MB-182 (BIOER) for DNA extraction. HPV DNA detection was performed using polymerase chain reaction (PCR) with degenerate primers MY09/11, which amplify fragments containing 450 bp from the L1 gene of a wide spectrum of HPV types. Then, typing was performed on HPV positive specimens using type-specific PCR, with primers targeting E6 region of HPVs 16, 18 and 33, and E7 region of HPV 45 (Table 1) [9]. Reactions were performed with the mixture of 25 μL solution (containing 100 ng of DNA, 1.5 mM of MgCl$_2$, 50μM of each dNTP, 20 pmol of each primer, and 1 unit of Taq DNA Polymerase and 1X buffer) and 5 μL of DNA extracted. PCR cycling steps (conditions) were: initial denaturation (95°C, 5 min), denaturation (35 cycles, 95°C, 30 sec), annealing (55°C, 1 min), elongation (72°C, 2 min), and final extension (72°C, 10 min). A negative control (without HPV DNA), and a positive control (Ca Ski cell DNA, HPV 16+) were introduced in each series. PCR products were analyzed on 2%

Table 1. Primer sequences used for High-Risk HPV genotyping.

HPV type	Primer sequences (5'-3')
HPV 16	GTTTGCAGCTCTGTGCATA CATTTTATGCACCAAAAGAGAACTGCAATG
HPV 18	GTGTTCAGTTCCGTGCACA TGAGAAACACACCACAATACTATGGCGCGC
HPV 33	GTCTCCAATGCTTGGCACA CATTTTGCAGTAAGGTACTGCACGACTATG
HPV 45	CCCACGAGCCGAACCACAG TCTAAGGTCCTCTGCCGAGC

agarose gel stained with ethidium bromide and visualized by UV transillumination.

Data analysis

Data were analyzed using SPSS® Statistics 23 software (IBM, Armonk, New York). Proportions were compared using χ^2 test for proportions. Differences with $P < 0.05$ were statistically significant. Data were presented as frequencies of distribution and proportions.

Ethical Approval

The study protocol and procedures were approved by the National Committee on Research Ethics for Human Health, Yaoundé, Cameroon (2020/12/81/CE/CNERSH/SP), and the study was performed in accordance with international ethical standards (Declaration of Helsinki and amendments). All participants were informed of their right to withdraw from the study at any time. Data were anonymized and kept confidential.

Table 2. Sociodemographic characteristics of the study participants.

Variables	N (%)
Age groups	
[20-29]	31 (22.63)
[30-39]	46 (33.57)
[40-49]	33 (24.08)
[50-59]	26 (18.98)
[60-65]	1 (0.7)
Marital status	
Single	65 (47.4)
Married	71 (51.8)
Widow	1 (0.7)
Puberty onset	
Normal	49 (35.76)
Late	88 (64.23)
Number of sexual partners	
[1-5]	67 (48.9)
[6-10]	54 (39.4)
[11-15]	14 (10.2)
[16-20]	1 (0.7)
History of abortion	
No	42 (30.6)
Yes	95 (69.3)
Parity	
Nulliparous	45 (32.8)
Primiparous	31 (23.3)
Pauciparous	32 (22.6)
Multiparous	22 (16.0)
Large multiparous	6 (4.3)
History of OCP use	
No	101 (73.7)
Yes	36 (26.2)
HIV status	
Negative	99 (71.3)
Positive	36 (27.2)

Results

Sociodemographic characteristics

Of the 616 women recruited to the study, 149 were HPV+. Cases with inflammatory or hemorrhagic smears (12 women) were excluded, reducing the sample size of HPV+ women to 137. Table 2 presents the sociodemographic characteristics of the participants of the study. Most HPV+ women were between 20 to 49 years old (80.28%). The participants were also distributed as HIV- (71.3%), with history of abortion (69.3%), Oral Contraceptive Pill (OCP) use (73.7%), married (51.8%), in monogamous marriages (69.9%), with late menarche (64.23%) and between 1 and 10 sexual partners (88.3%).

Distribution of HPV types

Figure 1 shows the distribution of HPV types among participants. Infections with HPV 16 represented 58.39% (with 51.25% of co-infections), HPV 18 represented 46.71% (62.50% of co-infections), HPV 33 represented 27.01% (62.16% of co-infections), and HPV 45, 12.41% (58.82% of co-infections).
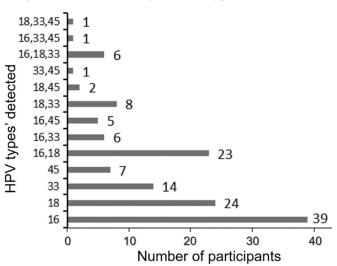
Precancerous cervical lesions

Of the 137 samples tested, 68 (49.63%) were classified as LSIL, 42 (30.6%) as NILM (Negative for intraepithelial lesion or malignancy), 21 (15.3%) as HSIL, 4 (2.9%) as ASC-US, and single cases of ASC-H and AGC (0.7%) were also observed (Figure 2).

