Original Research

Myofibroblasts as Important Diagnostic & Prognostic Indicators of Oral Squamous Cell Carcinoma

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Abstract

Background: Squamous cell carcinoma (SCC) accounts for approximately 94% of all oral malignancies, hence establishing oral squamous cell carcinoma (OSCC) as one of the top 10 most prevalent malignant tumors. Cells with several functions, such as macrophages and myofibroblasts, play a vital role in the biological behaviour of tumors. The aim of this study was to assess and evaluate the prevalence of myofibroblasts (MF) and macrophages in squamous cell carcinomas occurring in oral region.

Methodology: A total of 50 experimental subjects with well-differentiated oral squamous cell carcinoma (WDOSCC) ,moderately differentiated oral squamous cell carcinoma (MDOSCC), and poorly differentiated oral squamous cell carcinoma (PDOSCC) were taken .While 50 healthy subjects were taken as control group. The tissue samples were divided into sections that were 4 micrometers thick. These sections were subsequently subjected to both conventional staining using hematoxylin and eosin, as well as immunohistochemistry(IHC) staining using α-SMA. The comparative analysis of the expression levels of microRNAs was conducted across different stages of oral squamous cell carcinoma (OSCC). Statistical analysis was conducted on all of the outcomes.

Results: The findings revealed that the average final staining index score for patients with well-differentiated oral squamous cell carcinoma (WDOSCC) was 9.23, while it was 8.98 for those with moderately differentiated oral squamous cell carcinoma (MDOSCC), and 6.54 for individuals with poorly differentiated oral squamous cell carcinoma (PDOSCC). The control group, on the other hand, exhibited favourable cellular expressions.

Conclusion: The present investigation's results indicate that MFs play a significant role in the pathogenesis of OSCCs, and their evaluation may serve as a valuable tool for predicting the invasive characteristics of these malignancies. Consequently, we advocate for the utilization of MFs as a stromal marker to facilitate the identification of invasion and progression in patients with oral squamous cell carcinoma (OSCC).

Keywords: Alpha-smooth muscle actin, myofibroblast, oral squamous cell carcinoma.

Introduction

Oral cancer is a group of malignant diseases arising from the surface of the lips, gums, tongue, mouth, and palate. As keratinocytes are the major components of the epithelium over the oral cavity, squamous cell carcinomas (SCCs) account for 90–95% of patients with this subtype of head and neck malignant diseases in histology, followed by basal cell carcinomas, mesenchymal malignancies, hematologic tumors, and melanomas [1]. Oral

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SCC is a perennial major public health concern because of its high prevalence worldwide. According to 2018 statistics from the International Agency for Research on Cancer [2], approximately 350,000 cases of oral cancer are newly diagnosed each year, accounting for a cumulative incidence of 4.0 per 100,000 persons. In Taiwan, betel nut consumption has led to an incidence rate of 32.46 per 100,000 persons—the highest globally [3]. Therefore, several measures, including a population-based screening program, have been employed to prevent and control oral cancers in Taiwan [4].

Myofibroblasts are morphologically enlarged and irregular (star or web-shaped) fusiform cells with well-developed cell–matrix focal interactions and intracellular gap junctions [5,6]. The incorporation of α-SMA into actin stress fibres grants the myofibroblast contractile power, approximately 2-fold that of the force of fibroblasts, when cultured on substrates with high elastomer stiffness [7-9].

Hence, the current study was undertaken to assess the role of myofibroblasts as important diagnostic and prognostic indicators of oral squamous cell carcinoma.

Material and methods

The current study recruited 50 subjects with WDOSCC, MDOSCC, PDOSCC, and 50 healthy controls to evaluate the expression of myofibroblasts (MFs) using immunohistochemistry (IHC) utilizing a smooth muscle actin (SMA) antibody. The study sample consisted of 50 cases of well-differentiated oral squamous cell carcinoma (WDOSCC), moderately differentiated oral squamous cell carcinoma (MDOSCC), and poorly differentiated oral squamous cell carcinoma (PDOSCC), all of which were histologically confirmed. Additionally, 50 tissue samples from normal mucosa were included in the study, also with histological confirmation. The control group consisted of dental follicular tissue that was medically removed for orthodontic purposes, representing normal mucosa. Two slices, each with a thickness of 4 meters, were obtained from every tissue block. One tissue segment underwent staining using the conventional hematoxylin and eosin (H&E) method, while the other was submitted to immunohistochemical examination using the SMA marker. Hematoxylin and eosin (H&E) stained slides were employed as reference slides for the assessment and verification of oral squamous cell carcinoma (OSCC) cases.

