Original Article

Effect of fluoride on root resorption following heavy and light orthodontic force application for 4 weeks and 12 weeks of retention

Ersan I. Karadeniz[a]; Carmen Gonzales[b]; Tamer Turk[c]; Devrim Isci[d]; Aynur M. Sahin-Saglam[e]; Huseyin Alkis[f]; Selma Elekdag-Turk[d]; M. Ali Darendeliler[g]

ABSTRACT

Objective: To evaluate the null hypothesis that fluoride intake via drinking water has no effect on orthodontic root resorption in humans after orthodontic force application for 4 weeks and 12 weeks of retention.

Materials and Methods: Forty-eight patients who required maxillary premolar extractions as part of their orthodontic treatment were selected from two cities in Turkey. These cities had a high and low fluoride concentration in public water of $\geq$2 pm and $\leq$0.05 pm, respectively. The patients were randomly separated into four groups of 12 each: group 1HH, high fluoride ($\geq$2 ppm) and heavy force (225 g); group 2LH, low fluoride ($\leq$0.05 ppm) and heavy force; group 3HL, high fluoride and light force (25 g); and group 4LL, low fluoride and light force. Light or heavy buccal tipping force was applied on the upper first premolars for 28 days. At day 28, the left premolars were extracted (positive control side); the right premolars (experimental side) were extracted after 12 weeks of retention. The samples were analyzed with microcomputed tomography.

Results: On the positive control side, under heavy force application, the high fluoride groups exhibited less root resorption ($P = .015$). On the experimental side, it was found that fluoride reduced the total volume of root resorption craters; however, this effect was not statistically significant ($P = .237$). Moreover, the results revealed that under heavy force application experimental teeth exhibited more root resorption than positive control groups.

Conclusion: The null hypothesis could not be rejected. High fluoride intake from public water did not have a beneficial effect on the severity of root resorption after a 4-week orthodontic force application and 12 weeks of passive retention. (*Angle Orthod.* 2013;83:418–424.)

KEY WORDS: Fluoride; Root resorption; Micro-CT

[a] Research Assistant, Department of Orthodontics, Faculty of Dentistry, Karadeniz Technical University, Trabzon, Turkey.
[b] Senior Lecturer, Academic Advisor, and Acting Head, Department of Orthodontics, Faculty of Dentistry, University of Sydney, S & SWS LHDs Oral Health Services, and Sydney Dental Hospital, Sydney, Australia.
[c] Professor, Department of Orthodontics, Faculty of Dentistry, Ondokuz Mayis University, Samsun, Turkey.
[d] Assistant Professor, Department of Orthodontics, Faculty of Dentistry, Ondokuz Mayis University, Samsun, Turkey.
[e] Associate Professor, Inci-Dent Dental Hospital, Isparta, Turkey.
[f] PhD student, Department of Orthodontics, Faculty of Dentistry, Suleyman Demirel University, Isparta, Turkey.
[g] Professor and Chair, Discipline of Orthodontics, Faculty of Dentistry, University of Sydney; Department of Orthodontics, S & SWS LHDs Oral Health Services, and Sydney Dental Hospital, Sydney, Australia.

Corresponding author: Dr M. Ali Darendeliler, Discipline of Orthodontics, Faculty of Dentistry, The University of Sydney, Level 2, 2 Chalmers St, Surry Hills NSW 2010 Australia
(e-mail: ali.darendeliler@sydney.edu.au)

Accepted: August 2012. Submitted: July 2012.
Published Online: November 12, 2012

INTRODUCTION

Although orthodontically induced inflammatory root resorption is an inevitable pathologic consequence of orthodontic treatment, it is not an irreversible phenomenon. It has been suggested that small, shallow resorption lacunae may be completely repaired by deposition of new functional and nonfunctional cementum followed by reestablishment of a new periodontal ligament.[1,2] In the literature, it has been demonstrated that the repair of root resorption takes place in the central[3,4] and peripheral[2,5] parts of the resorption lacunae; therefore, healing seems to occur in all directions. Moreover, in the same active resorption lacunae, it has been shown that fibroblast-like cells, which are indicators of the reparative process, seem to invade the resorption zone from the circumference, while active resorption by multinucleated odontoclast-like cells takes place in the central part.[5] The histologic structure of this reparative cementum was demonstrated as a mixture of cellular and acellular cementum.[4]