HPV types and precancerous cervical lesions

Table 3 presents the associations between HPV types and precancerous cervical lesions. Mono-infections with HPV 16 were more common in cases with normal cytology (46.15%). On the other hand,

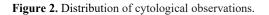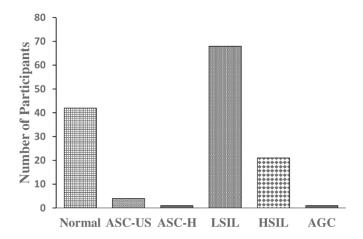**Figure 1.** Distribution of high-risk HPV types.

HPV 16 and 18 co-infections were found mostly in LSIL and HSIL cases , but were only significantly associated with HSIL lesions (OR: 5.4, P-Value=0.001)Instead, HPV18 as well as HPV 33, whether taken as a single or co-infection, were not significantly associated with any lesions.in mono-infection (HPV 18) and both mono-infection and multiple infection (HPV 33). All HPV 45 mono-infections were markedly associated with normal cytology (P-Value=0.001).

Risk factors, types of infection and precancerous lesions

Table 4 presents the influence of the risk factors studied on the occurrence of cervical lesions. It shows that both number of sexual partners and pill use have a significant association with the occurrence of precancerous cervical lesions, with P-Values of 0.02 and 0.001, respectively. Similarly, women using the pills were more likely (91.6%) to have precancerous lesions. In addition, the number of women with multiple infections increased with the number of sexual partners (*p*-Value=0.0001). The majority of HIV+ women presented precancerous lesions compared to HIV- women; therefore, a significant association was found between serological status and type of HPV infection (single/multiple infection), *p*-Value=0.04.

Figure 2. Distribution of cytological observations.

AGC: atypical glandular cells. ASC-US: atypic squamous cells of undetermined significance. ASC-H: atypic squamous cells cannot exclude high grade. LSIL: low grade squamous intra-epithelial lesion. HSIL: high grade squamous intra-epithelial lesion.

Table 3. Associations between HPV types and precancerous cervical lesions.

Lesions	HPV single infections				HPV Multiple infections									
	16	18	33	45	16,18	16,33	16,45	18, 33	18,45	33,45	16,18,33	16,33,45	18,33, 45	Total
None														
χ2	6.16	0.43	2.74	16.68	/	0.57	0.27	1.31	/	2.27	/	/	/	
P-Value	0.01*	0.5	0.09	0.00**	/	0.44	0.59	0.25	/	0.13	/	/	/	
Number	18	6	7	7	0	1	1	1	0	1	0	0	0	42
ASC-US														
χ2	0.93	3.009	/	/	/	/	/	/	/	/	/	/	/	
P-Value	0.33	0.08	/	/	/	/	/	/	/	/	/	/	/	
Number	2	2	0	0	0	0	0	0	0	0	0	0	0	4
ASC-H														
χ2	/	/	/	/	/	/	/	/	/	/	21.9	/	/	
P-Value	/	/	/	/	/	/	/	/	/	/	0.000	/	/	
Number	0	0	0	0	0	0	0	0	0	0	1	0	0	1
LSIL														
χ2	1.61	0.239	0.001	/	1.39	0.002	0.22	2.18	0.00	/	2.85	/	/	
P-Value	0.204	0.625	0.97	/	0.23	0.98	0.63	0.139	0.99	/	0.09	/	/	
Number	16	13	7	0	14	3	3	6	1	0	5	0	0	68
HSIL														
χ2	2.44	0.17	/	/	12.06	1.56	0.08	0.05	/	/	/	5.56	5.56	
P-Value	0.11	0.67	/	/	0.00**	0.21	0.76	0.81	/	/	/	0.01	0.01	
Number	3	3	0	0	9	2	1	1	0	0	0	1	1	21
AGC														
χ2	/	/	/	/	/	/	/	/	67.9	/	/	/	/	
P-Value	/	/	/	/	/	/	/	/	0.00	/	/	/	/	
Number	0	0	0	0	0	0	0	0	1	0	0	0	0	1
Total	39	24	14	7	23	6	5	8	2	1	6	1	1	137

*Correlation is significant at the 0.05 level (bilateral); **Correlation is significant at the 0.01 level (bilateral); AGC: atypical glandular cells; ASC-US: atypic squamous cells of undetermined significance; ASC-H: atypic squamous cells cannot exclude high grade; LSIL: low grade squamous intra-epithelial lesion; HSIL: high grade squamous intra-epithelial lesion.

