Signaling Pathway Genetic Variations In Apical Periodontitis

Athira Ajith[1], Usha Subbiah[1*], Deepika. P[2] and Minthami Sharon P[2]

Abstract

A better understanding of gene expression and variation in signaling pathways in apical periodontitis has resulted in identifying the host response and new therapeutic targets for inflammatory diseases. Inflammatory mediators can reduce cell signaling, interfering with transcription factor activation and inflammatory gene expression. In apical periodontitis (AP), genetic studies focusing on the host response and signaling pathways can establish a link between specific possible genes and the disease. This review aims to provide information on host response and genetic variations in major apical periodontitis signaling pathways.

Key Words: Periodontitis, Signalling Pathway, Inflammatory mediators

Introduction

Apical periodontitis (AP) a common infectious disease caused by endodontic etiological agents that leads to inflammation and periradicular tissue loss. AP is the most frequent inflammatory lesion affecting teeth in the jaws, which has a complex inflammatory and non-inflammatory cellular profile that helps regulate extremely complex disease processes. Bacterial infection of the tooth pulp, caries, trauma, or an iatrogenic injury can induce periapical inflammatory reactions, forming granulomas and cysts and bone loss. Endogenous mediators such as prostanoids, kinins, and neuropeptides can mediate immediate-type responses such as vasodilation, increased vascular permeability, and leukocyte extravasation in these inflammatory reactions (P. Stashenkoet al.,1998).

The host responds to the inflammatory response by developing an array of cells that includes intercellular messengers, antibodies, and effector molecules. Microbial factors and host defense forces collide with and destroy much of the periapical tissue, resulting in the creation of numerous AP lesions, the most frequent of which are reactive granulomas and cysts, as well as bone resorption around the roots of impacted teeth (I. Graunaite et al.,2011). Increased production of cytokines, proteases, and other proinflammatory mediators is a hallmark of inflammatory/infectious disorders. Host modulatory therapy aims to restore the body's balance of proinflammatory and anti-inflammatory mediators.

Significance | Review of signaling pathways of apical periodontitis

*Correspondence: Usha Subbiah
Email: ushat75@yahoo.com

Editor Fazlul Huq, Editor-in-Chief at Journal of Angiotherapy. and accepted by the Editorial Board December 08, 2021 (received for review November 23, 2021)

Author Affiliation:
[1]Human Genetics Research Centre, Sree Balaji Dental College and Hospital, Bharath Institute of Higher Education and Research (BIHER), Chennai, Tamil Nadu, India.
[2]Department of Paediatrics, Sree Balaji Medical College and Hospital, Bharath Institute of Higher Education and Research (BIHER), Chennai, Tamil Nadu, India.

Please cite this article:
Athira Ajith, Usha Subbiah, Deepika. P et al., (2021), Signaling Pathway Genetic Variations In Apical Periodontitis, Journal of Angiotherapy, 5(2), 1-12

There are several ways for controlling the host's response with AP. All cellular events, regardless of the host's innate or acquired immunity response, depend on the activation of several signal transduction pathways, which are affected by various microbial and host-derived factors such as lipopolysaccharide (LPS), proteases, cytokines, and other enzymes [M. E. Ryan,2003]. Phosphorylation of certain amino acid residues by kinases, which causes a conformational change in the protein's tridimensional structure, is the most prevalent modification linked with signal transduction. Since it does not involve de novo gene expression, phosphorylation is a very efficient method of transferring energy and regulating biological activity (J. A. C. de Souza,et al.,2012).

Targeting cell signaling pathways are essential for gene expression and are a modern strategy in regulating the levels of inflammatory mediators involved in the immune response, which can be done by activating a limited number of these signaling pathways. Indeed, the short-term modification of cell signaling pathways could affect cytokine gene expression without negatively influencing other fundamental cellular functions. As a result, it is crucial to consider individual signaling pathways in periodontal disease inflammation and tissue degradation (J. A. C. de Souza,et al.,20124). Therefore, this review provides current information on the essential signal transduction pathways and genetic basis in inflammatory periodontal disease.

