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Immunohistochemical expression of Cyclooxygenase 2 reflects the proliferative activity in the epithelium of odontogenic lesions

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Abstract

Purpose: Odontogenic cysts and tumors comprise a major component of lesions of the oral and maxillofacial region. The pathogenesis of these lesions involves the interaction between the odontogenic epithelium and the ectomesenchyme. However, the clinical behavior of these biological entities is unpredictable. The aim of this study was to evaluate the role of Cyclooxygenase 2 (COX-2) in the pathogenesis and prognostication of odontogenic lesions.

Materials and Methods: In this study formalin-fixed paraffin-embedded tissue section of Odontogenic Keratocyst (n=10) Dentigerous cyst (n=10), Radicular cyst (n=10) and unicystic ameloblastoma (n=10) were immunohistochemically stained with COX-2 (NCL2-COX-2-4H12) and with Ki 67 (Ki-67 GM001) using standard staining protocols. The cytoplasmic expression of COX-2 in all the lesions was semi-quantitatively assessed. The pattern of expression of COX-2 among the different odontogenic lesions was statistical analyzed using the ANOVA test and the chi-square test.

Results: All the 40 odontogenic lesions that were evaluated expressed COX-2 immunohistochemically. A high number of odontogenic epithelial cells expressed COX-2 in most of the odontogenic keratocyst, radicular cyst and unicystic ameloblastomas. The expression of COX-2 was significantly (p=0.036) higher in Unicystic Ameloblastomas and Radicular cyst compared to that of Odontogenic Keratocyst and the dentigerous cyst.

Conclusion: The recognition that expression of COX-2 by odontogenic epithelial cells may indeed shed a new light on the biological mechanisms involved in the development of these benign yet aggressive lesions of the jaws. An insight into the molecular interactions occurring in the odontogenic epithelium will aid in better management of these lesions.

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Introduction

Odontogenic lesions are the most common benign lesions that occur within the jaw bones. These lesions are derived from the epithelium associated with the formation of the hard and soft tissues of the tooth. Around 52.3– 70.7% of the cysts that are submitted to an oral pathology biopsy service are Radicular cysts, 16.6-21.3% are Dentigerous cyst 5.4% - 17.4% are Odontogenic keratocyst (5.4–17.4%) [1].

The Radicular cyst also called the periapical cyst is the most common odontogenic cystic lesion. The World Health Organization classification of head and neck tumors describes Radicular cyst as a cyst of inflammatory origin associated with non-vital teeth [2]. It arises due to activation of the cell rests of Malassez that are present in the periodontal ligament due to an inflammatory stimulus. Radicular cysts present as a swelling of the jaw and may be associated with pain/loosening of tooth, root resorption of the affected tooth and displacement of the adjacent teeth [3].

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The most common developmental odontogenic cysts are Dentigerous cysts. Dentigerous cyst is an odontogenic cyst that is attached to the cervical region of an unerupted tooth and envelops the crown [2]. This cyst arises due to the separation of the dental follicle from around the crown of unerupted teeth. The dentigerous cysts usually develop in relation to an impacted mandibular third molar tooth. Long standing dentigerous cysts can cause root resorption, bone expansion and local destruction [4]

The Odontogenic keratocyst is an odontogenic cyst characterized by a thin regular lining of parakeratinized stratified squamous epithelium with palisading hyperchromatic basal cells. [2]. Odontogenic keratocyst originates from the remnants of the dental lamina and exhibits typical cystic lining of 6 to 10 cell layer thickness. The odontogenic keratocyst is recognized for its intrinsic growth potential and its high rate of recurrence [5].

Unicystic ameloblastoma is a variant of intraosseous ameloblastoma that occurs as a single cystic cavity with or without luminal proliferation [2]. Unicystic ameloblastomas usually occur in younger individuals. Although a variant of ameloblastoma it possesses a less aggressive growth pattern compared to a solid ameloblastoma. On histologic examination they show a typical ameloblastomatous epithelium lining part of the cyst cavity, with or without luminal and/or mural tumor proliferation [5, 6].

Cyclooxygenase 2 is a 72 kda cytokine-inducible enzyme that is situated in the nuclear membrane and the rough endoplasmic reticulum of the human cell [7,8]. COX-2 is an important enzyme responsible for the synthesis of the prostanoids; prostaglandins, prostacyclin and thromboxane [9]. During inflammation, there is an abundance of COX-2 in macrophages and other inflammatory cells [10]. An upregulation of COX-2 is brought about by growth factors and inflammatory mediators such as lipopolysaccharides and tumor necrosis factors [8].

