

Human papillomavirus in tonsillar squamous cell carcinomas from Guatemala and Brazil



Alicia Rumayor Piña, MSc,^a Laísa Simakawa Jimenez, MD, student,^b Fernanda Viviane Mariano, PhD,^b Bruno Augusto Benevenuto de Andrade, PhD,^c Román Carlos, DDS,^d Albina Altemani, PhD,^b and Oslei Paes de Almeida, PhD^a

Objectives. A subgroup of tonsillar squamous cell carcinoma (SCC) is associated with human papillomavirus (HPV). Nevertheless, the prevalence of HPV seems to be variable in different regions and ethnic groups. There are no reports of HPV in tonsillar carcinomas in Guatemala, and data from Brazil are scarce. The aim of this study is to analyze and compare HPV presence in samples of tonsillar SCC from these countries.

Study Design. This study describes the histologic features, expression of p16 by immunohistochemistry (IHC), and HPV by in situ hybridization (ISH) in 13 Guatemalan and 13 Brazilian patients.

Results. All cases of tonsillar SCC from Guatemala were positive for p16, 92% expressed HPV by ISH, and 75% corresponded to the high-risk genotype 16/18. From the Brazilian patients, only four expressed p16, and all were negative for HPV.

Conclusions. Cases from Guatemala, which were mostly nonkeratinizing SCC and originated from the crypt/reticular epithelium of the tonsil, had high-risk integrated HPV, whereas in Brazilian cases, which were mostly keratinizing SCC that originated from the surface epithelium, there was no association with HPV. (Oral Surg Oral Med Oral Pathol Oral Radiol 2016;121:412-418)

Human papillomavirus (HPV)—related SCC is a subset of head and neck carcinomas presenting a distinct clinicopathologic profile, with increasing incidence worldwide. Besides the uterine cervix, penis, vulva, vagina, and anal region, the oropharynx is also a susceptible site for high-risk types of HPV, particularly HPV 16.^{1,2}

Information about the incidence and prevalence of oropharyngeal carcinoma is not well documented because it is usually grouped with oral carcinoma; less reliable information is available when dealing specifically with the tonsil because tonsillar carcinomas are almost always categorized as carcinoma of the oropharynx, together with carcinomas of the base of the tongue, lingual tonsil, soft palate, uvula, and oropharynx walls.³ Reports of these entities from

Guatemala are lacking; one recent report about the burden of HPV-related cancers did not have any data for tonsillar cancer.⁴ Data from Brazil are unclear because of the differences in the categorization of subsites. When reported only as oropharyngeal carcinoma, percentages varied from 16% to 23%, of all head and neck carcinomas.^{3,5} In studies specifically reporting tumors from the tonsil, percentages varied from 4% to 13% in three different cities from Brazil to 30% in Central and South America.⁶⁻⁹

HPV-associated oropharyngeal SCCs occur mainly in the tonsil of male patients, showing an increased survival rate compared with HPV-negative cases. This subset of carcinomas also show histologic and molecular features different from conventional SCCs, as these usually are nonkeratinizing, strongly express p16, and show a higher proliferation index and lower p53 expression. Some authors have stated that p16, by itself, is a good marker for favorable prognosis, regardless of HPV status.^{10,11}

It is probable that geographic/genetic differences exist in relation to the role of HPV in oropharyngeal carcinomas, and wide variation in prevalence has been observed among different regions and countries. In

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^aOral Pathology Section, Department of Oral Diagnosis, Piracicaba Dental School, University of Campinas (UNICAMP), Piracicaba, Brazil.

^bDepartment of Pathology, Medical Sciences Faculty, University of Campinas (UNICAMP), Campinas, Brazil.

^cOral Pathology Section, Department of Oral Diagnosis and Pathology, School of Dentistry, Federal University of Rio de Janeiro (UFRJ), Rio de Janeiro, Brazil.

^dCentro Clínico de Cabeza y Cuello/Hospital Herrera Llerandi, Ciudad de Guatemala, Guatemala.

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Statement of Clinical Relevance

In Guatemala, a high percentage of tonsillar squamous cell carcinoma is associated with human papillomavirus, whereas in Brazil, these tumors are more associated with classic risk factors, such as alcohol and tobacco.