Signaling pathways in periapical lesions

1. WNT signaling pathway

The WNT/-catenin (Wingless-Integrated/Beta-catenin) signaling system is one of the most critical intercellular signaling pathways in humans, and it is involved in cellular proliferation, differentiation, and regeneration, along with other functions (I. Ejaz S. Ghafoor 2019) . The pathway has been divided into non-canonical (beta-catenin-independent) and canonical (beta-catenin-dependent). WNT pathway ligands include a broad family of 350-400 amino acid glycolipoproteins such as WNT1, WNT2, and WNT3. Wnt will bind to the Frizzled (Fzd) and low-density lipoprotein receptor-related protein 5/6 (LRP5/6) receptors, enabling β-catenin to be translocated from the cytoplasm to the nucleus, where it will bind to T-cell factor/lymphoid enhancer factor (TCF/LEF) to regulate target gene transcription (L. C. de Souza 2019]. Various skeletal abnormalities, fibrotic diseases, inflammatory disorders, and malignant transformations of numerous malignancies are caused by misregulation of the WNT/-catenin system. Periapical lesions form due to bacterial infection of the dental pulp, which produces inflammatory substances, disrupting the triad of RANK-ligand (RANK), RANKL, and OPG receptors. WNT/b-catenin signaling regulation may provide better therapeutic aids for controlling bone loss in endodontic disease. Bone loss in periapical and periodontal lesions is affected by agonists and antagonists of the WNT signaling pathway. Active Wnt/-catenin signaling is important in bone, especially for osteoblast differentiation. The activation of the canonical Wnt pathway in osteoblasts has been found to prevent bone resorption by upregulating OPG expression while down-regulating RANKL expression. Variations in WNT3 have also been linked to changes in bone mineral density and have been linked to osteoporosis.

Differential expression of WNT proteins in AP tissues supports the evidence that these molecules play a role in AP development.WNT3, WNT3A, and WNT5A expression were shown to be considerably greater in AP lesions and WNT3A and WNT5A expression in chronic periodontal disease tissues as compared to healthy tissues (L. C. de Souza 2019).

1. Mitogen activated protein kinase (MAPK)

MAPKs are an evolutionarily conserved family of protein kinases that use diverse receptors to mediate basic biological processes and cellular responses to various external stimuli. MAPKs, or proline-directed serine/threonine kinases, play a significant role in signaling extracellular hormones, growth factors, cytokines, bacterial antigens, and environmental stressors (such as lipopolysaccharide) from gram-negative bacteria), as well as a variety of immune-mediated inflammatory responses.

The MAPK family is divided into three groups: ERK, JNK, and p38 MAPK, which control the activation of several transcription factors. MAPK is primarily active in inflammatory cells, which causes the manufacture of major inflammatory mediators such as tumor necrosis factor (TNF-), interleukin (IL)-1, IL-6, IL-8, and cyclooxygenase-2, either through direct gene transcription activation or messenger RNA stabilization(M. Suzuki *et al.*,2000, R. Beyaert *et al1996*. R. Winzen *et al.*,1999). Periapical lesions are caused by bacterial infection of the tooth pulp, which leads to inflammatory bone resorption. All three MAPK families, ERK, JNK, and p38, are activated by RANK in osteoclasts or osteoclast precursors. p38 MAPK also plays a key role in controlling IL-1 and TNF signaling networks and NF-κB ligand (RANKL)-induced osteoclastic bone resorption(C. Peifer et al.,2006).

p38 MAPK regulates osteoclast development by various factors, including prostaglandin E2, TNF-, and RANKL, according to recent research (G. L. Schieven2005),. Furthermore, studies using p38 inhibitors have confirmed that p38 MAPK is involved in osteoclastogenesis (R. Zhang,et al.,2008, T. Kaneko 2019). Studies have reported that p38 MAPK is important in osteoclast development and that its activation is linked to bone resorption in periapical lesions.

2. Nuclear factor kappa B (NF-κB)

The NF-κB transcription factor family has been involved in several pathways, and it plays an important role in regulating the

Table 1. Signalling pathway gene polymorphism in Apical periodontitis (AP)

