Research Article

Molecular Epidemiology of HPV infection linked to Cervical cancer in Congo: Current state of knowledge

Franck Gaëtan LOUBANOU TCHIBINDA[*(1)],
Luc Magloire Anicet BOUMBA[(2, 3,4)], Parfait Christy NGANGA [(2)],
Ghislain LOUBANO –VOUMBI [(2)], Eben EBATETOU-ATABOHO [(2)],
Rachel MOYEN [(1)], Donatien MOUKASSA [(2)]

[(1)] Faculty of Sciences and Techniques, University Marien NGOUABI, B.P. 69, Brazzaville, Congo.
[(2)] Faculty of Health Sciences, University Marien NGOUABI, B.P. 69, Brazzaville, Congo.
[(3)] Laboratory of Medical and Morphological Analysis, General Hospital of Loandjili, B.P. 8122, Pointe-Noire, Congo.
[(4)] National Institute for Research in Health Sciences. Research, Congo.

* **Corresponding author:** Franck Gaétan LOUBANOU TCHIBINDA,
Faculty of Sciences and Technologies, University Marien NGOUABI, PO Box: 69, Brazzaville- Congo.
E-mail: *loubanoutchibinda@gmail.com* Tel: +242 06 645 43 02.

Abstract

The cervical cancer is the second female cancer affecting Congolese women. Molecular epidemiology of HPV infection is not entirely well understood in Congo. The objective was to update knowledge on the molecular epidemiology of HPV infection in Congo. A bibliographic review was undertaken in the following research databases: PubMed-NCBI, Google Scholar, SCOPUS, Direct Science and Springer link. The articles in English and French were selected based on keywords. The selected articles are those highlighting at least three of the searched keywords. A total of 9 articles were selected out of 54 identified. The relative frequency of intraepithelial lesions was estimated at 15.36%, including 9.17% of low grade intraepithelial lesions (LSIL) and 6.19% of high grade intraepithelial lesions (HSIL). It shows that: thirteen (13) genotypes of HPV have been found in general. HPV16 with 27% overall infection was the most predominant genotype, followed by HPV 70 with 16.6%. Other genotypes are HPV 33 and HPV 6 (10.4%); HPV 31, HPV 35 and HPV 81 (6.2%), HPV 58 and HPV 83 (4.2%), HPV 18, HPV 67, HPV 66 and HPV 54 (2.0%). Young women under 40 are the most affected by HPV infection. Current knowledge of HPV infection remains poorly documented. National mapping of HPV infection becomes a public issue for a good mastery of the Congolese health system.

Keywords: HPV infection, molecular epidemiology, Immunity, precancerous lesions and cancer induced HPV.

1. Introduction

Human papilloma virus (HPV) infections are among the most prevalent in the world [Ferlay et al. 2019; Rapport OMS, 2018]. With a global incidence of over 30 million new cases per year, HPV infection is one of the most common viral infections sexually transmitted [Bruni et al. 2017; De Vuyst et al. 2013].

