# Association Between Human Papillomavirus Infection and Anus Cancer Precursor Lesions: A Review

**Ana Carolina Borges Monteiro, Yuzo Iano, Reinaldo Padilha França, Rangel Arthur, Pablo David Minango Negrete, Diego Pajuelo, Gabriel Gomes de Oliveira, and Lisber Arana Hisnostroza**

**Abstract** Human papillomavirus, commonly referred to as HPV, is a virus with tropism by differentiating tissues. Its pathogenesis is related to the disorder of genes that inhibit cell apoptosis and cell suppression, this fact favors its action and spread by an organism. Due to these characteristics, HPV is associated with cervical, anus, head, and neck cancer. Based on this, the present work has as objective the accomplishment of a bibliographical revision study of the main characteristics of this virus as well as its effects on the human organism. Studies such as this can be seen as key elements for understanding and preventing this sexually transmitted disease (STD), which has its highest incidence rates in underdeveloped and developing countries, where health and education policies are often scarce or nonexistent.

**Keywords** Human papillomavirus · Cytopathology · Molecular biology · Sexually transmitted disease · Diagnostic methods

## 1 Introduction

Human papillomavirus (HPV) is a 72 capsomer non-enveloped virus belonging to the Papovaviridae family, with a mean diameter of 55 nm. The genome of these

A. C. Borges Monteiro (✉) · Y. Iano · R. Padilha França · P. D. M. Negrete · D. Pajuelo · G. G. de Oliveira · L. A. Hisnostroza
School of Electrical and Computer Engineering (FEEC), University of Campinas – UNICAMP, Av. Albert Einstein - 400, Barão Geraldo, Campinas, SP, Brazil
e-mail: monteiro@decom.fee.unicamp.com.br

Y. Iano
e-mail: yuzo@decom.fee.unicamp.com.br

R. Padilha França
e-mail: padilha@decom.fee.unicamp.com.br

R. Arthur
School of Technology (FT), University of Campinas – UNICAMP, R. Paschoal Marmo - 1888, Jardim Nova Italia, Limeira, SP, Brazil



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viruses consists of eight to nine open reading frames and one regulatory region. This virus has double circular deoxyribonucleic acid (DNA) containing about 7900 base pairs and genes encoding eight proteins: E1, E2, E4, E5, E6, E7 (non-structural), and L1 and L2 (structural) that form the viral capsid with icosahedral symmetry. L1 protein when produced in a heterologous expression system has the ability to self-assemble. The genome of this virus is organized into three regions: early region (E), late region (L), and regulatory region (URR). The L1 and L2 genes are responsible for encoding viral capsid proteins and genes, as well as for the coding of proteins involved in viral replication and cell transformation. E1 and E2 genes are involved in viral replication, while E5, E6, and E7 genes encode proteins responsible for this cellular transformation. HPV infections can be divided into two types: cutaneous or mucosal [1, 2].

More than 200 types of HPV are currently described in the literature, which is classified according to their oncogenic potential. Approximately 45 types of this virus infect the male and female anogenital tract epithelium 3. HPV subtypes can be classified as low risk (types 6, 11, 42, 43, and 44) and high risk (types 16, 18, 31, 33, 35, 39, 45, 46, 51, 52, 56, 58, 59, and 68) [3, 4].

Since the 1970 s, German scientist Harald zur Hausen of the Cancer Research Center (Heidelberg—Germany) has devoted himself to the study of HPV. In her research, there was evidence of the association of subtype 16 Human papillomavirus with biopsies with positive reports of women with cervical cancer. Shortly after this discovery, Hausen also associated subtype HPV 18 with the same pathology. Both subtypes are found in approximately 70% of these biopsies. In the year 2008, Harald zur Hausen won the 2008 Nobel Prize in Medicine for his research on HPV. Based on these findings, further research on this virus has been boosted [3, 4].

This infection is due to unprotected sexual contact, which through micro-abrasions, facilitates the penetration of the virus in the deep layer of epithelial tissue. However, the infection can also occur through direct or indirect contact with lesions on other parts of the body. There is also the possibility of vertical transmission during pregnancy or at delivery. Low-risk oncogenic lesions are manifested as a common wart, genital wart, or condyloma [5].

