BJOS
Brazilian Journal of Oral Sciences

Volume 16
2017
e17090

Original Article

# Root morphology can be a risk factor for periodontal damage and root resorption in orthodontic movement

Ewerton Zaniboni[1], Mário Vedovello Filho[2], Mauro Pedrine Santamaria[3], Maria Aparecida Neves Jardini[4], Maria Fernanda Martins-Ortiz[5], Alberto Consolaro[6], Milton Santamaria Jr[7*].

[1] DDS. School of Dentistry, Heminio Ometto University Center, UNIARARAS. Dr. Maximiliano Baruto, 500. Araras-SP. 13607-339, Brazil.taozaniboni@hotmail.com

[2] PhD. School of Dentistry, Heminio Ometto University Center, UNIARARAS. Dr. Maximiliano Baruto, 500 Araras-SP 13607-339, Brazil mario@vedovelloeassociados.com.br

[3] PhD. Division of Periodontics, School of Dentistry, State University of São Paulo, UNESP. Av. Eng. Francisco Jose Longo, 777. São José dos Campos – SP. 12245-000, Brazil mauro.santamaria@fosjc.unesp.br

[4] PhD. Division of Periodontics, School of Dentistry, State University of São Paulo, UNESP.Av. Eng. Francisco Jose Longo, 777. São José dos Campos – SP. 12245-000, Brazil mauro.santamaria@fosjc.unesp.br

[5] PhD. Department of Oral Pathology, Bauru Dental School, University of São Paulo, USP. Alameda Otávio Pinheiro Brisolla, 9-75. Vila Universitária. Bauru-SP. 17012-901, Brazil. mfmartinsortiz@gmail.com

[6] PhD. Department of Oral Pathology, Bauru Dental School, University of São Paulo, USP. Alameda Otávio Pinheiro Brisolla, 9-75. Vila Universitária. Bauru-SP. 17012-901, Brazil.consolaro@uol.com.br

[7] PhD. School of Dentistry, Heminio Ometto University Center, UNIARARAS. Dr. Maximiliano Baruto, 500 Araras-SP 13607-339, Brazil santamariajr@ig.com.br

**Corresponding author:**
Milton Santamaria Jr
Heminio Ometto University Center, UNIARARAS. Dr. Maximiliano Baruto, 500 Araras-SP
13607-339, Brazil

**Aim:** The study evaluated, using histomorphometry, the percentage of hyaline area in periodontal ligament (PDL) and root resorption in orthodontic tooth movement (OTM). **Methods:** Ten rats were divided into two groups. G3 Group (n=5), with 3 days of OTM and G7 Group (n=5), with 7 days of OTM. A Control Group (n=5) consisted of contralateral teeth of each animal, which were not moved. Maxillary left first molar was moved, using stainless steel spring connected to the incisors with 40g force. Microscopic analysis was done in transversal sections of the mesiovestibular (MV) and distovestibular (DV) roots in the cervical level. **Results:** There was a PDL hyaline area in the DV root of 6.2% in G3 and 1.8% in G7. The root resorption area in G7 was 0.9%. On MV root and Control Group were not found occurrences of hyaline areas in PDL and no root resorption. **Conclusions:** Based on the results obtained, it might be concluded that smaller roots showed higher frequency of hyaline areas and root resorption.

**Keywords:** Tooth movement. Periodontal ligament. Root resorption. Orthodontics.

**Received:** January 14, 2017
**Accepted:** November 23, 2017

## Introduction

The complexity of events in orthodontic tooth movement (OTM) involves forces of compression and traction on the periodontal ligament (PDL) and alveolar bone, inducing morphological and microscopic reactions controlled by cytokines and growth factors, promoting tooth displacement[1-4].

In orthodontics, a mechanical stimulus induces tooth movement[5-6]. The orthodontic force causes an imbalance in PDL, an inflammatory process[7-8] where cytokines produced by osteoclasts, osteoblasts and osteocytes control bone remodeling around PDL[9-10].

Experimental studies allow clinical and microscopic evaluation of the tooth movement biology and root resorption. Different aspects may interfere in the effectiveness of OTM, such as the intensity and type of force applied, local and systemic diseases, bone alterations and the root morphology[11].

