Changes in the Periodontal Ligament After Experimental Tooth Movement Using High and Low Continuous Forces in Beagle Dogs

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Abstract: The aim of this study was to evaluate histological changes in the periodontal structures of beagle dogs after using high and low continuous forces during experimental tooth movement. An orthodontic appliance was placed on the second premolar and the first molar by exerting a continuous and constant reciprocal force of 25 cN on one side and 300 cN on the other side of the mandible. Tooth movement was recorded weekly. Dogs were sacrificed after one, four, 20, 40, and 80 days for histological evaluation. Hematoxylin and eosin (HE) staining was used for tissue survey, staining for alkaline phosphatase as a marker was used for active osteoblasts, and tartrate-resistant acid phosphatase staining was used for osteoclasts. After 24 hours, the remodeling process had already started at the pressure and tension side, and in some samples hyalinization was found. In contrast to earlier studies, hyalinization was found throughout the entire experimental period, both in molars and in premolars. In the periodontal ligament of some teeth, small patches of hyalinization were found at the pressure side, mostly located buccally or lingually of the mesiodistal plane, whereas others showed large areas of necrotic tissue. It is concluded that hyalinization limits tooth movement, but there is no relationship with the force level. (*Angle Orthod* 2004;74:16–25.)

Key Words: Periodontal ligament; Force magnitude; Histology; Orthodontics; Tooth movement

INTRODUCTION

Orthodontic tooth movement is the result of biological reactions within the periodontal ligament (PDL) and the alveolar bone, evoked by externally applied forces. It is generally believed that the use of an optimal force system is important for an adequate biological response in the peri-

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Accepted: June 2003. Submitted: March 2003.

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odontal system.¹ The hypothetical relation between the magnitude of the applied force and the rate of subsequent orthodontic tooth movement has received considerable attention in orthodontic research during the past decades.^{2–9} Also, in contemporary textbooks, optimal force levels are advocated.^{10,11} However, many of the authors cited above disregard the fact that force levels should be related to other parameters, such as the surface areas of the roots, the alveolar bone, and the geometry of the PDL over which the force is dissipated.

Cells are the motors for tissue remodeling, and they most probably react to changes in local stresses and strains that are the result of force application.^{12,13} This means that a thorough knowledge of these stresses and strains is essential for understanding the system; however, they are difficult to deduct from externally applied forces. First, the geometry of roots is rather complicated, and, therefore, estimates of their surface area show a large variation.¹⁴ Second, the biomechanical characteristics of the system are largely unknown and appear to change with time during orthodontic tooth movement.^{12,15} This means that to interpret experimental data, one has to deal with a rough approximation of the change in pressure at the so-called pressure side of the PDL induced by orthodontic forces. This change in pressure can be derived from the following equation,

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		Pressure		Type of		Duration of		
Authors	Year	P (kPa)	Force F (cN)	Force	Object	Force	Appliance	Species
Storey and Smith ²	1952	10–17	150–250	t	Cmax	21 d	Closing springs	Human
Reitan ²⁰	1960	2–10	40-140	t/b	P₁ max	27 d	Closing arch	Human
Lee ²¹	1964	10–17	150-260	t/b	Cmax	7 d	Torsion springs	Human
Reitan⁵	1957	5–6	50-60	t/b	Cmax	18	Closing spring	Human
Hixon et al ²⁶	1969	30-100	300-1000	t	Cmax	56 d	Closing springs	Human
Jarabak and Fizzell ³	1972	7–11	105–170	t/b	Cmax	60 d	Closing springs	Human
Boester and Johnston ⁶	1975	9–21	140–310	t	Cmax	70 d	Closing springs	Human
Quinn and Yoshikawa ⁷	1985	7–14	100-200	b	Cmax	?	Closing springs	Human
King et al ³⁸	1991	65–180	20-60	t	M₁ max	1–14 d	Closed coils	Rats
Lee ³⁹	1995	17–18	255-275	t/b	Cmax	50 d	Torsion springs	Human
Pilon et al ¹⁶	1996	10–40	50-200	b	P ₂ mand	120 d	Clastics	Dogs
Gibson et al ²²	1992	133	40	t	M₁ max	1–24 h	Closed coil	Rats
Rodriguez et al ²⁵	1996	166	50	b	M_1 max	?	Coil spring	Rats
Owman-Moll et al37	1996	5–20	50-200	t	P₁ max	49 d	Sectional	Humans
Verna et al ²⁴	2000	83	25	t	M₁ max	21 d	Coil spring	Rats
lwaski et al35	2000	4–13	18–60	b	Cmax	84 d	Coil spring	Humans
Kohno et al ²³	2002	4–11	1.2–10	t	M₁ max	2–14 d	Spring	Rats

