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Bone response to different strength orthodontic forces in animals with periodontitis

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Background: Occlusal alterations resulting from tooth movements caused by periodontitis-related bone loss are often corrected with orthodontic treatments. Although the outcome is usually satisfactory, a quantitative histomorphometric study of bone response would contribute to improving treatment planning and optimizing results.

Methods and Results: This study is a histomorphometric analysis of alveolar bone response to 51 and 75-g orthodontic forces applied to rat molars subjected to experimental periodontitis by placing a ligature around the neck of the molar during 48 h. The orthodontic device consisted of two bands with a tube welded to their palatine aspect, through which the arms of a helicoidal spring were threaded so as to exert force toward palatine. The device was placed immediately and 48 h after removing the ligatures. When applied 48 h post-removal of the ligature, both orthodontic forces caused an increase in bone volume in the periodontitis group.

Conclusions: Our study shows that application of orthodontic forces once periodontal infection has been controlled contributes to increasing alveolar bone volume, consequently improving bone quality.

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Periodontitis-related bone loss often causes tooth movements, which in turn alter occlusion. These occlusal alterations feature increased overjet and overbite, diastema, and extrusion, which are often corrected satisfactorily with orthodontic treatment. Although clinical (1–3) and experimental (4) studies combining periodontal and orthodontic therapy have been reported, there are no histomorphometric studies on bone response to date, nor has the effect of different strength orthodontic forces applied to teeth of animals suffering periodontal disease teeth been investigated. The effectiveness of combining these two therapies is supported only by radiographic and clinical evidence. Understanding bone response to different times of application and force strengths would allow more rational treatment planning, thus improving the outcome.

In view of the above, the aim of this experimental work was to perform histomorphometric and histologic studies on the effect of orthodontic forces on alveolar bone of rats suffering periodontitis-related bone loss, using different force strengths and times of application.

Material and methods

Forty-five male Sprague Dawley rats weighing 250 g, kept under regular laboratory feeding and housing conditions (water and hard diet), were assigned to one of nine groups (Table 1). The National Institutes of Health *Guidelines for the Care and Use of Laboratory Animals* (NIH Publication 85–23, Rev 1985) were observed.

Under ethyl urethane intraperitoneal anesthesia (1 g/kg body weight) a 4-0 diameter cotton thread was placed around the cervix of the first upper molars in order to induce periodontitis (5–11). The orthodontic force was generated by a device consisting of two stainless steel bands cemented to the first upper molars, with a tube welded to their palatine aspect and through which the arms of a helicoidal spring were threaded in order to exert force towards palatine (12). The animals were killed, and their upper maxillae were resected, fixed in 10% formalin, decalcified in EDTA (pH 7.2) and embedded in paraffin. Buccolingually

Table 1. Experimental groups

Groups	0 h	48 h	96 h	144 h
1	Control			
2	OF 51 g	Sacrifice		
3	OF 75 g	Sacrifice		
4	Ligature	Sacrifice		
5	Ligature	Removal of ligature	Sacrifice	
6	Ligature	Removal of ligature + OF 51 g	Sacrifice	
7	Ligature	Removal of ligature + OF 75 g	Sacrifice	
8	Ligature	Removal of ligature	OF 51 g	Sacrifice
9	Ligature	Removal of ligature	OF 75 g	Sacrifice

OF: orthodontic force.

oriented sections at the level of the central roots of the upper first molar were obtained and stained with hematoxylin and eosin. Paper tracings of projections of the sections were made, and the area used for the histomorphometric study was defined in the interradicular space (Fig. 1). The following histomorphometric parameters were measured according to stereological principles (13).

(a) Percentage of bone resorption surfaces were measured on the vestibular aspect of interradicular bone in the cervical and middle third of the interradicular space. For this purpose the resorption surfaces observed under the microscope were transferred to the corresponding zones of the tracings. The contour of the vestibular plate between the intersection of the bone outline and lines b and d was considered 100% (Fig. 1).

(b) Bone loss: percentage of the full height of the interradicular space corresponding to the periodontal space, measured from the root furcation to the interradicular bone crest (segment CG, Fig. 1).