An over expression of COX-2 has been associated with cell proliferation, apoptosis, angiogenesis and enhanced invasiveness [11,12]. The expression of C0X-2 has been associated with precancerous lesion and with the malignant tumors of the head and neck regions.[13]. However, the role of COX-2 in the clinical behavior of odontogenic lesions is less explored [14, 15, 16,17]. The aim of this study was to evaluate the expression of COX-2 in odontogenic lesions and assess its role in the clinical behaviour of these lesions.

Material and Methods

This study was approved by the Ethics Committee of Kasturba Medical College Manipal (108/2016). Forty formalin fixed paraffin embedded tissue specimens; Odontogenic Keratocyst (n=10) Dentigerous cyst (n=10) Radicular cyst (n=10) and Unicystic Ameloblastomas (n=10) were retrieved from the archives of the department. Five-micron thick sections were cut from each block and the sections were stained with hematoxylin and eosin. Tissue sections were reviewed to confirm the diagnosis and to determine the adequacy of the tissue for immunohistochemical staining.

Immunohistochemical Procedure

Four-micron thick sections were cut from the FFPE tissue blocks and were taken onto aminopropyl triethoxy silane (APES, Sigma-Aldrich Co. St. Louis, USA) coated slides.

Sections were deparaffinized through 3 changes of xylene and hydrated through descending grades of alcohol (100%, 95%, 70%). The endogenous peroxidase activity was quenched by incubating the sections in 3% H_2O_2 for 10 minutes.

Antigen retrieval was carried out using a domestic pressure cooker method with sodium citrate solution (pH 6.0). Following antigen retrieval, the tissue sections were allowed to cool at room temperature for 20 minutes. To block the non-specific binding sites the section were treated with protein block (Novacastra Lieca Biosystem, Newcastle) for 5 mins. The sections were then incubated in mouse monoclonal COX -2 primary antibody (NovacastraTM Mouse Monoclonal Antibody Cyclooxygenase-2: NCL-COX-2- 4H12)

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at 1:1000 dilution (with tris buffer, pH 7.6) for 60 minutes at 37°C in a moist humidifying chamber.

The antigen-antibody reaction was developed by incubating the section with secondary antibody (Secondary antibody Novolink polymer Anti-mouse IgG-Poly-HRP) for 30 minutes at room temperature in a moist chamber. The antigen-antibody reaction was detected using the chromogen Diaminobenzidine and its buffer. The sections were counterstained in Mayer's hematoxylin, were dehydrated through ascending grades of alcohol (70%, 95% and 100%) cleared in xylene and mounted with Dibutylphthalate Xylene (DPX).
For positive control tissue sections of normal buccal mucosa and colon carcinoma was used. For a negative control, a tissue section was stained in the similar manner except that instead of the primary monoclonal antibody, the tris buffer was used.

For Ki-67

Four-micron thick sections were cut from the FFPE tissue blocks (one each of Dentigerous cyst, Radicular cyst, Odontogenic Keratocyst and ameloblastoma) and were taken onto aminopropyl triethoxy silane (APES, Sigma-Aldrich Co. St. Louis, USA) coated slides. Sections were deparaffinized through 3 changes of xylene and hydrated through descending grades of alcohol (100%, 95%, 70%). Antigen retrieval was performed using tris EDTA buffer at pH 9.0, Endogenous peroxide was neutralized by treating the sections with pre-diluted 3% hydrogen peroxide. Following this, sections were incubated with pre diluted mouse monoclonal primary antibody-Ki -67 (clone Ki-67 GM001, Pathnsitu, USA) for 1 hour at room temperature in a moist humidifying chamber.

Then the sections were incubated with pre diluted primary target binder (PolyExcel Target Binder, PathnSitu, USA) at room temperature for 10 minutes. Slides were then incubated with secondary antibody pre diluted (PolyExcel Poly HRP, PathnSitu,USA), at room temperature for 10 minutes. The peroxidase activity was developed with Diamino Benzidine Tetra Hydrochloride (DAB). Finally, the sections were counter stained with Mayer's Haematoxylin, dehydrated, cleared, and mounted with Dibutylphthalate Xylene (DPX).

Assessment of COX-2 immunostaining

The expression of COX-2 was carried out in a semi-quantitative manner independently by two pathologists using a light microscope (Olympus BX41). Five high-power fields were observed for each case and the cells that showed a diffuse brown stain in the cytoplasm was considered positive for COX-2. The percentage of positive cells in the four fields was recorded, the mean percentage of positive cells in all the four fields was determined. The expression of COX-2 was scored as 0 (Absence of the stain), 1 (<25% of the cells were positive) 2 (25-50% of the cells were positive), 3 (51-75% of positive cells), and 4 (75-100% of positive cells).