The classic risk factors for both oncogenic and non-oncogenic development are early onset of sexual intercourse, multiplicity of lifelong sexual partners and high parity (non-surgical births), being young, being a smoker, and being of low socioeconomic status, prolonged oral contraceptive use, nutritional deficiencies, human immunodeficiency virus infection (HIV), and other genital infections caused by sexually transmitted agents, such as Chlamydia trachomatis, Herpes simplex virus) [6].

To understand the oncogenicity of HPV, it is necessary to consider that when the daughter cells immediately enter the G1 phase and are capable of either restarting the cell cycle or performing a temporary or permanent halt. The G1 phase has a checkpoint, G1/S, controlled by the pRb (retinoblastoma protein) pathway. Once the cell passes this point, it is compelled to replicate its DNA. If an incorrect copy of DNA occurs during S or DNA damage, then the cell will not pass the G2/M checkpoint, and gene-induced growth arrest and apoptosis will occur. In this process, several

inhibitor proteins can stop the advance of this cycle, such as p15 and p16, which block fundamental components for cell cycle progressions, such as cyclin-dependent kinases (CDK) and cyclins, preventing the advance of the phase cycle. G1 to S [7, 8].

Other inhibitors are p21, associated with the proto-oncogene ras, and p53, which are responsible for monitoring the quality, integrity of their chromosomes, and the correct execution of the different phases of the cycle. Human cells are equipped with mechanisms for controlling cell division. Mutations in the genetic content of these cells can overcome these defenses and contribute to cancer formation. One such mechanism of action is apoptosis, which occurs when fundamental components are damaged or production system control is unregulated. The development of tumor cells implies an escape from this mechanism. Protein p53, among its many functions, assists the onset of apoptosis, reducing the chance of genetically damaged cells being eliminated by initiating a carcinogenic process. Another mechanism for controlling cell division limits the number of times a given cell reproduces. In this mechanism, the tips of the chromosomes (telomeres) mark the number of divisions, and at the appropriate time initiate senescence and death at the time of telomerase. Activation of this enzyme induces cell immortalization, an indispensable event for carcinogenesis [7, 8].

Within this context in carcinogenesis two classes of genes are fundamental for the development of cancer, because under normal conditions they are responsible for cell cycle control. Proto-oncogenes stimulate, while suppressor genes inhibit the processes of cell division. Collectively, these two classes of genes are responsible for the uncontrolled proliferation found in cancers in humans. When mutations occur, proto-oncogenes become oncogenes, which are carcinogenic and cause excessive cell multiplication. These mutations cause the proto-oncogene to overexpress its growth-stimulating protein or to produce a more active form. In contrast, tumor suppressor genes when inactivated by mutations contribute positively to cancer when. The result of these combinations is the loss of action of functional suppressor genes, which deprives the cell of controls crucial for inhibiting inappropriate growth [7, 8].

Due to the dissemination of information about the incidence of STD, as well as its oncogenicity capacity, there is an increase in condom use among young people in some countries [9]. Therefore, the need to develop bibliographical review studies is noteworthy. Thus, the present study aims to present to the scientific community as well as the general population the main information about HPV that affects the anal regions so that this information is transmitted in a clear, concise, and updated way; after all, HPV in regions anal diseases is as risky as HPV housed in the uterine cervix region, whereas it is less addressed by the general media.

## 2 Methodology

The present study was performed based on the analysis of scientific works related to genital HPV, as well as diagnostic methods of this pathology. These studies were analyzed focusing on scientific publications conducted between 2014 and 2019.

## 3 Results and Discussion

The anal canal is histologically characterized by the presence of a multi-stratified squamous lining. The anal canal is located from the anal border to the pectineal line, which is characterized by being a transition zone between the squamous and non-squamous epithelium. In turn, the area that delimits the anal canal and rectum is made up of transition cells, which resemble the cells that make up the urogenital lining and rectal glandular mucosa. Thus, the appearance of tumors in the distal region of the pectineal line is called squamous cell carcinomas, or epidermoids, which are classified as keratinized. However, those originating from tissues above the dentate line are called epithelioid, which are non-keratinized. Women with cervical cancer are considerably more susceptible to the development of anus or rectal canal cancer [10].

