HPV INFECTION AND MALIGNANCY: CURRENT UNDERSTANDING AND FUTURE PERSPECTIVES
Infecção por HPV e malignidade: conhecimento atual e perspectivas futuras

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Abstract

Human papillomaviruses (HPV) are a group of genetically related organisms that commonly infect stratified squamous epithelium. Unlike many other viruses that infect epithelium and induce lysis of the cells they induce proliferative changes that result in both benign and malignant tumors. The aim of the present paper was to briefly review current understanding of HPV infection in relation to malignancy and also highlight perspectives with regard to the development of an effective vaccine.
Keywords: Human papillomavirus; Oral pathology; Malignancies.

Resumo

O papilomavírus humano (PVH) constitui um grupo de organismos geneticamente relacionados que freqüentemente infectam o epitélio escamoso estratificado. Ao contrário de muitos outros vírus que infectam o epitélio e induzem lise das células, eles induzem alterações proliferativas que resultam em tumores malignos e benignos. O objetivo do presente artigo foi revisar brevemente o atual conhecimento da infecção por HPV em relação com malignidade e também ressaltar as perspectivas com relação ao desenvolvimento de uma vacina efetiva.
Palavras chave: Papilomavírus humano; Patologia bucal; Neoplasias malignas.

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Introduction

To date, more than 100 types of human papillomavirus (HPV) have been identified. In the past 20 years, there has been an increasing interest in HPVs due to their potential role in the pathogenesis of malignant tumors. HPV infections are known to affect predominantly adult, sexually active age groups, whereas skin warts, at various anatomic sites, are usually associated with younger individuals (1).

In fact, HPV are long thought to be a common sexually transmitted viral infection. The virus targets injured cervical epithelial cells, gaining access at sites of microtrauma occurring during (sexual) intercourse. In addition, a subset, termed high risk types, may be strongly associated with the development of oral and cervical cancer (2,3). HPV16 is the most common HPV type found in ano-cervical cancers, where it is detected in over 50% of cases, while other HPV types commonly observed in cervical cancers include types 18, 45 and 31.(4) Similarly, HPV16 is the most commonly detected type in oral and pharyngeal carcinomas(5). In the mouth, HPV has been associated with cancer despite other risk activities, such the use of tobacco and/or alcohol (6). One possibility is that P53 (gene) inactivation is a major component of HPV-related carcinogenesis (7).

Furthermore, it has been shown that a single patient may concomitantly suffer from both cervical and oral cancer, although molecular biology techniques may prove useful to differentiate primary and secondary lesions. In one study, for instance, a patient with apparent mouth metastasis from cervical cancer was found to harbor two HPV sequences from different viruses (18 and 33, for the uterine cervix and the mouth, respectively), thus indicating the oral lesion as a synchronous second primary tumor.(8)

Viral structure

HPV's are small, nonenveloped DNA viruses that replicate in the nuclei of squamous epithelium cells. The viral genome is encased in a capsid layer, consisting of a major structural protein and a minor structural protein.(9) All of the potential coding regions, or open reading frames (ORFs), exist in one of the two DNA strands (10). This means that all of the genetic information is located in only one strand.

The genome is divided in three regions; a long coding region, and two regions consisting of the designated ORFs, called the early and late regions (11) The long coding region contains the viral origin for replication. It is responsible for the regulation of replication, and controls the transcription of some gene sequences in the early region. The early region, divided in eight coding regions, encodes important proteins in viral replication occurring 'early' in the viral life cycle. It is also responsible for maintaining high number of HPV and for high-risk types immortalization. The late region encodes for viral structural proteins necessary for capsid production (12).

HPV life cycle

HPV infection primarily infects the basal layer of the injured epithelium. The viral genomes are established in the host cell as unintegrated extrachromosomal elements or episomes. After infection, the first viral genes to be expressed are E1 and E2, which are the replication proteins. These proteins bind to the origin of DNA replication, located on sites within the long coding region (13). In the basal layer, the virus is in a nonproductive stage and is present in low copy numbers. The virus proliferates here, recruiting host factors for viral synthesis, and replicating its DNA enough to keep up with the mitosis of basal cells (9).

