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RESEARCH ARTICLES

Effect of force direction and masticatory force towards orthodontic tooth movement in rats

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ABSTRACT

The aim of the research is to investigate the influence of coil spring directions and masticatory force on the amount of OTM. Materials and Methods. Thirty-six male Wistar rats (n = 36) were divided proportionally into two groups with (M) or without masticatory force (NM), treated with palatal coil type (PD) or labial coil (LD) using a costumed stainless steel coil spring to deliver 35 cN force for separating the two incisors in 10 days. The examination dates were day 0, day 5, and day 10. The tooth distance values were calculated by subtracting the distance measured at day 0 from examination days and presented in 8 groups: PD5NM, PD10NM, PD5M, PD10M, LD5NM, LD10NM, LD5M, and LD10M. The study's results were analyzed using ANOVA followed by post hoc analyses. Result: All spring designs induced OTM. The OTM amounts from the lowest to the highest are PD5M, PD10NM, PD10M, LD5M, LD10M, LD5NM, PD5NM, and LD10NM, respectively: 0.26 mm; 0.06 mm; 0.25 mm; 0.44 mm; 0.58 mm; 0.9 mm; 0.97 mm; 1.03 mm, and 1.06 mm. The OTM distance was higher in the labial coil than in the palatal coil groups (p = 0.002). The amount of OTM in the masticatory group was lower than in the group without-masticatory force (p = 0.012), except in the day 10 palatal coil group. Conclusions: Masticatory force and force direction affected the amount of OTM. The labial coil induces more OTM than the palatal coil. Masticatory force decreased the OTM distance.

Keywords: chewing; labial coil; palatal coil; orthodontic force

INTRODUCTION

Tooth movement in orthodontic treatment is a biological response to mechanical forces applied to teeth. A remodeling process in the alveolar bone, involving both tension and pressure sides, is characteristic of the underlying biological responses. In the tension-pressure theory, there is a resorption process on the pressure side and an apposition process on the tension side.[1,2] Bone apposition and resorption by osteoblasts and osteoclasts, respectively, are essential to the effectiveness of orthodontic therapy, which is why these cells must be actively at work during the treatment.[3] The orthodontic tooth movement (OTM) process has been through various studies. Research using animal models of OTM is considered effective for studying bone remodeling induced by mechanical stress.[4] Provision of well-standardized orthodontic strength and easy-to-reproduce systems will be needed to obtain research results that can be learned. In addition, a precise morphological and biomechanical description of the nature of the tooth and its surrounding structures will allow estimation of the stress and strain on the periodontal ligament during OTM.[5] In recent decades, researchers have employed a variety of animal models, including rats, dogs, and primates, to explain how orthodontic tooth movement occurs.[6] Rats are considered good research models for studying OTM, with several practical advantages. First, mice are relatively inexpensive, making it easy to use large samples, and can be stored for long periods. Second, the preparation of histological preparations in mice is easier than in other animal models, for example, dogs. A third advantage is that most antibodies required for cellular and molecular biology techniques are available only for mice.[5]

The experimental design of orthodontic equipment frequently reveals flaws that have a substantial impact. Because the force's effect on the tissue depends on the size of the tooth involved, the magnitude of the force must be proportional to its root surface area.[7] Because mouse teeth are so little (rat molars are around 50 times smaller than human molars), designing efficient orthodontic appliances capable of providing steady and continuous stresses with an adequate strength range is difficult.[5] Achievement of optimal strength is essential to obtain maximum orthodontic tooth movement rates without damaging effects on the roots, periodontal ligament, and alveolar bone.[6] In addition, a standard methodology is very important to be able to do comparisons in various studies. Based on this description, the author is interested in researching the effect of the direction of the coil spring strength with and without masticatory force on the amount of tooth movement.