Precancerous lesions of the cervical epithelium are the basis for the development of this cancer following oncogenic viral strains of HPV. These lesions are easily detectable by cytological screening and genotyping of the HPV strains by molecular biology. However, about 70% of infections go away in about a year, and 90% of infections go away after two years. The spontaneous regression of the infection without any lesion developing and suggests very effective specific local immunity [SCHEURER et al. 2005]. However, 10% of women remain infected and develop precancerous alterations in the cervical epithelium and possibly cervical cancer, making persistent HPV infection the major risk factor for cervical cancer [Torres-Poveda et al. 2014; Ferlay et al. 2013]. In fact, types 16 and 18 are found in more than 90% of cases of cervical cancer [Torres-Poveda et al. 2014]. Over 120 different HPV genotypes infect humans, depending on their low-risk or high-risk oncogenic property [SOW, 2009; Émile C. 2009.; Fausch et al. 2005]. In Congo-Brazzaville, numerous studies had made it possible to build a factual basis for targeting women at high risk and to identify the epidemiological characteristics of these precancerous lesions of cervical cancer. Work carried out by Gombé Mbalawa in 1999 and the team from the International Cancer Research Center (IARC) in Lyon, cited by Parkin et al [Parkin et al. 1990], in the urban community of Brazzaville, focused on the correlation colposcopy - cytology and / or histology, had made it possible to reinforce the role of visual inspection with acetic acid (VIA) in the early detection of high-grade intraepithelial lesions (LEL). Molecular studies have brought a major breakthrough in the epidemiology of HPV linked to cervical cancer in Congo. On the other hand, most of the studies are carried out in Brazzaville and Pointe-Noire, the data on cervical cancer do not cover the whole republic due to the lack of anatomopathology structures and Molecular Biology laboratories. Finding that in Brazzaville and Pointe Noire, Noire, data on cervical cancer do not cover the entire republic due to the lack of pathology structures and Molecular Biology laboratories, located only in Brazzaville and Pointe Noire. Therefore, the objective of this study is to present a current state of knowledge on precancerous and cancerous lesions, and more precisely to describe the various strains circulating in Congo - Brazzaville, in order to better identify and strengthen awareness-raising capacities, prevention and control of cervical cancer in Congo.

2. Methodology

The bibliographic search was carried out in the following search engines: PubMed - NCBI, Google Scholar, SCOPUS, sciences direct and Springerlink, with the keywords, "HPV infection in Congo", "molecular epidemiology of cancers", "HPV and Immunity "," Precancerous and cancerous lesions "," HPV and genotyping ". Articles in French and English related to our theme have been selected. Finally, the articles published in the journals "Tropical Medicine", "Journal of Medical Virology", "International Journal of Sciences and Research" and "African Journal of Cancer" were also analyzed and then integrated. All articles have been the subject of a synthetic analysis, summarizing the useful information. A total of 54 articles were identified, after critical analysis, 8 were retained **(Table 1)** and were the subject of this work on the basis of the following criteria: articles containing at least 3 of 5 keywords entered for the search.

3. Epidemiological reminder on neck cancer in Congo

The Republic of Congo has an estimated population of 5,256,937 inhabitants (in 2020) of which 2,625,515 are women, or 49.9% [Rapport OMS, 2010]. Cervical cancer is a burden on the national health system. The lack of a national cancer registry at the national level says little about the documentation of cervical cancer. Statistical data is limited because the Brazzaville CHU hospital register only covers the city of Brazzaville and some data from the city of Pointe - Noire. According to the Brazzaville University Hospital Cancer Registry, cervical cancer also ranks second among cancers in women and represents 17% of female cancers behind breast cancer [Bruni et al. 2017]. Cervical cancer is therefore a real public health problem in Congo Brazzaville. In 2012, WHO estimates indicate an age-standardized incidence rate of 25.2 per 100,000 women. The age-standardized death rate is estimated at 13.0 per 100,000 women. The prediction by 2025 is 347 new cases for women under 65 and 110 new cases for women over 65 [Torres-Poveda et al. 2014].

Table 1: Articles retained

Titles	Parution	Authors
Results of a statistical study of cancer disease in the Republic of Congo-Brazzaville. About 502 cancer cases diagnosed in 12 years at A. Sice hospital in Pointe-Noire	1967	Charles REYMONDON
Estimates of the worldwide incidence of 25 major cancers	1999	D. Max Parkin
First Congolese observation of a clear cell adenocarcinoma of the cervix	2004	Jean Felix PECKO
Cervical intraepithelial neoplasia (CIN) in Brazzaville, Congo. Situation analysis	2004	Arthur NGOLET
Precancerous lesions of cervical cancer in Pointe-Noire: situation analysis	2007	Donatien MOUKASSA
Specific genotypes of the human papillomavirus in 125 high-grade squamous lesions and invasive cervical cancers in Congolese women	2014	M. L. Anicet BOUMBA
Prevalence of human papillomavirus infection in Congolese women with normal cervical cytology	2015	M. L. Anicet BOUMBA
Distribution of human papillomavirus genotypes by cervical cytological status in women hospitalized at Loandjili General Hospital, Pointe-Noire, South-West Congo (Brazzaville)	2015	M. L. Anicet BOUMBA
Evaluation of plasma il-7 concentration during HPV infection in women with cervical cancer in Congo	2019	F.G. LOUBANOU TCHIBINDA