As the host cells continue their normal life cycle or pattern of maturation, a subset of daughter cells detach and migrate from the basal layer. The infected host cells divide, and HPV DNA is divided between the daughter cells as they stratify and differentiate. Virus DNA travels with the host cells as they undergo their normal life cycle and mature. HPV does not encode a DNA polymerase and, hence, it is dependent on host cell differentiation to continue its own life cycle. The virions proliferate, moving with the host cells toward the terminally differentiating or keratinizing layers of the epithelium (9).

Differentiation

HPV-infected cells mature, stratify, and develop special characteristics during a process called differentiation. E4 is a protein expressed later in these terminally differentiating cells, and is also found in association with the viral capsids. Late region genes are also expressed in the differentiated cells near the surface of the epithelium, initiating the synthesis of the capsid layer for the genome. The surface epithelial cells do not divide, but their location offers an ideal site for viral transmission. The shedding of the naturally dying surface cells laden with HPV, also called koilocytes, serves as the vector of transmission. Once the capsid protein layer is formed, the genome is capable of infection and transformation(14).

Viral Integration

Incorporation of HPV genomes in the human host DNA, called integration, appears to be one of the final steps toward malignant transformation. This occurs in the HPV-16, and other high-risk types, but in low-risk types the viral genome remains as extrachromosomal plasmids.

Integration in the host cell DNA occurs in a break in the viral genome, usually at E1 and E2 region. The loss of host cellular control, and the persistence of the high risk virion may contribute to this occurrence. The disruption at E1/E2 plays a pivotal role in oncogenesis.(15) Integration at this region leads to the activation of p97, the normallysuppressed papillomaviral promoter that directly expresses E6 and E7. These two viral transforming genes possess oncogenic properties; they have the capacity to functionally inactivate the cellular tumor suppressors p53, and retinoblastoma protein (pRb). E6 complexes with p53, and E7 with pRb, thereby interrupting the existing cellular pathways that would normally lead to growth arrest and cell death. Integration causes the increased expression of E6 and E7, providing a selective growth advantage to the affected epithelial cell, but it alone is not sufficient for malignant transformation.(16)

HPV detection

Human papillomavirus is an ubiquitous or commensal entity responsible for skin infection that have a worldwide distribution with a broad spectrum of genotypes. The prevalence of HPV DNA varied from 42% in Zambia to 70% in Sweden (17). Two or multiple genotypes were frequently found in the same sample and the most prevalent HPV type is thought to be HPV-5, with an overall prevalence of 6.5 %. This was also the only type that was found in samples from all of the countries in that study.(17)

The prevalence of HPV DNA among children from the ages of 1 month to 4 years varies between 50 and 70%. The HPV DNA sequences commonly detected suggest a great diversity of genotypes and putative genotypes. In a study, (18) among 115 samples, a total of 73 HPV types or putative types were isolated. Of these, 26 putative HPV types had not been described before. Hence, asymptomatic HPV infections of normal skin seem to be acquired very early in infancy and are caused by a great multiplicity of HPV types.

HPV-DNA positivity in the absence of clinically or colposcopically detected lesions is a rare event. It has been shown that HPV DNA was detected in only one sample (3%) from men without visible lesions, in 5 samples (15%) from men with penile lesions but without urethral lesions, and in 16 men with urethral lesions (78%).(19) On the other hand, men aged 18-70 years attending a sexually transmitted disease clinic were screened for the presence of HPV infection. Penile skin swabs were assessed for HPV DNA using polymerase chain reaction with reverse line-blot genotyping and the prevalence of HPV was 28.2%. Oncogenic HPV types in that study were found in 12.0% of participants, nononcogenic types were found in 14.8% of participants, multiple types were found in 6.1% of participants, and unknown types were found in 5.9% of participants. The most prevalent subtypes were the nononcogenic varieties 6, 53, and 84. HPV positivity was not associated with age. These results indicate that HPV infection among men at high risk is common but that characteristics of male HPV infection may differ from those of female infection.(20) HPV infection can be detected not only by DNA amplification, but also using serological methods. However, the poor sensitivity of anti-HPV IgG would suggest the need of new methods of HPV detection: in fact, in a recent research, only

50% of HPV DNA positive individuals were also HPV-seropositive (21).