TABLE 1. Pressure values (in kPa) and force magnitudes (in cN) as reported in the literature. T indicates tipping force; b, bodily movement; max, maxillary; mand, mandibular; C, canine; P1, first premolar; P2, second premolar; M1, first molar; and d, days.

$$\Delta P (kPa) = \frac{\Delta F (in cN) \times 10^{-1}}{A (in cm^2)}$$

in which A is the effective root surface area that is supposed to be half the total root surface area. Application of this equation to data from literature leads to the pressure values presented in Table 1.

Assuming that a pressure of about 20 kPa would result in optimal tooth movement, Pilon et al¹⁶ performed a series of standardized experiments in dogs in which they used forces of 50, 100, or 200 cN to move mandibular second premolars in beagle dogs. These forces were supposed to result in local pressures of 10, 20, or 40 kPa. These conditions were supposed to represent low, moderate, or high pressures, respectively. Their results indicated that all these pressures evoke a similar tissue response. Large individual differences, however, were found in the rate of tooth movement irrespective of the applied force. The development of hyalinized areas could play an important role in this interindividual variation. Tissue necrosis (hyalinization) is caused by excessive compression of the PDL as a result of too much pressure.⁴ After the removal of the hyalinized tissue by neutrophil granulocytes and macrophages, and after undermining resorption by osteoclasts, the phase of acceleration begins and orthodontic tooth displacement proper starts.5

Quinn and Yoshikawa⁷ suggested four different models for the relation between force magnitude and subsequent orthodontic tooth movement. They suggested in model 3 that at low force levels a dose-response relation might exist between the force magnitude and rate of tooth movement. In that range, hyalinization would play only a minor role or even no role at all. Increases in pressure levels would lead to an optimal tissue response persisting over a wide range of pressures. The role of hyalinization on the indi-

vidual level would increase with higher pressures. Forces resulting in pressures beyond the advocated levels would result in slower tooth movement because of extensive hyalinization of the PDL.^{1,3,7} If this reasoning is correct, one has to assume that at forces below about 40 cN, no hyalinization will be found, whereas pressures beyond 275 cN would result in extensive hyalinization. The published literature is not conclusive on this subject because different types of orthodontic appliances were used, and the direction, duration, and type of tooth movement showed a huge variation, and, therefore, the comparison of the effects of different force levels on tooth movement is difficult.9 Therefore, the aim of the present study was to evaluate rate of tooth movement and tissue reactions after standardized application of low (25 cN) and high (300 cN) orthodontic forces that lead to low and high pressures in the PDL of different teeth within one experimental animal.

MATERIALS AND METHODS

Preparation of the dogs

A group of 15 young adult beagles with a complete permanent dentition was used. The experiment was approved by the Board for Animal Experiments of the University Medical Centre, University of Nijmegen, The Netherlands. Three months before the start of the experiment, the third and fourth premolars on both sides of the upper and lower jaw were extracted after hemisection. Before extraction, the dogs were premedicated with 1.5 mL Thalamonal (fentanyl 0.05 mg/ml and droperidol 2.5 mg/mL; Janssen Pharmaceutica, Beerse, Belgium) and anesthetized with 15 mg/kg Nesdonal (thiopental sodium 50 mg/mL; Rhone-Poulenc Pharma, Amstelveen, The Netherlands). Radiographic evaluation three months after extraction showed complete heal-