Masticatory pressure is rarely considered in the results of in vivo orthodontic research. Masticatory pressure is rarely considered in the results of in vivo orthodontic research. The research comparing the OTM in vivo of the hypofunction tooth on the tension side showed that the number of osteoblasts on the tension side was unaffected by the hypofunctional state. However, the number of osteoclasts on the pressure side was decreased during orthodontic tooth movement. The research comparing the OTM in vivo of the hypofunction tooth on the tension side showed that the number of osteoblasts on the tension side was unaffected by the hypofunctional state. However, the number of osteoclasts on the pressure side was decreased during orthodontic tooth movement.[8] Furthermore, clinicians frequently face cases requiring the movement of teeth that never have occlusal pressure or hypofunctional teeth, such as open bite, ectopic canine, palatoversion, linguoversion, and buccoversion.[9] Hypofunctional teeth result in atrophic alterations to the periodontal ligament, a reduction in the number of periodontal fibers and blood vessels, and a narrowing of the periodontal space.[10]

In vivo, tooth movement in rats uses molars as the gold standard. However, this model is often difficult to conduct; therefore, many researchers use incisors by separating the two incisors as an option. The orthodontic model can be cross-section histologically due to readily tipping incisors instead of bodily effects to limit the area measurement.[11] In addition, the incisors of rats erupt continuously, but in previous studies, it is known that incisors impede the eruption while used as the abutment.[12] There are two designs often found in vivo scenarios, palatally and labially; this study aimed to determine the effect of coil spring direction in rats with and without masticatory force on the number of orthodontic tooth movements.

MATERIALS AND METHODS

All study procedures were performed according to the approved protocol of the Institutional Animal Care and Usage Committee (ARRIVE guideline). The ethical permission of the research was approved by the Ethics and Advocacy Board, the Faculty of Dentistry of Universitas Gadjah Mada board No 00133/KKEP/FKG-UGM/EC/2019.

The study was conducted on 36 male Wistar rats, with body weights between 350-450 grams, by providing an orthodontic force using a coil spring to separate the two incisors randomly. A force of 35 cN was applied reciprocally, using 0.012 inches stainless steel coil spring (Ortho Prime Inc. USA; AW 85021201; Orthoshape SS 0.012”) to separate the two upper incisors. The orthodontic appliance was a 3-spin loop, 2 mm in diameter, with arms 12 mm in length, with “V” folds placed 9 mm from the coil region, A dynamometer (Correx gauge, Haag-Streit, Switzerland) was used to measure the 35 g of tension. The coil spring was deflected for 3.4 mm to deliver an orthodontic force of 35 cN reciprocally or 17.5 cN per upper incisor before they were installed. The palatal spring was placed parallel to the palatal surface (Figure 1B). Activation of the palatal spring caused the incisors to move radially in a distal and palatal direction following the movement of the palatal arm. Meanwhile, the labial (Figure 1A) spring which is mounted vertically parallel to the labial surface of the incisors,

(A) (B)

Figure 1. labial coil spring with the arm installed vertically on the labial surface of upper (A) incisors and the palatal coil spring installed horizontally with the arm on the surface of the palatal (B) to move the upper incisors distally

moved the teeth radially in a distal direction and downward. Experimental animals were divided proportionally into five groups: negative control (without treatment), palatal (P), or labial (L) coil types, with (M) or without masticatory (NM) force. In the group of rats without masticatory force, the incisal surfaces of the lower right incisors were cut every two days for 2 mm, and the contralateral teeth were left in contact.

The distance of the most distal upper margin of metal bands is measured simultaneously after installation and every day. The measurements were done twice, with 10 minutes different and in triplets repetitions with digital calipers (Mitutoyo, Japan), with an accuracy level of 0.01 mm by one calibrated independent observer. The OTM was consistently measured at the same point of the uppermost mesial side of the metal ring between incisors. The OTM values were calculated by subtracting the distance measured on day 0 from examination days (day five and day 10). The groups analyzed were named based on the treatment and the day of examination were PD5M, PD10M, PD10NM, PD5NM, LD5M, LD10M, LD5NM, and LD10NM (Table 1).

The data were normally distributed and eligible for the three-way ANOVA to assess differences and interactions between groups based on the results of this investigation. To establish whether there were significant differences between the groups, researchers used a post hoc LSD test. Statistical significance was defined as a p-value < 0.05.