Results

The present study involved the enrolment of 50 cases each of WDOSCC, MDOSCC, PDOSCC, as well as 50 controls. The investigation was carried out across three various grades of OSCC. Immunohistochemical analysis was conducted on the tissues using the SMA marker. The findings revealed that the average final staining index score for patients with well-differentiated oral squamous cell carcinoma (WDOSCC) was 9.23, while it was 8.98 for those with moderately differentiated oral squamous cell carcinoma (MDOSCC), and 6.54 for individuals with poorly differentiated oral squamous cell carcinoma (PDOSCC). The control group, on the other hand, exhibited favorable cellular expressions. The intergroup comparison of the final staining index score among different stages of oral squamous cell carcinoma (OSCC) showed no statistically significance difference($P > 0.05$). Similarly, the analysis of myofibroblast expression across different grades of OSCC showed uninteresting findings. There was a significant statistical difference ($P < 0.05$) observed in the final staining index score when comparing instances of oral squamous cell carcinoma (OSCC) with normal controls. Additionally, the expression of MF between the two groups also showed strong statistical significance.

Groups	P value
Well differentiated oral squamous cell carcinoma V/s Moderately differentiated oral squamous cell carcinoma	0.123
Well differentiated oral squamous cell carcinoma v/s poorly differentiated oral squamous cell carcinoma	0.101
Moderately differentiated oral squamous cell carcinoma v/s poorly differentiated oral squamous cell carcinoma	0.110

Table 1: Comparison of final staining index score between different grades of oral squamous cell carcinoma.

Discussion

Oral squamous cell carcinoma (OSCC) is the most frequent type of oral malignancy globally and is associated with a high mortality rate.[10] The progression of carcinomas has conventionally been attributed to a stepwise accumulation of genetic changes within the target epithelium. Such molecular progression has been demonstrated in the oral mucosa where it is initially reflected in the appearance of precursor lesions with epithelial hyperplasia and dysplasia followed later by the development of frank carcinoma, changes paralleled by increase in genetic alterations in the epithelium.[10] However, the focus on solely epithelial changes has begun to change, and a recent paradigm shift leads to increasing recognition that the micro-environment makes significant contributions to tumor progression.[11]

Concurrent with the conversion of nondiseased epithelial tissue to precancerous epithelium to carcinoma, the stroma also changes from normal to "primed" to "activated or tumor associated." Remodeling of the extracellular matrix (ECM) or "stromagenesis" is initiated by tumor cells, while stromal cells are responsible for the organization of this process. Fibroblasts are considered as one of the most important mesenchymal cells involved in tumor progression. Myofibroblasts are a unique group of cells phenotypically intermediate between smooth muscle cells and fibroblast.[12] In addition to their normal role in tissue homeostasis and repair, altered number and function of myofibroblasts have been implicated in diseases with increased ECM deposition and resultant fibrosis,[13] and now, researchers have started understanding their role in cancers. They modulate the tumor stroma through secretion of a myriad of factors such as chemokines, growth factors, and matrix-degrading enzymes like MMPs.MF are prominent feature of tumor stroma of many but not all OSCCs.[13] Hence, the current study was undertaken to assess the role of myofibroblasts as important diagnostic and prognostic indicators of oral squamous cell carcinoma.

The present study involved the enrolment of 50 cases each of WDOSCC, MDOSCC, PDOSCC, as well as 50 controls. The investigation was carried out across three various grades of OSCC. Immunohistochemical analysis was conducted on the tissues using the SMA marker. The findings revealed that the average final staining index score for patients with well-differentiated oral squamous cell carcinoma (WDOSCC) was 9.23, while it was 8.98 for those with moderately differentiated oral squamous cell carcinoma (MDOSCC), and 6.54 for individuals with poorly differentiated oral squamous cell carcinoma (PDOSCC). The control group, on the other hand, exhibited favorable cellular expressions. The intergroup comparison of the final staining index score among different stages of oral squamous cell carcinoma (OSCC) did not demonstrate statistical significance ($P > 0.05$). Similarly, the analysis of myofibroblast expression across different grades of OSCC showed uninteresting findings. There was a significant statistical difference ($P < 0.05$) observed in the final staining index score when comparing instances of oral squamous cell carcinoma (OSCC) with normal controls.