4. State of current knowledge on molecular epidemiology of HPV infection in Congo

4.1. Epidemiology of intraepithelial lesions

The first studies carried out on morbidity and mortality linked to cancer in the department of KOUILOU, precisely in Pointe –Noire by Reymondon et al. (1967) carried out in the medical department of the Adolphe Sice general hospital in Pointe-Noire, shows that cervical cancer was already at that time the number one cancer developed in women in this department [Reymondon et al. 1967].

In 2004, J.F. Peko et al, made a claim on a first Congolese observation of a clear cell adenocarcinoma of the cervix. It was for the first time that this observation was made on a 55-year-old woman, multiparous, menopausal for eight years and not taking an estrogen-progestogen substitute, had consulted the obstetric gynecology department of the Brazzaville University Hospital, for metrorrhagia and pelvic pain progressing for a month. The histological type reported was rare and this observation was the first in Congo [Peko et al. 2004].

The same year Arthur N'Golet et al. (2004), work on cervical intraepithelial neoplasia (CIN) in Brazzaville, Congo. This was a retrospective analysis of the results of 5,700 cervico-uterine smears in hospitals and in maternal and child health centers in Brazzaville [N'Golet et al. 2004]. The majority of women examined were between 21 and 30 years old (49%), with an obstetric profile dominated by single or multiple parity (83.20%), and a low socio-economic level. Out of 3,088 pathological smears, 200 cases of CIN predominantly in young people aged 20 to 29 had been identified. They were divided into 174 CIN I and 26 CIN II and CIN III. 88 invasive cancers predominated in women aged 50 to 60; a lag of around 10 years was thus noted between the latest observation period for CINs (30 to 39 years) and the age of onset of invasive cancers. On the other hand, the studies carried out by MOUKASSA et al. (2007), a situation analysis on precancerous lesions in cervical cancer in Pointe - Noire. They made it possible to collect 1,347 files from women who had a cervico-vaginal smear. Intraepithelial lesions were estimated at 15.36% for the relative frequency, including 9.17% of low grade intraepithelial lesions (LIE fg) and6.19% of high grade

intraepithelial lesions (LIE hg). The average age of high-grade LELs is 42.25 years, with extremes ranging from 17 years to 61 years. This preliminary study made it possible to establish the factual bases in relation to the epidemiological data of these precancerous lesions of cervical cancer in the Kouilou department in Congo-Brazzaville [MOUKASSA et al. 2007]. Still in the urban community of Pointe - Noire, the work carried out by BOUMBA et al. (2015), on the distribution of human papillomavirus genotypes by cytological status in women at the Loandjili General Hospital in Pointe-Noire. The entire study population had a mean age of 43.6 ± 9.5 years with extremes ranging from 16 to 72 years. Concerning the distribution of cytological lesions: of the 321 women included, 189 (58.9%) cases of normal cytology or benign cell changes (N / MCB) were diagnosed, 16 (4.9%) cases of undetermined cellular atypia (ASCUS), 63 (19.6%) cases of low grade intraepithelial lesions (LSIL), 47 (14.6%) cases of high grade intraepithelial lesions (HSIL) and 6 (1.9%) cases of cancer and based on an average age of 40 years, we summarized the results of this article in **Table 2.**

Table 2: Distribution of cytology according to the average age of 40 years

		Cytology			
Age	Number	ASCUS	LSIL	LSHL	ICC
< 40 ans	108	14	21	15	2
40 ans	81	2	42	32	4
Total	189(58.9%)	16(4.9 %)	63(19.6%)	47(14.66)	6 5(1.9%)

Regarding the work carried out by LOUBANOU et al. in relation to cytology and mean IL-7 concentrations, show that in normal cytology; ASCUS; LSIL and HSIL had shown that IL-7 concentrations were lower than those of invasive cervical cancers (ICC) [LOUBANOU TCHIBINDA et al. 2019].