RESULTS

All coil designs induced OTM in both with and without masticatory force groups. The OTM distance value was obtained from day 0 to day five, namely the day 5 group, or from day 0 to day 10, the day 10 group, namely the day 10 group, either in the labial and palatal coil spring group. Thel OTM amounts from the lowest to the highest are PD5M, PD10NM, PD10M, LD5M, LD10M, LD5NM, PD5NM, and LD10NM in 10 days are 0.26 mm; 0.06 mm; 0.25 mm; 0.44 mm; 0.58 mm; 0.9 mm; 0.97 mm; 1.03 mm and 1.06 mm, respectively.

The ANOVA analyses in Table 2 showed that the OTM group distance was significantly greater in the labial than in palatal coil spring types (p = 0.002). The amount of OTM in the group without

Table 1. The mean of OTM distance between palatal and labial springs with and without masticatory force

Group		OTM distance ± SD (mm)
PD5NM	Palatal spring day 5 without masticatory force	1.03 ± 0.270247
PD10NM	Palatal spring day 10 without masticatory force	0.25 ± 0.260064
PD5M	Palatal spring day 5 without masticatory force	0.06 ± 0.277128
PD10M	Palatal spring day 10 with masticatory force	0.44 ± 0.368646
LD5NM	Labial spring day 10 with masticatory force	0.97 ± 0.406325
LD10NM	Labial spring day 10 without masticatory force	1.06 ± 0.218251
LD5M	Labial spring day 5 with masticatory force	0.58 ± 0.055076
LD10M	Palatal spring day 10 with masticatory force	0.9 ± 0.134288

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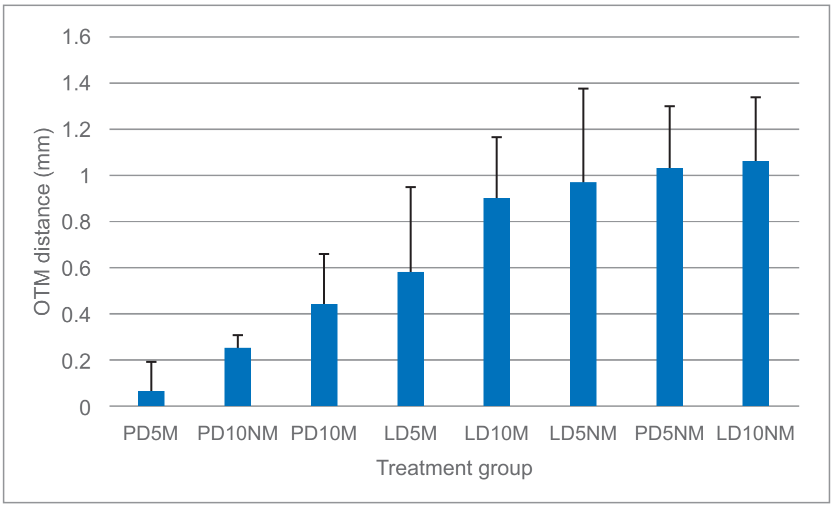

Figure 2. The OTM distance between palatal and labial springs with and without masticatory force. Orthodontic tooth movement distance values were calculated by subtracting the distance measured at day 0 from examination days. The graphic presents the data from the least and the widest separation of the upper incisors. P: palatal spring; L: labial spring; D5: Day 5; D10: Day 10; M: with Masticatory force; NM: without masticatory force.

masticatory force was greater than in the group with masticatory force (p = 0.012). There was no significant difference between days' observations, p = 0.77. There was no interaction between the types of spring and the day of observation (p = 0.78). There was an interaction between spring-type groups within the day observation group (p = 0.042) but not within the masticatory force group (p = 0.80). No interaction significantly was found in the OTM between the three variables.