 DOI: 10.2319/072512-606.1

After the recognition of its beneficial effects against caries prevention, fluoride became one of the most popular chemical agents in modern dentistry. In mineralized tissues, fluoride reacts by the hydroxyl group of calcium hydroxyapatite ($Ca_5[PO_4]_3OH$) crystals and forms calcium fluorapatite ($Ca_5[PO_4]_3F$) crystal,[6] which is less soluble[7] and therefore can be considered more resistant to demineralization. In the human body, fluoride is almost entirely stored in calcified tissues, including dental hard tissues such as cementum, dentin, and enamel.[6] In several studies, it has been shown that the concentration of fluoride in cementum is highest among the calcified tissues,[8–10] and the amount of accumulated fluoride in cementum increases by age.[11,12]

It has been shown that fluoride stimulates the formation of new bone, primarily via an increase in osteoblast number in vitro[13] and in vivo.[14] It is also believed that sodium fluoride directly inhibits osteoclastic activity by preventing calcium ion release and directly suppresses osteoclast acidity.[15] Previously, it was reported that the cells involved in the repair process of cementum were similar to the cells involved in bone metabolism.[4] Therefore, we hypothesize that fluoride may have a beneficial effect on root resorption 12 weeks after orthodontic force has ceased.

The aim of this investigation was to quantitatively evaluate whether high and low levels of fluoride intake via drinking water affects orthodontic root resorption repair in human subjects when heavy and light orthodontic forces are applied for 4 weeks and 12 weeks of retention.

MATERIALS AND METHODS

Forty-eight patients who required first premolar extractions as part of their orthodontic treatment and who had previously met strict patient selection criteria[16] were selected from two cities in Turkey that had a fluoride concentration of 0.05 ppm (low fluoride) and 2 ppm (high fluoride), respectively, in public water. Ethical approval was given by the Medical Faculty Ethics Committee of the Ondokuz Mayis University, Turkey and SSWAHS, NSW, Australia. All subjects and their guardians consented to participate in this study after receiving verbal and written explanations of the procedures.

Patients were randomly divided into four groups of 12 each according to their systemic fluoride intake via public water and the magnitude of force applied on upper first premolars. Maxillary first premolars were exposed to heavy (225 g) or light (25 g) buccal tipping orthodontic force for 4 weeks. The four groups were as follows: group 1HH, high fluoride and heavy force; group 2LH, low fluoride and heavy force; group 3HL, high fluoride and light force; and group 4LL, low fluoride and light force (Figure 1).

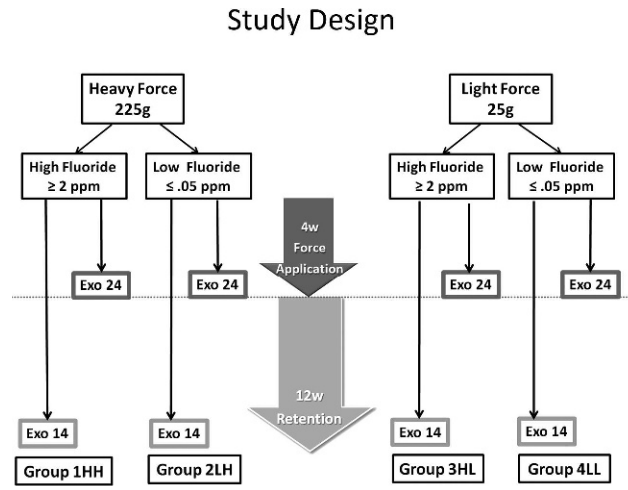

Figure 1. Study design.