Genes Screened for AP polymorphism	AP polymorphism Associated Genes	Authors /year
IL1B (+3954 C/T, rs1143634) IL6 (-174 G/C, rs1800795) IL10 (-1082 G/A, rs1800896) TNFA (-308 G/A, rs1800629 IL1B (+3954 C/T, rs1143634) IL6 (-174 G/C, rs1800795) IL10 (-1082 G/A, rs1800896) TNFA (-308 G/A, rs1800629 IL1B (+3954 C/T, rs1143634) IL6 (-174 G/C, rs1800795) IL10 (-1082 G/A, rs1800896) TNFA (-308 G/A, rs1800629 CD14,IL1B,IL 6,IL10,TNFA	genotype and allele distribution for the genetic polymorphism in IL6 was associated with symptomatic dental abscesses IL6 associated with symptomatic dental abscesses.	Sá et al. (2007)
FcgRIIA, FCgRIIIB, IL1A, IL1B	FcγRIIIb	Siqueira Junior et al. (2009)
IL1B	Associated with AP.	Morsani et al. (2011)
FcgRIIIA	FcγRIIIA not associated with AP	Siqueira Junior et al. (2011)
MMP2, MMP3, MMP9, MMP13, MMP14, TIMP2	MMP3 associated with AP. An altered transmission of MMP2 haplotypes	Menezes-Silva et al. (2012)
IL1A, IL1B	no association with AP	Salazar-Pelaéz et al. (2012)
IL1B, IL8/CXCL8, TNFA	IL8/CXCL8 -251 associated with	Amaya et al. (2013)
CD14 , TLR4	no association with AP	Rôças et al. (2014)
IL10, IL1B, TNF, IL6, OPG, RANKL,RANK	IL1B (rs1143643) showed association with AP	Dill et al. (2015)
MMP8	MMP-8 -799 C/T associated with AP.	Evrosimovska et al. (2015)
HSPA4, HSPA6, HSPA1L, HSPA4L, HSPA9	HSPA1L and HSPA6 showed association with AP	Maheshwari et al. (2016)
MMP1	The MMP1-1607 1G/2G and 1G/2G+2G/2G associated with AP	Trombone et al. (2016)
TLR4	Thr399Ile in TLR4 associated with the pathogenesis of acute apical abscesses	Miri-Moghaddam et al. (2017)
RANK, RANKL, OPG	RANK rs3826620 and RANKL rs9594738 associated with persistent apical periodontitis (PAP)	Petean et al. (2019)
WNT3, WNT3A, WNT5A, WNT8A, WNT9B and WNT11	WNT3, WNT3A associated with AP. WNT3-WNT9B-WNT3A HAPLOTYPES associated with AP.	LC de Souza et al. (2019)
BMP2, *BMP4*, *SMAD6*, and *RUNX2*	interactions between SNPs of *BMP2* and *RUNX2* and between in *BMP4* and in *SMAD6* - associated with PAP	Eriks Calvano Kuchler et al. (2021)

Table 2. Signalling pathway gene expression in AP

Genes Screened for gene expression in AP	Associated Genes	Authors /year
SERPINE1, TIMP1, COL1A1, COL5A1, VTN, CTGF, FGF7, TGFB1, TNF, CXCL11, ITGA4, and *ITGA5*	*SERPINE1, TIMP1, COL1A1, TGFB1,* and *ITGA4* upregulated in periapical granulomas *TNF* and *CXCL11* upregulated in active lesions	Gustavo Pompermaier Garlet et al (2012)
MMP-1, MMP-2, TIMP-1 and TIMP-2	MMP-1, MMP-2, TIMP-1, and TIMP-2 upregulated in periapical lesions	Naida Hadziabdic et al (2016)
CCL5 and ep300	CCL5 and ep300 upregulated in periapical lesions	Naida Hadziabdic et al. (2019)
WNT3, WNT3A, WNT5A, WNT8A, WNT9B WNT11	WNT3, WNT3A and WNT5A upregulated in AP.	LC de Souza et al. (2019)
Sequencing of 852 miRNAs	12- up-regulated, 94- down regulated miR-10a-5p highest expression levels in AP	Zhen Shen et al (2020)
MMP 1, 2, 7, and 9	MMP2 and MMP9 upregulated in AP.	Muhammad Adeel Ahmed et al. (2021)

expression of several genes that control innate and adaptive immune responses. REL-a (p65), NF-B1 (p50; p105), NF-B2 (p52; p100), c-REL, and REL-b24 are the five members of the NF-κB family [4]. Bacterial LPS, TNF-, IL-1, MMPs, COX2, and inducible nitric oxide synthase (iNOS) can activate the NF-κB pathway, producing a variety of proinflammatory mediators. When NF-κB is activated in a wide range of biologically active molecules, it can activate other signaling pathways such as MAPKs and TLRs. A recent study showed that NF-B (p50/p65) activation is significant in periodontally diseased tissues, suggesting that NF-κB inhibitors could treat periodontitis (E. Karl 2005).

