Case Reports and Reviews: Open Access

Research| Vol 1 Iss 2

Immunohistochemical Comparison of CD10 Expression in Dentigerous Cyst, Odontogenic Keratocyst, and Unicystic Ameloblastoma

Su Kyaw Myint*, Nandar Aung, and Zaw Moe Thein

Department of Oral Medicine, University of Dental Medicine, Yangon, Myanmar

***Corresponding author:** Myint SK, Department of Oral Medicine, University of Dental Medicine, Yangon, Myanmar; Tel: +9595028178; E-mail: sukyawmyint[AT]gmail.com

Received: November 12, 2020; **Accepted:** December 06, 2020; **Published:** December 13, 2020

Abstract

Odontogenic cysts and tumors are originated from clusters of cell rests of dental lamina retained in the tissues. The capacities with regards to additional expansion of these epithelial leftovers during cysts and tumors formation are at variance. CD10 is an enzyme endopeptidase, used to identify tumor cell differentiation and multiplication as well as prognosis of various types of tumors. Studies on behavior and aggressiveness of odontogenic lesions are still scarce. This article aimed to compare the immunohistochemical expression of CD10 in dentigerous cyst (DC), odontogenic keratocyst (OKC) and unicystic ameloblastoma (UA). In this laboratory based cross-sectional comparative study, immunohistochemical staining of CD10 was performed on histologically proven thirty-nine odontogenic lesions (13 DC, 13 OKC, and 13 UA). Results were statistically analyzed by using Chi-squared test, paired sample t-test, and independent t-test. Data showed mean percentage of CD10 immunoexpression was the highest in epithelial part of UA (63.85 ± 24.34) compared to DC (45.0 ± 23.28) and OKC (43.85 ± 28.44). CD10 immunoexpression was higher in stromal parts of UA and OKC (68.46 ± 17.25 and 64.23 ± 19.88 respectively) than DC (21.54 ± 15.73). There was statistically significant difference between CD10 epithelial immunoexpression and stromal immunoexpression within study groups (p = 0.021). Current data proposed that aggressiveness of UA and high recurrence rate of OKC might be related to increase in immunoexpression of CD10. It is indicated for surgical modification of these lesions compared to DC and recommended for regular follow-up.

Keywords: ***Dentigerous cyst; Odontogenic keratocyst; Unicystic ameloblastoma; CD10; Immunohistochemistry.***

1. Introduction

The jaws are host to a wide variety of cysts and neoplasms, to a great extent because of the tissues engaged with tooth development [1]. Odontogenic lesions have differing pattern of occurrence, recurrence and prognosis. Although most of the odontogenic lesions show benign histopathological characteristics, certain of those odontogenic lesions such as odontogenic keratocysts and ameloblastoma have neoplastic and destructive behavior [2]. Therefore, there needs to be studied more with appropriate markers to find out the underlying mechanism [3].

Citation: Myint SK, Aung N, and Thein ZM. Immunohistochemical comparison of CD10 expression in dentigerous cyst, odontogenic keratocyst, and unicystic ameloblastoma. Case Rep Rev Open Access. 2020;1(2):117.

Dentigerous cyst develops from accumulation of fluid around the coronal part or within the epithelium germinated by the reduced enamel epithelium [4]. This cyst type is harmless in nature and rarely recurs after removal [5]. Odontogenic keratocyst is a developmental cyst and has a high recurrence potential, with a recurrence rate of approximately 30% [6]. Unicystic ameloblastoma (UA), a subtype of intraosseous ameloblastoma, has been sub-classified into luminal, intraluminal, and mural based on the unfolding pattern of the ameloblastomatous epithelium [7]. It exhibits a recurrence rate of 6.7-35.7% [8].