4.2. HPV and genotyping Prevalence of infection

The work carried out by BOUMBA et al. (2015) on the prevalence of human papilloma virus infection in Congolese women [Boumba et al. 2015], shows a distribution of HPV infection according to the cytological diagnosis. High-risk HPV infection accounted for 7.4% in normal women with Benign Cell Modifications N / MCB); 18.7% in ASCUS women; 52.4% among LSILs; 76.6% in HSIL and 66.7% in cancer cases. Low risk HPV infection was 4.2% in normal cytologic cases; 18.8% in ASCUS and 14.3% in LSIL cases. No low-risk HPV infection was identified in HSIL and CHF. Infection with oncogenic genotypes accounted for 60.4% while that attributed to low-risk genotypes was only 39.6%. Thirteen genotypes had been identified of which, HPV16 with 27% overall infection was the most prevalent genotype followed by HPV 70 with 16.6% overall infection. The other genotypes are respectively in decreasing order are: HPV 33 and HPV 6 (10.4%), HPV 31, HPV 35 and HPV 81 (6.2%), HPV 58 and HPV 83 (4.2%),

HPV 18, HPV 67, HPV 66 and HPV 54 (2.0%). This work shows that the infection is significant in young women under 30, who alone accounted for 38.6% of the overall infection. A gradual inflection of the infection curve had been observed from the peak of 30 years to women over 60 years (11.1% of the overall infection). The prevalence of HPV was 41.50% in the work carried out by LOUBANOU et al. The mean IL-7 concentrations were 9.2 pg / ml for HPV positive and 5.9 pg / ml for HPV negative. This increase was therefore correlated with oncogenic HPV genotypes [LOUBANOU TCHIBINDA et al. 2019].

4.3. Genotypes

The same year, a study by BOUMBA et al. (2015) on the distribution of human papillomavirus genotypes by cytological status in women at Loandjili General Hospital, Pointe-Noire [19] is detailed as follows: On smears cervico normal, women infected with normal cytology were 11.6% or 22 out of 189 normal women. On indeterminate cytologies, women with HPV infection were 37.5%, or 6 out of 16 women. On low-grade lesions, 80.9% of women infected with a low-grade lesion were 51 out of 63. On high-grade lesions, and finally 100% of women diagnosed with a high-grade lesion or invasive carcinoma were HPV positive. And therefore Whatever the cytological status considered, the most prevalent genotype was

HPV16 followed by HPV 70 in cases of normal cytology and HPV33 in the rest of the cytological grades. It was noted that high-grade lesions and invasive cancer cases were exclusively infected with high-risk HPVs **(Figure 1).**
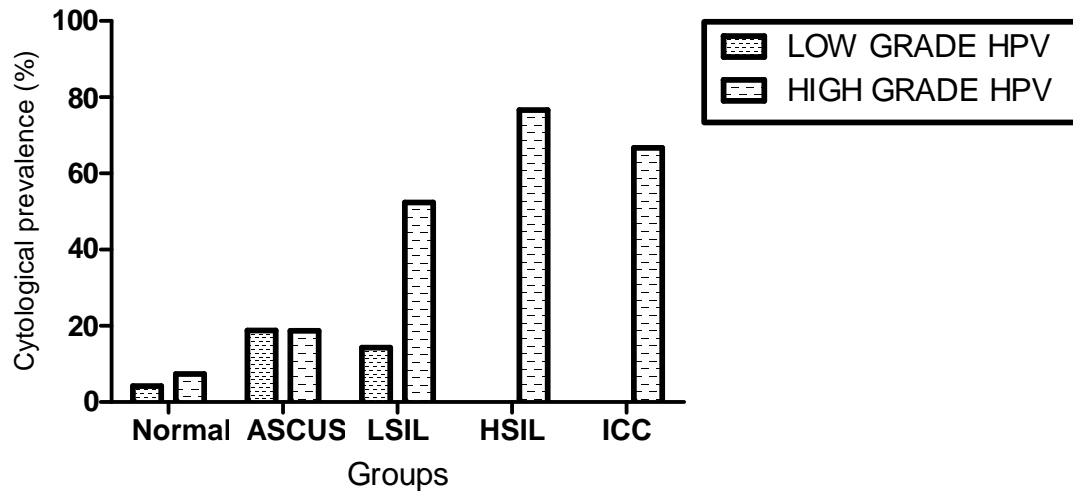

