Genetic and Other Factors Contributing to External Apical Root Resorption in Orthodontic Patients

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Abstract

Objective: External Apical Root Resorption (EARR) is a multifactorial disorder with adverse clinical outcomes in orthodontic practices often resulting in significant root shortening. This study examined the effect that specific single nucleotide polymorphisms (SNPs) have on the risk of developing EARR in orthodontic patients in Alabama. We also evaluated how other selected patient- and treatment-related factors may contribute to root resorption in these patients.

Setting/Sample: Patients included in this case-control study were treated at the University of Alabama at Birmingham School of Dentistry, Department of Orthodontics.

Methods: Panoramic radiographs were used to measure root resorption of the maxillary incisors. EARR was recorded when at least 20% of the root length had been lost with orthodontic treatment. Factors evaluated for association with EARR included ethnicity, sex, age, dental and skeletal classifications, ANB, U1-SN, overjet, treatment type and time, and SNPs in *IL-1A* (rs1800587), *IL-1B* (rs1143634), *IL-1RN* (rs419598), *P2RX7* (rs1718119 and rs2230912), *IRAK1* (rs1059703), and *CASP1* (rs530537, rs580253, and rs554344). Chi-square test, Student's t-test, Wilcoxon test, Benjamin-Hochberg false discovery rate (FDR) adjustment, and logistic regression were used to analyze the data. The significance level was defined as p<0.05.

Results: We found that extraction treatment protocol and dental classification displayed significant association with root resorption. Furthermore, the GG genotype of *IL-1A* rs1800587 variant (in individuals with an increased overjet) predisposed Caucasians to EARR. While *CASP1* (rs530537) variant may contribute to the risk of root resorption, it was not statistically significant after FDR adjustment (p=0.09).

Conclusions: Both patient- and treatment-related factors contributed to EARR.

Keywords

External Apical Root Resorption; risk factors; single nucleotide polymorphism; caspase 1; orthodontic treatment

Introduction

External Apical Root Resorption (EARR) is a multifactorial disorder with adverse clinical outcomes in orthodontic practices often resulting in significant root shortening[1]. Minor surface resorption on the root can be detected histologically as microscopic lacunae in the absence of EARR radiographic evidence[2,3]. This mild resorption can be repaired by cementoblasts. More aggressive progression and exposure of dentin increases the likelihood of permanent root resorption resulting in EARR. Although root resorption may occur in individuals who have never undergone orthodontic treatments, the incidence among treated individuals is quite high, about one-third of patients show signs of EARR[1,4].

A number of risk factors have been revealed related to EARR (e.g., genetics, tooth type and root morphology, bone structure, orthodontic therapy, periodontitis, trauma, and oral habits); however, the evidence is not clear[3,5]. Individuals vary in their susceptibility to EARR with Caucasian and Hispanic patients experiencing significantly more root resorption when compared to Asian orthodontic patiens[6,7]. This suggests that ethnic differences play an important role in individual's susceptibility to EARR. It is estimated that genetic factors can account for approximately 50-60% of EARR variations[8]. Analysis of sib-paired models reported high heritability estimate of 76% for maxillary central incisors[9]. Furthermore, monozygotic twins have shown concordance estimates for EARR that were approximately twice those seen in dizygotic twins[10]. A growing body of evidence suggests that variations in a number of possible genes contribute to the EARR phenotype. Many of these genes encode for proteins involved in the ATP/P2XR7/IL-1 inflammation modulation pathway[8].

The purinergic-receptor-P2X, ligand-gated ion channel 7 (P2RX7) is activated by the ATP released during compressive orthodontic forces[11-13]. This receptor is found to be involved in both osteoclasts, enabling their apoptosis, and osteoblasts, promoting their normal function. Murine studies have shown that when this receptor is inactivated, root resorption under orthodontic forces is increased[14]. In humans, three functional single nucleotide polymorphisms (SNPs) in the *P2RX7* gene have been investigated in relation to EARR: rs1718119, rs208294, and rs2230912[6,15-22]. Of these the +489 C allele (rs208294) and the +1068 G allele (rs1718119) were associated with root resorption.

P2RX7 also promotes the release of the interleukin-1 cytokines (IL-1α and IL-1β) by immune cells[15]. While IL-1α (encoded by the *IL-1A* gene) is an active pro-inflammatory cytokine released by macrophages during the early stages of inflammation, IL-1β (encoded by the *IL-1B* gene) is inactive and requires cleavage by caspase 1 (CASP1) to become active and contribute to the inflammatory process that results in removal of the necrotic tissue and eventually leads to root resorption[6,13,23,24]. However, the action of these cytokines is restrained by the interleukin-1 receptor antagonist (IL-1RN), which bind to the same

receptor as IL-1α and IL-1β. The high affinity binding of IL-1RN to the IL-1R1 receptor inhibits the IL-1 signaling activity. On the other hand, stimulation of IL-1R1 activates the IL-1 receptor-associated kinase 1 (IRAK1), a key modulator of the subsequent signal transduction pathways[25].