The intensity of the expression of COX-2 was graded as 0 (Absence of the expression), 1 (weak expression), 2 (moderate expression) and 3 (strong expression). The Total score was obtained by adding the score of percentage of positive cells and the score of staining intensity as suggested by Mendes et al (modified) [15]. A total score of 0 was given if there was complete absence of the expression of COX-2. And finally, the expression of COX-2 was regarded as low expression when the sum of the score for the percentage of positive cells and the intensity of the expression ranged from 1-4 and the expression of COX-2 was regarded as a high expression when the sum of the score for the percentage of positive cells and the intensity of the expression ranged from 5-7.

Statistical Analysis

All statistical analyses were performed using the Statistical Package for Social Sciences (SPSS) version 16.0.

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Descriptive analysis was carried out for patient characteristics. Inter-observer variability was assessed using Cohen's kappa coefficient and was found to be of good reproducibility (0.92). The association between expression of COX-2 and the odontogenic lesions was assessed using a one-way ANOVA analysis method and the Chi Square test. A *p* value of < 0.05 was regarded as statistically significant.

Table 1. Clinico-pathological details of the Odontogenic lesions and their COX-2 expression profile

No.	Age	Gender	Site FDI Numbering system)	Odontogenic lesion	Mean % of Cells expressing COX-2	Score for number of cells expressing COX-2	Intensity of the expression of COX-2	Total Score of the expression of COX-2
1	22	M	Right ramus of the mandible	Dentigerous cyst	60	3	1	4
2	38	M	11,21,22	Dentigerous cyst	0	0	0	0
3	12	M	47,46	Dentigerous cyst	25	1	1	2
4	10	M		Dentigerous cyst	11	2	2	4
5	49	F	48,47	Dentigerous cyst	40	3	1	4
6	33	F		Dentigerous cyst	22	2	2	4
7	20	M	47,46,45,44,43,	Dentigerous cyst	20	1	2	3
8	18	F	13,12	Dentigerous cyst	46	3	1	4
9	47	M	11,22	Dentigerous cyst	17	1	2	3
10	23	M	17----11--21	Dentigerous cyst	20	1	2	3
11	51	M	16	Radicular cyst	80	4	2	6
12	23	M	21	Radicular cyst	85	4	2	6
13	14	F	36	Radicular cyst	40	2	2	4
14	22	M	11	Radicular cyst	35	2	2	4
15	65	M	13.....21	Radicular cyst	79	4	2	6
16	42	M	41---------31,32,33,32,35,36	Radicular cyst	90	4	2	6
17	27	F	22----------25	Radicular cyst	30	2	2	4
18	25	F	43----------33	Radicular cyst	45	2	2	4
19	14	M	15,14,13	Radicular cyst	40	2	2	4
20	45	M	12,11	Radicular cyst	43	2	2	4
21	36	M	37,38	Odontogenic Keratocyst	20	1	1	2
22	35	F	47,48	Odontogenic Keratocyst	80	4	2	6
23	71	M	47---41	Odontogenic Keratocyst	25	1	2	3
24	33	M	38	Odontogenic Keratocyst	80	4	2	6
25	71	M	47---------41	Odontogenic Keratocyst	30	2	2	4
26	21	M	Right ramus of the mandible	Odontogenic Keratocyst	90	4	2	6
27	15	M	47,46	Odontogenic Keratocyst	48	2	2	4
28	13	F	85----------,42,,,,35	Odontogenic Keratocyst	50	2	2	4
29	45	M		Odontogenic Keratocyst	40	2	2	4
30	34	F		Odontogenic Keratocyst	22	1	2	3
31	28	M	32,33,34,35	Unicystic Ameloblastoma	25	1	1	2
32	31	F	48,47	Unicystic Ameloblastoma	75	3	3	6
33	17	M	48	Unicystic Ameloblastoma	80	4	2	6
34	29	M	38	Unicystic Ameloblastoma	66	3	2	5
35	42	M	48	Unicystic Ameloblastoma	22	1	1	2
36	36	M	48	Unicystic Ameloblastoma	40	2	2	4
37	17	F	37,38	Unicystic Ameloblastoma	25	1	2	3
38	24	F	48..	Unicystic Ameloblastoma	85	4	2	6
39	24	F	48	Unicystic Ameloblastoma	60	3	3	6
40	10	M	46	Unicystic Ameloblastoma	92	4	2	6

Results

In this study the expression of cyclooxygenase-2 was evaluated in 40 cases of odontogenic cysts and tumors that included 10 cases of dentigerous cysts, 10 cases of Radicular cyst, 10 cases of odontogenic keratocyst and 10 cases of Unicystic Ameloblastoma. The age of the patients ranged from 10 years to 71 years (Mean age 31.5 years). Among the patients 27 of them were males and 13 of them were females. The clinico pathological details of all the odontogenic lesions along with their COX-2 expression pattern are given in Table 1. All the odontogenic lesions except one case of dentigerous cyst that were evaluated showed a mild-severe expression of COX-2 (Fig 1).