CD10 is a member of matrix metalloproteases (MMPs) family characterized by the ability to degrade extracellular matrix proteins [9] that cleaves and inactivates neuropeptides and peptide hormones. Cell cycle is accelerated and cell motility is activated by interaction of tumor cells with CD10 positive stromal cells. Considering CD10 is fundamentally like other framework metalloproteinase and stromelysin, it may encourage the invasion and metastasis of tumor cells by forming a microenvironment [10]. CD10 might have an essential role in homeostasis, neoplastic changes and advancement of tumors [11]. Some studies have shown that a rise in CD10 expression might explain the part of mechanism of local tumor invasion in odontogenic lesions by proliferation of CD10 positive stromal cells [12]. High CD10 expression indicates poor prognosis in various tumors like breast carcinoma [13], cutaneous basal cell carcinoma [14] and oral squamous cell carcinoma [15].

In Myanmar, many studies that reveal the biological behavior of odontogenic lesions by using epithelial factors such as proliferation markers, tumor suppressor genes and other protein disclosing the pathway of cell cycle activity have been carried out. On the other hand, there was very few researches about pathogenesis and development of odontogenic lesions by using mesenchymal factors [16]. The present study was carried out to compare the epithelial and stromal expression of CD10 in dentigerous cyst, odontogenic keratocyst and unicystic ameloblastoma. Thus, CD10 might be useful in predicting tumor recurrence, evaluating tumor progression and behavior.

2. Materials and Methods

2.1 Selection of sample

Laboratory based cross sectional comparative study was conducted in Department of Oral Medicine, University of Dental Medicine, Yangon from September 2018 to August 2019. A total of 39 paraffin- embedded blocks of histologically proven odontogenic lesions (13 DC, 13 OKC and 13 UA) were selected along with biopsy requisition forms. Odontogenic tissue specimens with excessive tissue hemorrhage and intense inflammatory cells infiltrates, Ameloblastoma cases which were histologically follicular, plexiform, acanthomatous, granular, basaloid cell and desmoplastic and inadequate and unrepresentative specimens were excluded in this article.

2.2 Immunohistochemistry

Four Micrometer (4 μm) thick sections were cut from paraffin-embedded blocks and stained with routine Haematoxylin and Eosin (H&E) stain. All H&E stained tissue sections were thoroughly studied under binocular light microscope and diagnosed as DC, OKC and UA and then it was followed by immunohistochemical study by using monoclonal antibody against CD10 molecules.

Tissue sections 4 μm were mounted on poly-L-lysine coated slides and then were incubated for one hour at 65 °C on a slide warmer. Three changes of xylene, 5 minutes each were done to dewax the tissue slides. Microwave oven method was applied to bring back antigens. Three percent hydrogen peroxide was used to block endogenous peroxidase activity of the tissue for 15 minutes. The tissues were incubated with primary antibody (CD10) in a moist chamber at 4 °C overnight. Phosphate buffered saline was utilized three changes for 5 minutes each to wash primary antibody and then secondary antibody (goat antipolyvalent IgG) was used to incubate the tissue slides at room temperature for 30 minutes. The diaminoazobenzidine buffer was covered on the tissue slides and incubated for 5 to 20 minutes and was counterstained with Harris's hematoxylin.

2.3 Staining evaluation

Two pathologists reviewed all slides without knowing each other's score. CD10 positive cells were assessed the intensity and percentage of stained epithelial and stromal cells in 5 microscopic fields at 40 magnification. The number of positive cells was divided into the total number of cells counted in every field. The result was multiplied by 100 to find the percentage of positive cells. Scoring of the data was performed by the reference of Abdel-Aziz's study [17] as follows. If < 10% of epithelial and stromal cells show brown color in the membrane and cytoplasm of the cells, it was considered as negative. If those are stained > 10%, it was considered as positive. For the intensity, if 10-25% of epithelial and stromal cells show brown membranous and cytoplasmic staining, it was regarded as low intensity. If 26-50% of those cells stain, it was regarded as intermediate and if more than 50% of cells show brown membranous and cytoplasmic staining, it was considered as high intensity [17].

2.4 Statistical analysis

Analysis of the data was performed using Statistical Package for Social Science (SPSS software) version 18.0. Paired sample *t*-test was used to compare the epithelial expression and stroma expression of each lesion. Independent *t*-test was used to compare epithelial and stromal expression between studied groups. P-value < 0.05 was regarded as statistically significant.