FIGURE 1. Experimental appliance inducing bodily displacement of the premolar and molar in the direction of the arrows by a closed coil spring producing a reciprocal force of 25 or 300 cN. C indicates canine; P2, second premolar; and M1, first molar.

ing of the wounds and alveolar bone. Then, burr holes were prepared in the alveolar bone within the extraction areas, and custom-made titanium implants (height 10 mm, outer diameter 3.1 mm, sandblasted), with a locking screw on top, were placed and press fit into these holes. The soft tissues were closed with sutures (Vicryl absorbable 3-0; Ethicon, Brussels, Belgium) over the implants. Vinylpolysiloxane impressions (Express STD; 3M, St Paul, Minn) were made for construction of orthodontic appliances.

Construction of the appliance

Three months after implant placement, the implants were uncovered, the locking screws were removed, and a suprastructure was placed on the implants. A holder for a stainless steel sliding bar (ø 2.0 mm H6 type 316; Rijnvis, Rotterdam, The Netherlands) was attached with glass-ionomer cement (Ketac-Cem, ESPE, Seefeld, Germany) to this suprastructure. The rigid sliding bar was fixed into its holder by a small locking screw. Custom-made CoCr alloy crowns (Heraeus Kulzer, Hanau, Germany) were cemented on the mandibular second premolars and first molars with PanaviaEx Dental adhesive (Kuraray, Osaka, Japan). Polyacetal homopolymer tubes Ø 2.0 mm H7 (Vink Kunststof, Didam, The Netherlands), which were used as bearings, were glued into metal cylinders with bonding (Vitermer; 3M Dental Products). These cylinders in turn were soldered onto the crowns. The sliding bar, which was fixed to the implant suprastructure, ran freely through the low-friction polyacetal homopolymer tubes on the premolar and molar.¹⁷ To produce bodily displacement of the second premolar and the first molar, a Sentalloy® Closed Coil Spring (GAC International, New York, NY) producing a force of 25 cN was attached to buccal hooks on the crowns of the second premolar and the first molar (Figure 1). These springs exert a constant, continuous, reciprocal force on both teeth over

a wide range of activation, which means that reactivation is not needed. $^{\rm \scriptscriptstyle 17}$

Measurements

For each session, the dogs were sedated with 3 mL of a generic preparation containing 10 mg/mL oxycodon HCl, 1 mg/mL acepromazine (Vetimex Animal Health, Bladel, The Netherlands), and 0.5 mg/mL atropine sulfate (Centrafarm, Etten-Leur, The Netherlands). Once a week, the positions of the experimental teeth were measured with a digital caliper as the distance between a reference point on the tooth in question and a reference point on the implant construction. This technique has been shown to be accurate. In previous studies,^{16,17} the intraobserver differences appeared to be in the order of 0.01 mm, and the SD of the mean differences between two observers was 0.02 mm.

At each session, the coil spring and the sliding bar were removed to facilitate cleaning of the oral mucosa and the dentition with a toothbrush and 0.4 mg/mL chlorhexidine digluconate in water (Astra Chemicals, Rijswijk, The Netherlands). To minimize friction, the sliding bars were polished (Abraso-Star K50, Bredent, Senden, Germany) and vaseline was put in the polyacetal homopolymer tubes and on the sliding bar. Then, the sliding bar and the same coil springs were replaced and checked for friction and force delivery. By following this protocol, the force should be constant for a long period of time.

Time-displacement curves

Time-displacement curves of each tooth were constructed, based on the weekly intraoral measurements. For histological analysis, groups of three dogs were killed after one, four, 20, 40, and 80 days. Figure 2 shows an example of a time-displacement curve with the times of sacrifice indicated by arrows. The means and SD of the rate of



FIGURE 2. Time-displacement curve. Arrows indicate the time points of sacrifice.

movement of the experimental teeth during the preceding period (measurements once a week) were calculated for each animal after sacrifice. In this way, the rate of tooth movement could be related to the histological features.