Several SNPs in these genes have been extensively studied in orthodontic patients by various groups[6,12,15,17-22,26-30]. However, the results are inconclusive and their association to EARR varies with the population studied[13,31,32]. In some patients, though, the CC genotype of *IL-1B* rs1143634TT and the TT genotypes of *IL-1A* and *IL-1RN* polymorphisms (rs1800587 and rs419598, respectively) have been linked to an increase in root resorption[17,18,20-22,33]. On the contrary, the CC genotype of IRAK1 polymorphism rs1059703, has been suggested as protective for EARR; although, this variant was investigated only by one group[30].

In this study, we examined the effect that specific variants within the *P2RX7*, *IL-1A*, *IL-1B, IL-1RN*, *CASP1* and *IRAK1* genes, involved in ATP/P2XR7/IL-1 pathway, have on the risk of developing EARR in orthodontic patients in X. We also evaluated how other selected patient-related, as well as treatment-related factors may contribute to root resorption in these patients.

Materials and Methods

Patient Sample

This case-control study was approved by the University of Alabama at Birmingham Institutional Review Board (IRB160428005). It involved patients 10 years or older undergoing limited or comprehensive orthodontic treatments with fixed edgewise appliances at the University of Alabama at Birmingham School of Dentistry Orthodontic Clinic for at least 9 months, and had complete initial records as well as progress or final panoramic radiographs. All patients were treated by orthodontic residents under the supervision of various faculty members. Patients who had received prior orthodontic treatment from other clinics were excluded. We also excluded patients with a history of craniofacial syndromes, dental trauma, open apices or incomplete root formation. Furthermore, patients with unclear reference points on radiographs or heavily restored dentition were not included in this study. For the 195 patients that fitted these criteria we recorded their ethnicity, sex, age, dental and skeletal classifications (based on Angle's and Steiner's analysis, respectively), relative position of maxilla to mandible (ANB), maxillary incisor proclination (U1-SN), overjet, and treatment time and type (extraction/non-extraction). Each of these patients was also evaluated for EARR.

EARR diagnosis

Radiographs from two time points (initial records and progress/final records) were digitized using Dolphin Imaging software (Chatsworth, CA). We utilized the ruler on the cephalometric radiograph to measure the molar crown width and transferred that measurement to calibrate the panoramic radiographs. The calibrated panoramic radiographs were then used to measure the crown heights and root lengths of the maxillary incisors.

As previously described, this is a reliable method of measuring root to crown ratios[34]. The millimetric root resorption was computed using the $[(C_2 \times R_1)/C_1] - R_2$ formula; where R_1 and R_2 represent the root lengths at the first and second time point, respectively, and C_1 and C_2 the crown heights for each time point, respectively. The millimetric resorption was converted to % EARR in relation to the initial root length. EARR was recorded when at least 20% of the root length of at least one incisor had been lost with orthodontic treatment. The EARR measurements were completed by two examiners, who were calibrated by the same clinician.

Genetic analysis

Saliva samples collected from each patient were used to isolate genomic DNA utilizing Saliva DNA Collection, Preservation and Isolation Kits (Norgene Biotek, Ontario, Canada). SNP genotyping was performed on a LightCycler 480 Instrument (Roche Diagnostics, Indianapolis, IN) using TaqMan reagents (Thermo Fischer Scientific, Waltham, MA). The nine TaqMan SNP Genotyping Assays used in our study were purchased from Thermo Fischer Scientific (Waltham, MA) and are listed in Table 1.

Statistical analysis

In this study, 10% of the teeth (both central and lateral maxillary incisors) were measured by both examiners and the inter-examiner reliability was assessed using the kappa statistics. For descriptive statistics, we compared age, treatment time, overjet, ANB and U1-SN between EARR group and control group with a two-sided two sample Student's t-test or Wilcoxon test. The association between EARR disorder and gender was assessed using Pearson Chi-square test, while Mantel Haenszel Chi-square test general association statistics was used for ethnicity, and correlation statistic was used for dental classification, skeletal classification and extractions with standardized mid rank scoring. Chi-square test was also used to evaluate genotyping deviations from Hardy-Weinberg equilibrium, while the association between EARR disorder and genotypes or alleles was assessed using Mantel Haenszel Chi-square test general association statistics or Fisher's exact test for small sample size. Furthermore, logistic regression was used to assess the association between EARR disorder and each variable. Odds ratio and 95% confidence interval were obtained for group comparison to each variable. In addition, the interaction between extraction and the other variables as well as SNPs and other variables was assessed using logistic regression. The significance level was defined as $p < 0.05$ and all analysis was done using SAS software Version 9.4 (Cary, NC).

Results

The patients included in this study ranged in age from 9.5 to 51.2 years, with a mean age of 16.2 years. The sample consisted of Caucasians (74%), African Americans (16%), Hispanics (6%), Asians (3%) and South Asians (2%). Of the 195 patients examined, 53 of them were diagnosed with EARR. The interrater agreement on the EARR diagnosis of maxillary incisors was almost perfect with a kappa value of 0.85 and 95% confidence interval of 0.778 to 1.000.