3. Results

Of 39 samples, over all mean age of odontogenic lesions was 32.18 ± 16.07 where mean age of DC, OKC and UA were 27.62, 32.31 and 36.62 years respectively. DC, OKC and UA are the most common in 3rd and 4th decades. Among 13 samples of DC cases, 61.5% were male and 38.5% were female. In OKC cases, 53.8% were male and 48.2% were female. In UA cases, 46.2% were male and 53.8% were female. There was male sex predilection in DC and OKC. However, there was female sex predilection in UA. Of 13 samples of DC cases, 76.9% were occurred in maxilla and 23.2% were occurred in mandible. In OKC cases, 23.1% were detected in maxilla while 76.9% were detected in mandible. In UA cases, 7.7% were occupied in maxilla while 92.3% were occupied in mandible. DC was more commonly occurred in maxilla. In contrast, OKC, and UA were more commonly occurred in mandible (Table 1).

Table 1: Demographic Information of Study Groups.

Type of Lesions	Mean age	Gender		Site	
		Male	Female	Maxilla	Mandible
DC	27.62	8 (61.5%)	5 (38.5%)	10 (76.9%)	3 (23.2%)
OKC	32.31	7 (53.8%)	6 (48.2%)	3 (23.1%)	10 (76.9%)
UA	36.62	6 (46.2%)	7 (53.8%)	1 (7.7%)	12 (92.3%)

Based on Table 2, Regarding CD10 expression in epithelial part of DC, 23.1% showed low epithelial staining, 46.2% showed intermediate staining and 30.8% showed high epithelial staining. In OKC, 38.5% showed low epithelial staining, 23.2% showed intermediate staining and 38.5% showed high staining. In UA, 7.7% showed low staining, 23.1% showed intermediate and 69.2% showed high epithelial CD10 expression. Regarding to stromal CD10 expression, DC showed 69.2% of low staining and 30.8% intermediate staining. OKC showed 38.5% of intermediate staining and 61.5% high staining. UA showed 30.8% of intermediate staining and 69.2% high staining. There was statistically significant of CD10 expression in stromal part with p-value of < 0.001.

Table 2: CD10 Expression in Epithelial and Stromal Part of Dentigerous Cyst, Odontogenic Keratocyst, and Unicystic Ameloblastoma.

Epithelial expression	DC	OKC	UA	P-value*
No staining	-	-	-	0.165
Low	3 (23.1%)	5 (38.5%)	1 (7.7%)	
Intermediate	6 (46.2%)	3 (23.2%)	3 (23.1%)	
High	4 (30.8%)	5 (38.5%)	9 (69.2%)	
Stromal expression				
No staining	-	-	-	< 0.001*
Low	9 (69.2%)	-	-	
Intermediate	4 (30.8%)	5 (38.5%)	4 (30.8%)	
High	-	9 (61.5%)	9 (69.2%)	
* Chi-squared test				

According to Table 3, the mean epithelial CD10 expression of DC was higher than that of stroma. Mean stromal expression was higher than epithelial expression in OKC. CD10 expression in stromal part was higher than epithelial part of UA. The mean CD10 epithelial expression in UA was the highest followed by DC and OKC. The mean CD10 stromal expression in UA was the highest followed by OKC and DC. The CD10 expression was significantly different in epithelial and stromal part of DC, OKC, and UA with the p-value of 0.021.

Table 3: Comparison of CD10 expression in epithelial and stromal part of Dentigerous Cyst, Odontogenic Keratocyst and Unicystic Ameloblastoma.

Lesions	Epithelial expression	Stromal expression	P-value*
DC	45.0 ± 28.28	21.54 ± 15.73	0.021*
OKC	43.85 ± 28.44	64.23 ± 19.88	
UA	63.85 ± 24.34	68.46 ± 17.25	
*Paired sample t-test			

As Table 4 shows, the mean CD10 expression of OKC was significantly higher than DC in stromal part with p-value of < 0.001. The mean stromal CD10 expression in UA was significantly higher than that of DC with p-value of < 0.001.