Histology

Groups of three dogs were sacrificed after general anesthesia by a lethal dose of Narcovet (sodium pentobarbital 60 mg/mL, Apharmo, Arnhem, The Netherlands) after one, four, 20, 40, and 80 days. The mandibles were dissected, and each tooth and its surrounding bone were split into a mesial and a distal part, each containing one root for different methods of histological processing.

Mesiodistal paraffin sections and cryosections were prepared parallel to the long axis of the root for normal histological evaluation and enzyme histochemistry, respectively. The tissues for paraffin sections were fixed in a 4% buffered formaldehyde solution in 0.1 M phosphate-buffered saline for two weeks and then decalcified in 20% formic acid and 5% sodium citrate for approximately four weeks. The endpoint was determined by radiography (Philips Oralix, Eindhoven, The Netherlands). Furthermore, the sections were dehydrated and embedded in Paraplast (Monoject Scientific, Athy, Ireland). Serial mesiodistal sections of seven µm were prepared and stained with HE. The samples for cryosectioning and subsequent enzyme histochemical evaluations were rinsed in cold Tris-HCl buffer, containing 0.1 M Tris and 6% polyvinylpyrolidine (PVP) at pH 7.4. Then, the material was decalcified in cold 10% ethylenediamine-tetraacetic acid in Tris-HCl-PVP at pH 7.4. Decalcification was performed at 4°C, and the endpoint was determined by radiography (Philips Oralix). After 10 to 14 weeks of decalcification, the tissue was embedded in Tissue-Tek (Sakura, Zoeterwoude, The Netherlands) and kept at -80°C until sectioning. Serial mesiodistal sections of seven μ m were cut at -20° C on a cryostat microtome HM



FIGURE 3. Time-displacement curves of two dogs showing individual differences in the rate of tooth movement after the application of a force of 300 cN for 80 days. P indicates premolar; M, molar.

500 7 (Adamas Instrumenten, Leersum, The Netherlands). Selected sections were stained for alkaline phosphatase (AP) as a marker for active osteoblasts or tartrate-resistant acid phosphatase (TRAP) for differentiated osteoclasts and osteoclast precursors. For both enzymes, a modification of the staining techniques according to Van De Wijngaert and Burger¹⁸ were used.

RESULTS

All appliances were checked weekly, and at the time of sacrifice, all appliances were still in place and in good order. On the basis of the weekly measurements, the timedisplacement curves were constructed. The curves for most teeth could be divided into four phases of movement as has been described previously (Figure 2).¹⁶ In some curves, the transition from phase 1 to phase 2 was difficult to distinguish because of lack of data for the first phases. The rate of tooth movement showed large individual differences in the four phases after using low or high forces. As an example, in Figure 3, the time-displacement curves are given for two dogs, both killed after 80 days of tooth movement with 300 cN. In Figure 4, the time-displacement curves of a premolar and a molar in one dog, both moved by 25 cN at one side and 300 cN at the other, illustrate that the force level has no influence on the amount of tooth movement. In the following sections, the most common histological features will be described, after which the histological features will be related to the force level.

Histology

In the initial phase of tooth movement, after 24 hours of force application, cellular and tissue reactions had already started. At the pressure side, the fibers of the PDL were compressed (Figure 5B), whereas at the tension side the fibers were stretched (Figure 5A). Osteoclast and osteoblast



FIGURE 4. Time-displacement curves of a premolar and a molar in one dog moved by 25 cN at the left side and 300 cN at the right side for 80 days.

numbers were already increased at the pressure and tension side, respectively (not shown). The sections of teeth to which a high force (300 cN) was applied showed cell-free hyalinized areas of the PDL at the pressure side, mostly located in the cervical and apical part of the root. In the samples in which low forces were used, this phenomenon was less often present. TRAP and AP staining showed osteoclastic and osteoblastic activity, respectively (not shown).