DISCUSSION

Orthodontic treatment is treatment in the field of dentistry which aims to correct malocclusions in the teeth. Tooth movement in orthodontic treatment is obtained through remodeling the alveolar bone

Table 2. Anova analyses of the OTM distance between palatal and labial springs with and without masticatory force

Variable	Type III sum of squares	df	Mean square	F	Sig.	Partial eta squared
spring_type	0.948	1	0.948	14.137	0.002	0.469
day_observation	0.006	1	0.006	0.085	0.774	0.005
Mastiocation force	0.537	1	0.537	8.008	0.012	0.334
spring_type * day_observation	0.329	1	0.329	4.906	0.042	0.235
spring_type *Mastiocation forcei	0.005	1	0.005	0.076	0.786	0.005
day_observation * Mastiocation force	0.592	1	0.592	8.831	0.009	0.356
spring_type * day_observation * Mastiocation force	0.234	1	0.234	3.49	0.08	0.179

Note:
P: palatal spring; L: labial spring; D5: Day 5; D10: Day 10; M: with Masticatory force; NM: without masticatory force.
*p < 0.05; n = 3 for each group

Table 3. Post hoc analyzes OTM distance. Only tests with significantly different results ($p < 0.05$) between Palatal and Labial springs with and without masticatory force is presented

Group	Group	p value
PD5NM	PD10NM	0.002*
	PD5M	0.001*
	PD10M	0.013*
	LD5M	0.049*
PD10NM	LD5NM	0.004*
	LD10NM	0.002*
	LD10M	0.008*
PD5M	LD5NM	0.002*
	LD10NM	0.001*
	LD10M	0.004*
PD10M	LD5NM	0.023*
	LD10NM	0.01*
	LD10M	0.044*
LD10NM	LD5M	0.04*

and periodontal tissue in response to mechanical forces (orthodontic forces). Applying orthodontic force to the teeth causes alveolar bone resorption in the area of pressure in the periodontal ligament, while the tension area in the periodontal ligament experiences bone formation.[13,14]

The palatal spring was aligned with the palate surface. When the palatal spring was activated, the incisors moved radially in a distal and palatal manner in response to the movement of the palatal arm. Meanwhile, the labial spring, positioned vertically parallel to the incisors' labial surface, moved the teeth radially distal and downward. All groups of Wistar rats received the same amount of 35 gram force, which had been accepted as the optimal force for animal models in orthodontic studies.[11] Animals have been utilized extensively as models in a variety of scientific investigations, including orthodontic research.[15] Separation of the upper incisors is considered inaccurate for orthodontic research due to the tipping response to orthodontic forces.[5] Because of their relative physiological movement response to attain homogeneity of the same force exposure along the neck of the tooth to the apex, molars are sometimes believed to be more valid as an orthodontic tooth model. This study did not require uniformity of exposure to these forces. This study aimed to measure the distance response of the OTM that happens. According to one review, an animal orthodontic model can only test utilizing initial orthodontic force, regardless of the type of teeth used. Molar teeth that move to the mesial will be distally tipped to accommodate stronger or longer orthodontic forces. The condition of tipping motion to acquire a reasonable amount of force from one type of force direction can be overcome by transversely sectioning histological sections in absorption and apposition areas that expose a force with the same source, size, and direction of the force.[11]

In this study, antagonist lower incisors were cut 2 mm per 2 days, a cutting length determined based on a pilot study to free the upper incisors from masticatory forces. This result differed from previous research, which found that eruption rates of incisors decreased in both without appliance and with the appliance groups as well as the incisor inclination between the appliance and control groups on day 10.[16] However, the frequent tooth reduction in this experiment was concordant with the finding of another study that hypofunctional teeth caused periodontal ligament atrophy, decreasing the number of fibers and narrowing the periodontal space. The increasing transforming growth factor β increases alveolar bone apposition, narrowing the periodontal space and further causing tooth elongation.[10]

The OTM group distance was significantly greater in the labial than in palatal coil spring types, suggesting that the palatal coil force moves the incisor to the distal and palatal direction against wider palatal cortical bone than the labial spring against the thinner distal cortical alveolar bone.[17] Some bite raisers recently used for opening the bite freed the teeth from mastication force and unlocking the bite. Bite raisers treat special cases of malocclusion that impact the three planes of space, opening the occlusion for transverse or anteroposterior correction, preventing unwanted movement of certain teeth, and preventing function switching (due to premature contact).[18] The number of OTM in the group without masticatory force was greater than in the group with masticatory force. The results can be explained in two situations. First, liberating the masticatory force will open the bite locked so the teeth can move smoothly. Second, the synergistic effect of changes in the structure of the bone tissue around the teeth in the hypofunctional occlusal conditions results in loss of volume and mineral density together with structural changes around the alveolar bone by tooth movement.[19]

