Tooth movement characteristics in relation to root resorption in young and adult rats


The aim of this study was to investigate tooth movement characteristics in relation to root resorption in young and adult rats. Two groups of 30 rats each (aged 6 wk and 9–12 months, respectively) were used. Standardized orthodontic appliances were placed to move the maxillary molars mesially. Animals were killed 1, 2, 4, 8, and 12 wk after the beginning of the experiment and their jaws were processed for histomorphometric analysis. In parasagittal sections of ‘pressure zones’ the extent of root resorption was measured and expressed as percentage of total root length. Whereas a negative correlation was found between the velocity of tooth movement and the extent of root resorption, a positive correlation was noted between the amount/duration of tooth movement and root resorption, but only in adult rats. The duration of tooth movement showed the highest correlation coefficient and the smallest 95% confidence interval. In younger animals, correlations were weak, and other factors are likely to be involved in the individual susceptibility to root resorption.

The risk factors for orthodontically induced root resorption have been extensively researched and are generally divided into patient-related factors and treatment-related factors (1). Studies on the association between root resorption and orthodontic treatment regimen in humans showed consistent results in that heavy or constant forces were found to induce more resorption than light or dissipating forces (2–5) with no differences being observed between different treatment modalities (6, 7). However, studies on the association between root resorption and the amount of tooth displacement/duration of orthodontic treatment showed inconsistent results. Whereas some studies revealed a positive correlation (8–17), others did not (18–22). Lack of a significant correlation is often referred to as genetically based interindividual variations in susceptibility to root resorption (18, 23). However, experimental studies with a standardized orthodontic set-up showed that even in animals with a similar or homogenous genetic background, large interindividual differences occur in the velocity of tooth movement, in the recruitment of osteoclasts, and in the incidence/severity of root resorption (24–26). These results suggest that interindividual variations cannot exclusively be attributed to genetic factors.

In our opinion, two factors might have been overlooked: the effect of age; and the velocity of tooth movement. An age-categorized summary of the main results from human studies on the associations between root resorption and amount/duration of tooth movement indicates that growing subjects often do not show associations (6, 18–20, 22), whereas adult subjects often do (1, 9). Controversial results came exclusively from studies on mixed age groups (8, 11–13, 15–17, 21, 27). We therefore hypothesize that age plays an important role, which needs to be verified in standardized experimental studies. Regarding the velocity of tooth movement, hyalinization of periodontal ligament seems to be a key factor affecting the efficiency of tooth movement (28). The removal of hyalinized tissue is essential for tooth movement to occur (24, 29). Similarly, several lines of evidence have suggested that root resorption is closely associated with the removal of necrotic hyalinized tissue (30–36).

It was the aim of the present study to investigate whether a correlation exists between the severity of root resorption and tooth movement characteristics at the individual level and whether such a correlation is age related. For this purpose we established a standardized orthodontic set-up in two age groups of rats for time-periods of up to 12 wk.

Material and methods

Experimental procedures

Two groups of 30 male Wistar rats, aged 6 wk and 9–12 months, respectively, were used [the same material as described previously (25)]. Ethical permission was obtained from the Radboud University Nijmegen Medical Centre, the Netherlands. Briefly, a split-mouth design was used with the experimental side randomly chosen and the contralateral side as a control. An orthodontic appliance was placed at
the experimental side under general anaesthesia. A transverse hole was then drilled through the alveolar bone and maxillary incisors at the mid-root level. A ligature wire enclosing all three molars was bonded on the experimental side. A coil spring (GAC, New York, NY, USA) was attached to a ligature wire to move the molars mesially with a 10 cN force. At 1, 2, 4, and 8 wk, 5 or 6 rats from each group were killed, and at 12 wk the remaining animals were killed. These time intervals represented the duration of tooth movement for each group of rats. The distance between the most mesial aspect of the maxillary molar unit and the enamel–cementum junction of the ipsilateral maxillary incisor was measured at both experimental and control sides, and the difference between the two was taken as the amount of tooth displacement. The amount of tooth movement refers to tooth displacement, in mm, during the whole observation period. The velocity refers to the rate of tooth movement during the last period of observation; for example, a rat from wk 12: the velocity = (tooth displacement from wk 8 to wk 12)/4.

