Original Article

Comparison of the effects of three surgical techniques on the rate of orthodontic tooth movement in a rat model

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ABSTRACT

Objective: To evaluate the effect of corticotomy and corticision, with and without a full mucoperiosteal flap, on the rate of tooth movement and alveolar response in a rat model.

Materials and Methods: Sixty male, 6-week-old Wistar rats were divided into five groups based on surgical procedure, as follows: control (no tooth movement), orthodontic tooth movement (OTM) only, corticotomy, corticision, and corticision with full mucoperiosteal flap (corticision + flap). A force of 10–15g was applied from the maxillary left first molar to the maxillary incisors using nickel-titanium springs. Surgery was performed at the time of appliance placement (day 0), and tooth movement occurred for 21 days. Micro–computed tomography was performed on day 21 to evaluate the amount of tooth movement and alveolar bone parameters. Histomorphometry, including tartrate-resistant acid phosphatase staining, was performed to quantify the osteoclast parameters at day 21.

Results: No statistical differences in the amount of OTM, bone volume fraction, and tissue density and the osteoclast parameters were found among all experimental groups.

Conclusions: Corticotomy and corticision, with or without a full mucoperiosteal flap, did not show a significant effect on either the OTM magnitude or alveolar bone response. (*Angle Orthod.* 2017;87:717–724.)

KEY WORDS: Corticotomy; Corticision; Mucoperiosteal flap; Orthodontic tooth movement; Accelerated tooth movement

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INTRODUCTION

Rate of tooth movement limiting factors include bone turnover, bone density, and the degree of hyalinization of the periodontal ligament (PDL).¹ Surgical techniques have been used to modulate these biological processes. These modalities take advantage of the regional acceleratory phenomenon (RAP), in which a noxious stimulus to the bone can accelerate bone turnover and reduce regional bone density, leading to a transient osteopenia. The stimulus and RAP have been said to be proportional: that is, the larger the noxious stimulus, the greater the RAP.² Corticotomy and corticision are surgical procedures, with distinct degrees of invasiveness, shown in the literature as common techniques used to induce RAP and increase orthodontic tooth movement (OTM).

Corticotomy involves a full mucoperiosteal flap and fissures made through the buccal and/or lingual plates that surround the bone.³ Corticision uses a reinforced scalpel as a thin chisel to separate the interproximal cortices without raising a full flap.⁴ Studies^{5,6} have shown that both of these procedures can be efficient

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Figure 1. Corticotomy with a full-thickness mucoperiosteal flap and three .25 \times .25-mm holes were placed in the cortical bone with a No. $\frac{1}{4}$ round bur 5 mm mesial to the left maxillary first molar.

means of stimulating tooth movement by inducing RAP and accelerating bone remodeling.

A mucoperiosteal flap could induce ischemic events and cause the medullary bone to respond to the injuries, providing cells and calcium and other minerals for repair. Subsequently, this could cause a RAP response, with increased bone turnover and decreased bone density.^{7,8} Thus, elevating a full flap in conjunction with a surgical procedure would increase the trauma to the bone and increase the RAP response and possibly increase the rate of OTM.

Ruso et al.⁹ performed piezocision without a flap on the buccal alveolus in foxhound dogs and demonstrated that the piezocision group had significantly more OTM and less dense and mature bone than the control. However, the authors stated that perhaps the RAP effect would have been more marked if a flap had been elevated. In fact, Swapp et al.7 demonstrated that a flapless decortication procedure did not affect the rate of OTM in foxhound dogs when the third premolars were mesialized. It was suggested that to increase the RAP process, more damage may need to be created, and elevating a flap would induce the response needed. Therefore, the purpose of this study was to assess the effect of corticotomy and corticision, performed with and without a full mucoperiosteal flap, on the amount of tooth movement and changes in the

Figure 2. Corticision procedure: Corticision without a full mucoperiosteal flap procedure in the mesial-palatal and distopalatal gingiva adjacent to the left maxillary first molar.

alveolus in a rat model when a constant force was applied for 21 days.