Figure 1 : Distribution of genotyping according to cytology

In cases of multiple infections had been exclusively identified that high-grade genotypes in precancerous and cancerous lesions of the uterine cervix at 12.7% in LSILs, 23.4% in HSILs and 33.3% in the CCIs. These genotypes were in the following order, HPV 16, followed by HPV 33, HPV18, and HPV31 (Table 3).

Table 3: Correlation of cytology and genotyping in multi-infection cases

Grades	FCU Normal	ASCUS	LSIL	HSIL	ICC
Low grade HPV	4.10%	18.60%	14.30%	0.00%	0.00%
High grade HPV	7.30%	18.70%	52.50%	99.90%	100%

Other work by BOUMBA et al. (2014) on the specific genotypic profile of human papilloma viruses in 125 cases of high-grade epithelial lesions and invasive cervical cancer [24].

The molecular study involved 125 wax biopsies from patients with an average age of 44.3 ± 8.2 years and showed that:

In 81.5% of cases of high grade cervical lesions, HPV viral DNA was identified and the frequency was 98.3% (59/60) in invasive cervical cancers. On the other hand, in normal cytology, ASCUS and low-grade cervical lesions, high-risk HPV strains were absent among these cases of biopsies.

Out of a total of 10 genotypes identified, HPV 16 was the most infectious with 47.1% of high-grade lesion cases and 52.5% of invasive cervical cancer cases.

The other genotypes identified were, in decreasing order, HPV 33, HPV 18, HPV 31, HPV 69 and HPV 35. All genotypes identified in the two types of lesions were oncogenic types strongly involved in the occurrence of the disease cervical cancer.

5. Conclusion

In the Congolese population, the youngest women, if not less than 40 years old, are the most infected with HPV and present the precancerous and cancerous lesions of cervical cancer. On the other hand, HPV 16 and 33 are the most oncogenic and infectious strains of HPV encountered in this population. Innovative strategies based on efficient, rapid, inexpensive and mobile screening tools, including cytology and molecular biology at best, must be associated with vaccination programs at the national level, in all girls over 15 years of age who have not had sexual intercourse or, at the latest, within one year of the start of their sexual life, in order to protect them before they are exposed to the risk of infection with these viruses.

6. Conflict of Interest: The authors declare that they have no conflict of interest.

7. References

[1] Ferlay J, Colombet M, Soerjomataram I, Mathers C, Parkin D, Piñeros M, et al. Estimating the global cancer incidence and mortality in 2018: GLOBOCAN sources and methods. International journal of cancer 2019;144:1941-53.

[2] Rapport-OMD. Congo Brazzaville/(Congo) Population 2018. countrymetersinfo/fr/congo 2010.

[3] Bruni L, Barrionuevo-Rosas L, Albero G, Serrano B, Mena M, Gómez D, et al. ICO/IARC Information Centre on HPV and Cancer (HPV Information Centre). Human Papillomavirus and Related Diseases in the World Summary Report 2017;27.