Angiogenesis is assisted by NF-κB, which regulates the expression of pro-angiogenic chemokines CXCL1 and CXCL8 [S. P. Tabruyn, A. W. Griffioen2008]. Therefore, inhibiting NF-κB in the angiogenic pathway may reduce periapical lesions and suppress angiogenesis (J. N. Ihle and I. M. Kerr, 1995). The effects of NF-κB decoy ODN treatment on experimentally created periapical lesions and the suppression of NF-κB inhibits periapical lesion formation via blockage of angiogenesis are investigated in a study. The formation of experimentally produced periapical lesions decreased with a reduction in angiogenic responses in endothelial cells when decoy ODN inhibited the administration of NF-κB activity.

3. Janus tyrosine kinase-signal transducer and activator of transcription (JAK/STAT)

The JAK2-STAT3 (Janus kinase 2 / signal transducer and activator of transcription 3) pathway is a signaling pathway that involves activated enzymes known as Janus kinases (JAK1, JAK2, JAK3, and Tyk2) that are found in the cytoplasm of transmembrane receptors (C. Zhang *et al.*,2019). Its biological function is mainly related to cell proliferation, differentiation, apoptosis, and immune regulation, and it has been found to have a crucial role in the occurrence and development of various diseases, including cardiovascular disease, stroke, and malignancies. Activated JAKs will phosphorylate the receptor's cytoplasmic domain, activating its substrate protein, STATs (STAT1-4, 5a, 5b, and 6). STATs can form homo- or heterodimers after being phosphorylated, allowing them to enter the nucleus to regulate gene transcription. Although various ligands can activate individual STAT proteins, certain cytokines selectively activate specific STATs. For example, IFN- activates STAT1 predominantly via JAK1/JAK2, whereas IL-6 activates STAT3 via JAK1 (R. Starr and D. J. Hilton,1999).

Downregulation of the receptor/ligand complex, degradation of signaling intermediates, dephosphorylation of positive regulators (receptor, JAK, or STAT), and activation of particular suppressors [T. Berglundh and M. Donati 2005] are some of the regulatory mechanisms that control signal pathways. Many cytokines that are expected to play biologically important roles in rheumatoid arthritis (IFN-, IL-2, IL-4, IL-6, IL-7, IL-10, IL-12, and IL-15) and periodontal disease (INF-, TNF-, IL-1, IL-4, IL-6, and IL-10) signal through the JAK-STAT pathway. (E. A. Roberts et al.,1995, J. G. Walker 2006, S. Artavanis-Tsakonas et al.,1999)

4. Notch Signaling pathway

The Notch signaling pathway is a highly conserved ligand-receptor cell signaling system involved in many cell functions, including cell proliferation, survival, differentiation, and apoptosis (P. F. Christopoulos, et al., 2021). In vertebrates, four genes encoding Notch receptors (Notch 1-4) and five genes encoding Notch receptor ligands (Jagged 1 and 2 and also Delta 1, 3, and 4) have been discovered. Its role in bone homeostasis has also been identified (D. del Álamo et al.,2011).

The signaling is activated by directly binding the Notch receptor's extracellular domain on one cell to the relevant ligand on an adjacent cell. The Notch receptor is proteolytically cleaved by the gamma-secretase, releasing the Notch receptor intracellular domain that translocates to the nucleus and activates transcription of its target genes, such as the Heygene family of genes [R. A. Backer 2014]. Notch ligands are transmembrane proteins with large extracellular domains like their receptors. As a result, Notch signaling is a short-range signaling mechanism that allows the direct cell to cell communication. The Notch signaling pathway is a central regulator of inflammation, controlling the differentiation and activity of many cells in the innate and adaptive immune response (dendritic cells, natural killer cells, macrophages, B lymphocytes, and various T-cell types) (J. López-López 2012). Proinflammatory cytokines found near the site of inflammation stimulate the activity of the Notch signaling pathway in these cells. TNF-a and IL-1b act as Notch signaling pathway activators, inducing the expression of Notch ligands and receptors and promoting nuclear translocation of Notch receptor intracellular domains.

Genetics of periapical lesions

A complex signaling network determines the nature and extent of AP progression and the associated bone destruction process. Genetic predisposition can contribute to an individual's susceptibility to developing AP. Environmental and acquired risk factors such as diabetes, psychological stress, smoking, and genetic factors (such as gene polymorphisms) can increase the host inflammatory response and, as a result, periodontal disease susceptibility. The link between periodontal disease and higher levels of cytokines and proinflammatory mediators shows that endogenous pathways for negative regulation of these genes may be faulty or malfunctioning, which could be one factor contributing to increased periodontal disease susceptibility (A. Aminoshariae and J. C. Kulild 2015).