MATERIALS AND METHODS

Sixty male, 6-week-old (150-250 g) Wistar rats (Charles River Laboratories, Charles River, Wilmington, MA) were used for the experiments. All experiments were performed under an institutional approved protocol for animal research. The rats were randomly placed into five groups, which included the following: (1) control group (control), with no force or tooth movement (n = 5); (2) no surgical procedure and orthodontic spring (OTM) delivering 10-15g of force (n = 14); (3) corticotomy (corticotomy) and orthodontic force (n = 14); (4) corticision (corticision) and orthodontic force (n = 14); and (5) corticision with a full mucoperiosteal flap (corticision + flap) and orthodontic force (n = 13). A procedure per experiment was assigned to a group of five to eight rats in random order until all the groups were completed. A smaller number of rats was used for the control group, as less variation in the evaluated parameters was expected. Minimums of 11 rats per group were used based on a sample size calculation gathered from an initial analysis of five animals per group for differences in intermolar distance. The application of orthodontic



Figure 3. Micro-CT image and the region of interest for bone parameter measurements (shaded) tissue density and bone volume fraction. M indicates mesial; MB, mesiobuccal; MP, mesiopalatal; DB, distobuccal; and DP, distopalatal roots.

force and the surgical procedure were performed only at day 0, and OTM was performed for 21 days.

Animals were placed under general anesthesia with xylazine (13 mg/kg) and ketamine (87 mg/kg). The application of orthodontic force was performed using closed coil nickel-titanium springs with 10-15g of force for 1.5 mm of activation (Ultimate Wireforms Inc, Bristol, Conn) placed from the maxillary left first molar to the central incisors. The specific technique is described elsewhere.¹⁰

The corticotomy procedure was performed as previously described.11 Briefly, a full-thickness flap was elevated on the palate adjacent to the left first molar, extending mesially to allow for cortical perforations. Using a slow-speed hand piece and a No. $^{1\!/_4}$ round bur, three shallow perforations, each 0.25 mm wide and 0.25 mm deep, corresponding to the size of the bur, were made 5 mm mesial to the left first molar under water irrigation (Figure 1). Drilling was performed with a 0.25-mm bur, which was fully immersed into the bone for every perforation. The flaps were then sealed with cyanoacrylate tissue adhesive (Vetbond, 3M Unitek, St Paul, Minn). Primary closure was achieved for primary tissue healing.

Corticision was performed on the mesiopalatal and distopalatal aspects of the maxillary left first molars (Figure 2). A reinforced surgical blade (No. 11, Bard-Parker, Cherry Hill, NJ) capable of making an incision with a minimum thickness of 400 μ m was positioned on the mesiopalatal and distopalatal gingiva 0.5 mm from the corresponding tooth surface at an inclination of 45-60° to the long axis of the maxillary first molar. The blade was inserted, gradually penetrating the overlying gingiva, cortical bone, and cancellous bone to approximately 1 mm. To ensure the blade went consistently to a depth of 1 mm, a composite stop was placed 1 mm from the tip of the blade to stop it from penetrating past 1 mm.

In the corticision + flap experimental group, a flap was elevated similarly to the corticotomy procedure. The corticision was performed, excluding transmucosal entry, as described above. All surgical procedures were performed only once during the experimental period by one investigator.

After appliance insertion and surgical procedures, the rats were allowed to recover. The appliance was checked twice weekly, and additional bonding material was added if necessary. On days 7 and 14, following initial placement of the appliance, the animals were anesthetized and the springs reactivated to deliver 10-15g of force and were placed more gingivally on the incisors as a result of their continuous eruption pattern.