When the PDL is overly compressed, it can cause cell death, creating a hyaline area, cementoblast layer damage and root resorption[12]. Hyaline areas in PDL will delay tooth movement and will facilitate the presence of root resorptions[13-15].

Root resorption during OTM is a frequent phenomenon, but must not be considered normal or physiological. In some orthodontic treatments, apical root resorption is inevitable, but predictive factors can be defined[12-14]. It will depend on the magnitude, duration and type of force, which may cause many degrees of root resorption[16-18] in different root morphologies[17,19,20].

This study proposes to demonstrate the microscopic aspects of tooth movement in rats, by analyzing the hyaline areas in PDL and root resorptions in OTM, on roots with different types of morphology.

## Methods

### Sampling

The procedures of this research were performed in compliance with the ethical and legal recommendations specified by the Animals Ethics Commission (CEUA) of Fundação Hermínio Ometto - Uniararas (Report nº 030/2013).

The research used *Wistar* rats (Rattus norvegicus, albinos), male, three month old. They were kept at temperature of 25ºC, in plastic cages and were provided food (Nuvilab, Quimtia S.A, Colombo/PR, Brazil) and water *ad libitum*.

Ten animals were divided into three groups. Group G3 (n=5): three days of orthodontic tooth movement (OTM), Group G7 (n=5): seven days of orthodontic tooth movement (OTM) and Control Group (n=5): contralateral teeth not moved of each animal.

### Orthodontic movement

The animals were anesthetized with three parts of ketamine hydrochloride, 100mg/ml (Cetamin, Syntec do Brasil Ltda, Cotia/SP, Brazil) one part of the xylazine hydrochloride, 20mg/ml (Xilasin, Syntec do Brasil, Cotia/SP, Brazil) at the dose of 1ml/Kg, applied intramuscularly.

A closed stainless steel spring was placed between the maxillary left first molar (point of force application) and the maxillary incisors (point of anchorage), tipping forward the first molar, applying 40gf (Figure 1)[21].

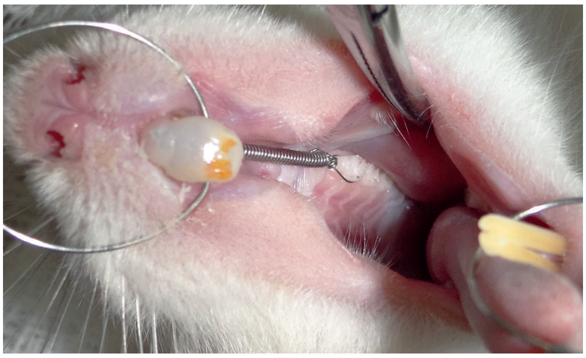

Figure 1. Device used in the movement of the first molar. Spring installed between the first molar and the incisors.

### Quantitative Histomorphometric Analysis

The animals were euthanized with an overdose of the anesthetic ketamine and xylazine mixture. The maxillae were placed in a 10% buffered formol solution for fixation during 2 days. They were demineralized with EDTA (monobasic sodium phosphate 4.4g; dibasic sodium phosphate 45g; EDTA 70g; Deionized $H_2O$, 1000 ml; Formaldehyde PA 50 ml) in a period of eight weeks.

The maxillae were embedded in paraffin and cut in transversal cross sections (5µm), stained with hematoxylin-eosin and the roots were analyzed in the cervical level[22]. The mesiovestibular (MV) and distovestibular (DV) roots were analyzed in optical microscope with an objective with 10x magnification (Zeiss KS 300, version 3.0). Five sections per animal were used for histomorphometrical quantification (Figure 2).

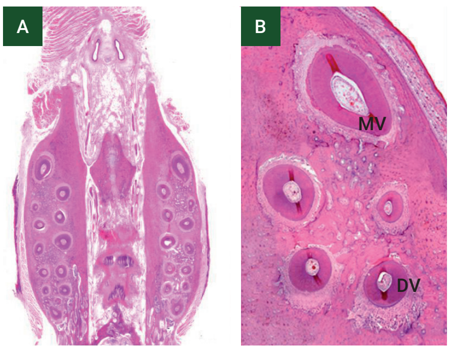


Figure 2. A - Microscopic cross section of the jaw (Staining - HE, magnification-4x). B - Microscopic cross section of the upper first molar, occlusal view. MV - mesiobuccal root. DV - distobuccal root (Staining - HE, magnification-10x).

