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# Evaluation of Human Papilloma Virus Infection Using Immunohistochemistry in Histologically Diagnosed Oral, Oropharyngeal and Laryngeal Cancers

**Reema Sachdev[1], Seema Baxi[2]**

[1]Third Year Resident, Deparment of Pathology, Sir-T hospital and Government College, Bhavnagar, India
Email: *reemasachdev17[at]gmail.com*

[2]Additional Professor, Deparment of Pathology, Sir-T hospital and Government College, Bhavnagar, India
Email: *seemabaxi[at]yahoo.com*

**Abstract:** ***Introduction: Oral, oropharynx and laryngeal cancers are reported to be associated with HPV. There is no data available regarding percentage of HPV induced head and neck cancers from most areas of Gujarat. Hence, this study was planned. Aims and Objectives: 1) To estimate overall propotion of HPV in oral, oropharyngeal and laryngeal squamous carcinomas. 2) To assess association of age, gender and site specific prevalence of oral, oropharyngeal and laryngeal squamous carcinomas related to HPV infection. Materials and Methods: A prospective hospital based study of HPV by immunohistochemistry was done on biopsies and surgical specimen of 24 consecutive cases of histologically proven oral, oropharyngeal and laryngeal squamous cell cancers. Inclusion criteria: All the oral biopises and surgical specimens received in pathology department which were histologically proven for oral, oropharyngeal, laryngeal squamous cell cancers. Exclusion criteria: Family history of oral, oropharyngeal and laryngeal squamous cell cancers and previously taken HPV vaccine. Results: Total Proportion of HPV in HNSCC was 25% in Bhavnagar region. Males were most commonly affected; most common age group was between 41-60 years and most common site of HPV associated HNSCCs was oropharynx. Conclusion: HPV is detected in HNSCCs in Bhavnagar region.***

**Keywords:** HPV, IHC.

## 1. Introduction

Oral, oropharyngeal and laryngeal squamous cell cancers are one of the four most common cancers in Bhavnagar district. Data suggests that 10%-46% of oral, oropharyngeal and laryngeal squamous carcinomas are due to HPV infection[1, 2]. HPV induced oral, oropharyngeal and laryngeal squamous cell cancers are seen in younger patients[3, 4], have a worse morphology of non-keratinizing type but have a better prognosis than non HPV cancers[5, 6]. No data is available regarding the percent of HPV induced oral, oropharyngeal and laryngeal squamous carcinomas from Gujarat and the region of this study. Hence, this study is planned with the following aims:

### Aims and Objectives

1) To estimate overall proportion of HPV in oral, oropharyngeal and laryngeal squamous carcinomas.
2) To assess association of age, gender and site specific prevalence of oral, oropharyngeal and laryngeal squamous carcinomas related to HPV infection.

## 2. Materials and Methods

### Material

A prospective hospital based study of HPV by immunohistochemistry was done on biopsies and surgical specimen of 24 consecutive cases of histologically proven oral, oropharyngeal and laryngeal squamous cell cancers.

### Method

1) Tissues received in lab were processed by routine methods for histopathology.
2) The demographic details and detailed history were entered in case record form after taking consent of patient.
3) After microscopic diagnosis, formalin fixed tissues of these cases were processed for immunohistochemistry for HPV.
4) Histological features of the tumor were noted and correlated with the result of HPV.

### Inclusion criteria

All the oral biopises and surgical specimens received in pathology department which are histologically proven for oral, oropharyngeal, laryngeal squamous cell cancers.

### Exclusion criteria:

1) Family history of oral, oropharyngeal and laryngeal squamous cell cancers.
2) Previously taken HPV vaccine

## 3. Results and Analysis

Total 24 HNSCC cases presented during the study period. From which 21 were biopsies and 3 were modified radical neck dissection specimens. From that, 6 cases were HPV positive. Hence total proportion of HPV in HNSCC is 25% in this study.

**Table 1:** Age and Sex wise split of HNSCCs and their relation to HPV infection

| Age Groups | Male | | Female | |
|---|---|---|---|---|
| | Total Number | HPV status | Total Number | HPV status |
| 20-40 Years | 02 | - | 01 | - |
| 41-60 Years | 08 | 02 | 04 | 02 |
| >60 Years | 06 | 02 | 03 | - |

Hence the Male: Female ratio of HNSCCs was 2: 1 and HPV positive cases were above 40 years of age.