Among the dentigerous cysts in 4/10 (40%) cysts the mean percentage of odontogenic epithelial cells that expressed COX-2 ranged between 25%-50%. Among the radicular cyst in 4/10 (40%) the mean percentage of COX-2 positive cells was above 75%. Among the odontogenic keratocysts in 3/10 (30%) cases of percentage of COX-2 positive cells was above 75%. Among the unicystic ameloblastomas in 3/10 (30%) cases of percentage of COX-2 positive cells was above 75% (Fig 2).

Among the dentigerous cysts in 5/10 (50%) cases the intensity of COX-2 expression was that of a moderate expression. All the 10/10 (100%) cases of radicular cysts showed a moderate intensity of the expression of COX-2. Among the odontogenic keratocysts in 9/10 (90%) cases showed a moderate intensity of the expression of the of COX-2. Among the unicystic ameloblastomas in 6/10 (60%) cases showed a moderate intensity of the expression of COX-2 (Fig 3)

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Based on the overall expression of COX-2 in the odontogenic lesions, all the dentigerous cysts 10/10 (100%) cases, 6/10 (60%) of the radicular cysts, 7/10 (40%) of the odontogenic keratocysts and 4/10 (40%) of unicystic ameloblastoma showed a low expression of COX-2. While 4/10 (40%) of radicular cysts. 3/10 (30%) of the odontogenic keratocysts and 6/10 (60%) of unicystic ameloblastoma showed a high expression of COX-2 (Fig 4).

The comparison of the mean percentage of odontogenic epithelial cells expressing COX-2 is given in Table 2. The radicular cyst showed a high expression of COX-2 compared to dentigerous cyst or odontogenic keratocyst (p=0.031) (Table 3). Among all the odontogenic lesions, Unicystic ameloblastoma showed a high expression of COX-2 (p=0.036) (Table 4). The comparison of the relative risk among all the odontogenic lesions is given in Table 5.

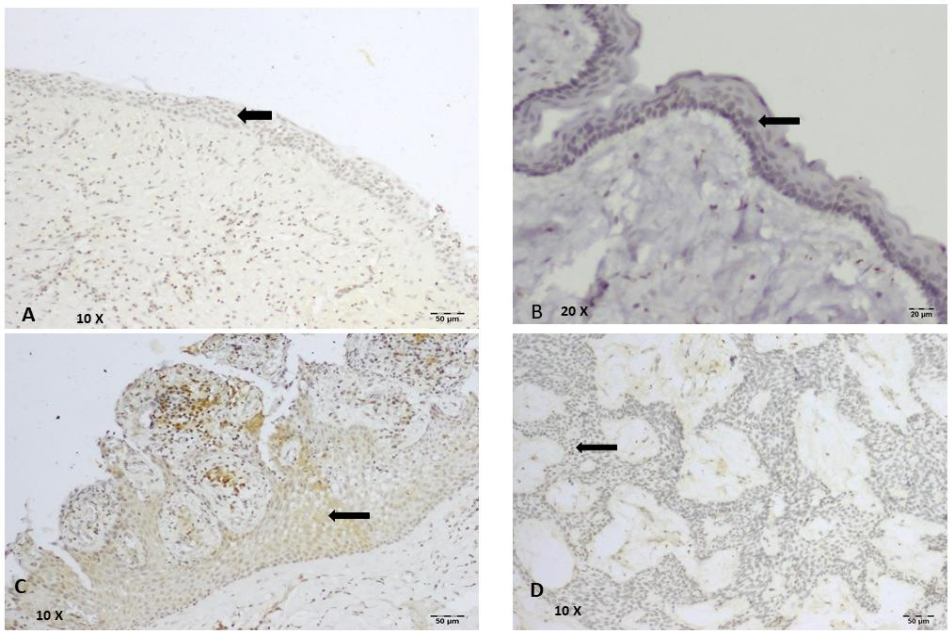

Fig 1. Expression of COX-2 in **A**. Dentigerous cyst, **B**. Odontogenic Keratocyst **C**. Radicular Cyst, **D**. Unicystic Ameloblastoma.