Speed brackets 0.022-in slot (Strite Industries, Cambridge, Ontario, Canada) were bonded on the buccal surfaces of the maxillary first permanent molars and first premolars. In groups 3HL and 4LL, 25 g of buccally directed force was induced by a 0.016-in beta-titanium-molybdenum alloy cantilever spring (Rematitan, Dentaurum, Ispringen, Germany) (Figure 2A). In group 2HH and group 4LH, 225g of buccally directed force was produced by a 0.017-in $\times$ 0.025-in beta-titanium-molybdenum alloy cantilever spring (Beta III Titanium, 3M Unitek, Monrovia, Calif) (Figure 2B). Force magnitude was measured with a strain gauge (Dentaurum). Light-cured band cement (Transbond Plus, 3M Unitek) was bonded on the occlusal surfaces of the mandibular first permanent molars to minimize occlusal trauma to the maxillary first premolars during the experiment (Figure 2C).

After 4 weeks of force application, the cantilever springs were removed in all the groups and the upper left first premolars were extracted as the positive control side. Maxillary right premolars (experimental side) underwent 12 weeks of retention with a passive 0.0175-in multistranded steel wire bonded on the palatal surface of the maxillary first, second premolars, and first molar (Figure 2D). After 12 weeks of retention, maxillary right premolars were extracted.

To assess cementum repair, volumetric changes of the resorption craters were scanned with a high-resolution X-ray microcomputed tomography (micro-CT) system (SkyScan 1172, Aartselaar, Belgium) above the cementoenamel junction to the root apex at 16.78 μm magnification and a resolution of 11.45 μm pixel size. The X-ray tube was operated at 60 kV and 165 mA.

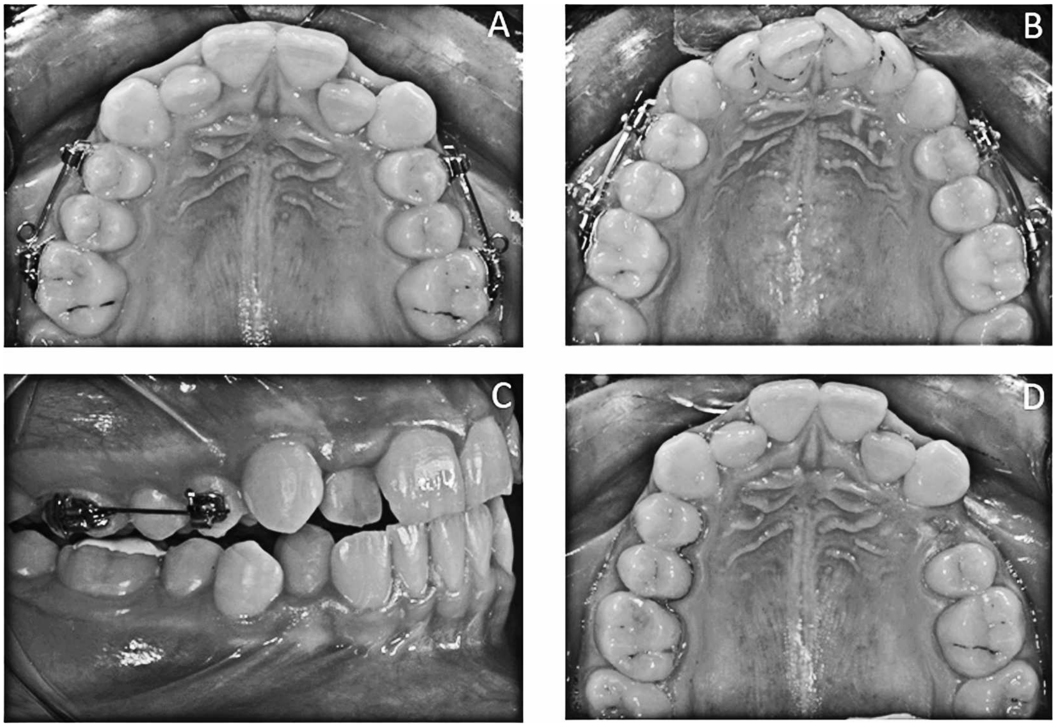

Figure 2. Intraoral images of the appliance. (A) Occlusal view, light force. (B) Occlusal view, heavy force. (C) Lateral view showing the band cement to avoid occlusal interferences. (D) Retention wire bonded on the upper first premolars and first molar (experimental side); the maxillary left premolar has been extracted after 4 weeks of force application (positive control side).

