Original Research Paper

VOLUME - 8 | ISSUE - 2 | April - 2024 | ISSN (O) : 2663-2888 | ISSN (P) : 2663-287X

IMMUNOEXPRESSION OF P16 AND KI67 MARKERS IN ORAL SQUAMOUS CELL CARCINOMA- A STUDY OF 56 CASES

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ABSTRACT

Background: Oral squamous cell carcinoma (OSCC), is the third most common form of cancer in the developing countries and sixth most common cancer in the world. Oral carcinogenesis is a multistage process arising from the accumulation of genetic events that disturb cell cycle control, proliferation, motility, survival and tumour-related angiogenesis. Hyper methylated p16 promoter tend to transform into oral cancers. Ki67 is a monoclonal marker and exclusively related to proliferation of cells and aggressiveness of malignant tumours. In this study, we evaluated the expression of p16, and Ki67 in different oral squamous cell carcinoma. **Material and method:** Prospective study was conducted on total of 56 oral biopsy received in the Department of Pathology, Indira Gandhi Institute of Medical Sciences, Patna. Inclusion criteria include patient above 18 years of age and oral biopsy received with a clinical suspicion of Oral squamous cell carcinoma. **Result:** We found that 46 cases (82.1%) was seen in male. Out of 56 cases, 22 cases (27.5%) were well differentiated, 31 cases (38.7%) were moderately differentiated and 3 cases (3.8%) were found to be poorly differentiated Oral squamous cell carcinoma. p16 was negative in 24 cases (42.9%) and positive in 32 cases (57.1%). Expression of p16 was increases with increasing tumor grade. Ki67 was positive in all cases and its expression increases with increasing tumor grade. **Conclusion:** The expression of cell cycle proteins p16 and proliferative marker Ki67 represents the significant marker to detect early oral and evaluate tumor grade of oral squamous cell carcinoma. These markers also help in deciding treatment protocol and prognostic assessment.

KEYWORDS

Oral squamous cell carcinoma, p16, Ki67

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Introduction

Oral squamous cell carcinoma (OSCC), is the third most common form of cancer in the developing countries and sixth most common cancer in the world.1Oral carcinogenesis is a multistage process arising from the accumulation of genetic events that disturb cell cycle control, proliferation, motility, survival and tumour-related angiogenesis.2 p16 is an inhibitor of cellular division, product of CDKN2A gene, located on the 9p21 chromosome.3 It has been found that p16 hyper methylation is frequent in pre-cancerous oral lesions and lesions with a hyper methylated p16 promoter tend to transform into oral cancers.4 Ki67 is a monoclonal marker used for proliferation. It is exclusively related to proliferation of cells and aggressiveness of malignant tumours.5 The aforementioned features make Ki67 as one of the best markers to calculate cell proliferation, and it can be used as a reagent that helps in determining a patient prognosis for several types of tumours.[6]

Materials and method

The present study was conducted a prospective study of 56 cases of Oral squamous cell carcinoma diagnosed in the period of September 2019 to august 2021 in the Department of Pathology, Indira Gandhi Institute of Medical Science, Patna. Inclusion criteria include patients above 18 years of age. Oral biopsy received for evaluation with a clinical suspicion of oral squamous cell carcinoma and slides received in Department of Pathology for review. A detailed history with special emphasis on addiction to Tobacco chewing and socioeconomic status in patients presenting with oral lesions was taken.Biopsy received was stained by using Haematoxylin & Eosin stain for histological typing and grading of the tumours was graded according to the WHO criteria.[7]

Immunohistochemistry

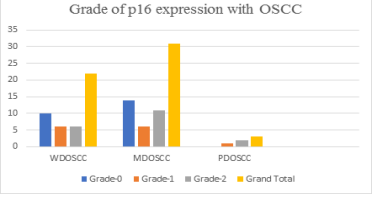
Immunohistochemical staining was done for p16 and Ki-67. 3–4 m thick sections taken on poly-L-Lysine precoated slides was dewaxed and stained with monoclonal mouse antibodies from master diagnostica. Adequate Positive and negative controls was taken. The results were recorded as according to the percentage of positive stained cells in 1000 counted cells in the ×40 microscope field. For p16 grade was assigned 0 (<10%), 1 (10%–50%) and 2 (>50%).For Ki67 grade was assigned 1+(10-30%),2+(30-50%) and 3+(>50%).For Ki67, nuclear positivity was considered as positive expression, and for p16, combined nuclear and cytoplasmic staining was considered as positive expression.