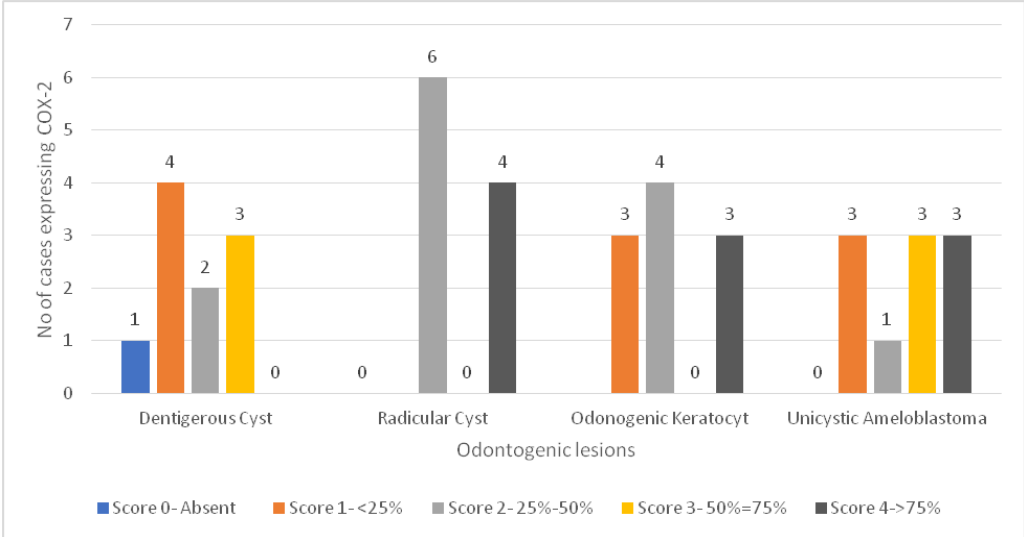

Fig 2. Comparison of the Percentage of COX-2 positive (Score) odontogenic epithelial cells in different odontogenic lesions

Discussion

COX-2, one of the isoforms of Cyclooxygenase is a key enzyme which mediates the process of inflammation. An up regulation of this enhances the synthesis of prostanoids especially Prostaglandin E [18, 19]. The COX-2/PGE2 pathway, by enhancing cell survival and growth, has been suggested to play a role in suppression of apoptosis and enhancing cell survival and growth [20]. This study evaluated the influence that COX-2 imparts on the proliferative activity of odontogenic epithelial cells. Dentigerous cysts develop due to accumulation of fluid (including glycosaminoglycan's) between reduced enamel epithelium of dental follicle and crown of unerupted teeth. In this study most of the Dentigerous cyst (9/10=90%) showed that 25%-50% of the reduced enamel epithelial cells expressed COX-2. About 50% of the cysts showed a moderate intensity of the expression of COX-2. In the comparative study by Seyedmajidi et al only 5/15 (33.3%) of the dentigerous cyst expressed COX-2, but only one case showed a high expression for COX-2. In the study by

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Alsaegh et al 13/16 cases expressed COX-2 [16].

In the later stages of the clinical course of dentigerous cyst, bone resorption is a common feature. Prostaglandin E 2 is a vital molecule that stimulates or inhibits bone metabolism and can increase the number of functionally active osteoclasts [21]. An upregulation of COX-2 in the cells of the reduced enamel epithelium will induce the synthesis PGE2 and facilitate bone resorption. Kumamoto et al demonstrated that sonic hedge hog (SHH) signaling molecule is expressed by the odontogenic epithelial cells of the dentigerous cyst [22]. The SHH pathway through activation of Ras/Raf/ERK pathway can in turn induce the expression of COX-2 in these cysts [16, 23].

Radicular cysts are categorized as inflammatory odontogenic cysts that occur as a sequel to dental caries. The infection in the periapical area stimulates the cell rests of Malassez in the periapical region to proliferate and result in a cyst formation.

In this study the expression of COX-2 was seen in more than 75% of the odontogenic rests in 4 out of the 10 cysts that were evaluated. Tsai et al in their study found that radicular cysts with more inflammation were associated with a high COX 2 expression.

Table 2. Comparison of the mean % of positive odontogenic cells among the different odontogenic lesions

Odontogenic lesion	Mean % of Positive odontogenic cells	Mean Difference	Std. Error	ANOVA test	95% Confidence Interval	
					Lower Bound	Upper Bound
Dentigerous Cyst	26.10±17.7	-30.600	10.683	.033*	-59.37	-1.83
Radicular Cyst	56.70±23.6					
Dentigerous cyst	26.10±17.7	-22.400	10.683	.174	-51.17	6.37
Odontogenic Keratocyst	48.50±26.2					
Dentigerous cyst	26.10±17.7	-30.900	10.683	.031*	-59.67	-2.13
Unicystic Ameloblastoma	57±26.9					
Radicular cyst	56.70±23.6	8.200	10.683	.868	-20.57	36.97
Odontogenic Keratocyst	48.50±26.2					
Radicular Cyst	56.70±23.6	-300	10.683	1.000	-29.07	28.47
Unicystic Ameloblastoma	57±26.9					
Odontogenic keratocyst	48.50±26.2	-8500	10.683	.856	-37.27	20.27
Unicystic Ameloblastoma	57±26.9					