After image acquisition, the raw data were processed with Nrecon software (version 1.4.2, Aartselaar, Belgium). Resorption craters were identified and isolated on three-dimensional (3D) reconstructions created by VG Studio Max software (version 1.2, Volume Graphics, GmbH, Heidelberg, Germany) (Figure 3A). The volume of the isolated crater images was quantified by Convex Hull 2D software (Chull 2D, University of Sydney, Sydney, Australia). In general, a convex hull creates a surface closure that is a local minimum in terms of its surface area. This effectively creates a flat closure that approximates the original tooth surface to a greater or lesser extent depending on the curvature of the tooth surface at the crater. A minor limitation associated with this volume estimation software is how the convex hull algorithm closes the craters. When a tooth surface is convex, flat closure will be more likely to underestimate crater volume. The

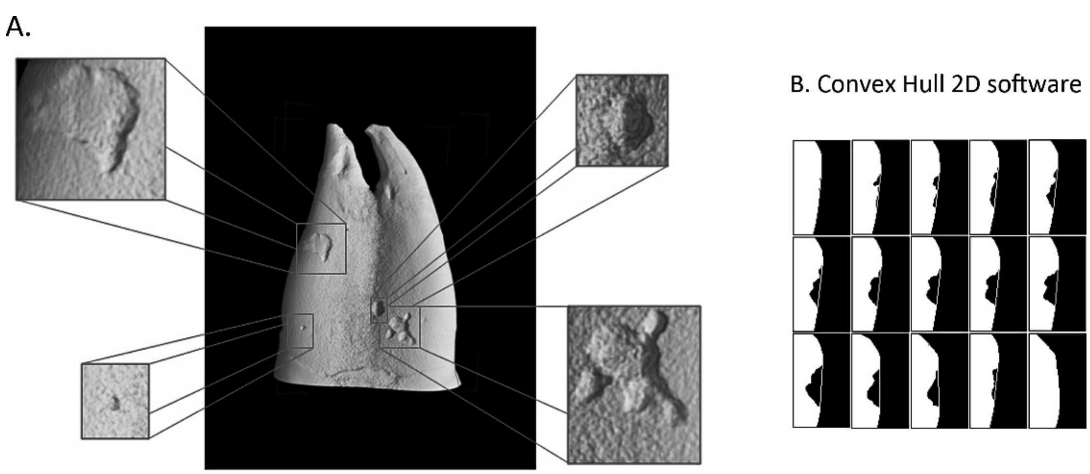

Figure 3. Microcomputed tomography analysis. (A) Identification and isolation of the crater. (B) Convex Hull two-dimensional software. This software draws a line connecting the edges of the resorption crater creating a subvolume for each axial slice. The total crater volume is calculated by summing the subvolumes of each closed cross section.

converse is also true: the software will overestimate volume in areas of concavity. This small error is mitigated, however, because this method of crater volume measurement is based on direct imaging of the tooth as a 3D object. Hence, the error in volume estimate is considered negligible (Figure 3B).

Statistical Analysis

All statistical analyses were undertaken with the Statistical Package for Social Sciences software (SPSS for Windows, version 16, SPSS, Chicago, Ill). Univariate analysis of variance and pairwise comparisons between the groups were performed. The results were depicted by using box plots. Bonferroni adjustments were made for multiple comparisons. Statistical significance was set at the $P \leq .05$ level. All measurements were done by the same blinded researcher.

RESULTS

On the positive control side (maxillary left premolars) under heavy force application, the high fluoride group exhibited less root resorption than the low fluoride group ($P = .015$) (Figure 4A, Table 1). However, under light force application there was no statistical difference between the high fluoride group and the low fluoride group ($P = .742$) (Figure 4A).

On the experimental side, root resorption continued after 12 weeks of passive retention and orthodontic force ceased. Interestingly, after 4 weeks of heavy force (225 g) application and 12 weeks of retention it was found that fluoride tended to reduce the volume of root resorption craters; however, this effect was not statistically significant ($P = .237$) (Figure 4B). The comparison between control and experimental sides is shown in Figure 4C. The results revealed that under heavy force application experimental teeth exhibited more root resorption than teeth in the positive control groups.