Table 4: Expression of CD10 between Dentigerous Cyst, Odontogenic Keratocyst, and Unicystic Ameloblastoma.

Lesions	**Epithelial expression**		**P-value***
	DC (45.00)	OKC (43.85)	0.918
	OKC (43.85)	UA (63.85)	0.666
	DC (45.00)	UA (63.85)	0.081
	Stromal expression		
	DC (21.54)	OKC (64.23)	<0.001*
	OKC (64.23)	UA (68.46)	0.568
	DC (21.54)	UA (68.46)	<0.001*
*Independent t-test			

4. Discussion

Odontogenic cysts and tumors comprise a large proportion of pathologic lesions in maxillofacial region, with a common origin from the odontogenic epithelium; however, these lesions exhibit different aggressive and biologic behaviors [18]. The pathogenesis of odontogenic lesion is determined that the growth and clinical behavior of pathoses are controlled by interrelating between epithelial and stromal cells [19].

CD10, also known as neprilysin, can be discovered around the fibroblast like tumoral stromal cells inside the invasive field of a variety of cancers suchlike prostate, colorectal, breast and lung carcinoma [20]. CD10 expression in intratumoral stromal cells may contribute either positively or negatively to tumor invasion, growth and metastasis [21]. During cancer cells colonize and metastasize, stromal cells multiply. Abundant stromal cells or formation of fibrotic focus is a predictor of clinical aggressiveness [22].

In the present study, CD10 immunoreactivity showed membranous and cytoplasmic staining mainly in the surface layers of epithelium of DC (Fig. 1). For the stromal part, most of DC cases (69.2%) showed low staining (Fig. 2). These results were found to be in accordance to those reported by Masloub et al. [23], Tadbir et al. [2] and Deepa et al. [3]. High expression of CD10 cytoplasmic and membranous immunoreactivity in epithelial lining indicated the neoplastic activity of epithelial lining of DC [24]. In dentigerous cyst, the mean CD10 epithelial expression (45.0 ±28.28) was higher than stromal expression (21.54±15.73). The results were similar to Tadbir et al. [2]. This was possibly due to effect of both CD10 stromal cells and inflammatory cells on the overlying epithelium [3].

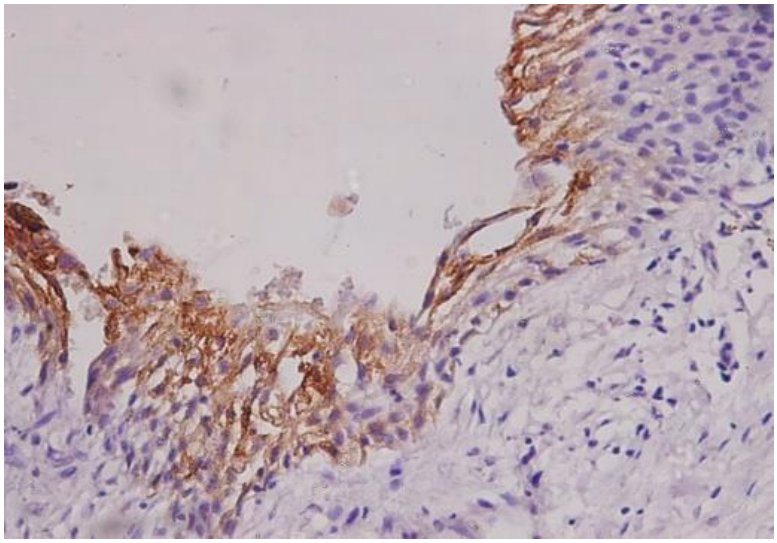

Fig.1. Immunohistochemical expression of CD10 in epithelium of DC x 400.

