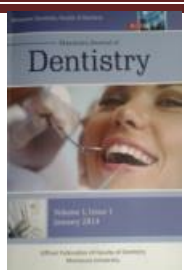
Mansoura Journal of DENTISTRY

Chronic periodontitis is defined as "an infectious disorder causes inflammation in the supportive tissues of the



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Abstract:

Angiogenesis is essential for the growth, invasion and metastasis of solid tumors. CD105 has been introduced as a marker of angiogenesis which stains vessels that are in the proliferating stage. The present study was carried out to investigate the immunohistochemical expression of CD105 in OSCC and correlate its expression in different histological grades as well as the available clinical parameters. Retrieved paraffin blocks of 30OSCC cases were used to evaluate CD105 immunohistochemical expression. Analysis revealed a statistically significant difference in CD105 expression among the different histologic grades and clinical stages.

Keywords: Oral squamous cell carcinoma. Angiogenesis. CD105. Microvessel density.

Introduction

Oral cancer is one of the most common cancers within the world with about 300,000 incident cases annually [1]. Oral squamous cell carcinoma (OSCC) represents 95% of all forms of head and neck cancer, and during the past decade its incidence has increased by 50%[2]. A variety of risk factors such as diet, alcohol, tobacco use, infections, genetic and environmental factors are related to oral cancer. Among these, tobacco smoking and alcohol use are wide researched and are universally considered as causative factors[3].

Angiogenesis is a process which new blood vessels develop from pre-existing blood vessels [0]. This process is a critical factor for cancer growth, progression and metastasis[4]. In addition, Microvessel density (MVD) is a parameter for evaluation of new angiogenetic patterns which has shown to be a prognostic factor for several malignant lesions including oral invasive carcinoma [5]. Overexpression of CD105 (endoglin) has been demonstrated in tumor vasculature and it has been suggested that the molecule has a role as a marker of proliferating endothelial cells [6,7] with no reaction with the normal tissue[5] . The purpose of our study was to study angiogenesis of OSCC by using CD105 and to investigate its correlation with the availabe clinico-pathological parameters.

Materials and Methods:

The present study was carried out on retrieved 30 paraffin embedded OSCC blocks. The slides cut at four µ thickness, were stained with hematoxylin and eosin for microscopical revaluation of the selected cases and were employed for immunohistochemical evaluation using anti CD105 antibody. A control group was formed by selecting five cases of oral pyogenic granulomas.

Evaluation of the immunohistochemical staining:

Angiogenic index for the OSCC cases were determined on the basis of immunoexpression of the biomarker CD105, using the microvascular count (MVC) technique, according to the procedure proposed by Maeda et al [8]. Each histological

section was examined with light microscopy at 40x magnification and five fields with the greatest degree of vascularization were identified. Next, the vessels in the areas selected were counted at 200x magnification. For each specimen, the MVC was expressed as the mean number of immunostained vessels per microscopic field. During this counting process, positively stained isolated endothelial cells and clusters of endothelial cells, with or without conspicuous lumen, were considered single vessels. The Immunoexpression of CD105 was classified as follows:

1) absence of staining; negative
2) focal staining; less than 50%
3) diffuse staining: more than or equal 50%

Descriptive data:

Descriptive statistics were calculated in the form of:

1. Mean ±Standard deviation (SD).
2. Frequency (Number-percent)

Analytical statistics:

Data were tabulated, coded then analyzed using IBM SPSS software package version 22.0**.** Qualitative data were described using number and percent. Quantitative data were described using mean, standard deviation for parametric data after testing normality using Shapiro-Wilk test. Significance of the obtained results was judged at the 5% level and all tests were 2 tailed. Chi-Square test and Monte Carlo test were used for categorical variables, to compare between different groups as appropriate.

Results:

This study was carried out on 30 cases of OSCC which were classified according to WHO classification system [9] as well differentiated (11 cases), moderately differentiated (11 cases), and poorly differentiated (8 cases). The results

showed that the age among the studied cases ranged from 23-88 years with a mean age of 61.37± 14.9 with a male to female ratio of 1:1. Regarding the immunohistochemical findings, there was no statistically significant difference in CD105 expression among the different age groups, gender, clinical presentation or anatomical site. However, there was statistically significant difference between CD105 expression among the histopathological grades and clinical stages (Table 1, 2).

Table (1) relation between CD105 expression and clinical staging

			Clinical staging		Total	p
			Stage I-II (Initial stage)	Stage III-IV (advanced stage)		
CD105 expression	Absence	n	5	0	5	0.007*
		%	41.7%	.0%	16.7%	
	Focal	n	5	9	14	
		%	41.7%	50.0%	46.7%	
	Diffuse	n	2	9	11	
		%	16.7%	50.0%	36.7%	
Total		n	12	18	30	

*statistically significant (p<0.05)

Table (2) relation between CD105 expression and histological grades (WHO)

			CD105 expression			Total	p
			absence	focal	diffuse		
Grade	Poorly differentiated	n	0	2	6	8	0.028 *
		%	.0%	14.3%	54.5%	26.7%	
	Moderate	n	1	6	4	11	
		%	20.0%	42.9%	36.4%	36.7%	
	Well differentiated	n	4	6	1	11	
		%	80.0%	42.9%	9.1%	36.7%	
Total		n	5	14	11	30	
		%	100.0%	100.0%	100.0%	100.0%	