**Table 2:** Cancer site and HPV status

| Cancer site | Number | HPV % as per cases in each site | Overall HPV status from HNSCC of respective site |
|---|---|---|---|
| Oral cavity | 16 | 01 | 6.25% |
| Oropharyngeal | 03 | 02 | 66.7% |
| Larynx | 05 | 03 | 60% |

HPV associated HNSCCs were seen predominantly in oropharynx (66.7%)

**Table 3:** Types of HNSCC malignancy and HPV status

| Histological Types | Total Numbers | % of variant from HPV positive HNSCC cases (6 cases) | HPV % as per cases in each variant |
|---|---|---|---|
| Squamous cell carcinoma | 20 | 66.7% | 04 (20%) |
| Basaloid variant of SCC | 02 | 16.7% | 01 (50%) |
| Verrucous carcinoma | 02 | 16.7% | 01 (50%) |

From the 6 cases of HPV detected, 67% were SCC, basaloid variant of SCC & verrucous cancer were 17% each.

**Table 4:** HPV status in different grades of SCC

| Histological grades | | Total Numbers (Out of 20 cases) | HPV % as per cases in each grade |
|---|---|---|---|
| As per Differentiation | As per keratinization | | |
| Well differentiated | Keratinizing SCC | 13 | 01 (7.6%) |
| Moderately differentiated | Non-keratinizing SCC | 05 | 03 (60%) |
| Poorly differentiated | | 02 | 00 |

HPV positivity was higher amongst cases of moderately differentiated (Non-keratinizing) SCC.