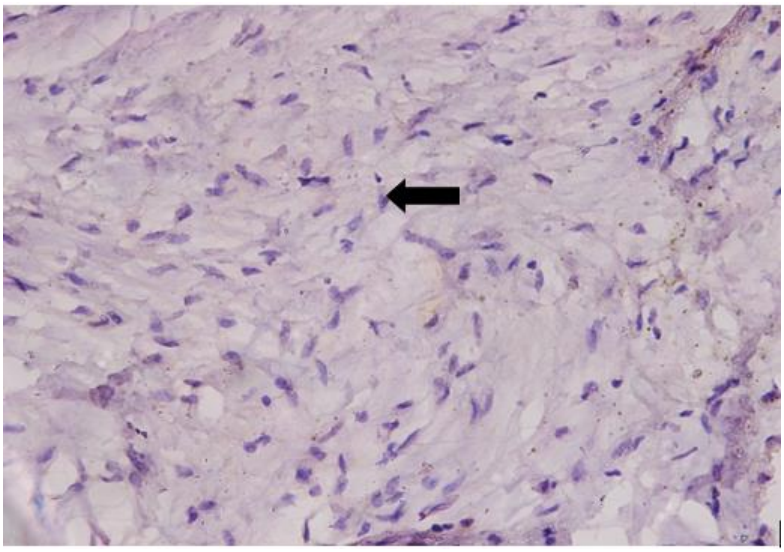

Fig. 2. Immunohistochemical expression of CD10 in stroma of DC x 400.

In odontogenic keratocyst, the expression of CD10 in the epithelial cell was observed especially in the basal and parabasal layers of the epithelial layer of OKC (Fig. 3). About one third of OKC cases (38.5%) showed high staining. However, the intensity of staining was high in majority of stromal cases (61.5%) (Fig. 4). These findings were indistinguishable from other findings of Deepa et al. [3] and Hormozi et al. [6]. The finding indicated the neoplastic potential of OKC epithelium and its tumor-like behavior, such as high rate of recurrence. For odontogenic keratocyst, the mean percentage of CD10 expressed cells in stromal part (64.23±19.88) was higher than that of epithelial part (43.85±28.44) (Table 3). This finding was consistent with Tadbir et al. [2]. Based on the results of previous studies, an increase in the expression of CD10 in the tumor stromal cells assisted in tumor progression and was further associated with poor prognosis in various tumors such as breast carcinoma, basal cell carcinoma, squamous cell carcinoma [14,25].

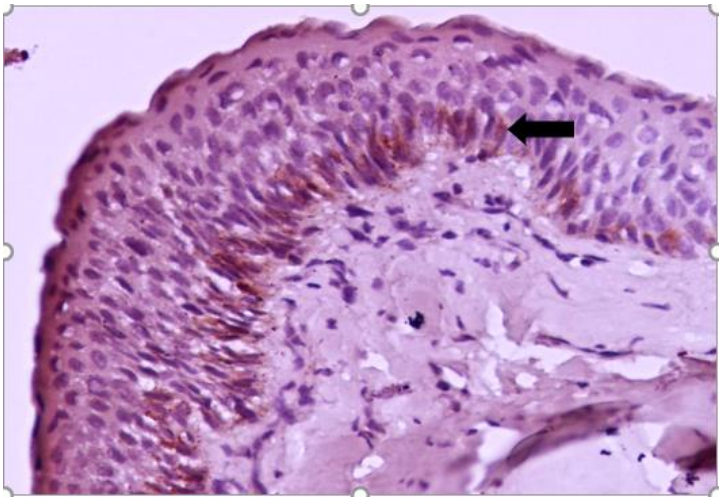

Fig. 3. Immunohistochemical expression of CD10 in epithelium of OKC x 400.