Prasad BV et al[14] evaluated the presence of myofibroblasts in OSCC, by immunohisto-chemistry using alpha smooth muscle actin (a-SMA) antibody. They evaluated a total of 50 biopsy specimens from the archives of the oral pathology, where 20 specimens out of 50 were of well-differentiated OSCC (WDOSCC), 20 were of poorly differentiated OSCC (PDOSCC), and 10 were of normal healthy controls. All the specimens were stained by immunohistochemically using with monoclonal antihuman α-SMA. Etemad-Moghadam et al method was used for assessing the myofibroblast distribution. Staining index was evaluated for the groups and compared. All the results were analysed by Statistical Package for the Social Sciences (SPSS) software. The mean percentage of myofibroblasts score for WDOSCC and PDOSCC were 2.88 and 2.92 respectively. The mean staining intensity score in WDOSCC and PDOSCC were 2.88 and 2.55 respectively. Statistically significant results were obtained while comparing the final staining index score between the OSCC group and normal control group. No significant correlation could be obtained while comparing the mean staining index score in between WDOSCC and PDOSCC. Malignant epithelium might induce the adjacent stromal tissue to produce myofibroblasts. These specialized cells may be utilized as therapeutic targets for the treatment of OSCC.

Conclusion

Based on the findings of the present study, it has been determined that MFs play a crucial role in the pathogenesis of OSCCs. Furthermore, the assessment of MFs may serve as a valuable tool in predicting the invasive behaviour of OSCCs. Hence, we advocate for the utilization of MFs as a stromal marker in patients with oral squamous cell carcinoma (OSCC) in order to facilitate the visualization of invasion and progression.

References

1. Dhanuthai K., Rojanawatsirivej S., Thosaporn W., Kintarak S., Subarnbhesaj A., Darling M., Kryshtalskyj E., Chiang C.P., Shin H.I., Choi S.Y., et al. Oral cancer: A multicenter study. Med. Oral Patol. Oral Cir. Bucal. 2018;23:e23–e29.
2. International Agency for Research on Cancer. [(accessed on 10 October 2020)]; Available online: http://gco.iarc.fr/
3. Cancer Registry Annual Report, 2016 (Taiwan) [(accessed on 20 January 2020)]; Available online: https://www.hpa.gov.tw/Pages/Detail.aspx? nodeid= 269&pid=12235
4. Chuang S.L., Su W.W., Chen S.L., Yen A.M., Wang C.P., Fann J.C., Chiu S.Y., Lee Y.C., Chiu H.M., Chang D.C., et al. Population-based screening program for reducing oral cancer mortality in 2,334,299 Taiwanese cigarette smokers and/or betel quid chewers. Cancer. 2017; 123:1597–1609.
5. Pakshir P., Hinz B. The big five in fibrosis: Macrophages, myofibroblasts, matrix, mechanics, and miscommunication. Matrix Biol. 2018;68–69:81–93.
6. Hinz B., Pittet P., Smith-Clerc J., Chaponnier C., Meister J.J. Myofibroblast development is characterized by specific cell-cell adherens junctions. Mol. Biol. Cell. 2004;15:4310–4320.
7. Wrobel L.K., Fray T.R., Molloy J.E., Adams J.J., Armitage M.P., Sparrow J.C. Contractility of single human dermal myofibroblasts and fibroblasts. Cell Motil. Cytoskelet. 2002;52:82–90.
8. Hinz B., Dugina V., Ballestrem C., Wehrle-Haller B., Chaponnier C. Alpha-smooth muscle actin is crucial for focal adhesion maturation in myofibroblasts. Mol. Biol. Cell. 2003;14:2508–2519.
9. Hinz B., Celetta G., Tomasek J.J., Gabbiani G., Chaponnier C. Alpha-smooth muscle actin expression upregulates fibroblast contractile activity. Mol. Biol. Cell. 2001;12:2730–2741.
10. Etemad-Moghadam S, Khalili M, Tirgary F, Alaeddini M. Evaluation of myofibroblasts in oral epithelial dysplasia and squamous cell carcinoma. J Oral Pathol Med. 2009;38:639–43.
11. Thode C, Jørgensen TG, Dabelsteen E, Mackenzie I, Dabelsteen S. Significance of myofibroblasts in oral squamous cell carcinoma. J Oral Pathol Med. 2011;40:201–7.
12. Angadi PV, Kale AD, Hallikerimath S. Evaluation of myofibroblasts in oral submucous fibrosis: Correlation with disease severity. J Oral Pathol Med. 2011;40:208–13.
13. Vered M, Dobriyan A, Dayan D, Yahalom R, Talmi YP, Bedrin L, et al. Tumor-host histopathologic variables, stromal myofibroblasts and risk score, are significantly associated with recurrent disease in tongue cancer. Cancer Sci. 2010;101:274–80.
14. Prasad BV, Kakatkar GS, Jain P, Jain M, Patel M, Khan J. Expression of Myofibroblasts in Oral Squamous Cell Carcinoma: An Immunohistochemical Study. J Contemp Dent Pract. 2016 Oct 1;17(10):857-860.