The demographics for both EARR and the control group are shown in Table 2. The EARR patients, on average, started orthodontic treatment at an earlier age (mean of 14.6 years) compared to the control group (mean of 16.8). However, this was not statistically significant, and the median age for the EARR and control patients were similar (13.1 and 13.3 years, respectively). Although not statistically significant, root resorption seemed to be more prevalent in females (58.5% compared to 54.2% in controls) than males. Furthermore, we found no statistical significance in the relationship between race/ethnicity and EARR diagnosis. There were also no statistically significant associations between EARR and other patient-related factors such as overall dental and skeletal classification, ANB, U1-SN, and overjet (Table 3). While we included patients that had been in treatments for at least 9 months, only 3.8% of the EARR group and 7.0% of the controls had received orthodontic treatment for less than 12 months (p>0.05). Furthermore, the average treatment times were comparable, 19.8 months and 19 months for EARR and controls, respectively (p>0.05). On the other hand, the extraction treatment type displayed significant association with root resorption (p=0.01) in the orthodontic patients in this study.

However, as the majority of our sample was comprised of Caucasians and there was not a good representation of the other races/ethnicities, a more in-depth study of the risk factors (including genetic variations) associated with root resorption could only be completed for Caucasians. Of the 144 Caucasian patients analyzed, 43 of them displayed root resorption with orthodontic treatment (Table 4). With this small sample size, an 80% power with α=0.05 could be obtained to detect an EARR difference of 15% if the proportion of control is 53%. If the proportion of control is 94%, then, the 80% power will detect the difference of 17% in EARR. Furthermore, about 46% of these patients had completed their orthodontic treatment at time of the evaluation, while only 34% of controls had finished their treatment. However, this was not statistically significant (p>0.05).

Similar to the data for the overall population, we identified no statistically significant association between EARR diagnosis in Caucasians and their sex, age, skeletal relationship, ANB, U1-SN, overjet and treatment time (p>0.05). Furthermore, extractions continued to be a significant risk factor for developing root resorption (p=0.02). We found that the odds ratio for root resorption in patients with extraction was 4.19 (95% confidence interval: 1.39 to 12.65) times the odds for non-extraction (p=0.01) (Table 5). However, unlike the whole population sample, in Caucasians, dental classification was significantly associated with EARR (p=0.03) and we found that patients with Class II or III malocclusion had an increased risk for root resorption (odds ratio of 2.55; p=0.02) compared to class I patients (Table 4 and 5). We also observed that for every one-year increase in starting age for orthodontic treatment, the odds to developing EARR decreased by 8%; although, this was not statistically significant (p=0.09; with 95% confidence interval of 0.83 to 1.101). Finally, logistic regression found no significant interactions between any of the variables mentioned above (p>0.05).

The genotype and allele frequencies of the nine SNPs analyzed are shown in Table 6. The IRAK1 rs1059703 genotype distribution in females, as well as the frequencies of all other SNPs followed the Hardy-Weinberg equilibrium. Since *IRAK1* is located in chromosome X, males could not be included in this calculation (they do not display a heterozygous

genotype). Of all the polymorphisms examined (*IL-1A* rs1800587; *IL-1B* rs1143634; *IL-1RN* rs419598; *P2RX7* rs1718119 and rs2230912; *IRAK1* rs1059703; *CASP1* rs530537, rs580253, and rs554344), only *CASP1* rs530537 was associated with EARR (p=0.005 and 0.01 for genotype and allele frequencies, respectively). Specifically, we found that the CC genotype for this variant increased the risk for root resorption with odds ratios of 3.90 and 3.51 times that of the CT and TT genotypes (p=0.003 and 0.01), respectively. However, after Benjamin-Hochberg false discovery rate adjustment, this association was only approaching significance (p=0.09).

Furthermore, logistic regression identified that the GG genotype in *IL-1A* rs1800587, when compared to GA+AA, interacted with overjet (p=0.02). For every unit increase in overjet, the odds ratio to develop EARR for the GG genotype was 1.39 with 95% confidence interval (1.05, 1.85). However, no other significant interactions were found between SNPs and sex, age, dental and skeletal classification, ANB, U1-SN, overjet, treatment type or treatment time. On the other hand, after controlling for significant genetic associations, we found that only in the CASP1 rs530537 did dental classification and treatment type remain significant predictors for EARR.

Discussion

External Apical Root Resorption is a complex disorder associated with orthodontic treatment. Since resorption could result in significant shortening of the roots and thus compromise the health of the dentition, understanding the risk factors that contribute to EARR is of great concern for the clinicians. In this study we evaluated the association of certain patient and treatment related factors in orthodontic patients in Alabama to the risk of developing EARR.

When looking at racial/ethnic differences, Sameshima and Sinclaire found that Caucasians and Hispanics displayed significantly more root resorption than Asians[35]. In our sample, the majority of the patients were Caucasians (74%) and the remaining 26% consisted of a combination of four races/ethnicities, which included African Americans (16%), Hispanics (6%), Asian (3%) and South Asians (2%). The number of patients of these later races/ethnicities were insufficient to identify statistical significance association with EARR. However, the trend showed Caucasians, Hispanics and South Asians exhibited more root resorption (30%, 27%, and 25%, respectively) than African Americans and Asians (19% and 0%, respectively). The other demographic factors we investigated were sex and age. Similar to other studies, we found that neither one of these factors was a good predictor of the risk of developing EARR with orthodontic treatment[5,36].