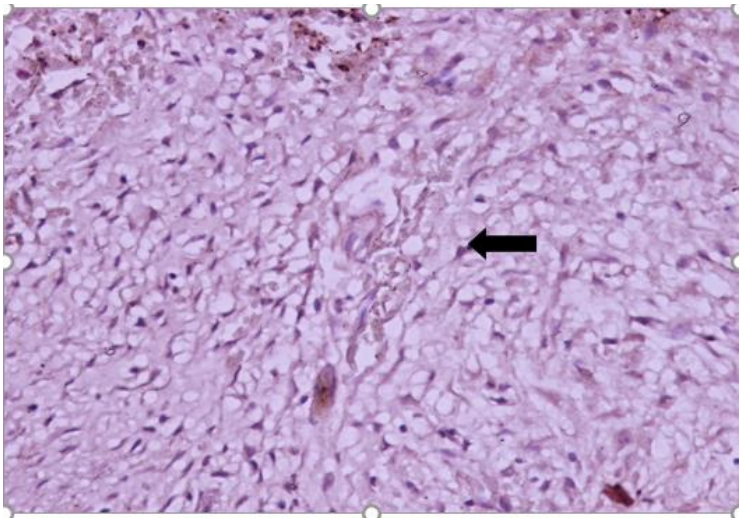

Fig. 4. Immunohistochemical expression of CD10 in stroma of OKC x 400.

Regarding unicystic ameloblastoma, the epithelial component CD10 immunoexpression was strongly positive in more than half of the cases (69.2%). Similarly, about 69.2% of cases was strongly positive in stromal component. This finding was slightly different from previous findings reported by Iezzi et al. [12], Ahlem et al. [26], Hormozi et al. [6] and Iqbal et al. [27] in which CD10 expression of UA only revealed low and intermediate immunoreactivity. In the current study, the intensity of both epithelium and stroma showed strong CD10 expression (69.2%) in UA (Fig. 5, and Fig. 6). The strong CD10 positivity in all components pointed out the aggressive behavior of UA. In this study, UA showed higher mean percentage of CD10 positive cells in stroma (68±17.25) than epithelium (63.85±24.34) (Table 3) but there was not much difference between these two values. This was similar to those studies conducted by Iezzi et al. [12], Tadbir et al. [2] and Ahlem et al. [26]. Increased immunoexpression of CD10 in stromal cells was related with nearby tumor invasion. It indicated that CD10 positive stromal cells were growing rapidly which was part of mechanism of invasive growth in ameloblastoma variants [13]. According to other studies, CD10 positivity in stromal cells was an indicator of worse prognosis; a significant correlation was found with lymph node metastases, local recurrences, and histological grade [15].

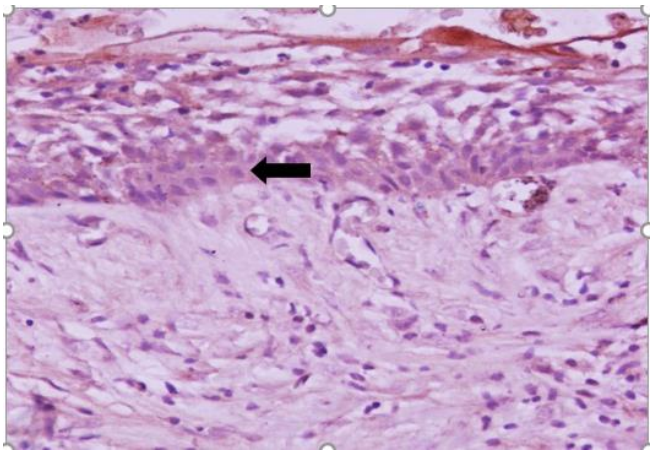

Fig. 5. Immunohistochemical expression of CD10 in epithelium of UA x 400.

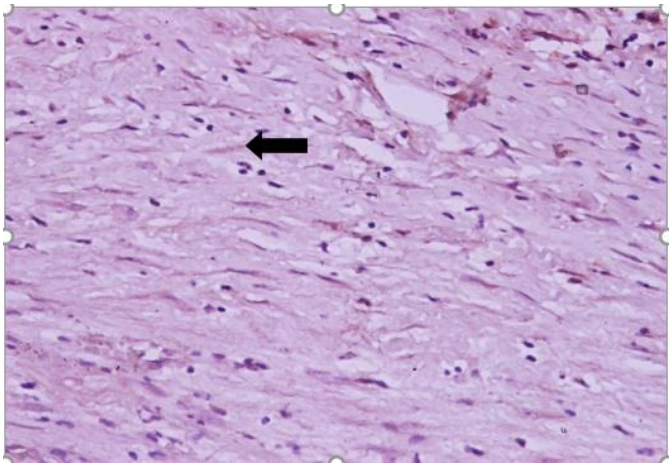

Fig. 6. Immunohistochemical expression of CD10 in stroma of UA x 400.