(c) Bone volume: fraction of total bone volume corresponding to trabecular volume, measured in the interradicular bone found in the cervical third of the interradicular space limited by line C (Fig. 1).

Overall statistical differences among groups were established using the Kruskall–Wallis non-parametric test, and differences within groups were determined using Bonferroni's test for multiple comparisons.

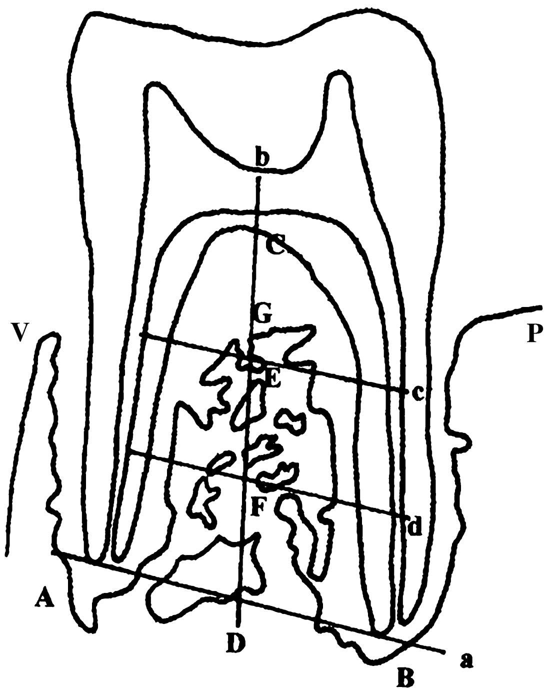

Fig. 1. The study area was defined on paper tracings of projections of the histologic sections as follows. Line a was drawn to determine segment AB, tangential to the apices of the vestibular and palatine roots. Line b was drawn from the middle of segment AB towards the middle point of the furcation, thus defining segment CD. The latter was in turn divided into three equal segments: CE, EF, and ED. Two lines parallel to AB were drawn through points E and F, dividing the interradicular space into the cervical, middle, and apical thirds. The intersection of segment CD and the crest of interradicular bone was defined as point G. Bone loss (BL) was measured on line b, using the following equation: $BL = CG/CD \times 100$. V: vestibular; P: palatine.

Results

Control sections (Group 1) exhibited the apical limit of the junctional epithelium at the cementoenamel junction, no signs of inflammation, and periodontal fibers clearly inserted between the cementum and the alveolar bone (Fig. 2).

The orthodontic forces applied to Groups 2 and 3 caused the first molars to move toward palatine, generating cervicolingual pressure zones and apicovestibular tension zones. The pressure zone exhibited narrowing of the periodontal ligament, extensive erosion surfaces on the periodontal bone, with no increase in the number of osteoclasts. The tension side of the periodontal ligament was found to have widened with no evidence of an increase in bone formation.

Group 4 exhibited periodontal pockets with inflammatory infiltrate among the epithelial cells. The infiltrate also occupied the gingival chorion and the periodontal ligament. Hemorrhage, necrosis, and granulation tissue were also evident. The periodontal bone surface exhibited increased erosion surfaces and number of osteoclasts (Fig. 3). Group 5 showed less inflam-

Fig. 2. Interradicular zone of a case from Group 1 (control). Bone formation areas with active osteoblasts can be observed on the vestibular surface of the interradicular bone, on the left side of the image (hematoxylin and eosin 10 ×).

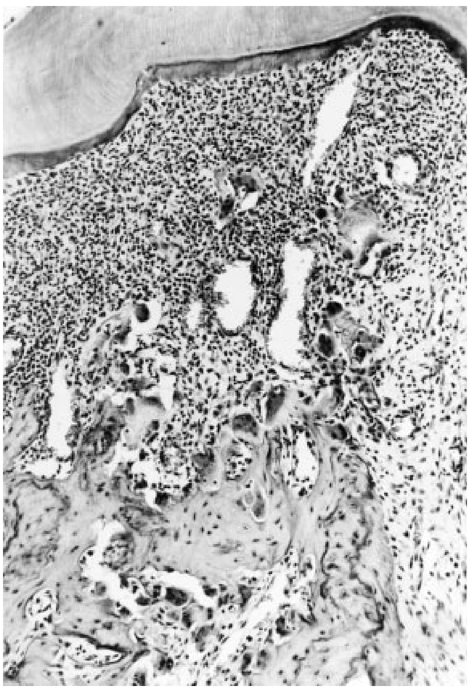

Fig. 3. Interradicular zone of a case from Group 4 (periodontitis). Note the presence of large quantities of inflammatory infiltrate in the furcation area. The interradicular bone shows bone loss and a large number of osteoclasts associated with erosion areas, both on the periodontal bone surface and in the medullary spaces (hematoxylin and eosin 10 ×).