Several kinds of research have been conducted to assess host risk factors such as age, gender, and systemic disorders (M. J. B. Silva 2011, J. F. Siqueira Jr,et al.,2009). On the other hand, the impact of

the host's genetic background on apical periodontitis (AP) development is a hot topic in endodontic research. Many genes are involved in AP pathogenesis, according to research involving knockout mice (KO) as an animal model [A. ElAyouti et al.,2011] and studies with human samples (E. C. Küchler et al.,2019) analyzing the complexity of endodontic biology(A. R. De Sá 2007). These investigations highlight candidate genes that could be genetic markers for AP establishment and development variations between individuals. Genes are candidate genes for AP etiology if a genetic variant is associated with the disease. A genome-wide association study approach, which uses a large sample size and a high density of genetic markers across the entire human genome, could identify new genetic variations. Furthermore, patients with AP may have one or more genetic variations in various genes, requiring the conversion of new genetic findings into clinical or public health applications.

Conclusion

Gene expression and variation in signaling pathways lead to identifying new therapeutic targets for inflammatory diseases. Since the WNT, MAPK, NF-B, JAK/STAT, and Notch signaling pathways are shared by various inflammatory mediators, blocking them may be more effective than targeting specific cytokines. However, inhibiting these pathways can have undesirable side effects because they are involved in various other physiological processes. Understanding the genetic basis of diseases will eventually lead to genetic tests to identify disease risks and develop modifying factors for treatments. Determining the genetic basis for apical periodontitis in signaling pathways could help researchers to understand how genetic variation and expression influence host response. More systematic genetic research, such as association study design with a larger sample size, twin studies, segregation studies, and linkage analysis, is required to understand genetic components implicated in signaling pathways connected to AP.

Author Contributions

Athira Ajith and Usha Subbiah conceived of the presented idea. **Deepika. P and Minthami Sharon P** encouraged and supervised the findings of this work. All authors discussed the results and contributed to the final manuscript.

Acknowledgment

None declared

Competing financial interests

The author(s) declare no competing financial interests.

References

P. Stashenko, R. Teles, and R. d'Souza, 1998. "Periapical inflammatory responses and their modulation," Crit. Rev. Oral Biol. Med., vol. 9, no. 4, pp. 498–521, [2]

I. Graunaite, G. Lodiene, and V. Maciulskiene, "Pathogenesis of apical periodontitis: a literature review," J. oral Maxillofac. Res., vol. 2, no. 4, 2011.

M. E. Ryan, 1995 "Clinical applications for host modulatory therapy.," Compend. Contin. Educ. Dent. (Jamesburg, NJ), vol. 23, no. 11A, pp. 1071–1076, 2002.

J. A. C. de Souza, C. R. Junior, G. P. Garlet, A. V. B. Nogueira, and J. A. Cirelli, 2012 "Modulation of host cell signaling pathways as a therapeutic approach in periodontal disease," J. Appl. Oral Sci., vol. 20, no. 2, pp. 128–138,.

I. Ejaz and S. Ghafoor, 2019."WNT signalling pathway in oral lesions," JPMA,

L. C. de Souza et al., 2019. "WNT gene polymorphisms and predisposition to apical periodontitis," Sci. Rep., vol. 9, no. 1, pp. 1–9,

M. Suzuki et al., 2000."The role of p38 mitogen-activated protein kinase in IL-6 and IL-8 production from the TNF-α-or IL-1β-stimulated rheumatoid synovial fibroblasts," FEBS Lett., vol. 465, no. 1, pp. 23–27,

R. Beyaert et al., 1996."The p38/RK mitogen-activated protein kinase pathway regulates interleukin-6 synthesis response to tumor necrosis factor.," EMBO J., vol. 15, no. 8, pp. 1914–1923,

R. Winzen et al., 1999."The p38 MAP kinase pathway signals for cytokine-induced mRNA stabilization via MAP kinase-activated protein kinase 2 and an AU-rich region-targeted mechanism," EMBO J., vol. 18, no. 18, pp. 4969–4980,

C. Peifer, G. Wagner, and S. Laufer, 2006. "New approaches to the treatment of inflammatory disorders small molecule inhibitors of p38 MAP kinase," Curr. Top. Med. Chem., vol. 6, no. 2, pp. 113–149,

G. L. Schieven, 2005."The biology of p38 kinase: a central role in inflammation," Curr. Top. Med. Chem., vol. 5, no. 10, pp. 921–928, [12] R. Zhang, L. Wang, and B. Peng, "Activation of p38 mitogen-activated protein kinase in rat periapical lesions," J. Endod., vol. 34, no. 10, pp. 1207–1210, 2008.