Material preparation

The rats received a lethal dose of anaesthetic before they were killed. They were then perfused with 4% paraformaldehyde solution in 0.1 M phosphate-buffered saline (PBS) at 37°C. The maxilla was dissected and immersed in 4% paraformaldehyde for 24 h at 4°C. After decalcification in 10% EDTA and paraffin embedding, serial parasagittal 7 μm sections were cut. Every 25th section was collected on SuperFrost/Plus slides (Menzel-Gläser, Braunschweig, Germany) and stained with hematoxylin and eosin (H&E).

Root resorption measurement

Three roots per section and three sections per molar block were selected. Root length was measured (the distance from the dentin–cementum junction to the root apex) and root resorptions were recorded in the ‘pressure regions’. Only dentinal resorptions were taken into consideration. The severity was expressed as the summed length of all resorption lacunae per root length (Fig. 1). The average severity was calculated as the mean of all measurements representing each individual animal.

**Statistics**

Distributions of the data were checked with the D’Agostino–Pearson normality test. Because the data did not pass the normality test, Spearman correlation tests were used. Correlations were considered significant if the P-value was < 0.05.

**Results**

Statistically significant positive correlations between duration of tooth movement and severity of root resorption were found only in the adult group (Fig. 2B). With respect to the amount of tooth movement, large individual variations were noted that tended to show a time-related increase in both young and adult rats (Fig. 3A,B). A significant, positive correlation between the duration of tooth movement and the severity of root resorption was observed only in the adult animals (Fig. 3C,D). Large individual variations were found in both age groups with respect to the velocity of tooth movement and severity of root resorption (Fig. 4A,B). A statistically significant negative correlation was found between the velocity of tooth movement and the severity of root resorption in the adult rats, but not in the younger rats (Fig. 4C,D).

**Discussion**

Root resorption is a frequent iatrogenic consequence of orthodontic treatment. Although several studies have been undertaken to find risk factors, few have been identified. The present results demonstrated that, in the rat, older animals respond differently to a standardized orthodontic protocol than younger animals. This indicates that, although we still have not been able to identify the underlying molecular and biological events, age in itself represents a factor to be reckoned with. We also noted considerable interindividual variation in the rate of tooth movement. Some animals were diagnosed as ‘slow movers’ whereas others were identified as ‘fast movers’. This is in agreement with previous studies in humans (18, 23) and animals (24–26). Our hypothesis, that age of the study subjects and the individual velocity of tooth movement are closely related to root resorption, was confirmed by our results showing that in older animals a negative correlation exists between velocity of tooth movement and root resorption.

It has to be acknowledged that the velocity of tooth movement, as seen in the present study, was related to the duration of tooth movement. The highest velocities were recorded in the first few weeks of treatment, in both age groups. Nevertheless, at each time-point there were ‘slow movers’ and ‘fast movers’. A negative correlation between velocity and root resorption seems logical if indeed formation and removal of hyalinized
tissue in the periodontal ligament space is the limiting factor for both the efficiency of tooth movement and the resorption of the roots (30–35). No causal relationship between the two phenomena could be established. Probably they have a common cause: extensive hyalinization leads to both root resorption and delay of tooth movement (36, 37).

Previous clinical studies on age-related differences in root resorption reported varying results: no difference between growing and adult subjects (9, 15, 38); a slightly higher prevalence of root resorption (39) in adults; or more root resorption, but only at limited areas, in adults (27). One drawback of these studies might be that age was studied as one of the many covariates. Experimental studies on root resorption with a standardized set-up are limited. MALTHA and colleagues (26) showed that prolonged tooth movement significantly increases root resorption in young adult Beagle dogs. This result was supported by the results of the present study, showing a positive correlation between duration of tooth movement and root resorption. Compared with velocity and amount of tooth movement, the duration showed the highest correlation with severity of root resorption. These results are in agreement with previous human studies on adults. As, in the group of younger rats, none of the correlations was statistically significant, we conjecture that other, yet-unknown, factors are also involved. These should be studied by using multivariate techniques.

The increase over time in interindividual variations in the amount of tooth movement and the severity of root resorption in both age groups suggests that the longer the orthodontic treatment, the less predictable root resorption is. Interestingly, the interindividual variations...
of the velocity of tooth movement tended to decrease with time in both young and adult rats.

In conclusion, the present study showed significant correlations between root resorption and the velocity, amount and duration of tooth movement in adult rats, but not in younger animals. This indicates that in various age groups different risk factors for root resorption may be of importance.

References
19. Owman-Moll P. Orthodontic tooth movement and root resorption with special reference to force magnitude and...