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<th>Force Magnitude and Duration Effects on Amount of Tooth Movement and Root Resorption in the Rat Molar</th>
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ABSTRACT

Objective: To test the hypothesis that there is no difference in the effect of different continuous moderate to very heavy forces on root resorption or amount of tooth movement.

Materials and Methods: In the study, 10, 25, 50 and 100 g mesial force were applied to the maxillary first molars of rat using nickel titanium closed-coil springs for 3 days, 14 days, and 28 days. The molars were extracted and the surface areas of the root resorption craters were measured using scanning electron microscope. The depths of the root resorption craters were measured using a three-dimensional laser scanning microscope. Tooth movement of the maxillary first molar was measured in relation to the maxillary second molar on digitized lateral cephalometric radiographs.

Results: Three days after force application, the tooth movement was not proportionally related to force magnitude. However, 14 days of force application resulted in significantly more tooth movement in the 10, 25, and 50 g force groups than in the 100 g force group. A force application of 10 g produced significantly more tooth movement at 28 days than all the other three force applications. The largest and deepest resorption craters were observed in the disto-buccal root followed by disto-palatal, middle-buccal, middle-palatal, and mesial root. Root resorption and tooth movement increased over time from 3 to 28 days. As heavier forces were applied, greater root resorption occurred.

Conclusion: The hypothesis is rejected. The light mesially oriented forces, as applied in this study, produced more tooth movement and less root resorption compared with heavier forces.

KEY WORDS: Root resorption; Heavy force; Depth; Area; Laser scanning microscope; SEM
heavy force magnitudes (10–100 g) and root resorption as well as to identify the location of the resorption sites, area, and depth on the five roots of the rat's upper first molars.

MATERIALS AND METHODS

Sixty 10-week-old male young adult Wistar rats (SLC, Shizuoka, Japan; body weight, 230–250 g) were used as experimental animals. The study was conducted under approval from the Animal Welfare Committee of Nagasaki University. The rats were divided into 12 groups (5 rats each) according to the magnitude and duration of the applied force. Three time groups were set at 3, 14, and 28 days. Nickel titanium (NiTi) closed-coil springs of 10, 25, 50, and 100 g (Figure 1A; Sentalloy, Tomy Inc, Fukushima, Japan) were used to move the maxillary left molar mesially. Contra lateral molars served as controls. The appliance was set under anesthesia (intraperitoneal injection of pentobarbital) with a dosage of 60 mg/kg body weight.

A NiTi closed-coil spring with an active length of 3 mm was inserted between the first and the second upper left molar as described previously. A coil spring was activated by pulling to a triple deflection (9 mm) (Figure 1A–D). This method of fixing the coil spring through incisors and alveolar bone suppresses tooth eruption and inhibits the tendency of the appliance to come loose. The force magnitude was measured at triple-spring length extension after setting back from four times extension. A tension gauge (DTN-30, -150, Teclock, Tokyo, Japan) was used in a water bath at 38°C. The procedure was accomplished when the appliance was set and at the end of the experimental time. The force magnitudes of 50 g coil spring measured before and after the experiment for 14 days were 56.1 ± 2.3 g and 60.6 ± 6.2 g, respectively (n = 10). At the end of the experiments the animals were sacrificed by an overdose of CO2.

To measure tooth movement, lateral cephalometric radiographs were taken. A cephalostat was specially constructed to standardize the rat's head position (Figure 1E and G). The distance between the X-ray tube (CMB-2, Softex, Kanagawa, Japan) and film was 50 cm. The cephalometric radiographs were digitized with a film scanner at 600 dpi. Tooth movement was measured on the digitized images with Scion Imaging software (Scion Corp, Frederick, Md).

The amount of tooth movement was determined by the change in the distance between the most posterior point of the posterior border of maxillary first molar crown and the most anterior point of the anterior border of the maxillary second molar crown during an experimental period (Figure 1F). To verify the accuracy and reproducibility, the rat was placed in the cephalostat under anesthesia and the measurement of amount of tooth movement was repeated 20 times. The value of rat tooth movement for 14 days by a force of 25 g was 44.4 ± 4.3 μm, and the range was ± 6 μm.

After the experimental tooth movements, the upper first molar, including its surrounding bone, was cut as a whole block, followed by delicately removing the alveolar bone to avoid any root surface damage. The resected molar was submerged in 1% sodium hypochlorite for 10 minutes to eliminate remaining periodontal ligament remnants. The five roots of a rat's upper molar were divided into three parts using a diamond disc: mesial root, middle roots (middle buccal and middle palatal), and distal roots (disto-buccal and disto-palatal) (Figure 1H, I, and J).