T. Kaneko et al., 2019. "Inhibition of nuclear factor kappa B prevents the development of experimental periapical lesions," J. Endod., vol. 45, no. 2, pp. 168–173,

E. Karl et al., 2005. "Bcl-2 acts in a proangiogenic signaling pathway through nuclear factor-κB and CXC chemokines," Cancer Res., vol. 65, no. 12, pp. 5063–5069,

S. P. Tabruyn and A. W. Griffioen, 2008. "NF-κB: a new player in angiostatic therapy," Angiogenesis, vol. 11, no. 1, pp. 101–106,

J. N. Ihle and I. M. Kerr, 1995. "Jaks and Stats in signaling by the cytokine receptor superfamily," Trends Genet., vol. 11, no. 2, pp. 69–74,

C. Zhang et al., 2019."JAK2/STAT3 is associated with the inflammatory process in periapical granuloma," Int. J. Clin. Exp. Pathol., vol. 12, no. 1, p. 190,

R. Starr and D. J. Hilton, 1999."Negative regulation of the JAK/STAT pathway," Bioessays, vol. 21, no. 1, pp. 47–52,

T. Berglundh and M. Donati, 2005. "Aspects of adaptive host response in periodontitis," J. Clin. Periodontol., vol. 32, pp. 87–107,

E. A. Roberts, K. A. McCaffery, and S. M. Michalek, 1997. "Profile of cytokine mRNA expression in chronic adult periodontitis," J. Dent. Res., vol. 76, no. 12, pp. 1833–1839,

J. G. Walker et al., 2006. "Expression of Jak3, STAT1, STAT4, and STAT6 in inflammatory arthritis: unique Jak3 and STAT4 expression in dendritic cells in seropositive rheumatoid arthritis," Ann. Rheum. Dis., vol. 65, no. 2, pp. 149–156,

S. Artavanis-Tsakonas, M. D. Rand, and R. J. Lake, 1999. "Notch signaling: cell fate control and signal integration in development," Science (80-.)., vol. 284, no. 5415, pp. 770–776,

P. F. Christopoulos, T. T. Gjølberg, S. Krüger, G. Haraldsen, J. T. Andersen, and E. Sundlisæter, 2021. "Targeting the Notch Signaling Pathway in Chronic Inflammatory Diseases," Front. Immunol., vol. 12,

D. del Álamo, H. Rouault, and F. Schweisguth, 2011. "Mechanism and significance of cis-inhibition in Notch signalling," Curr. Biol., vol. 21, no. 1, pp. R40–R47,

R. A. Backer et al., 2014. "A central role for Notch in effector CD8+ T cell differentiation," Nat. Immunol., vol. 15, no. 12, pp. 1143–1151,

J. López-López et al., 2012 "Tobacco smoking and radiographic periapical status: a retrospective case-control study," J. Endod., vol. 38, no. 5, pp. 584–588,.

A. Aminoshariae and J. C. Kulild, 2015. "Association of functional gene polymorphism with apical periodontitis," J. Endod., vol. 41, no. 7, pp. 999–1007,

M. J. B. Silva et al., 2011."The role of iNOS and PHOX in periapical bone resorption," J. Dent. Res., vol. 90, no. 4, pp. 495–500,

J. F. Siqueira Jr, I. N. Rôças, J. C. Provenzano, F. K. Daibert, M. G. Silva, and K. C. Lima, "Relationship between Fcγ receptor and interleukin-1 gene polymorphisms and post-treatment apical periodontitis," J. Endod., vol. 35, no. 9, pp. 1186–1192, 2009.

A. ElAyouti, M. I. Serry, J. Geis-Gerstorfer, and C. Löst, 2011 "Influence of cusp coverage on the fracture resistance of premolars with endodontic access cavities," Int. Endod. J., vol. 44, no. 6, pp. 543–549,.

E. C. Küchler, J. F. Mazzi-Chaves, L. S. Antunes, C. Kirschneck, F. Baratto-Filho, and M. D. Sousa-Neto, 2018 "Current trends of genetics in apical periodontitis research," Braz. Oral Res., vol. 32,.

A. R. De Sá et al., 2007 "Association of CD14, IL1B, IL6, IL10 and TNFA functional gene polymorphisms with symptomatic dental abscesses," Int. Endod. J., vol. 40, no. 7, pp. 563–572,