Discussion

Of 137 included participants, 38.7% presented multiple HPV infections, and the remaining presented single infections (HPV 16, HPV 18, HPV 33, and HPV 45). Precancerous lesions were found in 69.34% of participants, including low-grade squamous intra-epithelial lesion, high-grade squamous intraepithelial lesion, atypical squamous cells, and atypical glandular cells.

HPV 16, and in a lesser extent, HPV 18 are predominant and mainly found in co-infections with other HPV types. It is well-established that HPV 16 and 18 are the most common HPV types worldwide, accounting for more than 70% of cervical cancer cases, as well as other cancers associated with HPV infections [10–12]. The predominance of HPV 16 infections over other HPVs may result partly from the special ability of this HPV type to escape immune surveillance and become more virulent [4]. Our findings are in consistence with data indicating that HPV 16 infections may facilitate other HPV infections, particularly HPV 18 infections. Indeed, HPV 16 and 18 co-infections accounted for 54.72% of co-infections herein. The present hypothesis is however deserving further investigations, considering the potential to develop more efficient vaccines and therapeutics against HPV-associated cancers.

Precancerous lesions were observed in 69.3% of HPV+ participants. This is alarming considering the range of median age of participants (30 to 40 years), and the age of all the participants (22 to 64 years old). This finding highlights the need for raising the awareness of women, public health authorities, and other stakeholders about HPV role in cervical cancer development for urgent setup of HPV vaccination campaigns in Cameroon. The peak of HPV infections was found in women aged between 20 and 39. This observation correlates with many previous reports; It is widely accepted that higher infection rates in younger women may result from higher sexual activity, particularly between 25 and 35, which may increase their risk of exposure to HPVs [13,14]. On the same trend, in our study, the number of sexual partners was significantly associated with both infections with a type of HPV and the development of precancerous lesions. Women who had had more than 5 sexual partners were 3.27 x more likely to develop a precancerous lesion and 3.20x more likely to have multiple HPV infections compared to those who had had less partners. Other groups of women arguably with history of intense sex life, namely HIV+ and women with history of oral contraceptive pill (OCP) use, were also more likely to develop multiple HPV infections: 2.17x more for HIV+ women compared to HIV- and 6.9x for women with history of OCP compared to those using other

Table 4. Influence of risk factors on the type of lesion.

Variables	Lesions				Type of infection			
	No	Yes	P-value	OR (95% CI)	Mono-inf.	Co-inf.	P-value	OR (95% CI)
Puberty								
Normal	34.69	65.3	0.44		59.20	40.80	0.71	
Late	28.4	71.59			62.50	37.50		
Sexual Partners, N								
[1-5]	42.64	57.35		1.00 (Referent)	76.47	23.53		1.00 (Referent)
[6-10]	20.37	79.60	0.02*		55.55	44.44	0.000**	
[11-15]	14.28	85.71		3.20 (1.4-6.9)	21.43	78.57		3.20 (1.4-6.9)
[16-20]	0.00	100			0.00	100		
Abortion								
No	37.20	62.79	0.26		53.48	46.51	0.2	
Yes	27.66	72.34			64.89	35.10		
Parity								
Nulliparous	36.95	63.04			68.89	31.10		
Primiparous	25.8	74.19			54.83	45.16		
Pauciparous	34.37	65.62	0.51		65.62	34.37	0.53	
Multiparous	18.18	81.81			50	50		
Large Multip.	33.33	66.66			66.66	33.33		
OCP history								
No	38.61	61.38	0.001**	1.00 (Referent)	65.3T564	34.65	0.1	
Yes	8.33	91.6		6.9 (1.9-24.1)	50	50		
HIV serology								
Negative	34,34	65,65	0.13		66	34	0.04*	1.00 (Referent)
Positive	21,05	78,94			47.22	52.78		2.5 (1.2-5.5)

*Correlation is significant at the 0.05 level (2-sided); **Correlation is significant at the 0.01 level (2-sided). OCP: oral contraceptive pill.

contraceptive methods. The latter findings may also be explained at least partly by the fact that HIV infection may increase HPV pathogenesis [15,16], and women affected by HR HPV types that use OCP double or triple their risk of developing a precancerous or cancerous lesion [17,18].