Recent reviews of the literature found no evidence that malocclusion factors like dental and skeletal classifications, relative position of maxilla to mandible, maxillary incisor proclination and overjet, are associated with root resorption[5,36]. Our studies show similar results for the total patient population. However, unlike the whole population sample, we found that, in the Caucasian patients, deviation from class I molar relationship had a statistically increased risk for EARR compared to those with a class I relationship. While a recent clinical practice guideline for EARR stated that it wasn't clear if dental classification

was associated with root resorption[36], we found only one other study that identified class III patients with significantly more EARR[37].

In this study we also evaluated the length of time the orthodontic patients had been in treatment and the effect of extractions on the risk of EARR. Although the length of treatment has been presented as a risk factor for root resorption by various investigators[5,6,16,17,30,35,38,39], we did not identify any significant differences between the EARR and control groups related to treatment time. Since our study included patients that were in treatment for at least 9 months in our orthodontic clinic, it may be suggested that as these patients continue to be seen, a number of our control may develop EARR later in treatment. Even though, as described earlier, there were no significant differences in the distribution of patients who have completed orthodontic treatment between the EARR and control groups, we recognize this as a limitation of this study. Follow-up studies may give insight into the EARR timeline during orthodontic treatment. Furthermore, as described by Segal et al., prolonged treatment times do not necessarily equate with active orthodontic treatment[40]. Similarly, Sondeijker et al. concluded that length of treatment was not an independent predictor of amount of root resorption; however, they recommend avoiding prolonged orthodontic treatments[36].

On the other hand, our data supports previous studies related to premolar extraction being associated with EARR[5,36]. We found that root resorption was observed more often in patients with maxillary premolar extractions. However, extraction of maxillary and mandibular premolars may not necessary display a significant increase in EARR. This may be explained by the fact that an upper premolar extraction treatment protocol is often prescribed for patients with class II division 1 malocclusion, where the maxillary incisors need to be retracted. Thus, the incisor root resorption may be due, in part, to their apical displacement. However, in this study we did not measure the apical root displacements of the incisors.

Looking at other patient-related factors, the evidence linking genetic predisposition to EARR is continuing to grow. Several studies have investigated how variations in a number of genes that participate in one of the two pathways suggested to control osteoclasts activation during resorption: the ATP/P2XR7/IL-1 inflammation modulation pathway; or the OPG/RANK/RANKL (RANK ligand) pathway, contribute to the EARR phenotype[6,8,12,15,17-22,26-30]. In this study we analyzed polymorphisms in genes that encode for proteins involved in the ATP/P2XR7/IL-1 inflammation modulation pathway. This included genotyping for nine SNPs from six genes (*IL-1A* rs1800587; *IL-1B* rs1143634; *IL-1RN* rs419598; *P2RX7* rs1718119 and rs2230912; *IRAK1* rs1059703; *CASP1* rs530537, rs580253, and rs554344).

Members of the interleukin 1 family cytokines were some of the earliest molecules investigated in conjunction with root resorption. In some studies, the CC genotype of *IL-1B* rs1143634 and the TT genotypes of *IL-1A* rs1800587 and *IL-1RN* rs419598 polymorphisms have been linked to an increase risk of EARR[17,18,20-22,33]. However, we did not identify any significant associations with EARR for any of these three cytokines with one exception. In our Caucasian patients, an increase in overjet was associated with a higher risk for root

resorption in individuals carrying the GG genotype for rs1800587 variant of *IL-1A*. In fact, this was the only SNP that significantly interacted with any of the other patient or treatment related variables.

Concerning the *P2RX7* polymorphisms, three previous studies have examined their association with EARR[6,15,16]. Two of them found the GG genotype of the rs1718119 variant to be a significant risk for root resorption[15,16]. However, the second SNP (rs2230912) was not linked to EARR[6]. In this study, we did not find that any of the *P2RX7* polymorphisms tested were predictive of root resorption. Similarly, we discovered no association between the *IRAK1* rs1059703 polymorphism and EARR. This contrasts the study by Pereira et. al, who identified the CC genotype (C in males) as protective against root resorption[30].

Finally, we evaluated three SNPs of the *CASP1* gene (rs530537, rs580253 and rs554344). Similar to Sharab et al. study, we saw no association between EARR and the rs580253 and rs554344 polymorphisms[6]. On the other hand, our study demonstrated that the CC genotype for the rs530537 variant may increase the risk for root resorption. Furthermore, after controlling for significant genetic associations, we found that in patients with this SNP, dental classification and treatment type remained significant predictors for EARR. To our knowledge, this is the first study to demonstrate a potential link between a *CASP1* polymorphism and the EARR disorder. Further studies are needed to elucidate the possible mechanism of how this polymorphism may affect the function of CASP1 and, consequently, the activation of pro-inflammatory cytokines like IL-1B and the signaling pathway leading to root resorption.