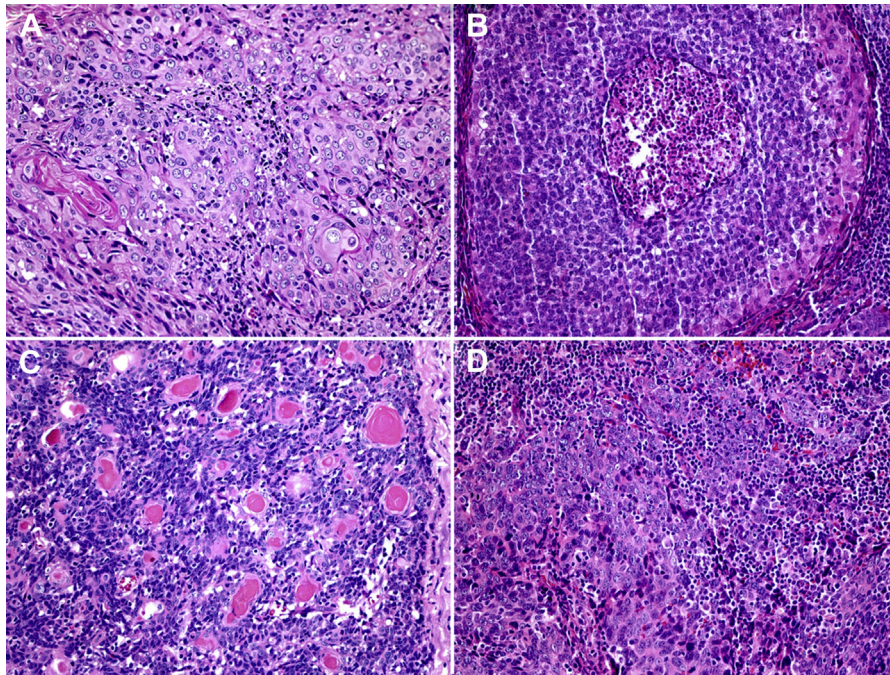


Fig. 1. Histologic features of tonsillar squamous cell carcinoma (SCC). **A**, Keratinizing SCC showing eosinophilic large squamous pleomorphic cells, and evident areas of keratin. **B**, Nonkeratinizing SCC. Nest of small basophilic undifferentiated cells with comedo-necrosis. **C**, Nonkeratinizing SCC with focal areas of keratin maturation. **D**, Nonkeratinizing SCC, showing lymphoepithelial/nasopharyngeal-like features (H&E, ×200).

North America and some European countries, HPV seems to be the primary cause of tonsillar carcinoma. In contrast, a study from China reported absence of HPV in their samples of 16 tonsillar SCC, attributing these results to cultural and behavioral differences.¹²⁻¹⁴

In the United States, the incidence of tonsillar carcinomas associated with HPV is highly variable, with percentages ranging from 22% to 90.9%, but in most reports, values are between 35% and 65%. In a worldwide systematic review of 969 oropharyngeal SCC cases, HPV was identified in 36%, with higher prevalence in North America (47%) and Asia (46%).¹⁵⁻¹⁷ In a recent Mexican study, which included 43 head and neck SCCs, 42% affected the oropharynx, and 35% the oral cavity. Of the oropharyngeal SCCs, 61% were associated with HPV when analyzed by polymerase chain reaction (PCR); an interesting finding was that risk habits for classic oral SCC, such as smoking, were less common in the HPV-positive group compared with the HPV-negative cases.¹⁸ A study of head and neck SCCs in Brazil, including various sites (pyriform sinus, larynx, tonsil, mouth and maxillary sinus), showed a low prevalence of HPV by PCR (11%), and two out of six cases of tonsillar SCC were positive for HPV.⁶

In recent meta-analysis, HPV prevalence in oropharyngeal SCC was low in South and Central Americas; however, data from these regions were limited.¹³⁻¹⁵ The association of known risk factors, such as tobacco and

alcohol, with oropharyngeal SCC has been found to be minimal in HPV-positive cases, whereas higher levels of consumption were reported in HPV-negative patients.^{19,20} The aim of this study was to determine the presence of HPV in tonsillar SCC in patients from Guatemala and Brazil.

MATERIALS AND METHODS

Formalin-fixed, paraffin-embedded tissue blocks of 13 tonsillar SCC were retrieved from the Centro Clínico de Cabeza y Cuello, Guatemala, and 13 from the Pathology Department, UNICAMP, Brazil. The clinical data for all cases were obtained from medical records. Smoking and alcohol drinking were measured and classified according to Ribeiro et al.²¹

All cases were histologically classified as keratinizing (K) and nonkeratinizing (NK) SCCs, and those with overlapping features were considered NK-type SCC, presenting focal areas of squamous maturation or keratinization (NK/focal K).

Immunohistochemistry (IHC) for p16^{INK4a} (CINtec Histology Kit, MTM laboratories, Heidelberg, Germany) was performed in 3-µm sections, according to the manufacturer's protocol. All cases were classified as positive or negative, the former when more than 80% of tumor cells presented strong, diffuse, nuclear, and cytoplasmic staining.²² HPV detection was performed by using ISH, with a wide spectrum probe (Y1404;

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