On day 21, the rats were euthanatized and the mandibles were removed. The maxilla was hemisected and placed in 10% formalin for 5 days at 4°C with constant agitation. Following fixation, micro-computed tomography (micro-CT) imaging and analysis on each animal was performed at 55 kV and 145 mA, collecting 1000 projections per rotation at 300-ms integration times. Three-dimensional images were constructed using standard convolution and back projection algorithms with Shepp and Logan filtering and were rendered within a 12.3-mm field of view at discrete density of 578,704 voxel/mm³ (isometric 12-mm voxels). The serial images were used for quantitative analysis of alveolar bone changes occurring in the region of interest on the maxillary first molar. The region of interest was defined vertically as the most occlusal point of the furcation to the apex of the maxillary roots. Transversely, it included the square with one side connecting the points of the most mesial part of the distobuccal root with the most mesial part of the distopalatal root and the other sides extending to the points of the most distal parts of the mesiobuccal and mesiopalatal roots (Figure 3). Parameters studied included bone volume fraction (BVF) and tissue density (TD).

OTM was evaluated by taking polyvinylsiloxane impressions of all maxillary molars on days 0, 7, 14, and 21. The impressions were used to fabricate a dental die stone. The models were placed on a table top with the occlusal plane made parallel. A millimeter ruler was placed adjacently and a digital photograph was taken parallel to the occlusal plane of the model and the ruler. This image was uploaded into ImageJ

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Figure 4. Magnitude of molar mesial movement (mean \pm SD) at each timepoint for all groups, # P < .05; ** P < .001.

(Image Processing and Analysis in Java, National Institutes of Health). Distance measurements were taken from the distal groove of the first molar to the mesial groove of the second molar with the millimeter ruler as reference for the distance. Intraexaminer reliability was measured by taking measurements twice, more than 2 weeks apart, on four randomly selected models from each surgical group in each day. The intraclass correlation coefficient was 0.99 (95% confidence interval: 0.984-0.994), indicating excellent reliability in the measurement technique.

After the analysis, the specimens were briefly washed with tap water and decalcified in 14% ethylenediamine tetraacetic acid for 2 weeks and then processed for standard paraffin embedding. Serial sagittal sections of the maxillae were performed and



Figure 5. Difference in molar mesial movement (mean \pm SD) between groups, P < .05 at each timepoint.

Table 1.	Distribution	of Data	for Bone	Volume	Fraction
	Distribution	or Data	IOI DOILC	volume	1 raction

Measure	Control	OTM	Corticotomy	Corticision	Corticision + Flap
No. of values	5	14	14	14	13
Mean %	78.04	76.58	73.35	80.27	80.01
SD	8.38	5.83	10.97	6.83	8.29
Minimum %	67.8	63.6	56	67.8	57.6
Maximum %	90.7	84.2	89.2	94.6	88.2
Percentiles %					
25th	71	73.43	65.18	74.75	78.55
50th	78.4	76.3	73.7	78.55	81.9
75th	84.9	81.35	82.63	85.95	85.4
Lower 95% CI %	67.73	73.21	67.02	76.33	75
Upper 95% CI %	88.45	79.95	79.68	84.22	85.02
D'Agostino and Pearson omnibus normality test	Sample size small	0.43	0.68	0.73	0.31

^a OTM indicates orthodontic tooth movement; SD, standard deviation; and CI, confidence interval.

stained for tartrate-resistant acid phosphatase (TRAP). Prior to histological staining, tissue sections were deparaffinized with xylene and rehydrated with decreasing concentrations of ethanol and then washed in deionized water.

Staining for TRAP activity was performed using an acid phosphatase leukocyte kit (386-A1, Sigma Chemical, St Louis, Mo) according to the manufacturer's instructions. Osteoclasts were considered as TRAP-positive multinucleated cells (2+ nuclei) and were counted on the alveolar bone surface of the compression side of the distobuccal root. Histomorphometry analyses were carried out using Osteomeasure Software (Osteometrics Inc, Decatur, Ga). Four sections that revealed the most pulp structure (mid-root sections) were used for measurements, and their means were used for statistical tests.

The area for measurement on the alveolar bone was identified as a square parallel to the sagittal axis of the distobuccal root with 200- μ m width and with the length extending from the bifurcation to the apex. Osteoclast surface was determined as the surface of an active osteoclast touching the alveolar bone and was then divided by total bone surface per defined area.