matory infiltrate in the juxtaepithelial gingival area and in the periodontal ligament, as compared with Group 4; the furcation area showed congestion, and the interradicular bone exhibited extensive erosion areas on the periodontal surface and in the medullary spaces. Molars of animals in Groups 6 (Fig. 4) and 7 tilted towards palatine, exhibited deep periodontal pockets and inflammatory infiltrate in the gingival epithelium, chorion, and periodontal ligament, hemorrhage and areas of necrosis, granulation tissue, and periodontal abscesses in the furcation area. The alveolar bone exhibited extensive erosion areas with increased number of osteoclasts.

Groups 8 (Fig. 5) and 9 exhibited substantially less inflammatory infiltrate in the gingival chorion and periodontal ligament. The vestibular plate and crest of the interradicular bone showed erosion surfaces. Unlike application of the 51-g force, the 75-g force was found to cause necrosis of the periodontal ligament on the pressure side.

The results of the histomorphometric study are shown in Table 2.

Discussion

Our study shows that application of orthodontic forces contributes to increasing alveolar bone volume, when bone volume has been decreased through experimental periodontitis.

It is well documented that alveolar bone response to orthodontic forces involves resorption on the pressure side and neoformation on the tension side. In this study, the force was applied to the bone modeling surface of the tooth alveolus, i.e. exerting a palatine oriented force that is opposite to the natural vestibular drift of the teeth in rats. Therefore, there were no resorption areas on the palatal plate of alveolar bone prior to applying the experimental force (12). For this reason, the vestibular aspect of interradicular bone, a natural bone modeling area, was used to measure bone activity.

Tooth response to the application of a force involves the coordinated action of periodontal ligament cells, osteoblasts, and osteocytes (14). The periodontal ligament is essential in alveolar bone response to a force applied to the tooth lodged in the alveolus, acting as a sensor of force, transmitting the force to the bone where osteocytes respond both to pressure (15) and to tension

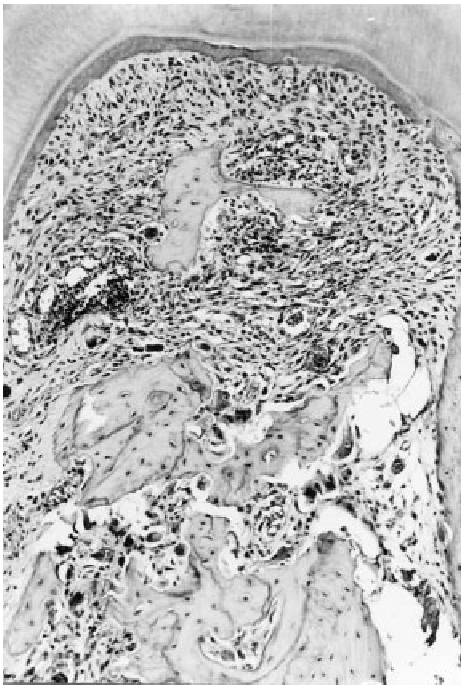

Fig. 4. Interradicular zone of a case from Group 6 (51-g orthodontic force applied immediately after removal of the cervical ligature used to induce periodontitis). Erosion areas and osteoclasts can be observed in the interradicular bone, both on the periodontal bone surface and in the medullary spaces. Note the presence of bone loss. The periodontal ligament exhibits granulation tissue presenting less inflammatory infiltrate than that observed in Group 4 (hematoxylin and eosin 10 ×).

