

## Association of Human Papilloma Virus (HPV) and Oral Cavity Squamous Cell Carcinoma (OSCC): Indian scenario

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### Abstract

**Introduction:** About 80-90% of HNSCC are associated with known risk factor e.g. smoking, tobacco chewing & alcohol abuse. The role of human papilloma virus (HPV) in the etiology of HNSCC has emerged in last few years. Many studies have proved HPV in oropharyngeal cancer but there are very few studies regarding role of HPV in oral cavity which constitutes the 90% of HNSCC. The purpose of this study was to look for the prevalence of HPV in HNSCC in India.

**Methods:** This was a prospective observational study. 50 cases of suspected malignant ulcers of oral cavity were biopsied. Having confirmed the lesion as SCC, HPV 16 and 18 analyses was done on frozen samples by PCR.

**Results:** The most common site of OSCC was oral tongue (50%). About 80% of the patients had some form of addiction. 36% of the patients had early stage disease whereas 64% had late stage disease with 76% cases of well differentiated SCC. Only 2 (4%) patients were found to be HPV positive.

**Conclusion:** HPV is an important etiological agent in HNSCC in western patients but its role in OSCC has yet not been significant. In India, tobacco and alcohol are commonly identified etiological agents of HNSCC. The rarity of HPV might be due to the differences in the cultural and sexual behavior. Also, HPV vaccine is predicted to reduce the incidence of oral cancer due to HPV. However, further studies are required to label HPV as etiological factor for OSCC proper.

**Keywords:** Oral Cavity Squamous Cell Cancer, Human Papilloma Virus, Oropharyngeal cancer

### Introduction

Head and neck cancers (HNC) represent the sixth most common malignancy worldwide, squamous cell carcinomas (SCC) constitutes the majority of them.<sup>1</sup> In India, it accounts for 30% of all cancers in males and 11-16 %

in females.<sup>2</sup> These are heterogeneous group of malignant tumors, arising from the epithelial lining of the upper aero digestive tract, i.e., the oral cavity, the pharynx and the larynx.<sup>3</sup>

They are characterized by a multiphasic and multifactorial etiopathogenesis.<sup>4,5</sup> An increasing number of patients that develop oral and oropharyngeal cancers are without any known local risk factors; implicating etiologic factors other than tobacco and alcohol as its cause. The role of chronic infection with high-risk types of human papilloma virus (HPV) in the etiology of head and neck squamous cell carcinoma (HNSCC) has gradually emerged during the past two decades.

This occurrence also justifies the increased incidence of HNC and the onset of the tumor in younger people, whether the most common risk factors (like tobacco and alcohol) are present or not.<sup>6</sup> There are very limited studies regarding role of HPV in oral cavity cancer which constitutes the majority i.e. 90% of the burden of HNSCC.

The purpose of this study was to look for the prevalence of HPV in HNSCC patients of Indian population.

### **Materials and methods**

This study was carried out in the Department of Surgical Oncology and Department of Microbiology, Nizam's Institute of Medical Sciences, Hyderabad from March 2012 to May 2014 after approval from institutional ethical and scientific committee. It was a prospective and observational study.

After informed written consent, 50 cases of histologically proven SCC of the oral cavity (lip, oral tongue, floor of mouth, alveolus, buccal mucosa and palate) were included in the study. Biopsy was taken from oral lesions of all fifty patients and sent for histopathological examination and microbiological analysis for molecular detection of HPV DNA. Immunocompromised patients, non squamous oral cancers, patients with past history of chemo/radiotherapy and patients in whom adequate biopsy specimen could not be taken due to technical reasons or

anatomical inaccessibility were excluded from the study.

Staging work up of all patients was done as per AJCC TNM staging guidelines and their treatment was carried out as per institution protocols.

### **Results**

Fifty patients (14 females and 36 males) of oral SCC were included in this study with age ranging from 25years to 87 years. The mean age was 51.06 years.

It was found that maximum number of cases affected the oral tongue i.e. 50% followed by buccal mucosa(30%), lower alveolar ridge (8%), upper alveolar ridge (6%), hard palate (4%) and lip (2%) in order.

Around 80% of the patients included in the study had some addiction out of which 50% were tobacco chewers, 8 patients (16%) were smokers and 4% patients did both. 10% patients had addiction to alcohol too with some form of tobacco exposure. However, 20% patients had no addictions. Other lesions associated with these cancerous lesions (submucous fibrosis, oral leukoplakia and oral melanosis) were seen in 4 patients.

