Original Article

KAI-1 ad p53 Expression in Odontogenic Cysts: An Immunohistochemical Marker Study

Abstract

Background: KAI-1/CD82 is a tumor suppressor gene with decreased gene expression being associated with increased invasive ability of oral squamous cell carcinoma and as hypothesized for various odontogenic cysts and tumors. p53 protein functions in G1-S phase of the cell cycle to allow the repair of damaged DNA. In the present study, p53 and KAI-1 expression was investigated using monoclonal antibodies in the various odontogenic cysts. **Aims:** To detect KAI-1 and p53 expression in radicular cysts, dentigerous cysts, and odontogenic keratocysts (OKCs) and to assess the relation between p53 and KAI-1 expression in the aforementioned cysts. **Materials and Methods:** The present study included histopathologically diagnosed cases of radicular cysts, dentigerous cysts, and OKCs for the expression of KAI-1 and p53 antibodies. **Results:** Among odontogenic cysts, radicular cysts expressed maximum positivity of KAI-1 (20.92%) while p53-positive cells were maximum in OKC (4.04%). The correlation between KAI-1 and p53 expression in the various odontogenic cysts was not found to be significant. **Conclusion:** The increased KAI-1 expression in the radicular cysts and its downregulation in OKCs may be indicative of aggressive clinical behavior and the fact that OKCs are hypothesized as neoplastic rather than being developmental in origin.

Keywords: *KAI-1, odontogenic cysts, p53*

Namrata N. Patil, Vijay Wadhwan[1], Abhishek Singh Nayyar[2], Minal Chaudhary[3], D. Santhosh Reddy[4], K. V. Chalapathi[5]

Departments of Oral Pathology and Microbiology and [2]Oral Medicine and Radiology, Saraswati-Dhanwantari Dental College and Hospital and Post-Graduate Research Institute, Parbhani, [3]Department of Oral Pathology and Microbiology, Sharad Pawar Dental College and Hospital, Wardha, [4]Department of Oral Pathology and Microbiology, Aditya Dental College and Hospital, Beed, Maharashtra, [1]Department of Oral Pathology and Microbiology, Subharti Dental College and Hospital, Meerut, Uttar Pradesh, [5]Department of Oral Pathology and Microbiology, Care Dental College and Hospital, Guntur, Andhra Pradesh, India

Address for correspondence:
Dr. Abhishek Singh Nayyar, 44, Behind Singla Nursing Home, New Friends' Colony, Model Town, Panipat - 132 103, Haryana, India.
E-mail: singhabhishekndls@gmail.com

Access this article online

Website: www.ccij-online.org

DOI: 10.4103/ccij.ccij_3_17

Quick Response Code:

Introduction

The term cyst is derived from the Greek word "Kystis" which means a bladder or sac. Kramer has defined cyst as "a pathological cavity having fluid, semi-fluid, or gaseous contents and which is not created by the accumulation of pus." Most cysts, but not all, are lined by epithelium. Like epithelium from elsewhere in the body, pathologic changes can and do occur within the cystic epithelium.[1] Odontogenic cysts are defined as those cysts which arise from the enamel organ or their remnants. Odontogenic cysts are lined by the epithelium derived from the remnants of the tooth forming organ, cell rests of Malassez, glands of Serres (cell rests of dental lamina), reduced enamel epithelium, and sometimes from the basal cell layers of the oral epithelia. During and after odontogenesis, these cell remnants remain as a common source of cystic changes within the jaw bones.[2,3] It is thought that the epithelial lining of the developmental odontogenic cysts has more proliferative potential than the epithelial lining of the inflammatory cysts. Radicular cyst is the most common cyst of inflammatory origin (about 52.3% of all diagnosed jaw cysts) that arises due to the effects of inflammation on epithelial remnants (cell rests of Malassez) situated in the apical portion of the periodontal ligament. Dentigerous cysts, on the other hand, are the most common developmental odontogenic cysts making up to 16.6% of all the jaw cysts reported. These cysts are associated with the crown of an impacted tooth caused by fluid accumulation between the reduced enamel epithelium and the enamel surface. Histologically, the cyst wall, in dentigerous cysts, is composed of connective tissue lined by low cuboidal, stratified squamous epithelium of two to three cell layer thickness; however, in the presence of inflammation, the thickness of the lining is bound to vary. Odontogenic keratocysts (OKCs) represent 11.2% of all developmental odontogenic cysts and are thought to arise from the derivatives of embryologic dental lamina or its remnants (glands of Serres) and extensions of basal cells from the overlying epithelium. Histologically, they are characterized by a regular epithelial lining of parakeratinized-stratified squamous epithelium, which is thin, ranging from six

How to cite this article: Patil NN, Wadhwan V, Nayyar AS, Chaudhary M, Reddy SD, Chalapathi KV. KAI-1 ad p53 Expression in Odontogenic Cysts: An Immunohistochemical Marker Study. Clin Cancer Investig J 2018;7:62-9.