Conclusion

Both patients and treatment factors contribute to development of EARR. In this study we showed that maxillary extraction treatment protocol was a risk factor for EARR. We also demonstrated that, in Caucasians, class II and III dental classifications and class III skeletal relationship was associated with root resorption. Furthermore, we found that the GG genotype of rs1800587 variant of *IL-1A* predisposed individuals with an increased overjet to root resorption. Finally, we showed that the rs530537 polymorphism of *CASP1* gene may contribute to the EARR phenotype and should be included as a candidate SNP in future studies.

Acknowledgment

List of Abbreviations

EARR	external apical root resorption
SNP	single nucleotide polymorphism

P2RX7	purinergic-receptor-P2X, ligand-gated ion channel 7
IL-1A	interleukin-1A
IL-1B	interleukin-1B
IL-1RN	interleukin-1 receptor antagonist
CASP1	caspase 1
IRAK1	interleukinin-1 receptor-associated kinase 1

References

1. Taithongchai R, Sookkorn K, Killiany DM. Facial and dentoalveolar structure and the prediction of apical root shortening. Am J Orthod Dentofacial Orthop. 1996;110(3):296–302. [PubMed: 8814031]

2. Abass SK, Hartsfield JK Jr. Orthodontics and External Apical Root Resorption. Seminars in Orthodontics. 2007;13(4):246–256.

3. Vlaskalic V, Boyd RL, Baumrind S. Etiology and sequelae of root resorption. Seminars in orthodontics. 1998;4(2):124–131. [PubMed: 9680910]

4. Killiany DM. Root resorption caused by orthodontic treatment: an evidence-based review of literature. Seminars in orthodontics. 1999;5(2):128–133. [PubMed: 10530286]

5. Sameshima GT, Iglesias-Linares A. Orthodontic root resorption. J World Fed Orthod. 2021;10(4):135–143. [PubMed: 34785166]

6. Sharab LY, Morford LA, Dempsey J, et al. Genetic and treatment-related risk factors associated with external apical root resorption (EARR) concurrent with orthodontia. Orthod Craniofac Res. 2015;18 Suppl 1:71–82. [PubMed: 25865535]

7. Sameshima GT, Sinclair PM. Predicting and preventing root resorption: Part II. Treatment factors. Am J Orthod Dentofacial Orthop. 2001;119(5):511–515. [PubMed: 11343023]

8. Hartsfield JK Jr., Everett ET, Al-Qawasmi RA. GENETIC FACTORS IN EXTERNAL APICAL ROOT RESORPTION AND ORTHODONTIC TREATMENT. Critical reviews in oral biology and medicine : an official publication of the American Association of Oral Biologists. 2004;15(2):115–122. [PubMed: 15059946]

9. Harris EF, Kineret SE, Tolley EA. A heritable component for external apical root resorption in patients treated orthodontically. Am J Orthod Dentofacial Orthop. 1997;111(3):301–309. [PubMed: 9082853]

10. Ngan DC, Kharbanda OP, Byloff FK, Darendeliler MA. The genetic contribution to orthodontic root resorption: a retrospective twin study. Aust Orthod J. 2004;20(1):1–9. [PubMed: 15233581]

11. Hartsfield JK Jr. Pathways in external apical root resorption associated with orthodontia. Orthod Craniofac Res. 2009;12(3):236–242. [PubMed: 19627526]

12. Iglesias-Linares A, Hartsfield JK Jr. Cellular and Molecular Pathways Leading to External Root Resorption. J Dent Res. 2017;96(2):145–152. [PubMed: 27811065]

13. Kalra S, Gupta P, Tripathi T, Rai P. External apical root resorption in orthodontic patients: molecular and genetic basis. J Family Med Prim Care. 2020;9(8):3872–3882. [PubMed: 33110782]

14. Viecilli RF, Katona TR, Chen J, Hartsfield JK Jr., Roberts WE. Orthodontic mechanotransduction and the role of the P2X7 receptor. Am J Orthod Dentofacial Orthop. 2009;135(6):694.e691–616; discussion 694-695.

15. Pereira S, Lavado N, Nogueira L, Lopez M, Abreu J, Silva H. Polymorphisms of genes encoding P2X7R, IL-1B, OPG and RANK in orthodontic-induced apical root resorption. Oral Dis. 2014;20(7):659–667. [PubMed: 24118270]

16. Borilova Linhartova P, Cernochova P, Kastovsky J, Vrankova Z, Sirotkova M, Izakovicova Holla L. Genetic determinants and postorthodontic external apical root resorption in Czech children. Oral Dis. 2017;23(1):29–35. [PubMed: 27493142]

17. Al-Qawasmi RA, Hartsfield JK Jr., Everett ET, et al. Genetic predisposition to external apical root resorption. Am J Orthod Dentofacial Orthop. 2003;123(3):242–252. [PubMed: 12637896]