Statistical Analysis

Simple descriptive statistics were used to summarize the data. Mean, standard deviation, percentile distribution, and confidence interval were computed for all variables. The D'Agostino and Pearson omnibus normality test was used to examine the normality of the data distribution. Osteoclast number and osteoclast surface area were not distributed normally; Wilcoxon signed rank tests were used to compare the osteoclast number and osteoclast surface area. Kruskal-Wallis test and Dunn's multiple comparison tests were used to compare the intermolar distance, BVF, and TD. All statistical tests were two-sided, and a P value of <.05 was deemed to be statistically significant. Statistical analyses were computed using Graph Pad software (La Jolla, Calif).

RESULTS

A total of 60 Wistar rats were included; all rats remained healthy during the experimental period. The magnitude of molar mesial movement followed a constant incremental pattern for all groups with significant differences between 7, 14, and 21 days (Figure 4). However, no significant differences were

Table 2. Distrib	ution of D	ata for Ti	issue Density ^a
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Measure	Control	OTM	Corticotomy	Corticision	Corticision + Flap
No. of values	5	14	14	14	13
Mean (mg/ccm HA)	1085	1050	1040	1046	1057
SD	14.34	14.72	34.44	36.59	46.8
Minimum (mg/ccm HA)	1069	1026	964	971	940
Maximum (mg/ccm HA)	1104	1076	1100	1099	1100
Percentiles (mg/ccm HA)					
25th	1073	1038	1023	1026	1028
50th	1080	1049	1039	1049	1070
75th	1100	1064	1065	1071	1094
Lower 95% CI (mg/ccm HA)	1067	1042	1020	1025	1029
Upper 95% CI (mg/ccm HA)	1103	1059	1060	1067	1085
D'Agostino and Pearson omnibus normality test	Sample size small	0.7268	0.5445	0.5743	0.28

^a OTM indicates orthodontic tooth movement; SD, standard deviation; and CI, confidence interval.



Figure 6. TRAP-stained sections. Arrows point to TRAP-positive cells lying on alveolar bone and dentin. a.b. indicates alveolar bone; De, dentin; P, PDL; NF, no flap; and WF, with flap.

found between groups at any of the timepoints (Figure 5).

Tables 1 and 2 show no significant differences in BVF (P = .18) and TD (P = .06) between the different experimental groups.

Osteoclast activity (Figure 6) was not significantly different between all groups, as observed in osteoclast numbers (P=.1321) or in osteoclast surface area (P=.1062) (Figure 7).

DISCUSSION

In the current study, no significant differences in OTM were observed between groups up to the conclusion after 21 days. These findings parallel those of Tsai et al.,¹² who found no significant difference in magnitude of OTM between corticision, microperforation, and control groups in rats after 3 weeks, although they reported a significant difference between both surgical and OTM groups at 2 weeks. Also in agreement with the current findings, Kurohama et al.¹³ found no difference between control (OTM) and different types of corticotomies (with different levels of invasiveness) in rats after 3 weeks. Similarly, Peron et al.¹⁴ did not find a difference between corticotomy, corticision, and OTM groups in male rats after 4 weeks.

The current results contrast with those of Teixeira et al.,¹¹ who demonstrated an increased rate of OTM with corticotomy. The positioning of the corticotomies in the current study was performed similarly to that study: 5 mm mesial to the first molar.¹¹ One plausible explanation for the different results could be related to the age

of the rats. In the current experiments, the rats were still growing, contrary to those used in the experiments of Texeira et al., whose rats were adults (17 weeks). In fact, the rate of OTM was quite different between the two studies. The control group (OTM) in the current study had approximately five times the rate of OTM when compared to their OTM group and close to 40% more movement than their corticotomy group.