to ten cell layer thickness and with a well-defined basal layer composed of columnar or cuboidal cells.[1-3] OKCs are aggressive cystic lesions that have a tendency to recur if not adequately treated and grow larger than other cysts with a mitotic activity observed in their epithelial lining which is more than that observed in dentigerous and radicular cysts.[4-6]

Genetic and molecular events underlying the development of metastasis have been studied extensively in the past. KAI-1 is a tumor suppressor gene which is inversely related to the progression and invasion of several tumors (metastasis) as was observed by Guo *et al*., who found upregulation of KAI-1 in early pancreatic carcinoma with its decreased expression in the presence of metastasis.[7] Wu *et al*. identified the role of KAI-1 in digestive tract carcinomas and predicted it to be a useful predictor of prognosis.[8] Farhadieh *et al*.[9] and Imai *et al*.[10] identified the role of KAI-1 in oral squamous cell carcinoma (OSCC) and suggested a decreased gene expression being associated with an increased invasive ability of OSCC. The expression of KAI-1/CD82 gene, inversely related to tumor progression, can, thus, be taken as a favorable prognostic indicator.[11]

At present, a number of oncogenes and tumor suppressor genes, including p53, RAS, β-catenin, and phosphatase and tensin homolog, have been implicated in various cancers.[12,13] p53 protein is a product of the tumor suppressor gene p53 which functions in the G1-S phase of the cell cycle to allow repair of the damaged DNA and to prevent the cell from entering the S phase or alternatively in guiding the damaged cells to apoptosis.[14-18] Furthermore, the high recurrence rate and clinically aggressive behavior of OKCs have caused several investigators to study the cause for the same, and expressions of KAI-1 and p53 protein in OKC have remained one of the most sought topics in this regard assuming it to be associated with cell proliferation in the same.[19,20] The aforementioned features seen in these cysts have, also, caused several authors to regard it as a benign neoplasm rather than a cyst.[5,21-23] In the recent WHO classification (2005), on the classification of odontogenic cysts and tumors, OKCs have been classified under benign tumors arising from epithelium and they have been renamed as keratinizing cystic odontogenic tumors.[21]

The expression of KAI-1 is supposed to decrease in cancer cell lines, also, OSCC. Cancer cells expressing KAI-1 attach to vascular endothelial cells through direct interaction between KAI-1 and DAR (an endothelial cell surface protein), leading to inhibition of tumor cell proliferation and induction of senescence.[22,23] The tumor metastasis is suppressed mainly by an inhibition of cancer cell motility and invasiveness.[11,24-26]

As OKCs have recently been categorized as benign neoplasms with high recurrence rates as high as 60%,[5,21-23] an attempt was made to explain the differences on the basis of its expression of KAI-1 and p53. In the present study, immunohistochemistry for KAI-1 and p53 was employed to evaluate the cell proliferation and aggressive behavior in the radicular and dentigerous cysts and OKC. There are no much studies as such, conducted in relation to odontogenic cysts, and this was first of its kind of studies. Before this, the expression of KAI-1 ad p53 has been studied in relation to the various malignancies. Hence, this study was designed to study the expression of KAI-1 ad p53 in odontogenic cysts and in particular to get an explanation for the neoplastic behavior of OKC, for whether any correlation existed.

In the present study, p53 and KAI-1 expression was investigated using monoclonal antibodies in the various odontogenic cysts. This study actually tries to compare the expression of the abovementioned genes in odontogenic cysts comparing the clinical implications as against OSCC, a condition wherein the expression of the aforementioned genes has already been discussed in detail with various hypotheses proposed as the possible role they might have in OSCC.

Materials and Methods

An immunohistochemical (IHC) study was carried out for the evaluation of KAI-1 and p53 expression in the various odontogenic cysts which comprised radicular cysts, dentigerous cysts, and OKC. Paraffin-embedded sections were obtained. The study sample consisted of 30 cases of radicular cysts, 27 cases of dentigerous cysts, 37 cases of OKCs, and 10 cases of normal buccal mucosa. The immunohistochemically stained tissue sections were evaluated by counting approximately 1000 cells in high power field wherever possible. The tissues which yielded insufficient epithelial lining as in some of the cases of dentigerous cysts, the total numbers of cells were counted and the labeling index was obtained. Staining was observed as nuclear and cytoplasmic membrane staining. Tissue sections positive for KAI-1and p53 were examined for the presence of brown-stained cytoplasm and evaluated by locating the epithelial linings most heavily labeled by scanning the sections at a ×100 magnification. Cell counts were made at ×400 magnification with a conventional light microscope in five randomly selected fields. KAI-1- and p53-labeled cell counting was done among all groups. The constituent cells of the lining epithelium were divided into basal, suprabasal/intermediate, and surface layers. Cuboidal/columnar cells located in one row at the basement membrane were considered as the basal layer. The surface layer constituted the flattened or polygonal cells consisting of one to five layers, localized just underneath the surface of the lining epithelium. The suprabasal/intermediate layer was composed of relatively large round cells between the basal and the surface

layers. The numbers of positively stained nuclei were expressed as a percentage of the total number counted for individual layer and in complete epithelium.

$$\text{KAI-1/p53 labeling index} = \frac{\text{Number of IHC positive cells (KAI-1/p53)} \times 100}{\text{Total number of cells observed}}$$

KAI-1 expression in the epithelium was converted into score defined by Farhadieh *et al.*[9] with score 1 assigned for <10% of cells with positive staining, 2 for 11%–30%, 3 for 31%–50%, and 4 for >51% of the total number of cells with positive staining. The cells which were positive for KAI-1 expression were divided according to their scores.