DISCUSSION

This investigation aimed to quantify how fluoride intake via drinking water affects the amount of root resorption under light (25 g) and heavy (225 g) force application for 4 weeks and after 12 weeks of retention. In brief, our results showed that fluoride reduced root resorption 4 weeks after force induction; however, after a retention period of 12 weeks the tendency toward decreased root resorption was not statistically significant.

The effect of fluoride on orthodontic root resorption was studied previously.[16,17–19] Foo et al.[17] postulated that fluoride reduced the average amount of root resorption in rats. However, this effect was not statistically significant ($P = .112$). On the other hand, Gonzales et al.[18] reported that not only fluoride exposure from drinking water but also the duration of this exposure has a significant effect on root resorption. Lim et al.[19] reported that rats exposed to 100 ppm fluoridated water exhibited a significant decrease in the depth and length of the root resorption lesions. In this study, we found that under heavy force application fluoride had a significant effect on reducing root resorption (Figure 4A). A decrease in the average rate of root resorption under light force application was also observed, but this effect was not statistically significant ($P = .742$).

Rygh[20] suggested that if the application of force is absent or is below a certain level repair will commence in the resorption sites with the deposition of cementum. Schwartz[21] stated that the minimum level of force that causes root resorption is between 7 g/cm^2 and 26 g/cm^2. According to the root surface area calculation in this study, 8 g/cm^2 and 70 g/cm^2 of force were applied in light and heavy force groups, respectively. Therefore, as expected, we observed less root resorption in the light force groups compared with the heavy force groups.

Brudvik and Rygh[1,5] demonstrated that root resorption continues until the hyalinized periodontal ligament is completely removed regardless of force termination.

A recent study by Cheng et al.[22] investigated root resorption repair 4 weeks and 8 weeks after the application of light and heavy forces showed a clear trend that root resorption continued for another 4 weeks after orthodontic force ceased. Moreover, they noted that the reparative processes were different between light and heavy orthodontic force application and resorptive activity was more pronounced after the application of heavy forces. It was previously shown that the resorptive process increased to peak 4 weeks after the force termination and slowed down to steady phase after 5, 6, 7, and 8 weeks of retention[2]; therefore, we selected 12 weeks of retention as the experimental time. The results of the present study showed that even after 12 weeks of retention the volumes of root resorption craters in the experimental side of the heavy force groups were larger than those in the positive control group (Figure 4B). On the other hand, less root resorption was observed in the experimental light force groups compared with the positive control groups (Figure 4C, Table 1). Our findings demonstrate that root resorption continues despite the cessation of orthodontic force, and this resorptive activity is more distinct under heavy force application as it was shown to cause larger and more persistent hyalinization in the periodontal ligament.[23]

Histologically, the physical properties of cementum are similar to those of bone,[6] and the cells involved in