18. Bastos Lages EM, Drummond AF, Pretti H, et al. Association of functional gene polymorphism IL-1beta in patients with external apical root resorption. Am J Orthod Dentofacial Orthop. 2009;136(4):542–546. [PubMed: 19815156]

19. Gulden N, Eggermann T, Zerres K, Beer M, Meinelt A, Diedrich P. Interleukin-1 polymorphisms in relation to external apical root resorption (EARR). Journal of orofacial orthopedics = Fortschritte der Kieferorthopadie : Organ/official journal Deutsche Gesellschaft fur Kieferorthopadie. 2009;70(1):20–38. [PubMed: 19194673]

20. Iglesias-Linares A, Yanez-Vico R, Ballesta-Mudarra S, et al. Postorthodontic external root resorption is associated with IL1 receptor antagonist gene variations. Oral Dis. 2012;18(2):198–205. [PubMed: 22035161]

21. Iglesias-Linares A, Yanez-Vico RM, Ballesta S, et al. Interleukin 1 gene cluster SNPs (rs1800587, rs1143634) influences post-orthodontic root resorption in endodontic and their contralateral vital control teeth differently. International endodontic journal. 2012;45(11):1018–1026. [PubMed: 22621214]

22. Iglesias-Linares A, Yanez-Vico RM, Ballesta-Mudarra S, et al. Interleukin 1 receptor antagonist (IL1RN) genetic variations condition post-orthodontic external root resorption in endodontically-treated teeth. Histology and histopathology. 2013;28(6):767–773. [PubMed: 23212299]

23. Lee DJ, Du F, Chen SW, et al. Regulation and Function of the Caspase-1 in an Inflammatory Microenvironment. J Invest Dermatol. 2015;135(8):2012–2020. [PubMed: 25815426]

24. Ferrari D, Pizzirani C, Adinolfi E, et al. The P2X7 receptor: a key player in IL-1 processing and release. Journal of immunology (Baltimore, Md : 1950). 2006;176(7):3877–3883. [PubMed: 16547218]

25. Akira S, Uematsu S, Takeuchi O. Pathogen recognition and innate immunity. Cell. 2006;124(4):783–801. [PubMed: 16497588]

26. Tomoyasu Y, Yamaguchi T, Tajima A, Inoue I, Maki K. External apical root resorption and the interleukin-1B gene polymorphism in the Japanese population. Orthodontic Waves. 2009;68(4):152–157.

27. Iglesias-Linares A, Yanez-Vico RM, Ortiz-Ariza E, et al. Postorthodontic external root resorption in root-filled teeth is influenced by interleukin-1beta polymorphism. Journal of endodontics. 2012;38(3):283–287. [PubMed: 22341060]

28. Linhartova P, Cernochova P, Holla LI. IL1 gene polymorphisms in relation to external apical root resorption concurrent with orthodontia. Oral Diseases. 2013;19(3):262–270. [PubMed: 22882407]

29. Guo Y, He S, Gu T, Liu Y, Chen S. Genetic and clinical risk factors of root resorption associated with orthodontic treatment. Am J Orthod Dentofacial Orthop. 2016;150(2):283–289. [PubMed: 27476361]

30. Pereira S, Nogueira L, Canova F, Lopez M, Silva HC. IRAK1 variant is protective for orthodontic-induced external apical root resorption. Oral Dis. 2016;22(7):658–664. [PubMed: 27250598]

31. Pinheiro LHM, Guimaraes LS, Antunes LS, Kuchler EC, Kirschneck C, Antunes LAA. Genetic variation involved in the risk to external apical root resorption in orthodontic patients: a systematic review. Clinical oral investigations. 2021;25(10):5613–5627. [PubMed: 34392402]

32. Nieto-Nieto N, Solano JE, Yanez-Vico R. External apical root resorption concurrent with orthodontic forces: the genetic influence. Acta Odontol Scand. 2017;75(4):280–287. [PubMed: 28358285]

33. Iglesias-Linares A, Sonnenberg B, Solano B, et al. Orthodontically induced external apical root resorption in patients treated with fixed appliances vs removable aligners. The Angle orthodontist. 2017;87(1):3–10. [PubMed: 27504821]

34. Wang J, Rousso C, Christensen BI, et al. Ethnic differences in the root to crown ratios of the permanent dentition. Orthod Craniofac Res. 2019;22(2):99–104. [PubMed: 30650237]

35. Sameshima GT, Sinclair PM. Predicting and preventing root resorption: Part I. Diagnostic factors. Am J Orthod Dentofacial Orthop. 2001;119(5):505–510. [PubMed: 11343022]

36. Sondeijker CFW, Lamberts AA, Beckmann SH, et al. Development of a clinical practice guideline for orthodontically induced external apical root resorption. Eur J Orthod. 2020;42(2):115–124. [PubMed: 31087032]

37. Kaley J, Phillips C. Factors related to root resorption in edgewise practice. The Angle orthodontist. 1991;61(2):125–132. [PubMed: 2064070]

38. Brezniak N, Wasserstein A. Root resorption after orthodontic treatment: Part 2. Literature review. Am J Orthod Dentofacial Orthop. 1993;103(2):138–146. [PubMed: 8427218]