According to staging, 10% of the patients had stage I disease, 26% had stage II disease whereas major bulk of the patients i.e.64% presented later in the disease progression (38% in stage III and 26% in stage IV).

Histopathologically, 38 patients (76%) had well differentiated SCC while 12 patients (24%) were moderately differentiated with no cases of poorly differentiated carcinoma. Lymphovascular invasion was seen in 3 patients (6%) and 7 patients (14%) had perineural invasion too.

The study used PCR kit to find out the presence or absence of HR HPV (HPV 16 and 18) in these fifty patients. Only 2 (4%) patients were found to be positive for HPV infection. Both were less than 45 year of age, one being 41 and other 26 years old. Both were gutkha chewer and alcoholic.

Tumor was located in buccal mucosa and presented in stage III in both.

About 39 patients (78%) underwent surgery as the major modality of treatment. Adjuvant radiotherapy was given to 17 patients (34%) most of which were belonging to the stage III and IV. 9 patients (18%) were treated by radiotherapy and chemotherapy as primary mode of treatment. Two patients were lost from follow up before starting the treatment.

Nine patients presented back as recurrence of the disease within the study period. Out of this 1 patient was HPV positive, rest were HPV negative -all of whom presented initially in stage III or IV.

### Discussion

HNC are a biologically heterogeneous group of cancers involving different subsites with similar risk factors and pathological features.<sup>1</sup> The majority of HN malignancies are squamous cell carcinomas originating from the epithelium of the oral cavity in 90% of cases,<sup>7</sup> being the most common cancer in males and third most common cancer in Indian females.<sup>8</sup>

Although HNC are the most easily observable type of cancers, second only perhaps to skin cancers in potential ease of detection, there have not been any significant advances in the control of this disease and mortality rate. Globally, the main risk factors for oral cancer are tobacco and alcohol, the combined effect being multiplicative rather than additive. However, approximately 20% of oral cancers occur in people lacking these established risk factors and not everyone exposed to tobacco and alcohol develop the malignancy.<sup>9</sup>

The participation of HPV in oral and oropharyngeal carcinogenesis was first proposed by Syrjanen et al. HPV-16 and -18 are the major high-risk types and are associated with a wide range of oral lesions, from benign to premalignant and malignant.<sup>10</sup>

HPV-related HNSCCs are distinct from smoking related cancers. Not only do they arise independent of smoking activity, but they involve younger patients and are associated with more favorable clinical outcomes.<sup>11, 12</sup> Perhaps one of the most important developments in head and neck oncology of the past decade is the demonstration that patients with HPV-mediated oropharyngeal cancers have significantly improved clinical outcomes, compared to patients with HPV-negative oropharyngeal carcinomas. The possibility of offering patients with HPV-mediated cancers less aggressive adjuvant therapy is especially relevant given the potential for devastating radiation toxicities. Clinical trials are underway to investigate the impact of “treatment deintensification” for these patients.

Lingen et al studied 409 cases of OCSCC and found that the etiologic fraction for HR-HPV in OCSCC was only 5.9%.<sup>13</sup>

In two large case-control studies by Ribeiro et al in relative high incidence of HNSCC region of north America and Latin Europe, tumour DNA from 196 fresh tissue biopsies were analyzed for multiple HPV types and it was found that HPV16 E7 DNA prevalence was 3.1% (6/196), including 4.4% in the oropharynx (3/68), 3.8% in the hypopharynx/larynx (3/78) and 0% among the 50 cases of oral cavity carcinomas.<sup>14</sup>

In another study by Poling et al published in 2014 on HPV status of non-tobacco related SCC of the lateral tongue, HPV was detected in just 1 (1.3%) of the 78 HNSCCs. This singular case was from a 62 year old white male who was a current smoker and a moderate consumer of alcohol at the time of diagnosis.<sup>15</sup> However, HPV was not detected in any patients under 40 years of age (n = 10), or in patients who never smoked (n = 44), had quit smoking (n = 15) and/or were light consumers of alcohol (n = 55), even though this profile depicts the prototypic patient with HPV related HNSCC.<sup>15</sup>

Liang et al also found the incidence of HPV to be very low in oral tongue cancer across a wide range of patient ages including patients <45 years and hence concluded that the rising trend of oral tongue cancer in patients without traditional risk factors, seemingly, cannot be attributed to HPV.<sup>16</sup>

Van rensburg et al, in 1995, studied 66 pts for HPV and found only 1 patient to be positive. Another study by same authors in 1996 among 146 patients of OCSCC could demonstrate only 2 cases i.e. 1.4%<sup>17</sup> positive for HPV infection.