The study was approved by the Institutional Ethics Committee before the commencement of the study. The results were presented in two sections with detailed analysis of KAI-1 expression as descriptive statistics and significant differences in the group. Similar results were applied for p53 expression, and the statistical analysis was carried out.

Principle of immunohistochemical staining

Sections were hydrated with increasing grades of alcohol and brought to distilled water and treated with hydrogen peroxide (H_2O_2) to eliminate endogenous peroxidase activity. The tissues were then incubated sequentially with:

- Primary antibody (KAI-1, C-16, sc-1087, primary antibody, rabbit polyclonal anti-human antibody, Santa Cruz Biotechnology, Inc., p-53, clone DO-7, primary antibody, mouse monoclonal anti-human antibody, DAKO), which binds to specific tissue antigens
- Secondary antibody (biotinylated secondary antibody, DAB chromogen, DAB substrate buffer, hematoxylin, DAKO), which binds to the primary antibody; it is a polyvalent antibody that will bind to primary antibodies derived from rabbit, mouse, rat, and guinea pig
- Addition of peroxidase substrate (H_2O_2) and chromogen results in the formation of a colored precipitate at the tissue antigen sites. Counterstaining with hematoxylin aided in visualization.

Positive and negative controls

Normal oral mucosa samples showing KAI-1 labeling for p-53 expression acted as a positive control. One positive control was included for each IHC cohort. One section from each positive control was used as the negative control by omitting the primary antibody and by incubating with tris-buffered saline.

Statistical analysis

Statistical analysis was performed with SPSS (version 13, SPSS Inc., Chicago, IL, USA) package. The statistical tests used for the analysis of the results were as follows:

- Descriptive statistical analysis
- One-way analysis of variance (ANOVA)
- Least square difference (LSD) method
- Chi-square test for intergroup comparisons
- Independent *t*-test.

Results

KAI-1 counts were observed in decreasing order in the normal buccal mucosa with a mean of 24.28 ± 4.15, followed by radicular cysts with a mean of 20.92 ± 4.68, dentigerous cysts with a mean of 18.38 ± 4.17, and OKCs with a mean of 11.01 ± 13.01. Descriptive statistics was, also, performed for the expression of p53 in the aforementioned cysts and control groups. Statistically, significant variations of means of p53-labeling indices were found among all groups. p53 counts were observed in decreasing order in OKCs with a mean of 4.04 ± 4.13, normal buccal mucosa 3.44 ± 2.32, radicular cysts 0.45 ± 0.72, and dentigerous cysts 0.16 ± 0.37. One-way ANOVA showed highly significant variation (P = 0.00) of the mean squares 4104.89 between groups and 128.66 within groups with the degree of freedom (df) between groups (df = 4.00) and within groups (df = 129.00) in case of KAI-1 expression in epithelium of radicular cysts, dentigerous cysts, OKCs, and normal mucosa. F =31.91 was obtained showing that statistically, significant variations of means of the labeling indices were found among all the groups [Table 1]. One-way ANOVA in case of p53 expression in epithelium of radicular cysts, dentigerous cysts, OKCs, and normal buccal mucosa, also, revealed highly significant variation (P = 0.00) of the mean squares 20,266.40 between groups and 49.07 within groups with the df between groups (df = 4.00) and within groups (df = 129.00) [Table 1]. The LSD *post hoc* test for KAI-1 expression in radicular cysts, dentigerous cysts, OKCs, and normal buccal mucosa, also, came out to be highly significant in all the compared groups, except between the expression in radicular and dentigerous cysts, radicular cysts and normal buccal mucosa, and dentigerous cysts and normal buccal mucosa (P > 0.05) [Table 2]. The LSD *post hoc* test in case of p53 expression was though not found to be significant in the compared groups with

Table 1: One-way analysis of variance for KAI-1 and p53 expression in the epithelial lining of radicular cysts, dentigerous cysts, odontogenic keratocysts, and normal buccal mucosa

	ANOVA				
	Sum of squares	df	Mean square	*F*	*P*
KAI-1					
Between groups	16,419.58	4.00	4104.89	31.91	<0.0001**
Within groups	16,596.84	129.00	128.66		
p53					
Between groups	81,065.60	4.00	20,266.40	413.00	<0.001**
Within groups	6330.18	129.00	49.07		

**Highly significant at P<0.001. ANOVA: Analysis of variance

$P > 0.05$ [Table 3], 100% positivity was noted for KAI-1 expression in radicular and dentigerous cysts while OKCs showed a positivity in 22 (59.45%) of the cases and 15 cases were found to be negative for KAI-1 staining. There were significant differences observed in the radicular and dentigerous cysts and OKCs. Regarding p53 expression, 10 cases of Radicular cysts out of 30, 6 of Dentigerous cysts out of 27 and 31 of OKCs out of 37 were positive (83.78%). OKCs showed the highest positivity of 83.78 % as compared to Radicular and Dentigerous cysts which showed a positivity of 33.33% and 22.22% respectively. Also, KAI-1 and p53 expressions were compared between each group and came-out to be statistically significant (P=0.00) [Table 4].