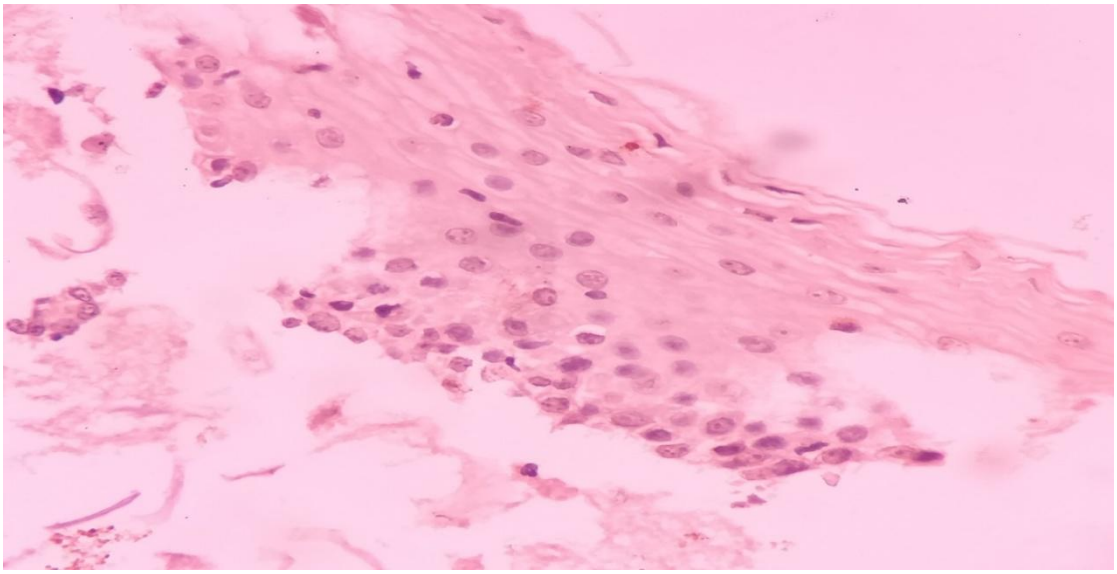

**Figure 1:** Shows moderately differentiated SCC of Vallecula, <5% cells Showing 1+ Nuclear staining (HPV IHC 40x)

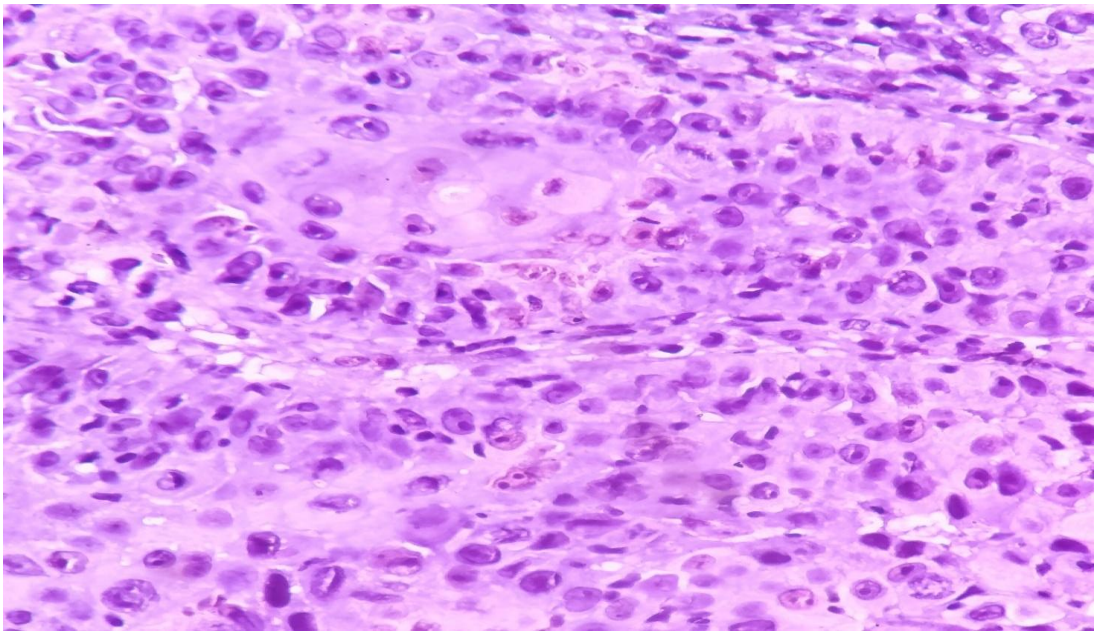

Fig 2 shows Well Differentiated SCC of Base of tongue, 5% cells are showing 1+ Nuclear staining (HPV IHC 40x)

## 4. Discussion

Despite extensive literature survey, no publications or material could be found on HPV detection by IHC. For discussion, data of other studies of HPV detected by other measures (ISH and PCR) have been used.

**Table 5:** Total HPV proportion of HNSCC in Gujarat comparison with different studies

| Studies | Sample size & Method | Total HPV positive | Total HPV proportion |
|---|---|---|---|
| Patel K. et al[7] (2013) (Gujarat) | 97 (PCR) | 00 | 00 |
| Goswami et al[8] (2017) (GCS, Gujarat) | 100 (PCR) | 20 | 20% |
| Present study | 24 | 06 | 25% |

It can be seen from table 1 that in Patel K et al study there is no association found between HNSCC and HPV infection. However, in Goswami et al total proportion of HPV in HNSCC is 20% which is consistent with result of this study.

## 5. Conclusion

1) Proportion of HPV infection detected by IHC was 25% in histologically diagnosed HNSCCs in hospital based population in Bhavnagar.
2) HPV associated HNSCCs were seen most commonly in male, age group 41-60 years and most common site involvement was oropharynx.
3) Classical SCC of moderately differentiated grade was the most common variant associated with HPV infection.

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## Author Profile

**Dr. Reema Sachdev,** 3rd Year resident, Department of Pathology, Sir-T hospital and Government medical College, Bhavnagar.
**Postal address:** Room No.30, Pg 1 hostel, Sir-T hospital campus, Jail road, Kalanala, Bhavnagar-364001

**Dr. Seema Baxi,** Additional Professor, Deparment of Pathology, Sir-T hospital and Government medical College, Bhavnagar.
**Postal address:** SUMIRAN, Plot No-2201-A1/2, Near Fulwadi Chowk, Hill Drive, Bhavnagar-364002