Journal of Advanced Medical and Dental Sciences Research

@Society of Scientific Research and Studies ***NLM ID:*** *101716117*

Journal home page: www.jamdsr.com doi: 10.21276/jamdsr Index Copernicus value = 91.86

(e) ISSN Online: 2321-9599; (p) ISSN Print: 2348-6805

Original Research

Association of oxidative stress and production of inflammatory mediators matrix metalloproteinase-9 and interleukin 6: Systemic events in radicular cysts

[1]Gunveen Kour, [2]Andleeb Manhas, [3]Harshy, [4]Swati, [5]Divya, [6]Divyani

[1]Consultant Oral Pathologist, Private Practioner, Jammu, India;
[2]Senior Resident, SKIMS Srinagar, J& K, India;
[3]Senior Lecturer, Department of Oral Pathology and Microbiology, Swami Devi Dyal Hospital and Dental College, Panchkula, Haryana, India;
[4]Associate Professor, Department of Oral Pathology and Microbiology, Swami Devi Dyal Hospital and Dental College, Panchkula, Haryana, India;
[5,6]PG Student, Department of Oral Pathology and Microbiology, Swami Devi Dyal Hospital and Dental College, Panchkula, Haryana, India

ABSTRACT:
Background: Reactive oxygen species induce molecular damage and disturbed redox signaling, that result in the loss of bone homeostasis, increased pro-inflammatory mediators, and MMP overexpression and activation, leading to apical tissue breakdown. On the other hand, oxidative stress has been strongly involved in the pathogenesis of atherosclerosis, where a chronic inflammatory process develops in the arterial wall. **Aims and objectives:** To determine the association of oxidative stress and the production of inflammatory mediators MMP-9 and interleukin 6 (IL-6) in systemic events in radicular cyst growth. **Materials and methods**: Patients with periapical granulomas and Radicular cysts with clinical attachment loss > 3 mm, probing depth > 5 mm, bleeding on probing, round, well-defined radiolucencies of periapical tissues in radiograph were included into the present study. **Results:** The mean ages of patients and controls were 46 and 49 years, respectively. Mean patient BMI (24.30 ± 3.60 kg/m2) were higher than mean control BMI (28.02 ± 3.60 kg/m2). Levels of serum MDA ($p \leq 0.031$), IL-6 ($p \leq 0.033$), TNF-alpha ($p \leq 0.003$), and MMP-9 ($p \leq 0.032$) were significantly increased in patients as compared to those in the control group. However, the levels of SOD ($p \leq 0.004$) and GPx ($p \leq 0.032$) were significantly decreased in patients compared to controls. **Conclusion:** Proinflammatory cytokines and the bone-resorbing mediators plays a well-established role in the development of radicular cysts. Moreover, an increase in the lipid peroxidation level and the rise in the reactive oxygen species has an important role in the pathogenesis of radicular cyst.
Keywords: pro-inflammatory mediators, Superoxide Dismutase, Malondialdehyde, Glutathione Peroxidase.

Received: 12 October, 2021 Accepted: 15 November, 2021

Corresponding author: Andleeb Manhas, Senior Resident, SKIMS Srinagar, J& K, India

This article may be cited as: Kour G, Manhas A, Harshy, Swati, Divya, Divyani. Association of oxidative stress and production of inflammatory mediators matrix metalloproteinase-9 and interleukin 6: Systemic events in radicular cysts. J Adv Med Dent Scie Res 2021;9(12):31-34.

INTRODUCTION

Oxidative stress is involved in the pathogenesis of a variety of inflammatory disorders. Radicular cyst usually results in the formation of an apical lesion, caused by the immune response to endodontic infections.[1] Reactive oxygen species (ROS) produced by phagocytic cells in response to bacterial challenge represent an important host defense mechanism, but disturbed redox balance results in tissue injury. During endodontic infection, ligation of Toll-like receptors (TLRs) on phagocytes' surface triggers activation, phagocytosis, synthesis of ROS, activation of humoral and cellular responses, and production of inflammatory mediators, such as, cytokines and matrix metalloproteinases (MMPs).[2] ROS induce molecular damage and disturbed redox signaling, that result in the loss of bone homeostasis, increased pro-inflammatory mediators, and MMP overexpression and activation, leading to apical tissue breakdown. On the other hand, oxidative stress has been strongly

involved in the pathogenesis of atherosclerosis, where a chronic inflammatory process develops in the arterial wall.[3] Cystic growth involves the provision of stimulus by antigens, microbes, fibroblasts, and growth factors that causes the inflammatory response by releasing various cytokines which leads to the explosive cell division and the epithelial proliferation in a cystic cavity continues, cells in the middle of the mass become deficient of nutrients which leads to the central necrosis.[4] These degenerative necrotic tissues in the cystic lumen are chemotactic for neutrophils that causes the influx into the cystic lumen and these contents have a higher osmotic load as compared to the surrounding tissues. The MMPs usually expressed in infectious and necrotic odontogenic lesions[5] and the ROS signaling may activate MMPs like MMP-2 and MMP-9.