The lack of sensitivity may also explain the reason for the low detection rate of HPV antibodies in a large retrospective serological cohort, where only 47% of sera from women with cervical cancer and 33% of individuals with oropharyngeal disease were HPV16 L1 positive.(22)

Nevertheless, it has been estimated that HPV seroprevalence in Thailand women varies from
3.9 to 9.1%, with the highest prevalence rates associated with younger age, herpes simplex type 2
(HSV-2) infection and for females being married with partners who have extra-marital sexual partners.(23)

HPV and other malignancies

HPV has been more controversially associated with prostate cancer;(24) squamous cell and adenosquamous carcinoma of the colon and rectum;(25;26) ovarian carcinoma;(27) squamous cell carcinoma of the fingers;(28;29) non-melanoma skin malignancies such as basal cell carcinoma;(30) anal cancer and its precursor lesion, anal squamous intraepithelial lesions(31) and transitional cell carcinoma (TCC) of the urinary bladder (32;33), although these results are not supported by others who have not found association of HPV types 6, 11, 16, 18, 31, 33 and 51 with carcinoma of the bladder.(34) In addition, HPV DNA has been detected in a small proportion of cases of bronchopulmonary carcinoma, and thus HPV infection appears to play a limited role in the tumorigenesis of most lung carcinomas.(35).

There is an increasing evidence supporting the role of HPV in the development of squamous cell carcinoma of the head and neck (SCCHN). Recently, some large sample (36-41) size studies showed that SCCHN and potentially malignant oral lesions, histologically diagnosed, contain DNA of high-risk HPV genotypes (e.g. HPV-18 and -16).

HPV and cancer prognosis

HPV has been found to be an indicator of the severity of oral cancer.(42) In fact, it had already been observed that HPV DNA was associated with all confirmed grade 3 cervical intraepithelial neoplasia and primary cervical cancers.(43) It may be that the histological grading of cervical lesions are not only associated with HPV type, but also to HPV DNA viral load.(44)

In addition, it is possible that polymorphisms in human leukocyte antigen (HLA) genes are implicated in the risk for developing human papillomavirus (HPV)-associated cervical neoplasia.(45) Also, HPV DNA loads of six oncogenic HPV types were measured in cervical scrapes of human immunodeficiency virus (HIV)-infected and uninfected women. In both groups, HPV loads increased with the grade of cervical disease. In the same study, HIV infection did not affect HPV loads in low-grade lesions but was associated with significantly higher HPV loads in severe dysplasia; highest loads were found in advanced HIV disease.(46)

HPV vaccine

HPV infection may be prevented by neutralizing antibodies specific for the viral capsid proteins. In clinical trials, vaccines comprised of HPV virus-like particles (VLPs) have shown great promise as prophylactic HPV vaccines.(47) However, due to the fact that capsid proteins are not expressed at detectable levels by infected basal keratinocytes, vaccines with therapeutic potential must target other non-structural viral antigens. Two HPV oncogenic proteins, E6 and E7, are important in the induction and maintenance of cellular transformation and are co-expressed in the majority of HPV-containing carcinomas. Therefore, therapeutic vaccines targeting these proteins may have potential to control HPV-associated malignancies.(48) Various candidate to therapeutic HPV vaccines are currently being tested whereby E6 and/or E7 is administered in live vectors, in peptides or protein, in nucleic acid form, as components of chimeric VLPs, or in cell-based vaccines.(49;50) Encouraging results from experimental vaccination studies in animal models have led to several prophylactic and therapeutic vaccine clinical trials.(51-53) Should they fulfill their promise, these vaccines may prevent HPV infection or control its

potentially life-threatening consequences in humans.

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