Similar studies carried out by Sand et al<sup>18</sup> in Sweden and Smith et al<sup>19</sup> in USA in 2004 among OSCC patients found 3/24 (24%) and 10/106 (9.4%) patients positive for HPV respectively.

Moreover, Rivero et al in 2006 found no patient positive for HPV out of a group of 23 patients included in their study in Brazil.<sup>20</sup>

The lowest prevalence of HPV infection has been recognized in Africa. Van Rensburg et al. and Boy et al. have reported low HPV prevalence in South African patients with OSCC, varying from 0% and 11.9%. Also, in Sudanese patients, Ibrahim et al. have failed to detect HPV DNA in OSCC specimens.<sup>21, 22, 23</sup>

These findings suggest that HPV infection in this area plays a marginal role in oral carcinogenesis and that different cofactors participate in the development of OSCC.

There is not much data available regarding the incidence of HPV-induced oral cancers in the Indian scenario as well. A few studies carried out in the Indian population reported HPV prevalence ranging from 17 to 67%.

In a study by Jalouli et al. performed in Indian population, HPV prevalence was 24% (15/62) in the 62 samples from OSCC patients.<sup>24</sup> In another study of 110 patients from Eastern India by Nagpal et al. in 2002 with tobacco-induced OSCC, out of 37 patients who tested positive for HPV, HPV-16 was the most frequently encountered type

(22.7%), followed by HPV-18 (14.5%) and HPV-16/18 coinfection (10%).<sup>25</sup>

The HPV prevalence of 16.8% was reported by Pillai et al. in 1999 in South Indian population.<sup>26</sup> Similarly, Agarwal et al studied 20 patients of OSCC & had 7 cases positive for HPV.<sup>27</sup>

Barwad et al identified 47.1% HPV prevalence in OCSCC patients in their study group. However this high prevalence may be attributed to nonspecific detection of HPV instead of concentrating on high risk types.<sup>28</sup>

As types other than high risk HPV may be just bystander infection than carcinogenic, nothing can be proven in this regard.

In another study by Kopikar et al at Tata Memorial Hospital, high risk HPV was detected in only 6% of oral cavity cancers.<sup>29</sup>

There are three large reviews in the literature reporting association of HNSCC with HPV worldwide. McKaig et al. in 1998 noted that HPV prevalence in HNSCC detected by PCR was 34.5%, by ISH 15.8%, and by Southern blot 24.5%.<sup>30</sup>

In 2008, a meta-analysis (1988–2007) of 62 studies by Termine et al reported a total of 4852 samples of HNSCC. The papers selected denoted a marked degree of heterogeneity with respect to the sample size: the majority of these papers concerned had <100 cases (45 of 62, 72.6%). Regarding geographical location, the majority of the studies regarded patients from America, Europe and Asia (21, 20 and 17 studies, respectively), while the remaining four studies were from Africa. The pooled prevalence of HPV DNA in the overall samples was 34.5%, while it was 38.1% in OSCC and 24.1% in the non site-specific HNSCC group.<sup>31</sup>

Similarly, Kreimer et al. in a review of 5,046 cases of HNSCC found an overall HPV prevalence of 25.9% and 23.5% in OSCC.<sup>32</sup> In both these meta-analysis, PCR-based studies reported a higher prevalence of 34.8% when compared with ISH based studies.

## Conclusion

Head and neck cancer is one of the common cancers associated with large morbidity and mortality. This is especially true for the Indian subcontinent where it is the most common cancer in males and the third most common in females.

Human Papilloma Virus has been proved beyond doubt, to be an important etiological agent in the HNSCC/ oropharyngeal carcinomas. However, its role in the oral cavity SCC proper has yet not been certified. As far as India is concerned, tobacco and alcohol still top the list of etiological agents of OSCC, making HPV a minor causal factor or just an additive agent for OSCC. It might be possible to explain this on the basis of the cultural differences and the differences in the sexual behavior.

However, studies with larger number of patients and longer duration of follow up are required to establish HPV as a definitive etiological agent for OSCC proper.

**Conflicts of interest:** None

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