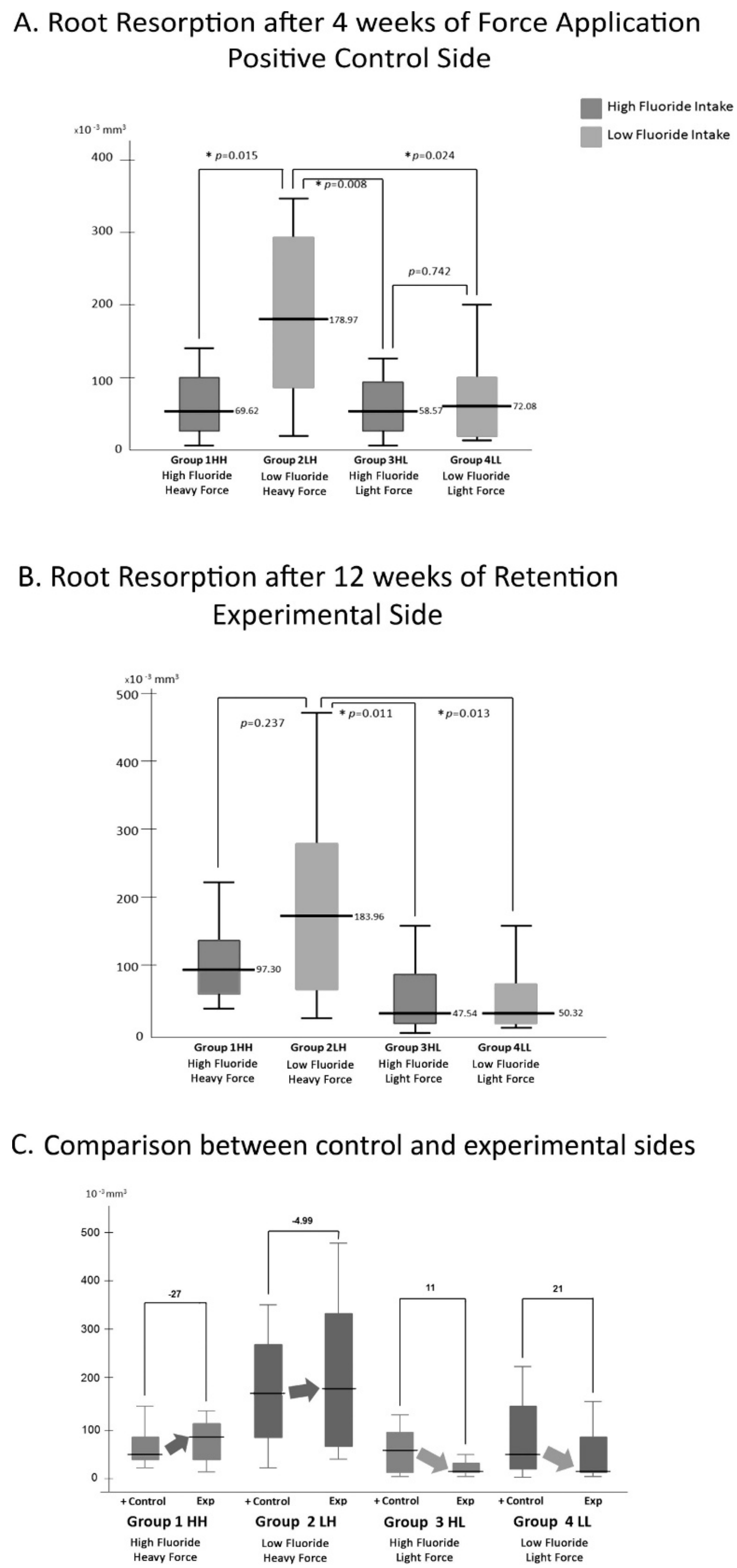

Figure 4. (A) Root resorption after 4 weeks of force application (positive control side). (B) Root resorption after 12 weeks of retention (experimental side). (C) Comparison between positive control and experimental sides in each group.

Table 1. Descriptive Statistics of Sex, Age and Root Resorption[a]

		Sex		Age		Root Resorption ($\times 10^{-3}$ mm^3) 4 Weeks of Force		4 Weeks of Force + 12 Weeks of Retention		Difference Between Experimental and Control Groups	
Group	N	Male	Female	Average	Range	Mean	SD	Mean	SD	Mean	SD
Group 1HH	12	5	7	14.9	11.7–20.0	69.62	65.72	97.3	68.29	−27.67	94.22
Group 2LH	12	6	6	15	14.0–17.0	178.97	119.64	183.96	149.91	−4.99	170.82
Group 3HL	12	6	6	15.4	13.1–17.8	58.57	47.53	47.54	51.75	11.02	67.01
Group 4LL	12	6	6	15.1	13.5–16.5	72.08	77.41	50.32	81.58	21.75	95.31

[a] Group 1HH, indicates high fluoride intake and heavy force; group 2LH, low fluoride intake and heavy force; group 3HL, high fluoride intake and light force; group 4LL, low fluoride intake and light force; SD, standard deviation.