Division of hyaline area by the total PDL area gives the percentage of periodontal hyaline area (Figure 3). The percentage of root resorption was calculated at the same way, with the division of resorption area by the total root area (Figure 4).

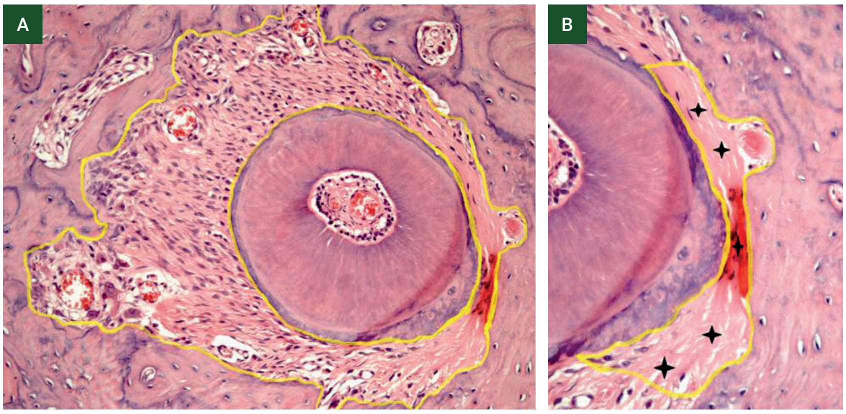


Figure 3. Cross section of distobuccal root (DV) with 3 days of orthodontic movement. A - Section area of the PDL. B - * Periodontal hyaline area (Staining - HE, magnification-40x).

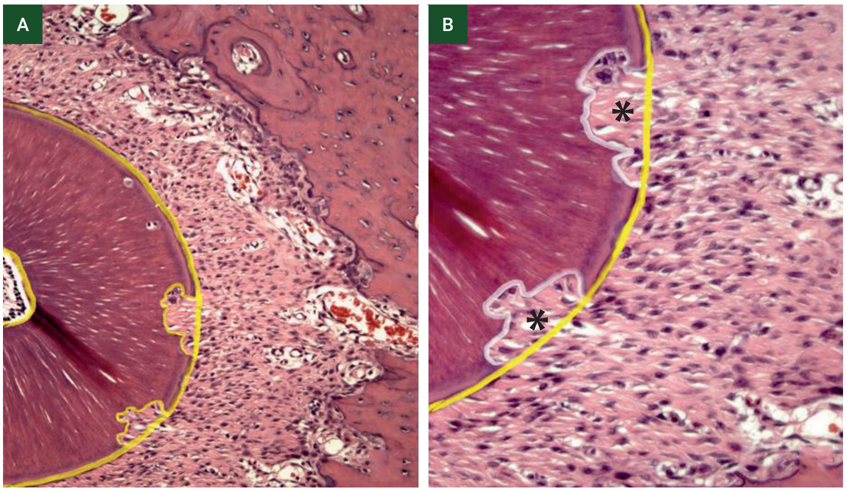


Figure 4. Cross section of distobuccal root (DV) with 7 days of orthodontic movement. A - Section area of the root, including cementum and dentin (Staining - HE, Lens - 4x). B - * Root resorption area (Staining - HE, magnification-40x).

The frequency of root resorption events was also verified in each animal by total in group (n=5) in the different periods of OTM and roots.

The mean of hyaline area percentage of PDL and percentage of root resorption were compared by ANOVA and the Tukey post-test ($p<.05$).

## Results

Hyaline area percentage in PDL and root resorption percentage, according to the OTM period and analyzed root are presented in Table 1.