[4] De Vuyst H, Alemany L, Lacey C, Chibwesha CJ, Sahasrabuddhe V, Banura C, et al. 2013. The burden of human papillomavirus infections and related diseases in sub-saharan Africa. Vaccine31:F32-F46.

[5] SCHEURER ME, Tortolero-Luna G, Adler-Storthz K.2005. Human papillomavirus infection: biology, epidemiology, and prevention. International Journal of Gynecologic Cancer15:727-46.

[6] Torres-Poveda K, Bahena-Román M, Madrid-González C, Burguete-García AI, Bermúdez-Morales VH, Peralta-Zaragoza O, et al.2014. Role of IL-10 and TGF- 1 in local immunosuppression in HPV-associated cervical neoplasia. World journal of clinical oncology5:753.

[7] Ferlay J, Soerjomataram I, Ervik M, Dikshit R, Eser S, Mathers C, et al. 2013. GLOBOCAN 2012 v1. 0. Cancer incidence and mortality worldwide: IARC CancerBase;11.

[8] SOW AA. 2018.Faculté de Médecine et d'' Odontostomatologie.

[9] Émile C. 2009. Infection à papillomavirus (HPV), cancer de l'utérus et vaccination. Option/Bio;20:10-1.

[10] Fausch SC, Fahey LM, Da Silva DM, Kast WM.2005. Human papillomavirus can escape immune recognition through Langerhans cell phosphoinositide 3-kinase activation. The Journal of Immunology174:7172-8.

[11] Parkin DM, Pisani P, Ferlay J. 1999. Estimates of the worldwide incidence of 25 major cancers in 1990. International Journal of Cancer;80:827-41.

[12] Reymondon C RE, Bourges M, Deme J.1967. Résultats d'une étude statistique de la maladie cancéreuse en République du Congo- Brazzaville. A propos de 502 de cancer diagnostiqués en 12 ans à l'hôpital A. Sice de Pointe-Noire. Médecine tropicale ;;27 : 417-22.

[13] Peko J, Buambo S, Kokolo J. 2004. Première observation congolaise d'un adénocarcinome à cellules claires du col de l'utérus. Bull Soc Pathol Exot;97:113-4.

[14] N'Golet A, Koutoupot BR, Lubuélé L, Moukassa D, Etoka SE.2004. Les néoplasies cervicales intra épithéliales (CIN) à Brazzaville, Congo. Analyse de situation. Annales de Pathologie: Elsevier;. p. 324-8.

[15] Moukassa D, N'Golet A, Lingouala L, Eouani M, Samba J, Mambou J, et al. 2007. Lesions precancereuses du cancer du col uterin a Pointe-Noire: analyse de situation. Médecine tropicale;67:57-60.

[16] Boumba LMA, Qmichou Z, Mouallif M, Attaleb M, Mzibri ME, Hilali L, et al. 2015. Human papillomavirus genotypes distribution by cervical cytologic status among women attending the General Hospital of Loandjili, Pointe-Noire, Southwest Congo (Brazzaville). Journal of Medical Virology;87:1769-76.

[17] LOUBANOU TCHIBINDA FG , BOUMBA AL, EBATETOU-ATABOHO E, LOUBANO-VOUMBI G, THOUASSA AJC, MOUKASSA D.2019. EVALUATION DE LA CONCENTRATION PLASMATIQUE DE L'IL7 AU COURS DE L'INFECTION A HPV CHEZ LES FEMMES AYANT LE CANCER DU COL DE L'UTERUS AU CONGO. Annale des Sciences de la Santé;19.

[18] Boumba LMA, Mouallif M, Hilali L, Moukassa D, Ennaji MM. 2015. Prevalence of Human Papillomavirus infection among Congolese Women with Normal Cervical Cytology. IJSR;4:2319-7064.

[19] Boumba LMA, Hilali L, Mouallif M, Moukassa D, Ennaji MM. 2014. Specific genotypes of human papillomavirus in 125 high-grade squamous lesions and invasive cervical cancer cases from Congolese women. BMC Public Health;14:1320.