*Significant association

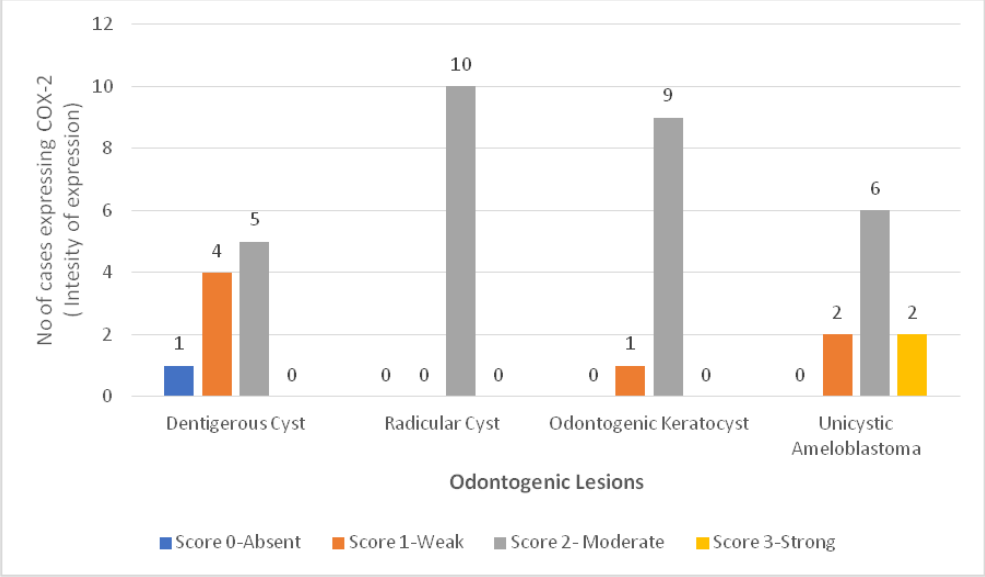

Fig 3. Comparison of the intensity of the expression of COX-2 by the odontogenic epithelial cells in different odontogenic lesions

In their study they found that COX-2 was expressed by the odontogenic epithelial layer, in the subepithelial fibroblasts, macrophages and endothelial cells [24].

In healthy tissue COX-2 is not detected or detected in only in low levels and is not expressed by normal epithelial or normal odontogenic epithelial cells. Growth factors, hormones and cytokines induce the expression of COX-2 [25, 26].

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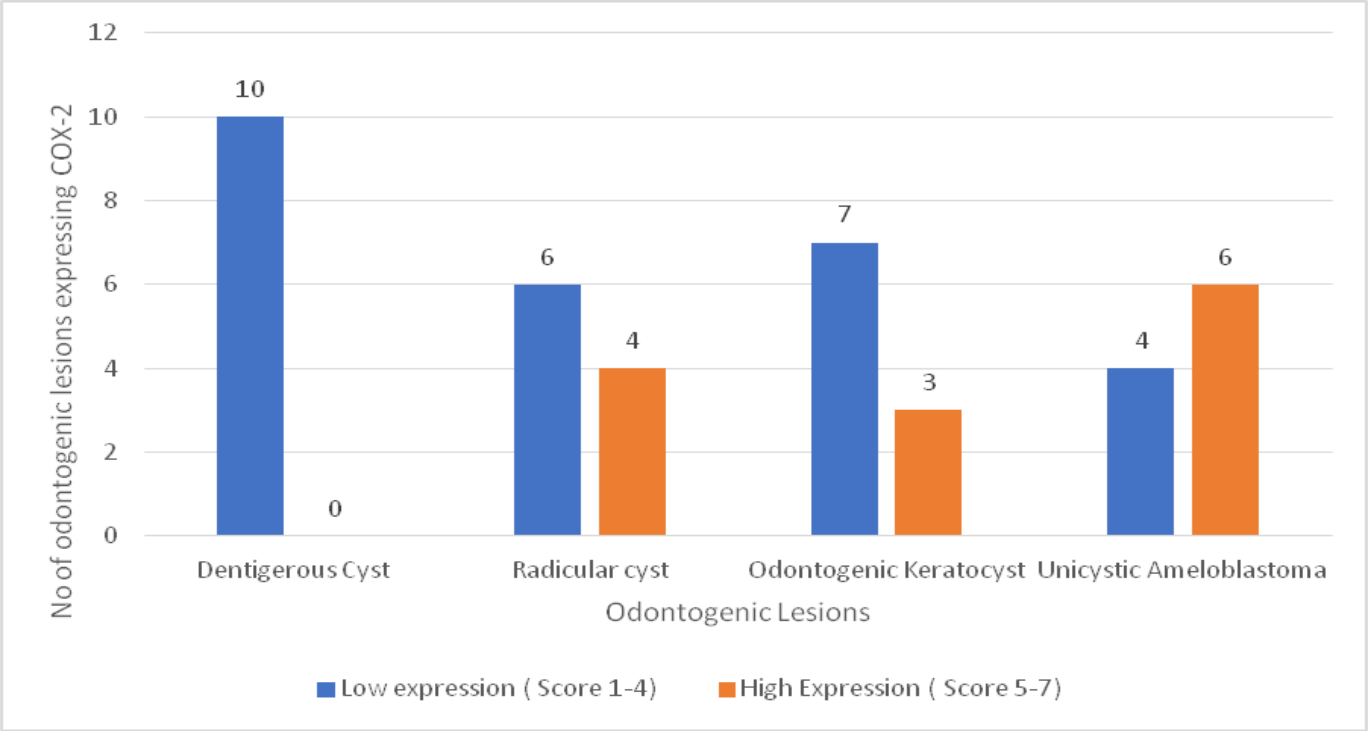