ROS constitute an important host defense mechanism against invading pathogens. Hence, the combination of bacterial phagocytosis and secretion of proteolytic enzymes and immuno-modulatory compounds that assist in the killing and digestion of bacteria.[6] Oxidants can cause tissue injury via damage to deoxyribonucleic acid (DNA) and proteins, peroxidative injury to lipid membranes, activation of proinflammatory cytokines, and proteases like MMPs and interleukins.[7] RCs arise to control the infection as the low intensity chronic stimulus triggered by bacterias and their products provides conditions to the organism, confining the aggression to the periapical region. During these events of defense, different cells produce cytokines, mainly interleukin 1 (IL-1), IL-6 and tumor necrosis factor alpha (TNF-α), which are involved in this immunoinflammatory response and play a role in bone resorption and in the stimulation of mitogenesis.[8] So, the present study was conducted to determine the association of oxidative stress and the production of inflammatory mediators MMP-9 and interleukin 6 (IL-6) in systemic events in radicular cyst growth.

MATERIALS AND METHODS

Total 40 patients were included into the study in which 30 men and 10 women, aged between 20 to 40 years were included into the study from the Department of Dentistry of a private dental college. The ethical clearance was obtained from the ethical committee of the institution.

INCLUSION CRITERIA

- Patients with periapical granulomas and Radicular cysts
- clinical attachment loss > 3 mm, probing depth > 5 mm
- bleeding on probing
- round, well-defined radiolucencies of periapical tissues in radiograph

EXCLUSION CRITERIA

- patients receiving antibiotic therapy
- compromised periodontal status
- pregnant and lactating women
- any chronic infection or depression.

Our control group consisted of 15 subjects (9 men and 6 women) with,

- healthy, attached gingiva
- no signs of periodontal disease nor bleeding on probing.

Method of execution involves the assessment of body mass index (BMI) and collected blood samples to evaluate MMP-9, IL6, Superoxide Dismutase **(**SOD), Malondialdehyde (MDA), and Glutathione Peroxidase (GPx) levels via a chemical assay. Levels of TNF-alpha and IL-6 were measured via enzyme-linked immunosorbent assay. Statistical analysis was done by using SPSS Version 21.0 and the variables were shown as mean ± standard deviation (SD). Patient and control values were compared using the student 't' test and $p \leq 0.05$ was considered significant

RESULTS

The mean ages of patients and controls were 46 and 49 years, respectively. Mean patient BMI (24.30 ± 3.60 kg/m2) were higher than mean control BMI (28.02 ± 3.60 kg/m2). Levels of serum MDA ($p \leq 0.031$), IL-6 ($p \leq 0.033$), TNF-alpha ($p \leq 0.003$), and MMP-9 ($p \leq 0.032$) were significantly increased in patients as compared to those in the control group (Table 1). However, the levels of SOD ($p \leq 0.004$) and GPx ($p \leq 0.032$) were significantly decreased in patients compared to controls.

TABLE 1: Levels of circulating biochemical variables in patients with a periapical cyst

VARIABLES	CONTROLS (N=15)	SUBJECTS (N=40)	P-VALUE
MDA (nmol/ml)	1.26 ± 0.02	4.20 ± 0.18	0.031
SOD (nmol/ml)	0.12±0.02	0.02±0.01	0.004
GPx (nmol/ml)	7.76±1.68	5.79±1.89	0.032
IL-6 (pg/ml)	4.87±0.51	7.64±1.89	0.033
TNF-α (pg/ml)	27.86±4.12	31.67±5.69	0.003
MMP-9 (pmol)	42.55±3.64	155±6.93	0.032

DISCUSSION

Immune-mediated damage of periodontal supporting tissues causes periodontitis which leads to the loss of teeth. Apical lesions can progress to form an radicular cysts which is mainly due to the increased levels of MMP-9.[9] In our study we found the significantly increased levels of serum MDA, IL-6, TNF-alpha, and MMP-9 in patients when compared to patients of

control group. While the previous studies suggested that RC formation is mainly due to the degradation of collagen via MMPs [9]. However, another study says that the primary factor in RC formation is due to collagenases.[10] Henrique et al. says that the separation of epithelium from the connective tissues leads to the progression of lesion and recurrence.[11]

An experimental study reported a significant elevation in the levels of MDA, which may be an indicator of oxidative stress.[12] The production of reactive oxygen species after the activation of polymorphonuclear leukocytes may help in the formation of inflammatory lesions. The imbalance of oxidative species in the periapical part may cause the development of asymptomatic periapical lesions.[13] Non-toxic levels of ROS increase proinflammatory mediators, and enzymes of the extracellular matrix that leads to the destruction of apical tissue and the formation of apical lesions.[14] According to the present study the levels of SOD and GPx were significantly decreased in patients when compared to controls. The total antioxidant status indicates the ability of antioxidants to scavenge free radicals and the reduced activity of SOD and GPx may cause the development of the lesion further which may results into the oxidative stress.[15] Oxidative non-proteolytic activation of MMP causes the periodontal inflammation. The exact mechanism of involving MMP-9 in intracellular signaling is unclear. MMP-9 may have an important role in the angiogenesis of odontogenic cysts and ROS can directly activate MMP-8 and MMP-9 in periodontal tissues via oxidizing enzymes. Signaling molecules like chemokines, cytokines, and growth factors may be handled by the active form of MMPs, regulating their bioavailability and function.[16,17]

CONCLUSION

Proinflammatory cytokines and the bone-resorbing mediators plays a well-established role in the development of radicular cysts. Moreover, an increase in the lipid peroxidation level and the rise in the reactive oxygen species has an important role in the pathogenesis of RC.