Result

In the present study, majority of the cases were males and most of the cases belongs to age group of 51-60 years. Out of 56 cases of OSCC, 22 cases of WDOSCC, 31 cases of MDOSCC and 3 cases of PDOSCC were found. In this study, majority of the cases were from lower socioeconomic class. Among the 56 cases, 46 (82%) cases had history of tobacco chewing or smoking habits. In respect to histological grading 54.5% of well differentiated, 54.8% of moderately differentiated and 100% of poorly differentiated were p16 positive. Similarly expression of Ki67 positivity increases with increasing histological grade of squamous cell carcinoma.

Grade of p16 expression with OSCC

Grade of p16 expression with OSCC			
	WDOSCC	MDOSCC	PDOSCC
Grade-0	10	14	0
Grade-1	6	6	1
Grade-2	6	11	2
Grand Total	22	31	3

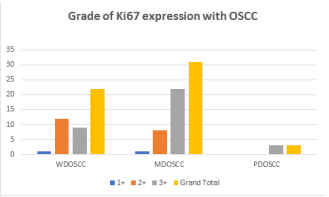


Grade of Ki67 expression with OSCC

Grade of Ki67 expression with OSCC			
	WDOSCC	MDOSCC	PDOSCC
1+	1	1	0
2+	12	8	0

VOLUME - 8 | ISSUE - 2 | April - 2024 | ISSN (O) : 2663-2888 | ISSN (P) : 2663-287X

3+	9	22	3
Grand Total	22	31	3



Discussion

The present study was conducted on 56 cases of oral biopsy specimens. This study was done to evaluate the detailed clinical history and sociodemographic profile of patients with malignant oral squamous epithelial lesions. In the current study the expression of p16 and Ki67 in malignant oral squamous epithelial lesions was assessed. In this study further comparison of the expression of p16 and Ki-67 with the histological type and tumour grade was done.

In present study mean age was found to be 40.5 years. The present study shows most common site of lesion is buccal mucosa (47.5%) followed by tongue (16.3%). Our study is similar to Kiran G et al. (2012)8. In the present study 78.3% of cases show personal history of Tobacco chewing /drinking/smoking which is similar to Tsai KY et al. (2009)9. In the present study it was observed that out of 80 cases, 22 cases (27.5 %%) were WDOSCC, 31 cases (40%) MDOSCC and 3 cases (3.75%) PDOSCC which is almost similar to Azizi SA et al. (2016).[10]

In case of OSCC where 12 cases (54.2%) of WDOSCC, 17 cases (54.8%) of MDOSCC and 3cases (100%) of PDOSCC shows p16 positive expression which is observed as increasing trend. Angiero F et al. have demonstrated an increase in p16 expression in higher grades of oral squamous cell carcinoma.11 It is also found that in 9 cases (40.9%) of WDOSCC, 23 cases (71.8%) of MDOSCC and 3 cases (100%) of PDOSCC shows 3+ Ki67 immunoexpression.

Conclusion

To conclude, we found that Ki-67 and p-16 can be useful as a marker of degree of dysplasia and transformation to malignancy. Expression of p16 and Ki67 increases with increasing histological grade of Oral squamous cell carcinoma. Ki-67 in addition can also serve as a marker of degree of differentiation of tumours. Thus, these immunohistochemical marker panels could be integrated with clinicopathological parameters for better assessment of patients with oral lesions. They can aid to detect early lesions. We recommend further evaluation of these markers for prognostic assessment. Such information will further enhance the judgment of clinicians for deciding the treatment protocol in the interest of patients. Further, studies with large sample size are required to establish a clear and definite conclusion in terms of clinical application of these molecular markers.

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