Discussion

Odontogenic cysts comprised an unusually diverse group of lesions because odontogenesis is a complicated process in which cells in various stages of differentiation participate in a complex, predetermined manner, constituting a group of frequent intraosseous lesions in the jaw bones.[1-3] Unlike the radicular and dentigerous cysts, the OKC can assume a clinically aggressive and destructive behavior.[4,5] If inadequately treated, these cysts cause considerable expansion within and damage the jaw bones. A significant clinical problem is the high recurrence rate (12.65%) observed, following surgical enucleation in these cysts.[6,19]

When a cyst grows, multiple cytokines are liberated, such as interleukins (ILs), tumor necrosis factor (TNF), matrix metalloproteinases, tenascin, fibronectin, and parathyroid-hormone-related proteins (PTH-rPs), which modulate the function of other cell types and are involved in cellular immune and inflammatory responses through auto- and para-crine signaling, leading to extensive bone damage.[24] OKCs are characterized by their epithelial lining that has some intrinsic growth potential added to which inflammation alters not only the morphology but also the

Table 2: Least significant difference *post hoc* test analysis between KAI-1 expression in radicular cysts, dentigerous cysts, odontogenic keratocysts, and normal buccal mucosa

Variable 1	Variable 2	Mean	SE	*P*	95% (CI)	
					Upper bound	Lower bound
Radicular cysts	Dentigerous cysts	2.54	3.01	0.40	−3.41	8.49
Radicular cysts	Normal buccal mucosa	9.91	2.79	<0.001**	4.40	15.42
Dentigerous cysts	OKCs	−3.36	4.14	0.42	−11.55	4.84
Dentigerous cysts	Normal buccal mucosa	7.37	2.87	0.01*	1.69	13.05
OKCs	Normal buccal mucosa	−5.90	4.20	0.16	−14.21	2.41

*Significance at P<0.05, **Highly significant at P<0.001. SE: Standard error, CI: Confidence interval, OKCs: Odontogenic keratocysts

Table 3: Least significant difference *post hoc* test analysis between p53 expression in radicular cysts, dentigerous cysts, odontogenic keratocysts, and normal buccal mucosa

Variable 1	Variable 2	Mean	SE	*P*	95% (CI)	
					Upper bound	Lower bound
Radicular cysts	Dentigerous cysts	0.28	1.86	0.88	−3.39	3.96
Radicular cysts	Normal buccal mucosa	−2.99	2.56	0.24	−8.05	2.07
Dentigerous cysts	OKCs	−3.88	1.77	0.03	−7.39	-0.37
Dentigerous cysts	Normal buccal mucosa	−3.27	2.59	0.21	−8.40	1.86
OKCs	Normal buccal mucosa	0.61	2.50	0.81	−4.33	5.55

SE: Standard error, CI: Confidence interval, OKCs: Odontogenic keratocysts

Table 4: Comparison between KAI-1 and p53 expression in different groups by independent *t*-test

Sample	Variables	Mean±SD	SE	*t*	*P*	95% (CI)	
						Lower bound	Upper bound
Radicular cysts	KAI-1	20.92±4.68	0.86	23.66	<0.001**	18.71	22.24
	p53	0.45±0.72	0.13				
Dentigerous cysts	KAI-1	18.38±4.17	0.80	22.59	<0.001**	16.56	19.87
	p53	0.16±0.37	0.07				
OKCs	KAI-1	11.01±13.01	2.14	3.10	<0.001**	2.44	11.49
	p53	4.04±4.13	0.68				
Normal buccal mucosa	KAI-1	24.28±4.15	1.31	13.86	<0.001**	17.62	24.06
	p53	3.44±2.32	0.74				