*statistically significant (p<0.05)

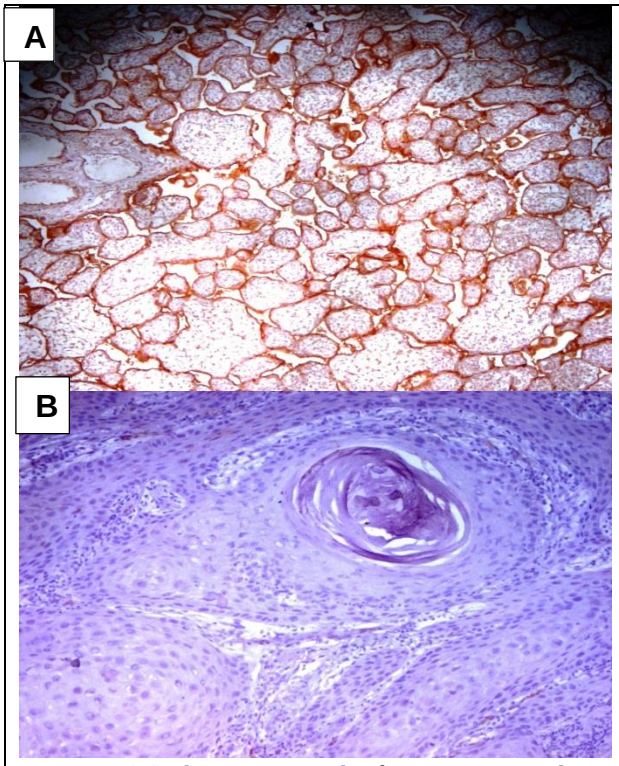

Figure 1: (A) Photomicrograph of pyogenic granuloma was used positive reaction (CD105, PAP-DAB ×200). (B) Photomicrograph of well differentiated OSCC showing absence CD105 expression (CD105, PAP-DAB×200).

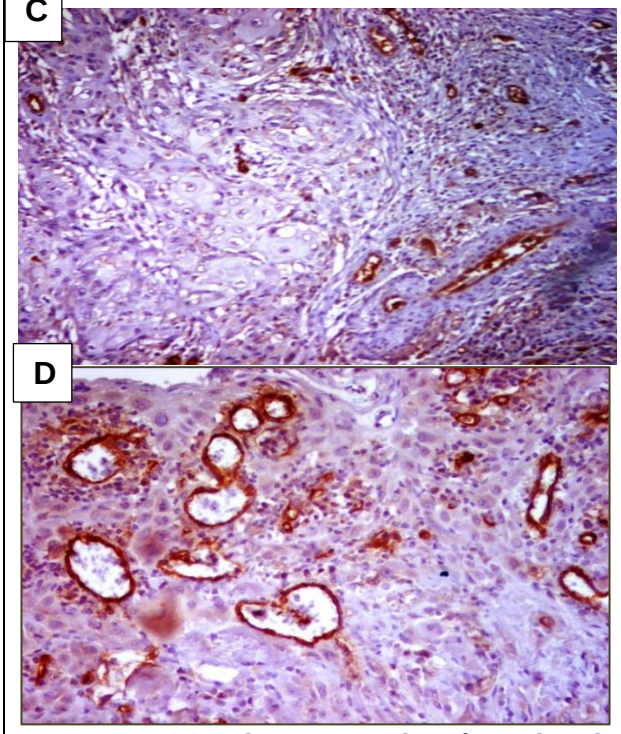

Figure 2 :(C) Photomicrograph of moderately differentiated OSCC showing focal CD105 expression (CD105,PAP-DAB×200).
(D)Photomicrograph of poorly differentiated OSCC showing diffuse CD105 expression (CD105, PAP-DAB x200).

Discussion :

In the present study, the immunohistochemical expression of CD105 showed no statistically significant difference among the various age groups, gender, shape of clinical presentation, and the anatomical sites of the studied OSCC cases. This results were consistent with studies performed by other investigators [10, 11]. Similar to Marinescu I (2014), [12] a positive association was found between CD105 expression and the histopathologic grades. This finding disagreed with other investigators[13,14,15,16,17]. Regarding the clinical staging, the current series revealed significantly higher CD105 expression in the cases of higher stages. This results is in accordance with previous studies that reported the higher the clinical , the more the CD105 expression[18,7,19,16]. Our results stands contradicting other findings that failed to reveal any significant association between Clinical stage and CD105 expression[15, 17].

In the current work, each of the clinical parameters of the TNM system showed significant association with the CD105 expression. Moreover, the individual parameters forming the TNM system (tumor size, lymph node involvement, distant metastasis) also revealed significantly different CD105 expression. This finding was in accordance with several studies considering the tumor size[16], [10,18], the stage of nodal metastasis[10]. This finding was in disagreement with Szafarowski T et al 2018[17] upon considering the tumor size. and in partial disagreement with Lee S et al [18] who found negative association between lymph node metastases group and CD105 expression. Therefore, based on the present results, the expression of CD105 may be used as a prognostic predictor of aggressiveness of OSCC cases and may aid in decisions on cancer diagnosis and therapy. However, the small sample size may be the reasons for differences between our results and previous studies. Accordingly, further studies with a larger sample size evaluating the CD105 in relation to prognosis are required to confirm the suggested role of CD105 in OSCC progression.

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