REFERENCES

1. Neville BW, Day TA: Oral cancer and precancerous lesions. CA Cancer J Clin. 2002, 52:195215. 10.3322/canjclin.52.4.195
2. Khot K, Deshmukh SB, Alex S: Comparative analysis of the immunohistochemical expression of vascular endothelial growth factor and matrix metalloproteinase-9 in keratocystic odontogenic tumor, dentigerous cyst and radicular cyst. J Cancer Res Ther. 2015, 11:635-640. 10.4103/0973-1482.144591
3. Pechalova PF, Bakardjiev AG: Cysts of the jaws: a clinical study of 621 cases [Article in Croatian]. Acta Stomatol Croat. 2009, 43:215-224.
4. de Oliveira Rde C, Beghini M, Borges CR, et al.: Higher expression of galectin-3 and galectin-9 in periapical granulomas than in radicular cysts and an increased toll-like receptor-2 and tolllike receptor-4 expression are associated with reactivation of periapical inflammation. J Endod. 2014, 40:199-203. 10.1016/j.joen.2013.10.031
5. Stamenkovic I: Extracellular matrix remodelling: the role of matrix metalloproteinases. J Pathol. 2003, 200:448-464. 10.1002/path.1400
6. Belmar MJ, Pabst C, Martínez B, Hernández M: Gelatinolytic activity in gingival crevicular fluid from teeth with periapical lesions. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2008, 105:801-806. 10.1016/j.tripleo.2007.12.002
7. Binker MG, Binker-Cosen AA, Gaisano HY, de Cosen RH, Cosen-Binker LI: TGF-β1 increases invasiveness of SW1990 cells through Rac1/ROS/NF-κB/IL-6/MMP-2. Biochem Biophys Res Commun. 2011, 405:140-145. 10.1016/j.bbrc.2011.01.023
8. Graves DT, Oates T, Garlet GP: Review of osteoimmunology and the host response in endodontic and periodontal lesions. J Oral Microbiol. 2011, 3:[Epub]. 10.3402/jom.v3i0.5304
9. Halliwell B, Gutteridge JMC: Free Radicals in Biology and Medicine . Oxford University Press, Oxford; 2015. 10.1107/S2059798317004533
10. Carocho M, Ferreira IC: A review on antioxidants, prooxidants and related controversy: natural and synthetic compounds, screening and analysis methodologies and future perspectives. Food Chem Toxicol. 2013, 51:15-25. 10.1016/j.fct.2012.09.021
11. Henriques ÁC, Vasconcelos MG, Galvão HC, de Souza LB, de Almeida Freitas R: Comparative analysis of the immunohistochemical expression of collagen IV, MMP-9, and TIMP-2 in odontogenic cysts and tumors. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2011, 112:468-475. 10.1016/j.tripleo.2011.05.033
12. Strycharz-Dudziak M, Kiełczykowska M, Drop B, Swiatek L, Kliszczewska E, Musik I, Polz-Dacewicz M: Total antioxidant status (TAS), superoxide dismutase (SOD), and glutathione peroxidase (GPx) in oropharyngeal cancer associated with EBV infection. Oxid Med Cell Longev. 2019, 2019:5832410. 10.1155/2019/5832410
13. Ayala A, Muñoz MF, Argüelles S: Lipid peroxidation: production, metabolism, and signaling mechanisms of malondialdehyde and 4-hydroxy-2-nonenal. Oxid Med Cell Longev. 2014, 2014:360438. 10.1155/2014/360438
14. Molavian H, Madani Tonekaboni A, Kohandel M, Sivaloganathan S: The synergetic coupling among the cellular antioxidants glutathione peroxidase/peroxiredoxin and other antioxidants and its effect on the concentration of H2O2. Sci Rep. 2015, 5:13620. 10.1038/srep13620
15. Cavalla F, Osorio C, Paredes R, et al.: Matrix metalloproteinases regulate extracellular levels of SDF-1/CXCL12, IL-6 and VEGF in hydrogen peroxide-stimulated human periodontal ligament fibroblasts. Cytokine. 2015, 73:114-121. 10.1016/j.cyto.2015.02.001
16. Glowacki AJ, Gottardi R, Yoshizawa S, Cavalla F, Garlet GP, Sfeir C, Little SR: Strategies todirect the enrichment, expansion, and recruitment of regulatory cells for the treatment of disease. Ann Biomed Eng. 2015, 43:593-602. 10.1007/s10439-014-1125-2

17. Dahiya P, Kamal R, Gupta R, Bhardwaj R, Chaudhary K, Kaur S: Reactive oxygen species in periodontitis. J Indian Soc Periodontol. 2013, 17:411-416. 10.4103/0972-124X.118306