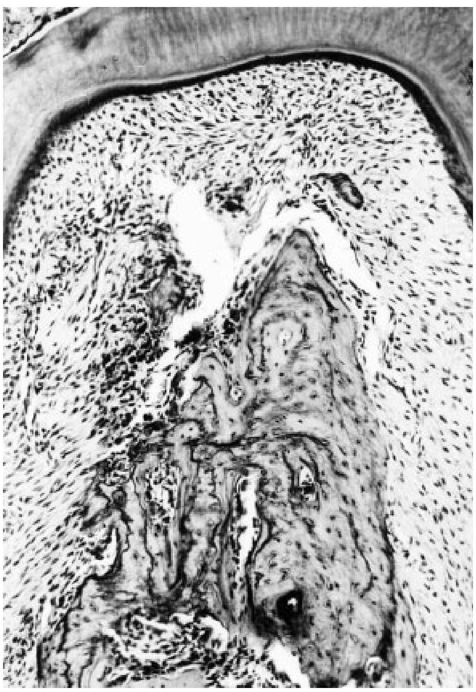

Fig. 5. Interradicular zone of a case from Group 8 (51-g orthodontic force applied 48 h post-removal of the cervical ligature used to induce periodontitis). Inflammatory infiltrate in the periodontal ligament is negligible. The interradicular bone presents few erosion areas, which are localized on the periodontal surface of the crest. Note the marked density of the interradicular bone (bone volume) (hematoxylin and eosin 10 ×).

(14), in other words serving as mechanosensor and mechanotransductor (14, 16, 17). A mechanism of this sort may account for the increase in bone volume observed in animals with periodontitis-associated bone loss and subjected to orthodontic forces. It must be pointed out that in histomorphometric terms, bone volume indicates the trabecular density of bone.

The slight variation in bone volume observed between animals with periodontitis and those subjected to orthodontic forces immediately upon withdrawal of the ligature may occur because the signals triggered by the force that stimulate bone formation are masked by the signals triggered by the inflammation that stimulate bone resorption. It has been demonstrated that resorption of alveolar bone during periodontitis is associated with inflammatory cell release of different cytokines, such as interleukin-1 (IL-1), IL-3, IL-6, α and β tumor necrosis factor, and prostaglandin E2 (18, 19). These factors activate osteoclasts and inhibit osteoblasts, resulting in progressive loss of alveolar bone. However, the intensity of the force also plays an essential role. The lighter 51-g force used in our study increased bone volume even under conditions of inflammation.

Application of the forces 48 h post-removal of the ligature resulted in increased bone volume compared to animals subjected only to periodontitis. This increase in bone volume may have taken place because the bone remodeling signals were triggered by the force once the inflammation was under control. Thus, the concentration of bone resorption stimulating factors released by immunologic cells had decreased, and the bone was able to respond adequately to the modeling and remodeling mechanisms and adapt its structure to the functional requirements.

Based on our results and on the studies in the literature reporting the clinical observation of patients successfully treated with combined periodontal and orthodontic therapy (1–3), we recommend initiating orthodontic therapy of periodontal patients once

Table 2. Histomorphometric determinations

	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6	Group 7	Group 8	Group 9	$*p < 0.05$
BR%	9 ± 7	75 ± 14	67 ± 10	30 ± 22	61 ± 17	89 ± 19	78 ± 19	74 ± 23	62 ± 41	4 vs. 1,2, 3, 6, 7, 8, 9
BL%	8 ± 1	12 ± 3	16 ± 6	24 ± 5	10 ± 3	15 ± 6	23 ± 7	24 ± 5	19 ± 9	4 vs. 1, 2, 5
BV	0.69 ± 0.04	0.66 ± 0.23	0.53 ± 0.11	0.33 ± 0.07	0.33 ± 0.1	0.57 ± 0.22	0.25 ± 0.15	0.44 ± 0.17	0.51 ± 0.21	4 vs. 1, 2, 3, 6, 9

BR: bone resorption. Percentage of bone resorption surfaces on the vestibular aspect of the interradicular bone found in the interradicular space.
BL: bone loss. Percentage of the full height of the interradicular space corresponding to the periodontal space, measured from the root furcation to the interradicular bone crest.
BV: bone volume. Fraction of total bone volume corresponding to trabecular volume, measured in the interradicular bone, found in the cervical third of interradicular space.
± standard deviation.
$*p < 0.05$ Bonferroni's test.

the inflammation has been controlled. Our experimental histomorphometric results lend support to this recommendation, given that the increase in bone volume is greater when forces are applied once the periodontal inflammation is under control and remnant bone quality is consequently enhanced.