Fig 4. Comparison of the pattern of expression of COX-2 among the different odontogenic lesions.

During the inflammatory process there is an abundance of chemical mediators including tumor necrosis factor and interlukin-1. Interlukin 1 alpha has been found to increase the expression of COX-2 in fibroblasts [27]. When COX-2 expressed, it catalyzes the synthesis of prostaglandin from arachidonic acid [28].

Odontogenic keratocysts have a reputation of being aggressive in its clinical behaviour. In this study the expression of COX-2 was seen in more than 75% of the odontogenic cystic lining cells in 3/10 (30%) odontogenic keratocysts. Seyedmajidi, et al in their study found that all (15/15) of the odontogenic keratocyst that they evaluated, expressed COX-2, but only 5/15 (33%) cysts showed a strong expression of COX-2.They suggested that the COX-2 is a major acting marker in the aggressive behavior of odontogenic keratocysts [16].

In a study by Mendes et al all the 20 cases of odontogenic keratocysts that they evaluated expressed COX-2, and 18 of these 20 odontogenic keratocyst showed a strong expression of COX-2.

They also found that the cysts that showed a showed a strong expression of COX-2 also expresses p53 and Ki 67.COX-2 may contribute to the biological regulation of its epithelial lining [15]. In another study by Kaczmarzyk et al 37/41 odontogenic keratocyst that they evaluated expressed COX-2. They also found that the expression of COX-2 correlated with the expression of bcl-2 in these odontogenic keratocysts [17].

Table 3. The association between the Odontogenic cysts and the expression (Total Score) of COX-2

Odontogenic Lesions	Expression of COX-2			
	Low n (%) (Score 1-4)	High n (%) (Score 5-7)	X^2(df)	p Value
Dentigerous Cyst (n=10)	10 (100%)	0 (0%)	4.845(2)	0.031*
Radicular Cyst (n=10)	6 (60%)	4 (40%)		
Odontogenic Keratocyst (n=10)	7 (70%)	3 (30%)		

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Among the Unicystic ameloblastomas in 3/10 (33%) cases the expression of COX-2 was seen in more than 75% of the odontogenic epithelial cells. Earlier, studies have evaluated the expression of COX-2 in solid ameloblastoma than in unicystic ameloblastomas[13,15].

In the study by Seyedmajdi et al 3/15 of the ameloblastomas showed a high expression of COX-2. They found that the pattern of expression of COX-2 in the ameloblastomas was similar to that of odontogenic keratocysts. But there was a significant difference between the expression of COX-2 in ameloblastomas and dentigerous cysts [16].

To validate the role of COX-2 in proliferation of odontogenic epithelium, one case each of dentigerous cyst, radicular cyst, odontogenic keratocyst and unicystic ameloblastoma was immunohistochemically stained with Ki-67. The case of unicystic ameloblastoma and the case of odontogenic keratocyst stained positive for Ki-67, whereas Ki 67 was not expressed by the Radicular cyst and Dentigerous cyst (Figure 5). The association of COX-2 and Ki-67 in these odontogenic lesions will be evaluated in future.

This study along with other studies in literature strengthens the notion that COX-2 plays a vital role in confirming an aggressive nature to odontogenic lesions. As a limited number of odontogenic lesions were evaluated in this study, further studies on a larger cohort of odontogenic lesions and by evaluating the expression of COX-2 with other proliferative markers is necessary to determine the molecular mechanism by which COX-2 functions in the odontogenic epithelial cells.