**Highly significant at P<0.001. SE: Standard error, CI: Confidence interval, OKCs: Odontogenic keratocysts, SD: Standard deviation

proliferative potential of the epithelial lining.[11,25] The keratinocytes synthesize IL-1 and IL-6 and these cytokines and TNF account for raised levels of PGs and collagenase synthesis by uninflamed cystic linings. Furthermore, PTH-rPs have been hypothesized to be expressed in high levels in OKCs and to play a possible role in the growth of these cysts and bone resorption seen by acting synergistically with IL-1.[24] Different markers such as proliferating cell nuclear antigens (PCNAs), p53, Ki-67, and silver nucleolar organizer regions have been studied in odontogenic cysts with PCNAs denoting aggressiveness in such lesions and their potential to proliferate.[26,27] One of the newest markers studied in odontogenic cysts is KAI-1.[28] KAI-1 has been detected in normal human tissues and is a regulator of cell behavior. The expression of KAI-1 has been seen to decrease in cancer cell lines with high propensity for metastasis.[9-12] Cancer cells expressing KAI-1 attach to vascular endothelial cells through direct interaction between KAI-1 and DAR (an endothelial cell surface protein), leading to inhibition of tumor cell proliferation and induction of senescence.[22,23] The tumor metastasis is suppressed mainly by an inhibition of cancer cell motility and invasiveness.[11,24-26] As OKCs have recently been categorized as benign neoplasms with high recurrence rates as high as 60%,[5,21-23] an attempt was made to use immunohistochemistry for KAI-1 and p53 in the aforementioned cysts to evaluate cell proliferation and the aggressive behavior shown by them in the present study.

In the present study, OKCs showed only 11.01% KAI-1 positivity as against radicular and dentigerous cysts which showed 100% positivity for KAI-1. In radicular cysts, KAI-1 expression was found with strong and/or intermediate positivity in the epithelial lining with approximately, the entire epithelial lining exhibiting KAI-1 reactivity, and it was intense in areas of inflammation [Figure 1] indicating KAI-1 expression to be upregulated in lesions with inflammation. In dentigerous cysts, KAI-1 positivity was seen throughout the thickness of the epithelium. Staining was seen as distinct membranous or cytoplasmic involving the epithelium lining [Figure 2] clearly evident even in areas of minimal inflammation and places of diffuse inflammation indicating KAI-1 upregulation being not just a phenomenon associated with inflammation but also other factors including the variable cytokines liberated by the cystic linings. In OKC, only 83.78% of the cases showed positivity for KAI-1. Positivity was noticed in the superficial layers and tended to be focally or diffusely positive and was least in the basal cell layers and suprabasal layers which exhibited the absence of KAI-1 expression [Figure 3]. Some of the keratocystic linings in which inflammation was minimal exhibited negligible or absence of staining. Not much has been reported about the KAI-1 expression in odontogenic cysts, and there has been a paucity of studies in the literature regarding the expression of KAI-1 in the odontogenic apparatus. Extensive search revealed one study conducted by Iezzi *et al*.[28] where the authors found a positive KAI-1 expression in radicular and dentigerous cysts but very little expression in OKC among which none of the parakeratinized OKCs showed positivity and only four out of 16 orthokeratinized OKCs revealed KAI-1 positivity. Our results are in partial agreement with their study as KAI-1 expression in dentigerous and radicular cysts was nearly comparable with their studies although OKCs exhibited KAI-1 positivity to a lesser extent.

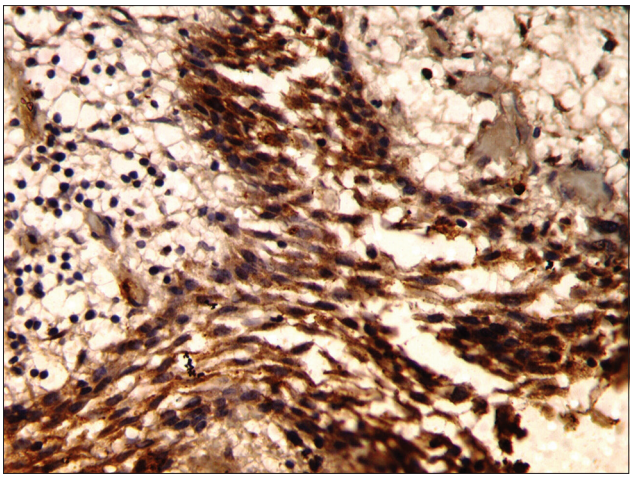

Figure 1: KAI-1 expressivity in radicular cyst (×400)

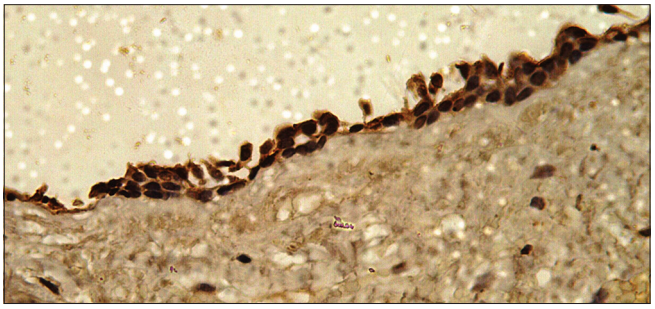

Figure 2: KAI-1 expressivity in dentigerous cyst (×400)