39. Linge L, Linge BO. Patient characteristics and treatment variables associated with apical root resorption during orthodontic treatment. Am J Orthod Dentofacial Orthop. 1991;99(1):35–43. [PubMed: 1986524]

40. Segal GR, Schiffman PH, Tuncay OC. Meta analysis of the treatment-related factors of external apical root resorption. Orthod Craniofac Res. 2004;7(2):71–78. [PubMed: 15180086]

Table 1:

TaqMan SNP Genotyping Assays

SNP ID	Gene	Assay ID	Probe Sequence and Polymorphism
rs1800587	*IL-1A*	C_9546481_30	GATTTTTACATATGAGCCTTCAATG[**G/A**]TGTTGCCTGGTTACTATTATTAAAG
rs1143634	*IL-1B*	C_9546517_10	CATAAGCCTCGTTATCCCATGTGTC[**G/A**]AAGAAGATAGGTTCTGAAATGTGGA
rs419598	*IL-1RN*	C_8737990_10	ATCTGAGGAACAACCAACTAGTTGC[**C/T**]GGATACTTGCAAGGACCAAATGTCA
rs1718119	*P2RX7*	C_11704039_10	CGCTTGTCTGCATTCTCCCCAGGCC[**A/G**]CTGTGTTCATCGACTTCCTCATCGA
rs2230912	*P2RX7*	C_15853715_20	ATTCCTGGACAACCAGAGGAGATAC[**A/G**]GCTGCTTAGAAAGGAGGCGACTCCT
rs1059703	*IRAK1*	C_8966368_10	AGGGGGGATGCAGCTGGCGGCCTCC[**A/G**]AATGCCCGGGCACCCCCGCCACCAC
rs530537	*CASP1*	C__27136093_10	AAAGATATCGTGGAGACAGATATCT[**C/T**]TGCAAATGTTTTGTGAAATTAAGGA
rs580253	*CASP1*	C_7498638_10	ATGTTAAAGATTGCATTGAGTTGTA[**A/G**]TATATCTGGGACTTGCTCAGAGTGT
rs554344	*CASP1*	C_27136072_10	CCTCCCCATCTCCCTGTCATCTTAC[**C/G**]CTTTGATTGGAGACAGTTCTGAAGG

Table 2:

Demographics of the EARR and Control Patients

	EARR		Controls		Significance (p-value)
Race/Ethnicity (N, %)					0.85
Caucasian	43	(81.1%)	101	(71.1%)	
African American	6	(11.3%)	25	(17.6%)	
Hispanic	3	(5.7%)	8	(5.6%)	
Asian	0	(0%)	5	(3.5%)	
South Asians	1	(1.9%)	3	(2.1%)	
All races/ethnicities	53	(100%)	142	(100%)	
Sex (N, %)					0.59
Female	31	(58.5%)	77	(54.2%)	
Male	22	(41.5%)	65	(45.8%)	
Age (years)					0.06
Mean (SD)	14.6	(5.6)	16.8	(9.8)	
Median (range)	13.1	(9.5-51.2)	13.3	(10.6-48.9)	

SD: standard deviation

Table 3:

Analysis of Cephalometric and Treatment Variables in EARR and Control Patients

Variable	EARR (N=53)		Controls (N=142)		Significance (p-value)
Patient-related Factors					
Dental Classification (N, %)					0.07
Class I	18	(34.0%)	74	(52.1%)	
Class II	29	(54.7%)	59	(41.6%)	
Class III	6	(11.3%)	9	(6.3%)	
Skeletal Classification (N, %)					0.16
Class I	23	(43.4%)	75	(52.8%)	
Class II	24	(45.3%)	61	(43.0%)	
Class III	6	(11.3%)	6	(4.2%)	
ANB (°, SD)	2.3	(3.4)	2.7	(2.7)	0.52
U1-SN (°, SD)	106.8	(15.7)	106.4	(9.1)	0.87
Overjet (mm, SD)	4.4	(3.5)	4.3	(2.2)	0.81
Treatment-related Factors					
Treatment Type (N, %)					0.001 [*]
Non-extraction	42	(79.2%)	128	(90.1%)	
Upper premolar extraction	7	(13.2%)	3	(2.1%)	
Upper/lower premolar extraction	4	(7.6%)	11	(7.8%)	
Treatment Time (months, SD)	19.8	(5.8)	19.0	(7.0)	0.44

SD: standard deviation; *p<0.05

Table 4:

Evaluation of Patient and Treatment Characteristics in Relation to the EARR Diagnosis in the Caucasian Population