Anal epithelial and anal canal neoplasms, besides presenting differences in embryonic origin, also present different segments for lymphatic drainage, according to their location before the pectineal line. Just above the pectineal line, the lymph flows toward the paravertebral lymph nodes and the perirectal spaces, which are indications for the diagnosis of rectal adenocarcinoma. Just below the pectineal line, lymph flows toward the nodes of the inguinal region, being characteristic for the detection of inguinal adenopathies [11].

Tumor severity and clinical presentation depend on anal size and location. Lesions less than 2 cm are classified as small and have a probability of cure in 80% of cases. However, tumors of 5 cm or more are usually invasive and compromise the lymph nodes, thus presenting less than 50% chance of cure [12].

Just as HPV has tropism in the transitional zones of the cervix, HPV in the anal canal has a preference for the anal transition zone (ZTA). Based on this feature, the anal Pap smear test began. This exam consists of the removal of anal cells for cytological studies by introducing anal swab or endocervical brush in the anal canal. For this, the swab should be introduced between 3 and 4 cm inside the anal canal. This occurs because the length of the anal canal varies from 2.52 to 2.93 cm in women and from 3.27 to 3.4 cm in men. After the introduction, the swab or brush should be rotated 360º around the axis. The material should be uniformly deposited on a glass slide and subsequently fixed and stained properly [13].

The cytological analysis performed on anal smears aims to search for columnar epithelium cells, squamocolumnar cells (coming from the transition zone) and squamous cells. High-grade anal intraepithelial neoplasias (AIS) are considered precursors of anal carcinomas, presenting a high relationship with infection with high oncogenic HPV risk subtypes, mainly by subtypes 16 and 18. These subtypes have their DNA identified between 35 and 90% of tumor cells [13, 14].

The risk of progression is associated with the severity of dysplasia, and HPV virus classification is directly linked to premalignant and malignant lesions, without correlation with the recurrence rate of lesions in the anal region. Thus, the treatment of precursor lesions aims to prevent the evolution of squamous cell carcinoma [15].

AIS are classified as synonymous with severe dysplasia, severe atypia, and in situ carcinoma. This pathology has a higher incidence in squamous cells, being responsible for the cytological picture of binucleation or multinucleation, the presence of koilocytes and atypical paraceratocyte cells in addition to moderate dysplasia. It is noted that most anal carcinomas follow a cytological pattern similar to that found in cervical carcinomas [18].

The biological effects caused by infections with other STDs are related to the presence of persistent inflammation accompanied by damage through oxidative metabolites. Infected cells are prone to increased cytokine secretion. When this infection becomes chronic there is the induction of tissue damage by indirect production of reactive oxygen species, thus triggering the inflammatory cascade. As a consequence of this process, there is a decrease in immunity. In addition, if the patient is a smoker, immunosuppression will be even higher, as nicotine is a determining factor for Langerhans cell depression. Thus, the somatization of immunosuppressor cofactors further facilitates the installation of the HPV virus [18].

The anal canal is more affected than the anal margins, and the transition zone between the anus and rectum is the most affected by tumors. The transition zone has areas of normal rectal mucosa and the presence of squamous epithelium, with the presence of cells of various sizes arranged in arrangements, also has microvilli arranged in columns. This region is affected by high-grade squamous intraepithelial lesions, which are associated with infections caused by high-risk oncogenic HPV subtypes. When untreated, the lesions may progress to invasive carcinoma [17, 18].

About 20% of affected patients are asymptomatic, the others usually have anorectal bleeding or pain usually accompanied by the sensation of mass occupying the rectal region. This symptomatology, however, is often mistakenly mistaken for hemorrhoidal diseases. Anal oncogenic low-risk HPV infection in the latent form initially manifests itself through anal pruritus, with later onset of warts. However, the presence of anal pruritus is not a determining factor for the closure of this diagnosis, since pruritus has a multifactorial etiology such as inflammatory bowel disease, uremia, parasitic diseases, dermatopathies, inadequate hygiene habits, inadequate clothing, the presence of STDs, among others [19].