As shown in Table 4, there was no statistically significant difference in mean epithelial expressions among study groups. However, there was significant difference in mean stromal expressions between DC and OKC ($p < 0.001$) and between DC and UA ($p < 0.001$). The statistically difference was not observed between mean stromal CD10 expression of OKC and UA. This was consisted with the study of Tadbir et al. [2] and Hormozi et al. [6] which reported that the absence of any significant difference of CD10 expression in stroma of OKC and UA was probably be related to similar biological aggressiveness, neoplastic potential and high recurrence rate of OKC and UA. Since a high rate of recurrence had been reported for OKC [28] and one of the pathologic mechanisms for OKC, in a manner similar to UA, was related to the matrix metalloproteinases [29]. Many studies had been reported intense stromal CD10 expression in recurrent ameloblastoma. It can be concluded that tumor invasion of the extracellular matrix, part of mechanism of the invasive figure of ameloblastoma, was associated with CD10 expression by stromal cells [12,17].

5. Conclusion

This article, in line with the various studies published in the literature, showed that higher CD10 immunoexpression in stromal cells appear to be significant markers of local invasiveness and recurrence of UA and OKC. It can be observed that connective tissue cells also take part in significant role in the biological behavior of these lesions the same as

epithelial cells. When CD10 immunoreactivity is highly expressed, radical surgery is recommended and usual observation is required after surgery. Further studies with considerable number of samples and different odontogenic lesions are necessary to be explicit about the role of CD10 in tumorigenesis and to develop new approaches to treatment.

6. Acknowledgements

The authors would like to thank Rectors, Ethic, and Research Committee members and staffs from Department of Oral Medicine, University of Dental Medicine, Yangon. The authors would like to thank authorities and responsible persons from Ministry of Health and Sport, Myanmar for their financial support (External Grant 80/2019).

REFERENCES

1. Regezi JA. Odontogenic cysts, odontogenic tumors, fibroosseous, and giant cell lesions of the jaws. Mod Pathol. 2002;15**:**331–331.
2. Tadbir A, Geramizade B, Ranjbaran H. CD10 expression in dentigerous cyst, odontogenic keratocyst and ameloblastoma. Asian J Biol Sci. 2013;6(4):221-227.
3. Deepa K, Munisekhar MS, Charu S, et al. Comparision of immunohistochemical expression of CD10 in odontogenic cysts. J Clin Diagn Res. 2014;8(11):35-38.
4. Kramer IR, Pindborg JJ, Shear M. Histological typing of odontogenic tumours. 2nd ed. Berlin: Springer; 1992.
5. Kechik KA, Siar CH. Spatial distribution of osteopontin, CD44v6 and podoplanin in the lining epithelium of odontogenic keratocyst, and their biological relevance. Ann Diagn Pathol. 2018;32:17-22.
6. Hormozi E, Fard VN, Naseri MA, et al. Comparison of immunohistochemical expression of CD10 in keratocystic odontogenic tumor and ameloblastoma. Dent Res. 2016;13:110-116.
7. Philipsen HP, Reichart PA. Unicystic ameloblastoma: A review of 193 cases from the literature. Oral Oncol. 1998;34(5):317-325.
8. Black CC, Addante RR, Mohila CA. Intraosseous ameloblastoma. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2010;110(5):585-592.
9. Malemud CJ. Matrix metalloproteinases (MMPs) in health and disease: an overview. Front Biosci. 2006;11:1696-1701.
10. Iezzi G, Piatelli A, Rubini C, et al. CD10 expression in stromal cells of ameloblastoma variants. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2008;105(2):206-209.
11. Kim HS, Kim GY, Kim YW, et al. Stromal CD10 expression and relationship to the E-cadherin/B-catenin complex in breast carcinoma. Histopathology. 2010;56(6):708-719.
12. Huang WB, Zhou XJ, Chen JY, et al. CD10-positive stromal cells in gastric carcinoma: Correlation with invasion and metastasis. Jpn J Clin Oncol. 2005;25:245-250.
13. Iwaya K, Ogawa H, Izumi M, et al. Stromal expression of CD10 in invasive breast carcinoma: a new predictor of clinical outcome. Virchows Arch. 2002;440:589-593.
14. Aiad H, Hanout H. Immunohistochemical expression of CD10 in cutaneous basal and squamous cell carcinomas. J Egyptian Nat Cancer Inst. 2007;19:195-201.