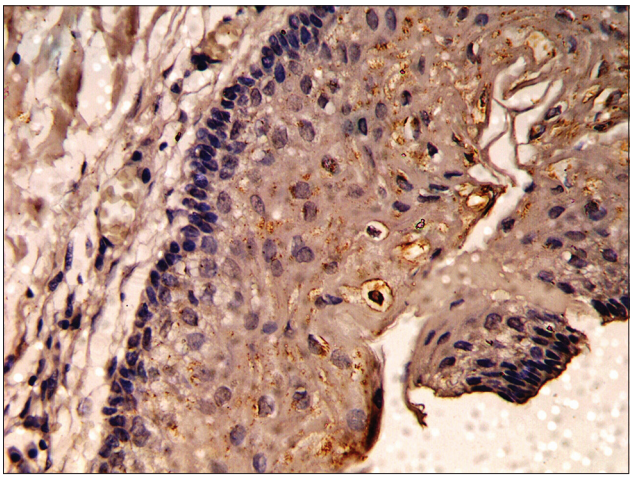

Figure 3: KAI-1 expressivity in odontogenic keratocysts (×400)

There is a documented proof that downregulation of KAI-1 gene is associated with increased metastasis. Hence, taking the reverse, also, to be true, any lesion which is benign in nature is supposed to express KAI-1, depending on the level of its aggressiveness. The term OKC designates a cyst with a characteristic histological appearance and a specific clinical behavior. The epithelium of OKCs is believed to have an intrinsic growth potential and shows strong evidence of being neoplastic rather than developmental origin.[5,21-23] OKCs also share allelic loss of the same loci that have been implicated in the development of OSCCs providing further proof regarding its being neoplastic in nature.[22,23] Thus, results of our study show that although in radicular [Figure 1] and dentigerous cysts [Figure 2], there was positivity to KAI-1, in OKCs [Figure 3], there was significantly less expression. This lack of KAI-1 expression in OKCs could help explain the differences in the clinical and pathological behavior of OKCs, and according to what seems to be the pattern in several types of epithelial tumors could be related to the increased aggressive behavior, invasiveness, and high frequency of the recurrences found in OKCs. An increase in cell proliferation plays an important role in the development of odontogenic cysts.

p53 protein is a product of the tumor suppressor p53 gene which functions in G1-S phase of the cell cycle to allow repair of the damaged DNA. p53 gene has a shorter life in normal cells and cannot be detected immunohistochemically; however, when mutated, the p53 protein becomes more stable and detectable. Therefore, p53 protein is expressed in actively proliferating cells. While positive staining for p53 may be correlated with genetic mutation, the wild protein can, also, be retained in the tissues by, for example, binding to other proteins or due to some defects in the normal degradation pathway and can, therefore, be identified by immunohistochemistry. Wild-type p53 protein acting as a tumor suppressor downregulates cell growth, but mutation in p53 can inactivate its tumor suppression activity allowing the dominant oncogenic factors to lead to malignant transformation.[14-18]

In the present study, OKCs showed 83.78% p53 positivity as against radicular and dentigerous cysts which showed 100% positivity for p53. p53 immunolabeling was dense and scattered in the basal and suprabasal cell layers in OKCs [Figure 4], whereas very few densely stained cells were located in the basal cell layers in radicular [Figure 5] and dentigerous cysts [Figure 6] and normal oral mucosa. p53 expression was highest in OKCs [Figure 4]. In radicular [Figure 5] and dentigerous cysts [Figure 6], most of the p53-positive cells were located in the basal and suprabasal cell layers. This was in agreement with the study conducted by Ogden *et al*.,[29] who concluded that most of the p53-positive cells were located in the basal cell layers in OKCs, whereas radicular and dentigerous cysts were negative for p53. Slootweg[30] and Li *et al*.[31] however reported that positive cells were detected in all odontogenic cysts though to variable extents. The findings of the present study were in accordance with these authors as well as with the various other studies present in the literature.[20,26,27,32]

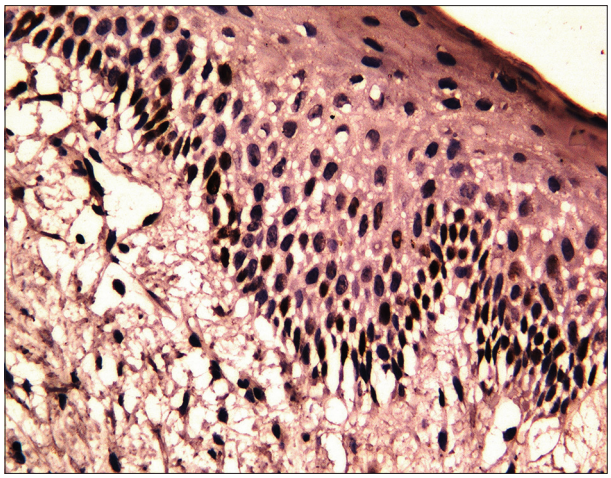

Figure 4: p53 expressivity in odontogenic keratocysts (×400)