Table 1. Mean of hyaline percentage area of PDL and percentage of root resorption, found in each group, according to the OTM period and root analyzed.

| Root | Hyaline area (%) | | | Root resorptions area (%) | | |
|---|---|---|---|---|---|---|
| | Control | 3 days | 7 days | Control | 3 days | 7 days |
| MV | 0 | 0 | 0 | 0 | 0 | 0 |
| DV | 0 | 6.2* | 1.8* | 0 | 0 | 0.9* |

* Statistically significant compared to the Control Group on each root analyzed ($p \leq 0.05$).
MV - mesiovestibular root.
DV - distovestibular root.

The MV root presented no segmental hyaline areas in PDL. There were hyaline areas in OTM Groups of the DV root, and were statistically significant in comparison of the Control and MV Groups ($p \leq 0.05$). The hyaline areas were greater in 3rd day (mean of 6.2%) than in 7th day (mean of 1.8%). On MV root, there was no root resorption. On the DV root resorption appeared in the 7th period of OTM (mean of 0.9%).

The frequency of root resorption occurred in the period of 7 days of OTM. This frequency of root resorption was 3:5 (60% of the animals) in the DV root (Table 2).

Table 2. Frequency of root resorption (animals by total in group), according to the OTM period and root analyzed.

| Root | Control | 3 days | 7 days |
|---|---|---|---|
| MV | (0:5) 0% | (0:5) 0% | (0:5) 0% |
| DV | (0:5) 0% | (0:5) 0% | (3:5) 60% |

## Discussion

Animals as experimental models allows clinical reproducibility[22]. This *in vivo* research provides relevant data on physiological and pathological conditions that may be useful for establishing more effective clinical interventions[23]. Orthodontic tooth movement (OTM) causes resorption of alveolar bone on the compression side and osteogenesis on the tension side. Studies investigating the mechanisms involved in this process are important to improve orthodontic treatment[24].

This OTM experimental design[21], tips forward the first molar, without interfering on the rat craniofacial structure. It is efficient to study bone remodeling and root resorptions in Orthodontics. This model allowed studies such as auxiliary therapies[25] and drug interference[26] on bone remodeling in orthodontic movement.

Transversal sections give a direct view of the alveolar bone between the roots, and also allow evaluation of the cortical bone in the same section. Thus, all the structures could be analyzed simultaneously, and compared with the same structures on the contralateral control side, without OTM, being an important control of the biologic reactions found. In the longitudinal sections it is not possible to visualize all the roots at the same time and neither to simultaneously analyze the different anatomic regions, such as cortical and medullary bone[27].

Mesiovestibular (MV) and distovestibular (DV) roots are exposed to moderate forces and intense forces, respectively, during OTM. The root morphology has an influence on the orthodontic force intensity and PDL tissue reaction, and in the production of root resorptions.

In this research, two roots of the rat first molar were compared and evaluated simultaneously. The MV root, bigger, presented no root resorption. Bigger roots with larger dimensions present better distribution of the applied forces. This fact was confirmed given that no hyalinization of the PDL was found in the MV root in the OTM groups. The presence of hyalinized tissue within the PDL is a microscopic sign of excessive compression, resulting from the intense application of forces.

When these forces are intense and prolonged, the cells that line the tooth roots, the cementoblasts are injured. The cementoblasts protect the root surface, because they do not have receptors for the mediators that participate in bone remodeling[12-14]. The majority of external root resorptions, as an initial phenomenon, present large destruction of the layer of cementoblasts, denuding the mineralized dentin surface and exposing it to the action of the bone remodeling cells[12-14]. Therefore, excessive orthodontic forces may result in undesired root resorptions.

Root resorption could be seen with abundance in the DV root, in the period of 7 days of movement, in which the percentage and the frequency were greater than in the MV root. This result is explained because its morphology is smaller in comparison with those of the MV root. On the 3$^{th}$ day of movement, there was clear evidence of the high incidence of forces on the mesial surface of this DV root, with the presence of large segmental PDL hyaline areas.

The results demonstrated the anatomic influence on root resorptions. The DV root dimension is smaller, with its conical shape, as in a single-rooted tooth, promoting greater pressure and less distribution of forces on PDL walls. Thus, smaller roots may present evident effects, such as root resorption and hyaline area, because they have poor ability to dissipate compressive force during tooth movement.[20]

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