The mesial surfaces of the roots were scanned with an SEM (TM-1000, Hitachi, Tokyo, Japan). The area of the resorption craters was measured by means of commercial software (Mimics program, DICO, Tokyo, Japan), and the deepest point of the root resorption craters was evaluated with a three-dimensional (3D) laser scanning microscope (VK-9500, Keyence, Kyoto, Japan) (Figure 2). The same investigator performed all measurements, and every measurement was repeated three times. The mean value was used as the final measurement. The Mann-Whitney test was used to compare pairs of groups.

RESULTS

Tooth Movement

Tooth movement was not force sensitive before 14 days. However, at the end of 28 days, 10 g of light force application produced significantly more tooth movement compared with heavier force application. The amount of tooth movement increased significantly from 0 to 28 days in all force level groups (Figure 3).

In the 3-day experimental period the greatest amount of tooth movement (0.16 mm) was observed when 50 g of force was applied. In the 14-day period animals the 100-g group showed the least amount of tooth movement (0.20 mm). Finally, in the 28-day period the greatest amount of tooth movement (0.79 mm) was observed when the 10 g coil spring was used compared to the amount of tooth movement obtained when 25 g (0.65 mm), 50 g (0.64 mm) and 100 g (0.66 mm) coil springs were used (P < .05).

Observation by SEM

Most of the control roots exhibited areas covered by undamaged cementum with a characteristic smooth surface (Figure 4). The apical half of the roots was covered with thick cementum with a rough surface that, in some cases, contained resorption craters.
Figure 1. (A) Load-deflection curve of 3 mm nickel-titanium closed-coil springs (10, 25, 50, 100 g) measured by a loading cell of 0.5 kgf (Model-1350D and -1012, Aikoh Engineering, Osaka, Japan) in a chamber at 37°C. The extension range between 8 and 9 mm was used in this study. (B) Intraoral picture of the appliance. (C) Schematic representation of the appliance set on the rat's dry skull, occlusal view. (D) Lateral view of the appliance showing the location of the hole made on the alveolar bone and the incisors. (E) A cephalostat. (F) Lateral cephalometric radiograph showing a 10-g closed-coil spring after 28 days of force application. The arrow indicates the measurement portion.
Figure 2. Mesial view of the mesial-buccal root after 25 g of force application for 28 days. (A) Scanning electron micrograph (×60); (B) and (C) Three-dimensional image obtained with the laser scanning microscope; (D) A graph of the depth of resorption pit on which the deepest point of the crater was measured.

Root resorption craters with well-defined margins were observed in the mesial surface of all the roots in the 14- and 28-day group (Figure 4). Three different types of craters were clearly identified: isolated lacunae, wide shallow resorption pit, and deep resorption craters. Small isolated lacunae were mainly seen scattered on the mesial roots (cervical half of its mesial surface). Wide shallow and deep resorption craters were observed mostly on the middle and distal roots (mesial surface) of all experimental roots. Similar to the control group, the 3-day experimental group showed no clear resorption craters.

Resorption Area Measured from SEM Image

The resorption area varied in size from one root to another (Figure 5). The largest resorption area was found in the disto-buccal root followed by the disto-palatal root, the middle-buccal root, middle-palatal root, and mesial root for both the 14- to 28-day groups. Because of its small size, the middle-buccal root was almost destroyed when a 100-g force was applied for 28 days. The predominant location of resorption craters was noted in the cervical half of the mesial and between upper first and second molars. (G) Design of cephalostat; left, a view from tail side; right, a view from abdominal side. (H) Buccal view (left) and palatal view (right) of upper right first molar. (I) Apical view and cutting lines (upper) and separated parts (lower). (J) Mesial views of mesial root (left), middle roots (center, middle-buccal and middle-palatal) and distal roots (right, disto-buccal and disto-palatal). m indicates mesial; b, buccal; d, distal. A white bar indicates 2 mm of length.
Figure 4. Scanning electron micrograph (×60) of the upper right left molar, mesial view. M indicates mesial root; MB, middle-buccal root; MP, middle-palatal root; DB, disto-buccal root; DP, disto-palatal root. Control, 14-day, and 28-day experimental groups are shown. White arrow heads indicate resorption craters.
ROOT RESORPTION AND MAGNITUDE OF HEAVY FORCES

middle roots. On the distal roots (buccal and palatal), the resorption craters extended over the cervical and part of the apical halves of the roots. These findings were similar in the 14- and 28-day experimental period (10 g, 25 g, 50 g, and 100 g).