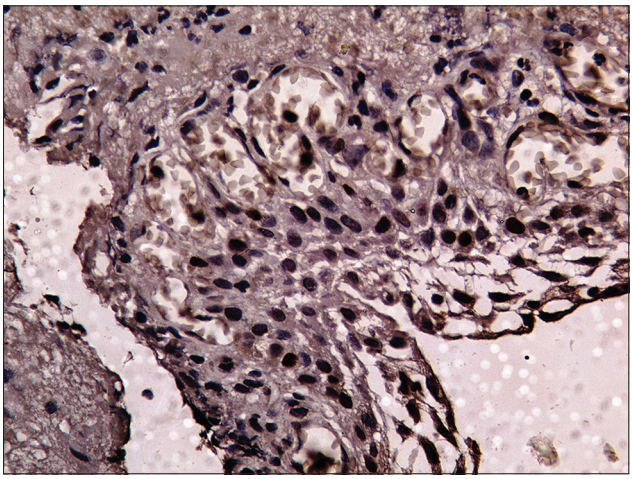

Figure 5: p53 expressivity in radicular cyst (×400)

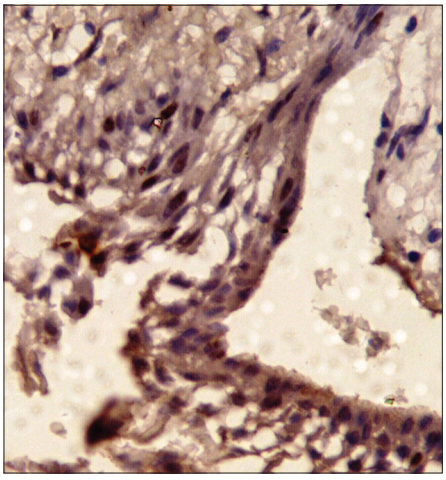

Figure 6: p53 expressivity in dentigerous cyst (×400)

Recent studies have, also, indicated that p53 alteration occurs at a greater frequency in invasive than in noninvasive carcinomas.[15,33] The high reactivity of p53 protein in

OKCs could, thus, be related to the factors peculiar to the extensive cystic lesions including their locally aggressive behavior, high mitotic activity of their cells, and their tendency to recur, although the rate of recurrence might depend on the method and adequacy of their treatment. This p53 reactivity, thus, indicates the possible role, it carries, in the high intrinsic growth potential and biological aggressiveness of these lesions.

Conclusion

Among odontogenic cysts, radicular cysts expressed maximum positivity of KAI-1 while p53-positive cells were maximum in OKC. The increased KAI-1 expression in the radicular cysts and its downregulation in OKCs may be indicative of aggressive clinical behavior and the fact that OKCs are hypothesized as neoplastic rather than being developmental in origin. Furthermore, the high reactivity of p53 protein in OKCs could be related to the factors peculiar to the extensive cystic lesions including their locally aggressive behavior, high mitotic activity of their cells, and their tendency to recur.

Although a large body of work exists regarding the significance of p53 expression, the significance of increased or decreased KAI-1 expression in the aggressiveness in nonneoplastic lesions remains as yet unclear. This study, thus, paves the way for further studies to be conducted to investigate, if any, correlation existed between KAI-1 and p53 expressions in these odontogenic cysts and numerous other odontogenic lesions with suspect clinical behavior as very limited studies, till date, have been conducted in this regard.

Acknowledgement

We would like to acknowledge all the patients who contributed in the study without whom this study would not have been feasible.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