the repair process of cementum were suggested to be similar to the cells involved in bone metabolism.[4] It has been shown that fluoride reduced the number of the osteoclasts in bone.[24] Additionally, it has been demonstrated that fluoride decreases the number of resorption lacunae made by individual clast cells,[25] the resorbed area per clastic cells,[16,19,25] and the depth of the clastic cell demineralization pit.[16,19,26] In the present study, high fluoride groups exhibited a decreased amount of root resorption after the application of light and heavy forces for 4 weeks. (Figure 4A) However, after a retention period of 12 weeks, root resorption was more distinct in the high fluoride groups as the difference between the control and experimental sides in the high fluoride groups is less than in the low fluoride groups (Figure 4C). One possible explanation might be the possibility of a reduction in the osteoclast population and activation that might reduce bone remodeling, leading to an increase in the hyalinization zone, which might amplify the resorptive processes.[17] This approach could explain why the experimental side of the high fluoride groups underwent a more pronounced resorptive activity during the retention period, especially under heavy force application.

Individual susceptibility or resistance to root resorption seems to be a significant factor affecting the results of this study (Table 1). Although strict selection criteria were followed during patient selection, some specific factors, such as genetics, individual susceptibility, or resistance to root resorption, cannot be controlled or predicted by practitioners. A significant contribution of individual response to orthodontic force activation was also noted in several previous studies.[21,22]

Although the high spatial resolution provided by micro-CT allows the identification and measurement of the volume of the root resorption lesions with high accuracy, micro-CT scan does not allow researchers to distinguish between normal and repaired cementum. Therefore, the amount of repaired cementum in resorption craters could not be calculated. This can only be investigated histologically or with nano-computed tomography technology. However, with these methods the number of histologic sections is usually limited to a small number per tissue sample, and it is not possible to obtain a fully 3D volumetric evaluation of repaired cementum. In this study, it might be assumed that the intraindividual differences in the volume of the resorption craters between the positive control and experimental groups were estimates of the amount of reparative cementum. However, this needs to be confirmed histologically.

Further investigation is still required to elucidate whether fluoride will have a similar effect in patients undergoing a complete duration of orthodontic treatment.

CONCLUSION

- The null hypothesis could not be rejected. Although patients with a high fluoride intake exhibited significantly less root resorption after 4 weeks of heavy force application, this effect was not statistically significant 12 weeks after passive retention and orthodontic force ceased.
- Therefore, high fluoride intake from public water was not found to have a beneficial effect on the severity of root resorption after heavy and light orthodontic force application for 4 weeks and 12 weeks of retention.

REFERENCES

1. Brudvik P, Rygh P. Transition and determinants of orthodontic root resorption-repair sequence. *Eur J Orthod*. 1995; 17:177–188.
2. Vardimon AD, Graber TM, Pitaru S. Repair process of external root resorption subsequent to palatal expansion treatment. *Am J Orthod Dentofacial Orthop*. 1993;103: 120–130.
3. Barber AF, Sims MR. Rapid maxillary expansion and external root resorption in man: a scanning electron microscope study. *Am J Orthod*. 1981;79:630–652.
4. Cheng LL, Turk T, Elekdag-Turk S, Jones AS, Yu Y, Darendeliler MA. Repair of root resorption four and eight weeks following the application of continuous light and heavy forces on human premolars for four weeks: a histology study. *Am J Orthod Dentofacial Orthop*. 2011 In Press.
5. Brudvik P, Rygh P. The repair of orthodontic root resorption: an ultrastructural study. *Eur J Orthod*. 1995;17:189–198.