There was no significant difference between the day's observations and no interaction between the types of spring and the day of observation. There was an interaction between spring-type groups within the day observation group but outside the masticatory force group. No interaction significantly was found in the OTM between the three variables, which can be explained with the post hoc analyses in the following paragraph. The result obtained from the smallest to the largest OTM distances are PD5M, PD10NM, PD10M, LD5M, LD10M, LD5NM, PD5NM, and LD10NM, but the OTM distances between the group sequences were the same. Orthodontic tooth movement in the labial spring group increased together with the length of observation days.

In contrast to the labial spring group, the palatal spring group, the PD5NM group was significantly larger than the PD10NM, and there were indications of decreased tooth movement from day 5 to day 10. That might be explained as follows. The group with mastication in this study inhibited OTM. PD5NM is a palatal spring group without masticatory force known not to inhibit OTM, which can be interpreted as the palatal spring's resistance that causes the rapid decay of the tooth movement. PD5M was the same as on day 10 in both groups with and without mastication, confirming the direction factor of the palatal spring type against the palatal bone's thickness to be the cause.[17]

In the same experiment, the osteoblast was not decreased from day 5 to day 10, but the osteoclast number did. The situation of the current experiment was simulant to the OTM, which was not decreased from day 5 to day 10. Using palatal spring in normal occlusion reduces the OTM and likely the osteoclast number formation on the pressure side, while comparable osteoblast number on the tension side.[8] Labial springs move teeth at a larger distance as the teeth are moved through an area of thin bony support. The amount of OTM distance of the three palatal spring groups was smaller than that of the labial spring group. Only the PD5NM group had the same number of OTM as all the labial groups except for the LD5M group, which had a masticatory force with the potential for OTM inhibition, combined with a variable labial spring which relatively had no bone density barriers.

Wrapping together the masticatory force has to be accounted for in orthodontic studies since the hypofunction tooth responds differently when orthodontically moved, especially compared to the normal teeth. Hypofunctional teeth, when orthodontically moved, have less heparan sulfate proteoglycan exposure, which plays a role in osteoclastic activity compared to normal teeth. The study observed chondroitin sulfate (CS) throughout the extracellular matrix, and HS was observed on the endothelial cells and the osteoclastic cells on the compressive side. CS and HS were detected in fewer numbers in the hypofunctional group without an orthodontic appliance. Applying a larger force causes CS to appear in compression areas with no cells and fibers, whereas HS is on the periphery of this area. However, in the smaller force, the distribution of CS and HS was similar to that of normal controls. These findings indicate that CS and HS are affected by orthodontic forces and suggest their different functions in tissue remodeling.[20] Compressive force rapidly caused apoptosis of the PDL and vascular endothelial cells in hypofunctional teeth but not normal teeth. Vascular Endothelial Growth Factor (VEGF) is important in alveolar bone's resorption and apposition processes because it affects the proliferation and differentiation of osteoblasts and osteoclasts in vitro. The Expression of

VEGF in hypofunctional teeth also decreases during orthodontic movement leading to vascular constriction and endothelial cell apoptosis.[21] The VEGF decreasing were confirmed by the study on blood vessel numbers in the current experiments.[22] However, the number was not influenced by the hypofunction duration, the pressure or the tension sides, or the interaction between all the variables. The number of blood vessels in the tension side of the normal teeth group on day 5 was higher than on day 0 and day 10, as well as on both the same days of the pressure side group, but all the intra and inter-hypofunction groups are not significantly different

CONCLUSION

The results showed that the group distance was significantly greater in the labial than in palatal coil spring types. The distance of tooth movement in the group without masticatory force was greater than in the group with masticatory force. There was no significant difference between the days of observation. There was an interaction between spring-type groups within the day observation group but not within the masticatory force group.

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