References

1. Shear M. Cysts of the Oral Regions. 3rd ed. Oxford: Butterworth-Heinemann; 1992.
2. Shafer WG, Hine MK, Levy BM. Cysts of odontogenic origin. In: Textbook of Oral Pathology. 4th ed. Philadelphia, PA: Elsevier; 2004.
3. Neville BW, Damm DD, Allen CM, Bouquot JE. Odontogenic cysts. In: Neville BW, Damm DD, Allen CM, Bouquot JE, ed. Oral and Maxillofacial Pathology. 2nd ed. Philadelphia, PA: WB Saunders; 2005.
4. Shear M. Odontogenic keratocysts: Clinical features. Oral Maxillofac Surg Clin North Am 2003;15:335-45.
5. Shear M. The aggressive nature of the odontogenic keratocyst: Is it a benign cystic neoplasm? Part 2. Proliferation and genetic studies. Oral Oncol 2002;38:323-31.
6. Blanas N, Freund B, Schwartz M, Furst IM. Systematic review of the treatment and prognosis of the odontogenic keratocyst. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2000;90:553-8.
7. Guo X, Friess H, Graber HU, Kashiwagi M, Zimmermann A, Korc M, *et al*. KAI1 expression is up-regulated in early pancreatic cancer and decreased in the presence of metastases. Cancer Res 1996;56:4876-80.
8. Wu Q, Ji Y, Zhang MQ, Chen YQ, Chen F, Shi DL, *et al.* Role of tumor metastasis suppressor gene KAI1 in digestive tract carcinomas and cancer cells. Cell Tissue Res 2003;314:237-49.
9. Farhadieh RD, Smee R, Ow K, Yang JL, Russell PJ, Crouch R, *et al.* Down-regulation of KAI1/CD82 protein expression in oral cancer correlates with reduced disease free survival and overall patient survival. Cancer Lett 2004;213:91-8.
10. Imai Y, Sasaki T, Shinagawa Y, Akimoto K, Fujibayashi T. Expression of metastasis suppressor gene (KAI1/CD82) in oral squamous cell carcinoma and its clinico-pathological significance. Oral Oncol 2002;38:557-61.
11. Liu WM, Zhang XA. KAI1/CD82, a tumor metastasis suppressor. Cancer Lett 2006;240:183-94.
12. Solomon MC, Carnelio S, Gudattu V. Molecular analysis of oral squamous cell carcinoma: A tissue microarray study. Indian J Cancer 2010;47:166-72.
13. Fabbrocini G, Russo N, Pagliuca MC, Delfino M, Staibano S, Molea G, *et al.* p53, cyclin-D1, PCNA, AgNOR expression in squamous cell cancer of the lip: A multicenter study. Photodermatol Photoimmunol Photomed 2000;16:172-7.
14. Levine AJ, Finlay CA, Hinds PW. P53 is a tumor suppressor gene. Cell 2004;116 2 Suppl: S67-9.
15. Matlashewski G, Banks L, Pim D, Crawford L. Analysis of human p53 proteins and mRNA levels in normal and transformed cells. Eur J Biochem 1986;154:665-72.
16. Foulkes WD. p53 – Master and commander. N Engl J Med 2007;357:2539-41.
17. Bradley G, Trembly S, Irish J, MacMillan C, Baker G. The expression of p53 induced protein with death domain (Pidd) and apoptosis in OSCC. Cancer Res 2007;36:415-24.
18. Shin DM, Lee JS, Lippman SM, Lee JJ, Tu ZN, Choi G, *et al.* p53 expressions: Predicting recurrence and second primary tumors in head and neck squamous cell carcinoma. J Natl Cancer Inst 1996;88:519-29.
19. Chirapathomsakul D, Sastravaha P, Jansisyanont P. A review of odontogenic keratocysts and the behavior of recurrences. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2006;101:5-9.
20. Tosios KI, Kakarantza-Angelopoulou E, Kapranos N. Immunohistochemical study of bcl-2 protein, Ki-67 antigen and p53 protein in epithelium of glandular odontogenic cysts and dentigerous cysts. J Oral Pathol Med 2000;29:139-44.
21. Shetty DC, Urs AB, Godhi S, Gupta S. Classifying odontogenic keratocysts as benign cystic neoplasms: A molecular insight into its aggressiveness. J Maxillofac Oral Surg 2010;9:30-4.
22. Anand VK, Arrowood JP Jr., Krolls SO. Malignant potential of the odontogenic keratocyst. Otolaryngol Head Neck Surg 1994;111:124-9.
23. Partridge M, Towers JF. The primordial cyst (odontogenic keratocyst): Its tumour-like characteristics and behaviour. Br J Oral Maxillofac Surg 1987;25:271-9.
24. Yoshida BA, Sokoloff MM, Welch DR, Rinker-Schaeffer CW. Metastasis-suppressor genes: A review and perspective on an emerging field. J Natl Cancer Inst 2000;92:1717-30.

25. Tonoli H, Barrett JC. CD82 metastasis suppressor gene: A potential target for new therapeutics? Trends Mol Med 2005;11:563-70.
26. Jackson P, Millar D, Kingsley E, Yardely G, Ow K, Clark S, *et al*. Methylation of a CpG island within the promoter region of the KAI-1 metastasis suppressor is not responsible for down-regulation of KAI-1 expression in invasive cancers or, cancer cell lines. Cancer Lett 2000;157:169-76.
27. Oliveira G, Lluxen S, Chaves AC, Rados PV, Sant'Ana Filho M. Immunohistochemical analysis of patterns of p53 and PCNA expression in odontogenic cystic lesions. Med Oral Patol Cir Bucal 2008;13:E275-80.
28. Iezzi G, Piattelli A, Artese L, Goteri G, Fioroni M, Rubini C. KAI-1 protein expression in odontogenic cysts. J Endod 2007;33:235-8.
29. Ogden GR, Chisholm DM, Kiddie RA, Lane DP. p53 protein in odontogenic cysts: Increased expression in some odontogenic keratocysts. J Clin Pathol 1992;45:1007-10.
30. Slootweg PJ. p53 protein and Ki-67 reactivity in epithelial odontogenic lesions. An immunohistochemical study. J Oral Pathol Med 1995;24:393-7.
31. Li TJ, Browne RM, Prime SS, Paterson IC, Matthews JB. p53 expression in odontogenic keratocyst epithelium. J Oral Pathol Med 1996;25:249-55.
32. Gaballah ET, Tawfik MA. Immunohistochemical analysis of P53 protein in odontogenic cysts. Saudi Dent J 2010;22:167-70.
33. Crosthwaite N, Teale D, Franklin C, Foster GA, Stringer BM. p53 protein expression in malignant, pre-malignant and non-malignant lesions of the lip. J Clin Pathol 1996;49:648-53.