Age-related effects of OTM in rats were previously studied by Ren et al.,¹⁵ who found that young and adult rats had different rates of OTM. In fact, that study reported a major enhancement in the rate of OTM of the young rats (6 weeks old) compared to the adult rats (9-12 months old) in the first 3 weeks. This magnitude of OTM in the 6-week-old group was similar to that of the current experiments for the first 3 weeks. Furthermore, their adult group rats had 0.6 mm of mesial movement after 4 weeks of OTM, which corresponds to the same magnitude of OTM in the corticotomy group and almost twofold magnitude higher rate in the control group (OTM) in the study of Texeira et al. Taking all of these findings into consideration, perhaps the effects of corticotomy on the rate of OTM in growing rats may not be as robust as those seen in adult rats.

Alveolar changes were evaluated using micro-CT and histomorphometry analyses to correlate the presence of osteoclasts with the RAP process, increased bone turnover, and increased tooth movement. BVF, TD, osteoclast number, and osteoclast surface were not significantly different when all groups were compared. This correlated with the insignificant OTM differences between the groups at all timepoints.



Figure 7. (a) Osteoclast number: Comparison of osteoclast number on day 21 measured using histomorphometry. Comparison of the groups demonstrated no significant difference (P = .1321; n = 7). (b) Osteoclast surface area: Comparison of osteoclast surface area on day 21 measured using histomorphometry. Comparison of the groups demonstrated no significant difference (P = .1062; n = 7).

This agrees with the findings of Tsai et al.,¹² who found no difference in BVF between flapless surgical procedures and control (OTM) groups at 3 weeks. However, they reported increased numbers of osteoclasts in the surgical groups in 8-week-old rats. The current results contrast with those of Teixeira et al.,¹¹ who demonstrated a significant decrease in BVF and a significant increase in the osteoclast numbers in the corticotomy group compared to the control. Again, this may be an age-related effect, as previously described. Interestingly, Kurohama et al.¹³ found a significant decrease in BVF with different degrees of bone removal with corticotomies; however, reduction in BVF did not translate to an increased magnitude of OTM in 10-week-old rats.

An objective of this study was to observe the effects of a full mucoperiosteal flap on the rate of OTM. Significant resorption of the alveolus was seen in studies by Yaffe et al.8 when flaps were raised on the buccal and lingual surfaces, demonstrating an alveolar response to separating the periosteum from the bone. Swapp et al.7 suggested that elevating a flap could contribute to the RAP response when they observed no significant difference in OTM in flapless surgical procedures compared to the control. The present study consisted of a corticotomy group, which traditionally requires a flap, and a corticision group, which does not require a flap and is less invasive. The corticision + flap group was added to compare the flap's effect. It was shown that there was no significant difference in OTM or alveolar response, contrary to the findings by Swapp et al.7 This suggests that alveolar bone damage may need to be more extensive and consist of damaging the medullary bone beneath the cortical bone to induce more of a response. Kim et al.4 used corticision without a flap in a feline model, and the surgical blade was driven to depths of up to 10 mm into the bone, providing further damage to the underlying trabecular bone. They observed a significant increase in OTM, suggesting that the procedures need to be deeper in bone than was the case in the current study. However, recent findings from Kurohama et al.¹³ contradict this hypothesis, as corticotomies with different degree of invasiveness did not influence the magnitude of OTM in rats.

This study had some limitations. There was a considerable amount of variation in all the outcomes evaluated between animals in the same surgical groups. This could be due to the strain of rats that were outbred, thereby causing variability in growth and bone metabolism between the animals. Nonetheless, this heterogeneity better reflects the orthodontic population generally. Histomorphometrical analysis at days 7 and 14 could have provided more information on the biological effects of these procedures. However, since rates of OTM were not significantly different, the clinical relevance of these findings would be limited.

CONCLUSIONS

- No significant differences between the surgical groups and the control group in the magnitude of OTM were observed at any of the timepoints evaluated.
- No significant differences in osteoclast numbers and surface area and bone parameters (BVF and TD) was

found between surgical and control groups after 21 days of OTM.

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