LSIL precancerous lesions were more common than other lesion types (49.63%), possibly due to the fact that it is a mandatory passage for all higher-grade precancerous lesions and cancers. In addition, many high-grade lesions (HSIL, AGC) would regress to LSIL before returning to normal, provided the subject is not exposed to risk factors that would significantly increase the risk of malignant progression of a cervical lesion [19]. The analysis of associations between HPVs and precancerous lesions indicated that although HPV 16 and 18 are generally (alone and in combination) predominant in advanced precancerous lesions (LSIL and HSIL), their oncogenicities are only revealed when they are associated with each other HPV (HPV 16, 18). In addition, analyses revealed that women infected with the HPV 16, 18 combination were 5.46 times more likely to develop an HSIL lesion (p-Value = 0.001) compared to those infected with other genotypes. Moreover, 100% of HPV 45 mono-infections were found in samples with normal cytology, indicating that HPV 45 would be very weakly associated with the development of precancerous lesions in women in Yaoundé. Notably, in accordance with previous reports, and notably with a study performed in Cameroon [7], women with multiple HPV infections, which constituted 38.7% of the participants were 12.5x more likely to develop a precancerous lesion than those infected with a single HPV type. This could be explained by the combined action of HPVs that would accelerate the precancerous lesion development to cancer. Future studies addressing the risk factors that favor the most common HPV infections and HPV co-infections that accelerate carcinogenesis may contribute to fight against cervical cancer in Cameroon.

Although the eight most common types found in 89% [20]of cancer cases worldwide are HPV 16, 18, 45, 31, 33, 52, 58, and 35, this study was limited to HPV 16, 18, 45, and 33, which are generally the most common in sub-Saharan Africa [20]. Further studies are needed to determine the prevalence of HR-HPV in Cameroon.

Conclusions

Globally, our results confirm that HPV 16, and in a lesser extent, HPV 18 are the most common HR HPV types in Cameroon. Our findings indicate that HPV 16

infections may facilitate other HPV infections, in particular HPV 18 infections, partly explaining why they are the most common infections worldwide. Moreover, women with multiple HPV infections would be more likely to develop a precancerous lesion than those infected with a single HPV type. The peak of HPV infections was found in women aged between 20 and 39. These observations highlight the need for raising the awareness of women, public health authorities, and other stakeholders about the urgent need for setting up HPV vaccination campaigns in Cameroon.

Authors' contribution
TSR, DN, AG and NNY conceived and performed the study and contributed to the acquisition of the reagents and to the preparation of the manuscript. NKAH, SEPF, HPDF, and PBT contributed to data analysis and to the preparation of the manuscript. FBK contributed to the acquisition of reagents and materials, and to data analysis.

References
1. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D (2011) Global cancer statistics. CA Cancer J Clin 61: 69-90.
2. Zhao S, Zhao X, Hu S, Lu J, Duan X, Zhang X, Chen F, Zhao F (2019) Distribution of high-risk human papillomavirus genotype prevalence and attribution to cervical precancerous lesions in rural North China. Chin J Cancer Res. 31: 663-672.
3. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A (2018) Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 68: 394-424.
4. Mudini W, Palefsky JM, Hale MJ, Chirenje MZ, Makunike-Mutasa R, Mutisi F, Jemal A (2018). Human Papillomavirus Genotypes in Invasive Cervical Carcinoma in HIV-Seropositive and HIV-Seronegative Women in Zimbabwe. J Acquir Immune Defic Syndr. 79: e1-6.
5. Bosch FX, de Sanjosé S (2007) The epidemiology of human papillomavirus infection and cervical cancer. Dis Markers 23: 213-27.
6. Unckell F, Streeck RE, Sapp M (1997) Generation and neutralization of pseudovirions of human papillomavirus type 33. J Virol 71: 2934-2939.
7. Catarino R, Vassilakos P, Tebeu P-M, Schäfer S, Bongoe A, Petignat P (2016) Risk factors associated with human papillomavirus prevalence and cervical neoplasia among Cameroonian women. Cancer Epidemiol 40: 60-66.
8. Nayar R, Wilbur D (2015) The Pap Test and Bethesda 2014. Acta Cytologica 59: 121–132
9. Gurgel APAD, Chagas BS, Amaral CMM do, Albuquerque EMB, Serra IGSS, Silva Neto J da C, Cartaxo MMT, Freitas de CA (2013) Prevalence and Genetic Variability in Capsid L1 Gene of Rare Human Papillomaviruses (HPV) Found in

Cervical Lesions of Women from North-East Brazil. BioMed Research International. Biomed Res Int 2013: 546354.