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References

1. Melsen B, Agerbaek N, Markenstam G. Intrusion of incisors in adult patients with marginal Browe-Loss. *Am J Dento Facial Orthop* 1989;**96:**232–241.
2. Polson AM, Subtelny JD, Meitner SW *et al.* Long term periodontal status after orthodontic treatment. *Am J Dento Facial Orthop* 1988;**93:**51–58.
3. Vanarsdall R, Musich D. Adult orthodontics: diagnosis and treatment. In: Graber TM, Swain BF, eds. *Orthodontics; current principles and techniques.* St. Louis: C.V. Mosby Co, 1992: 791–856.
4. Ericsson I, Thilander B, Lindhe J, Okamoto H. The effects of orthodontic tilting movements on the periodontal tissues of infected and non infected dogs. *J Clinic Periodontol* 1977;**4:**278–293.
5. Carranza FA Jr, Simes RJ, Mayo J, Cabrini RL. Histometric evaluation of periodontal bone loss in rats. I. The effect of marginal irritation, systemic irradiation and trauma from occlusion. *J Periodont Res* 1971;**6:**65–72.
6. Ubios AM, Costa OR, Cabrini RL. Early steps in bone resorption in experimental periodontitis: a histomorphometric study. *Acta Odontol Lationoam* 1993;**7:**45–50.
7. Shoji K, Horiochi H, Shinoda H. Inhibitory effects of biphosphonate (risendronate) in experimental periodontitis in rats. *J Periodont Res* 1995;**30:**277–284.
8. Mandalunis PM, Costa OR, Ubios AM. Dynamics of bone loss in experimental periodontitis. *Acta Odontol Latinoam* 1998;**11:**27–35.
9. Mitsuta T, Horivich H, Shinoda H. Effects of topical administration of chlordronate on alveolar bone resorption in rats with experimental periodontitis. *J Periodontol* 2002;**73:**479–486.
10. Alencar VB, Bezerra MM, Lina V, *et al.* Disodiumchlodronate prevents bone resorption in experimental periodontitis in rats. *J Periodontol* 2002;**73:**251–256.
11. Mandalunis PM, Steimetz T, Castiglione JL, Ubios AM. Alveolar bone response in an experimental model of renal failure and periodontal disease: a histomorphometric and histochemical study. *J Periodontol* 2003;**74:**1803–1807.
12. Steimetz T, Mandalunis PM, Ubios AM. Effects of compressive forces on a bone modeling surface. *Acta Odontol Latinoam* 1997;**10:**111–115.
13. Weibel ER, Kistler GS, Scherle WF. Practical stereological methods for morphometric cytology. *J Cell Biol* 1966;**30:**24–37.
14. Warshawsky NM, Su M, Broke JL, *et al.*Expression of conexin 43 in rat mandibular bone and periodontal ligament cells during experimental tooth movements. *J Dent Res* 1997;**76:**1357–1360.
15. Mullender MG, van Rietbergen B, Riiegsegger P, Huiskes R. Effects of mechanical control set point of bone cells on mechanical control of trabecular bone architecture. *Bone* 1998;**22:**125–131.
16. Bozal CB, Fiol JA, Ubios AM. Early osteocyte response to bone resorption stimuli. *Acta Odontol Latinoam* 2001; **14:**24–29.
17. Noble BS, Reeve J. Osteocyte function, osteocyte death and bone feature resistance. *Mol Cell Endocrinol* 2000;**159:**7–13.
18. Schwartz Z, Goultschin J, Dean DD, Boyan BD. Mechanisms of alveolar bone destruction in periodontitis. *Periodontol 2000* 1997;**14:**158–172.
19. Baker P. The role of immune responses in bone loss during periodontal disease. *Microbes Infect* 2000;**2:**1181–1119.