In turn, high-risk anal oncogenic HPV presents difficulties in its diagnosis, since the lesions are subclinical and their detection is dependent on anoscopy exams. As the lesions are inapparent and usually asymptomatic, the individual seeks medical attention only when the lesions have progressed. Anal border tumors are classified as dermatological lesions and may develop as verrucous carcinoma, Kaposi's sarcoma, Bower's disease, or squamous cell carcinoma. However, those that affect the rectal transition zone or rectal canal are considered malignant and have four origins histological: squamous cell carcinoma, malignant melanoma, adenocarcinoma, and sarcomas [19, 20].

## 4 Diagnostic Methods

The most suitable diagnostic methods for detecting the presence of HPV in the anal region are anoscopy and anal Pap smear. Anuscopy consists of applying 5% acetic acid to the anorectal mucosa for two minutes. Subsequently, the excess is removed with a physiological solution, followed by toluidine blue. The examination should be performed using a colposcope. The result is observed through the presence of ketoacid reactions, which appear in coloration whitish when positive, indicating HPV infection [20].

Polymerase chain reaction (PCR) is a high sensitivity test and is used to prove the existence or the absence of HPV DNA. The PCR reaction is the amplification of viral DNA (HPV) using conserved sequences from specific HPV regions as primers. This test also enables the identification of the HPV genotype by amplifying specific regions for each of the high- or low-grade viruses [21, 22].

Recent studies show that increased expression of miR-16, miR-25, miR-92a, and miR-378 and reduced expression of miR-22, miR-27a, miR-29a, and miR-100 are attributed to viral oncoprotein. E6 or E7. In the future, miRs are expected to be powerful tools for early detection of HPV regardless of their subtype and site of infection [23].

Researchers have developed a methodology for the detection of urine high-risk oncogenic HPV. This technique is based on the detection of virus DNA present in the samples. The results obtained were 90.9% accurate in identifying cervical lesions with abnormal cells [24].

In recent years, there have been a growing number of researches related to pathology detection through the digital processing of medical images [25–27]. In this context, it is expected that in the coming decade's methodologies will be developed that are capable of detecting precursor lesions and cancerous lesions of HPV, similarly to blood cell detection and counting research. These studies have shown high sensitivity, accuracy, specificity, low computational and runtime time consumption, and low cost. Cost reduction is paramount for poor people's access to quality health [25, 26].

In addition, it is important to note that in the not too distant future, the results of cytopathological examinations, patient anamnesis, and data from health institutions can be transmitted and shared by real-time telecommunication channels without data loss and low memory consumption of the devices involved regardless of the type of operating system involved (Android, Windows, Linux, iOS, among others) [27, 28]. Such action is paramount, because the faster the doctor has access to the medical reports, the faster the patient is referred for treatment, and consequently the chances of cure increase.

## 5 Conclusions

The association between HPV and malignant neoplasms shows that this virus is responsible for the precursor lesions that direct individuals to cancer development. However, with recent advances in molecular biology coupled with cytopathology practices, they have led to the possibility of early and more accurate diagnosis of the subtype involved. The classification of the HPV subtype involved in lesions is paramount for the correct management of the disease. Even with such advances, prevention through education focused on adolescents, youth, and adults is the best way to ensure the health of individuals. Only with massive awareness activities can we avoid future HPV complications with other STDs. Importantly, not all STDs are curable, some should only be followed and controlled throughout the patient's life. Such information is often not disseminated as it should. In addition to prevention, the establishment of telemedicine-based data sharing networks is important to promote better treatment and follow-up of patients who live in remote regions or where there is a shortage of professionals specialized in this type of clinical management. In addition, it is necessary to create complex databases that, using artificial intelligence techniques, are able to compile data on cytopathology, molecular biology, dermatoscopy, cytology, and histology of patients affected by HPV. Thus, the medical diagnosis may become increasingly accurate and less likely to fail to identify and classify the HPV subtype; after all, an incorrect medical diagnosis may lead to the patient's death or irreversible damage.

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