10. Sanjose de S, Quint WG, Alemany L, Geraets DT, Klaustermeier JE, Lloveras B, Tous S, Felix A, Bravo LE, Shin HR, Vallejos SC, Ruiz de PA, Lima MA, Guimera N, Clavero O, Alejo M, Llombart-Bosch A, Cheng-YC, Tatti AS, Kasamatsu E, Iljazovic E, Odida M, Prado R, Seoud M, Grce M, Usubutun A, Jain A, Suarez HAG, Lombardi EL, Banjo A, Menéndez C, Domingo JE, Velasco J, Nessa A, Chichareon BCS, Qiao LY, Lerma E, Garland MS, Sasagawa T, Ferrera A, Hammouda D, Mariani L, Pelayo A, Steiner I, Oliva E, Meijer JC, Al-Jassar FW, Cruz E, Wright CT, Puras A, Llave LC, Tzardi M, Agorastos T, Garcia-Barriola V, Clavel C, Ordi J, Andújar M, Castellsagué X, Sánchez IG, Nowakowski MA, Bornstein J, Muñoz N, Bosch XF (2020) Human papillomavirus genotype attribution in invasive cervical cancer: a retrospective cross-sectional worldwide study. Lancet Oncol. 11: 1048-1056.

11. Forman D, de Martel C, Lacey CJ, Soerjomataram I, Lortet-Tieulent J, Bruni L, Vignat J, Ferlay J, Bray F, Plummer M, Franceschi S (2012) Global burden of human papillomavirus and related diseases. Vaccine 5: F12-23.

12. Paz-Zulueta M, Álvarez-Paredes L, Rodríguez Díaz JC, Parás-Bravo P, Andrada Becerra ME, Rodríguez Ingelmo JM, García RMM, Portilla J, Santibañez M (2018) Prevalence of high-risk HPV genotypes, categorised by their quadrivalent and nine-valent HPV vaccination coverage, and the genotype association with high-grade lesions. BMC Cancer. 18: 112.

13. Carozzi F, De Marco L, Gillio-Tos A, Del Mistro A, Girlando S, Baboci L, Trevisan M, Burroni E, Grasso S, Rossi GP, Ronco G (2014) Age and geographic variability of human papillomavirus high-risk genotype distribution in a large unvaccinated population and of vaccination impact on HPV prevalence. J Clin Virol 60: 257-263.

14. Poynten IM, Machalek D, Templeton D, Jin F, Hillman R, Zablotzska I, Prestage G, Holt M, Grulich A (2016) Comparison of age-specific patterns of sexual behaviour and anal HPV prevalence in homosexual men with patterns in women. Sex Transm Infect 92: 228-231.

15. Liu G, Sharma M, Tan N, Barnabas RV (2018) HIV-positive women have higher risk of human papilloma virus infection, precancerous lesions, and cervical cancer. AIDS 32: 795-808.

16. Rohner E, Sengayi M, Goeieman B, Michelow P, Firnhaber C, Maskew M, Bohlius J (2017) Cervical cancer risk and impact of Pap-based screening in HIV-positive women on antiretroviral therapy in Johannesburg, South Africa. Int J Cancer. 141: 488-496.

17. Mejía L, Muñoz D, Trueba G, Tinoco L, Zapata S (2016) Prevalence of human papillomavirus types in cervical cancerous and precancerous lesions of Ecuadorian women. J Med Virol. 88: 144-152.

18. Tachezy R, Smahelova J, Salakova M, Arbyn M, Rob L, Skapa P, Jirasek T, Hamsikova E (2011) Human papillomavirus genotype distribution in Czech women and men with diseases etiologically linked to HPV. PLoS One. 6: e21913.

19. Solomon D, Davey D, Kurman R, Moriarty A, O'Connor D, Prey M, Raab S, Sherman M, Wilbur D, Wright T, Young N (2002) The 2001 Bethesda System: terminology for reporting results of cervical cytology. JAMA 287: 2114-2119.

20. Aromseree S, Chaiwongkot A, Ekalaksananan T, Kongyingyoes B, Patarapadungkit N, Pientong C (2014) The three most common Human Papillomavirus oncogenic types and their integration state in Thai women with cervical precancerous lesions and carcinomas. Journal of Medical Virology. 86: 1911–1919.

Corresponding author

Dr. Richard TAGNE SIMO, PhD
Department of Biomedical Sciences,
Faculty of Science, University of Ngaoundere,
PO Box 454 Ngaoundere, Cameroon.
Tel: 00237 698 61 32 39
E-mail: rstagne@gmail.com

Conflict of interests: No conflict of interests is declared.