6. Robinson C, Kirkham J, Weatherell JA. Fluoride in teeth and bone. In: Fejerskov OEJ, Burt BA, eds. *Fluoride in Dentistry*. Copenhagen, Denmark: Munksgaard Textbook; 1996: 69–83.
7. Grynpas MD. Fluoride effects on bone crystals. *J Bone Miner Res*. 1990;5(suppl 1):S169–S175.
8. Gedalia I, Nathan H, Schapira J, Haas N, Feldmann J. Fluoride concentration of surface enamel, cementum, lamina dura, and subperiosteal bone from the mandibular angle of Hebrews. *J Dent Res*. 1965;44:452.
9. Yardeni J, Gedalia I, Kohn M. Fluoride concentration of dental alculus, surface enamel and cementum. *Arch Oral Biol*. 1963;8:697–701.
10. Singer L, Armstrong WD. Modified diffusion method for analysis of fluoride. *J Dent Res*. 1962;41:910.
11. Nakata TM, Stepnick RJ, Zipkin I. Chemistry of human dental cementum: the effect of age and fluoride exposure on the concentration of ash, fluoride, calcium, phosphorus and magnesium. *J Periodontol*. 1972;43:115–124.
12. Stepnick RJ, Nakata TM, Zipkin I. The effects of age and fluoride exposure on fluoride, citrate and carbonate content of human cementum. *J Periodontol*. 1975;46:45–50.
13. Farley JR, Wergedal JE, Baylink DJ. Fluoride directly stimulates proliferation and alkaline phosphatase activity of bone-forming cells. *Science*. 1983;222:330–332.
14. Turner CH, Garetto LP, Dunipace AJ, et al. Fluoride treatment increased serum IGF-1, bone turnover, and bone mass, but not bone strength, in rabbits. *Calcif Tissue Int*. 1997;61:77–83.
15. Krieger NS, Tashjian AH Jr. Parathyroid hormone stimulates bone resorption via a Na-Ca exchange mechanism. *Nature*. 1980;287:843–845.
16. King AD, Turk T, Colak C, Elekdag-Turk S, Jones AS, Petocz P, Darendeliler MA. Physical properties of root cementum: part 21. Extent of root resorption after the application of 2.5 degrees and 15 degrees tips for 4 weeks: a microcomputed tomography study. *Am J Orthod Dentofacial Orthop*. 2011;140:e299–e305.
17. Foo M, Jones A, Darendeliler MA. Physical properties of root cementum: part 9. Effect of systemic fluoride intake on root resorption in rats. *Am J Orthod Dentofacial Orthop*. 2007;131:34–3.
18. Gonzales C, Hotokezaka H, Karadeniz EI, Miyazaki T, Kobayashi E, Darendeliler MA, Yoshida N. Effects of fluoride intake on orthodontic tooth movement and orthodontically induced root resorption. *Am J Orthod Dentofacial Orthop*. 2011;139:196–205.
19. Lim E, Belton D, Petocz P, Arora M, Cheng LL, Darendeliler MA. Physical properties of root cementum: part 15. Analysis of elemental composition by using proton-induced x-ray and gamma-ray emissions in orthodontically induced root resorption craters of rat molar cementum after exposure to systemic fluoride. *Am J Orthod Dentofacial Orthop*. 2011; 139:e193–e202.
20. Rygh P. Orthodontic root resorption studied by electron microscopy. *Angle Orthod*. 1977;47:1–16.
21. Schwartz AM. Tissue changes incident to orthodontic tooth movement. *Int J Orthod*. 1932;18:331–352.
22. Cheng LL, Turk T, Elekdag-Turk S, Jones AS, Petocz P, Darendeliler MA. Physical properties of root cementum: part 13. Repair of root resorption 4 and 8 weeks after the application of continuous light and heavy forces for 4 weeks: a microcomputed-tomography study. *Am J Orthod Dentofacial Orthop*. 2009;136(3):320.e1–e10; discussion 20–21.
23. Thilander B, Rygh P, Reitan K. Tissue reactions in orthodontics. In: Graber T, Vanarsdall RL, eds. *Orthodontics: Current Principles and Techniques*. St Louis, Mo: Mosby; 2005:145–219.
24. Hellsing E, Hammarstrom L. The effects of pregnancy and fluoride on orthodontic tooth movements in rats. *Eur J Orthod*. 1991;13:223–230.
25. Okuda A, Kanehisa J, Heersche JN. The effects of sodium fluoride on the resorptive activity of isolated osteoclasts. *J Bone Miner Res*. 1990;5(suppl 1):S115–S120.
26. Taylor ML, Maconnachie E, Frank K, Boyde A, Jones SJ. The effect of fluoride on the resorption of dentine by osteoclasts in vitro. *J Bone Miner Res*. 1990;5(suppl 1):S121–S130.