Variable	EARR (N=43)	Controls (N=101)	Significance (p-value)
Sex, (N, %)			0.66
Female	23 (53.5%)	50 (49.5%)	
Male	20 (46.5%)	51(50.5%)	
Age (years, SD)	13.3 (2.0)	16.2 (9.4)	0.43
Dental Classification (N, %)			0.03 [*]
Class I	13 (30.2%)	53 (52.5%)	
Class II	24 (55.8%)	43 (42.5%)	
Class III	6 (14.0%)	5(5.0%)	
Skeletal Classification (N, %)			0.14
Class I	19 (44.2%)	53 (52.5%)	
Class II	19 (44.2%)	45 (44.5%)	
Class III	5(11.6%)	3 (3.0%)	
ANB (°, SD)	2.2 (3.5)	2.6 (2.5)	0.56
U1-SN (°, SD)	105.2 (16.1)	104.3 (9.1)	0.72
Overjet (mm, %)	4.6 (3.8)	4.4(2.1)	0.67
Treatment Type (N, %)			0.02 [*]
Non-extraction	34 (79.1%)	95 (94.0%)	
Upper premolar extraction	6 (14.0%)	3 (3.0%)	
Upper/lower premolar extraction	3(6.9%)	3 (3.0%)	
Treatment Time (months)	20.2 (6.2)	19.5 (7.1)	0.61

SD: standard deviation; *p<0.05

Table 5:

Odd Ratios of Specific Treatment and Patient Related Factors Associated with EARR in Caucasians

Variable	Odds Ratio	95% Confidence Interval		P-value
Sex				0.66
Female vs male	1.17	0.57	2.40	
Age	0.92	0.83	1.01	0.09
Dental Classification				0.03*
Class II+III vs I	2.55	1.19	5.44	0.02*
Skeletal Classification				0.14
Class II +III vs I	1.39	0.68	2.86	0.47
ANB	0.96	0.84	1.09	0.50
U1-SN	1.01	0.98	1.04	0.65
Overjet	1.04	0.91	1.19	0.59
Treatment Type				0.02*
Ex vs non-ex	4.19	1.39	12.65	0.01*
Treatment time	1.01	0.96	1.07	0.60

Ex: extraction; non-ex: non-extraction; *p<0.05

Table 6:

Distribution of SNP Genotypes in EARR and Control Caucasian Patients

Gene	SNP	Genotype/Allele	EARR (N, %)	Controls (N, %)	Significance (p-value)	Adjusted p-value[**]
		GG	16 (37.2)	53 (52.5)		
		GA	22 (51.2)	40 (39.6)	0.24	0.54
IL-1A	rs1800587	AA	5 (11.6)	8 (7.9)		
		G	54 (62.8)	146 (72.3)	0.11	0.54
		A	32 (37.2)	56 (27.7)		
		GG	19 (44.2)	56 (55.4)		
		GA	20 (46.5)	35 (34.7)	0.40	0.54
IL-1B	rs1143634	AA	4 (9.3)	10 (9.9)		
		G	54 (67.4)	147 (72.8)	0.36	0.54
		A	32 (32.6)	55 (27.2)		
		CC	4 (9.3)	7 (6.9)		
		CT	12 (27.9)	43 (42.6)	0.25	0.54
IL-1RN	rs419598	TT	27 (62.8)	51 (50.5)		
		C	20 (23.3)	57 (28.2)	0.38	0.54
		T	66 (76.7)	145 (71.8)		
		AA	13 (30.2)	20 (19.8)		
		AG	16 (37.2)	45 (44.6)	0.40	0.54
P2RX7	rs1718119	GG	14 (32.6)	36 (35.6)		
		A	42 (48.8)	85 (42.1)	0.29	0.54
		G	44 (51.2)	117 (57.9)		
		AA	25 (58.1)	70 (69.3)		
		AG	15 (34.9)	25 (24.8)	0.42	0.54
P2RX7	rs2230912	GG	3 (7.0)	6 (5.9)		
		A	65 (75.6)	165 (81.7)	0.24	0.54
		G	21 (24.4)	37 (18.3)		
		AA	34 (79.0)	75 (74.3)		
		AG	6 (14.0)	16 (15.8)	0.86	0.86
IRAK1	rs1059703	GG	3 (7.0)	10 (9.9)		
		A	74 (86.0)	166 (82.2)	0.42	0.54
		G	12 (14.0)	36 (17.8)		
		CC	17 (39.5)	15 (14.8)		
		CT	16 (37.2)	55 (54.5)	0.005[*]	0.09
CASP1	rs530537	TT	10 (23.3)	31 (30.7)		
		C	50 (58.1)	85 (42.1)	0.01[*]	0.09
		T	36 (41.9)	117 (57.9)		
		AA	1 (2.3)	4 (3.9)		
CASP1	rs580253	AG	14 (32.6)	23 (22.8)	0.52	0.59
		GG	28 (65.1)	74 (73.3)		

Gene	SNP	Genotype/Allele	EARR (N, %)	Controls (N, %)	Significance (p-value)	Adjusted p-value[**]
		A	16 (18.6)	31 (15.3)	0.49	0.59
		G	70 (81.4)	171 (84.7)		
		CC	1 (2.3)	5 (4.9)		
		CG	14 (32.6)	23 (22.8)	0.39	0.54
CASP1	rs554344	GG	28 (65.1)	73 (72.3)		
		C	16 (18.6)	33 (16.3)	0.64	0.68
		G	70 (81.4)	169 (83.7)		

[*]
p<0.05

[**]
p-value after Benjamin-Hochberg false discovery rate adjustment