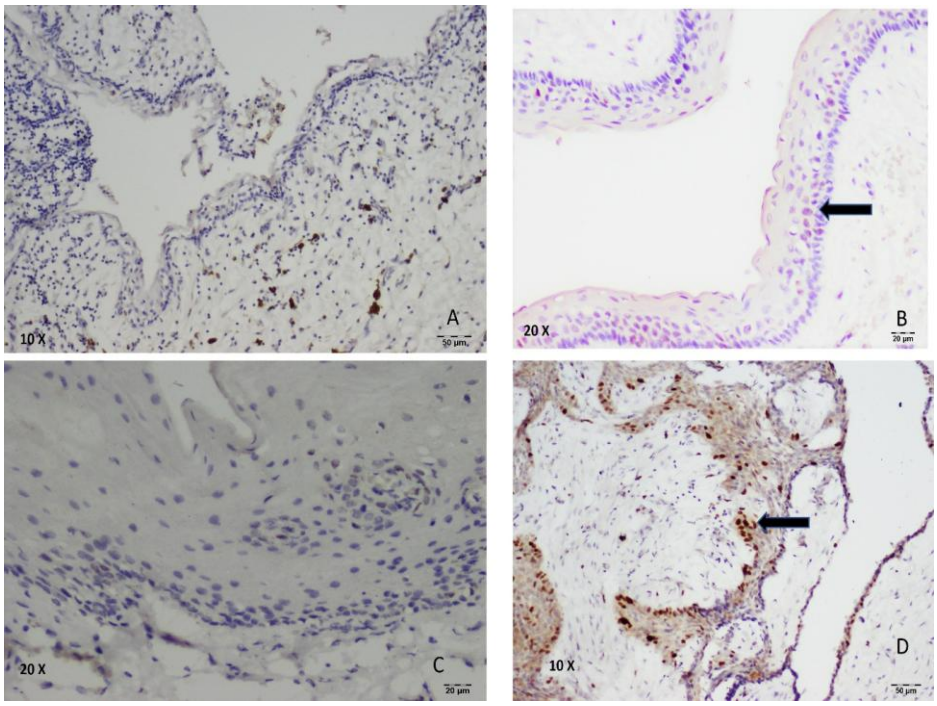

Figure 5. A. Dentigerous cyst (Absence of expression of Ki 67) **B.** Odontogenic Keratocyst (Ki-67 expression in the basal and supra basal cells of the odontogenic epithelial lining) **C.** Radicular Cyst (No expression of Ki 67). **D.** Unicystic Ameloblastoma (Expression of Ki-67 by the ameloblast-like cells)

Table 4. Association of the expression of COX-2 (Total Score) among the different odontogenic Lesions

Odontogenic Lesions	Expression of COX-2		X^2(df)	p Value
	Low score (1-4) n (%)	High Score (5-7) n (%)		
Dentigerous Cyst (n=10)	10 (100%)	0 (0%)	8.547 (3)	0.036*
Radicular Cyst (n=10)	6 (60%)	4 (40%)		
Odontogenic Keratocyst (n=10)	7 (70%)	3 (30%)		
Unicystic Ameloblastoma	4 (40%)	6 (60%)		

*Significant association

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Conclusion

The present study evaluated the expression of COX-2 in the odontogenic lesions; Dentigerous cyst, Radicular cyst, odontogenic keratocyst and Unicystic ameloblastoma. The expression of COX-2 was higher in Radicular cyst, odontogenic keratocyst and Unicystic ameloblastoma compared to dentigerous cyst. COX-2 being an inflammatory marker, showed a high expression among the Radicular cysts. This study emphasizes the fact that COX-2 plays a vital role in biological behavior of odontogenic cysts and tumors. Thus, incorporating anti-inflammatory medicaments and chemopreventive inhibitors of COX-2 will prove to be beneficial in the management of Radicular cysts, Odontogenic keratocysts and Unicystic ameloblastomas.

Conflicts of interest

The authors deny any conflicts of interest in regards to the current study.

Acknowledgement

Dr Aditi Deshpande for providing us with Cox-2 monoclonal antibody for the study.

Table 5. Association of the expression of COX-2 (Total Score) and risk ratio among the odontogenic lesions

Odontogenic lesion	COX 2 Expression Low n (%)	COX2 Expression High n (%)	Chi Square test	95% Confidence Interval		Risk Estimate
				Lower Bound	Upper Bound	
Dentigerous Cyst	10 (100%)	0 (0%)	.043	1.005	2.765	1.667
Radicular Cyst	6 (60%)	4 (40%)				
Dentigerous cyst	10 (100%)	0 (0%)	.105	.952	2.143	1.429
Odontogenic Keratocyst	7 (70%)	3 (30%)				
Dentigerous cyst	10 (100%)	0 (0%)	.005	1.170	5.341	2.500
Unicystic Ameloblastoma	4 (40%)	6 (60%)				
Radicular cyst	6 (60%)	4 (40%)	0.500	.101	4.097	.643
Odontogenic Keratocyst	7 (70%)	3 (30%)				
Radicular Cyst	6 (60%)	4 (40%)	.329	.376	13.465	2.250
Unicystic Ameloblastoma	4 (40%)	6 (60%)				
Odontogenic keratocyst	7 (70%)	3 (30%)	.185	.549	22304	3500
Unicystic Ameloblastoma	4 (40%)	6 (60%)				

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