Root resorption was dependent on force magnitude in all the groups except for the mesial root (14- and 28-day group), and the middle palatal root (28-day group). Time-related root resorption was clearly seen in all roots \( P < .05 \) except for the mesial root with 10, 25, 50 g, and the middle palatal root with 100 g (Figure 5). The 10-g force group demonstrated lesser amount of root resorption compared with the heavier force level groups.

**Resorption Depth Measured by 3D Laser Scanning Microscope**

The deepest resorption craters were found in the disto-buccal root for both experimental periods (14- and 28-day groups). The depth of resorption craters decreased gradually in the following order: middle-buccal root, disto-palatal root, middle-palatal root, and finally mesial root (Figure 6). The depth of resorption craters was mostly dependent on the magnitude of force in all the roots except for the mesial roots.

**DISCUSSION**

In this study, we studied the effect of various magnitudes of force and different durations of force application on root resorption. Since the human molar is approximately 20 times larger than a rat molar,\(^1\) the coil springs of 10, 25, 50, and 100 g used in a rat should correspond to 200, 500, 1000, 200 g in a human molar. 2000 g of force is comparable to a rapid expansion screw that produces 1300–4500 g by a single activation (0.25 mm).\(^2\) though the force produced by expansion screw is different from a simple pulling force in this study. Our results showed that 3 days after force application, the tooth movement was not related proportionally to the force magnitude. However, 14 days of force application resulted in significantly more tooth movement in the 10, 25, and 50 g force groups than in the 100-g force group. When a 10-g closed-coil spring was used, tooth movement was achieved significantly greater than 25, 50, and 100 g after 28 days, but root resorption area and depth were the least. On the other hand, root resorption was obviously dependent on the force magnitudes up to 100 g of super heavy force.

Although there are many reports about force magnitude relating to tooth movement or root resorption, comprehensive studies including force magnitude, tooth movement, and root resorption are limited. Ideal tooth movement is described as maximum amount of tooth movement with minimum pathologic side effects and minimum patient discomfort.\(^3\) Our study showed that 10 g of light force application produced significantly larger tooth movement with significantly less root resorption over a period of 28 days in relation to heavier force application in rats.

There are many reports about the effect of force magnitude on tooth movement. Kohno et al\(^4\) examined the rate of tooth movement under light orthodontic forces of 1.2, 3.6, 6.5, and 10 g of force for 14 days to move rat molars. They showed tooth movement depended on the force magnitudes of this range. King et al\(^5\) demonstrated that the effective tooth movement of rat molars ranged from 20 to 40 g, and its velocity did not increase over 40 g. Many investigators have reported that root resorption was aggravated by increasing force magnitudes.\(^6\) Although there have been some contradictory reports that heavy forces did not increase root resorption,\(^7\) heavy force is generally considered to be harmful, and our results strongly support this. Collectively, the optimum force for the

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\(^{1}\) Reference 1
\(^{2}\) Reference 2
\(^{3}\) Reference 3
\(^{4}\) Reference 4
\(^{5}\) Reference 5
\(^{6}\) Reference 6
\(^{7}\) Reference 7
The reason why root resorption in each of the five roots appeared quite different is still unknown. One possible factor could be the distribution of mechanical stress on different root and alveolar bone surfaces. For instance, the mesial root of the maxillary first molar grows obliquely to the occlusal plane, whereas the rest of the roots grow perpendicular to it. Also, the sizes of the roots differ from each other.

From a mechanical point of view, the first reaction to the application of an orthodontic load is an alteration in the strain-stress distribution within the periodontal ligament and the surrounding alveolar bone. Tooth movement in our rat model consisted of a mesial tipping, extrusion, and rotation of the maxillary first molar. The compressed zone appeared inter-radicularly below the crown on the mesial side of the distal roots (buccal and palatal roots) and on the cervical half of the middle and mesial roots (mesial sides). To elucidate further details of the resorption pattern in this study model, we are progressing with an investigation involving 3D tooth movement and a finite element method analyses.

CONCLUSION

- Light mesially oriented 10 g forces on rat molars, as applied in this study, produced more tooth movement and less root resorption than 25, 50, and 100 g of heavier force applications.

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