15. Piattelli A, Fioroni M, Iezzi G, et al. CD10 expression in stromal cells of oral cavity squamous cell carcinoma: a clinic and pathologic correlation. Oral Dis. 2006;12:301-304.
16. Nadalin M, Fregnani E, Silva-Sousa Y, et al. Presence of myofibroblasts and matrix metalloproteinase 2 in radicular cysts, dentigerous cysts, and keratocystic odontogenic tumors: A comparative immunohistochemical study. J Endod. 2012;38(10):1363-1367.
17. Abdel-Aziz A, Amin MM. EGFR, CD10 and proliferation marker Ki67 expression in ameloblastoma: Possible role in local recurrence. Diagn Pathol. 2012;7:14.
18. Xie L, Moroi Y, Tsuji G, et al. CD 10 bearing fibroblast inhibits matrigel invasive potency of interleukin-I a producing squamous cell carcinoma by diminishing substance P level in the tumor microenvironment. Cancer Sci. 2010;101:2570-2578.
19. Ravikanth M, Soujanya P, Manjunath K, et al. Heterogeneity of fibroblasts. J Oral Maxillofac Pathol. 2011;15:247-250.
20. Truffi M, Mazzucchelli S, Bonizzi A, et al. Nano-strategies to target breast cancer-associated fibroblasts: Rearranging the tumor microenvironment to achieve antitumor efficacy. Int J Molecular Sci. 2019;20:3.
21. Takahara M, Chen S, Kido M, et al. Stromal CD10 expression, as well as increased dermal macrophages and decreased Langerhans cells, are associated with malignant transformation of keratinocytes. J Cutan Pathol. 2009;36:668-678.
22. Hasebe T, Tsuda H, Hirohashi S, et al. Fibrotic focus in invasive ductal carcinoma: an indicator of high tumor aggressiveness. Jpn J Cancer Res. 1996;87:386-394.
23. Masloub SM, Azim AM, Elhamidet SA. CD10 and osteopontin expression in dentigerous cyst and ameloblastoma. Diagnostic Pathol. 2011;6:44.
24. Anjum R, Naseem N, Nagi AH. Clinicopathological characteristics and expression of CD10 in soft tissue lesions associated with impacted third molar. Oral Health Dent Manag. 2014;13(2):165.
25. Oba J, Nakahara T, Hayashida S, et al. Expression of CD10 predicts tumor progression and unfavorable prognosis in malignant melanoma. J Am Acad Dermatol. 2011;65:1152-1160.
26. Ahlem B, Wided A, Amani L, et al. Study of Ki67 and CD10 expression as predictive factors of recurrence of ameloblastoma. Eur Ann Otorhinolaryngol Head Neck Dis.2015;132:275-279.
27. Iqbal F, Bashir N, Khan MM, et al. CD10 expression in ameloblastoma variants. Pakistan Oral & Dental Journal. 2018;38(2):151-155.
28. Neville BW, Damm DD, Allen DD, Bouquot JE. Odontogenic cysts and tumors. 4th ed. St. Louis, Elsevier; 2016.
29. Regezi JA, Sciubba JJ, Jordan RCK. Oral pathology: Clinical pathologic correlations. 7th ed. St. Louis, Saunders; 2017.

Citation: Myint SK, Aung N, and Thein ZM. Immunohistochemical comparison of CD10 expression in dentigerous cyst, odontogenic keratocyst, and unicystic ameloblastoma. Case Rep Rev Open Access. 2020;1(2):117.