In the second phase, the phase of arrest, several sections of both force levels showed areas of hyalinization at the pressure side after four and 20 days of force application (Figures 6 and 7). In these regions, distortion of the normal periodontal fiber arrangement was encountered. Deviating periodontal fiber arrangement was also seen in areas without apparent hyalinization. These areas were mostly not located at the pressure side proper but more to the buccal or to the lingual side. Some sections showed a normal periodontal structure, whereas in other sections the periodontal



FIGURE 5. (A) Photomicrograph showing the stretched fibers of the PDL of a second premolar at the tension side after a force of 25 cN was applied for 24 hours. HE, bar = 50 μ m; B indicates bone; PDL, periodontal ligament; and T, tooth. (B) Photomicrograph showing the compressed PDL at the pressure side of a second premolar after a force of 25 cN was applied for 24 hours. HE, bar = 50 μ m; T indicates tooth; PDL, periodontal ligament; and B, bone.



FIGURE 6. Photomicrograph showing focal hyalinization of the PDL at the pressure side of a second premolar to which a force of 300 cN was applied for four days. HE, bar = $50 \ \mu$ m; T indicates tooth; B, bone; H, hyalinization; OC, osteoclast; and R, undermining resorption.

FIGURE 7. Photomicrograph showing osteoclasts (OC) in the vicinity of a hyalinized area of the PDL at the pressure side of a second premolar to which a force of 25 cN was applied for 20 days. TRAP, bar = 50 μ m; T indicates tooth; B, bone; and H, hyalinization.

fibers were oriented parallel to the root or even completely disorganized. Adjacent to hyalinized areas, TRAP-positive cells were often found, either within the periodontal space or in the bone marrow cavities, related to direct or undermining resorption (Figure 7). At the end of the phase of arrest (after 20 days), the number of osteoclasts seemed to increase in both force groups in comparison with the start of that phase (after four days), but it was slightly higher after using high forces. At the tension side, the collagenous fibers connected the tooth to the bone, and an increased number of osteoblasts were arranged along the alveolar bone. In some cases, a thin layer of osteoid had deposited in which Sharpey's fibers were embedded. AP-positive cells were less prominent in the 25 cN group than in the 300 cN group (Figure 8). During the phase of arrest, root resorption was rare.

The third and fourth phases were reached after 40 and 80 days of orthodontic force application, respectively. In these acceleration and linear phases, pressure sides of teeth subjected to both forces showed collagenous fibers without a clear orientation and irregular bone surfaces due to direct bone resorption. At some pressure sides, however, hyalinized areas were still/again present. In these areas, the structure of the PDL was completely lost, and cells could not be distinguished. This phenomenon was slightly more prominent after using high forces (300 cN). The dimensions of these hyalinized areas differed considerably. Large areas covering 600 μ m of the length of the root surface (Figure 9) as well as focal patches measuring less than 150 μ m (Figure 10) were found. As in the earlier phases, these areas were mostly not located at the pressure side proper but more to the buccal or to the lingual side (Figure 11). Adjacent to the focal patches of hyalinization, a bone spicula was sometimes remaining (Figure 10). In general, an accumulation of osteoclasts was present in the vicinity of the hyalinized areas. These cells were in some samples involved in direct bone resorption but more often in root resorption. At the tension sides, bone deposition had taken place, and the bone surface was mainly covered with APpositive osteoblastic cells (Figure 8).



FIGURE 8. Photomicrograph showing osteoblasts (OB) covering the bone surface at the tension side of a second premolar to which a force of 300 cN was applied for 20 days. Alkaline phosphatase, bar = 50 μ m; T indicates tooth; B, bone.

FIGURE 9. Photomicrograph showing a large area of hyalinization within the PDL at the pressure side of a second premolar to which a force of 300 cN was applied for 40 days. The section was taken 100 μ m away from the central section lingually of the mesiodistal plane. HE, bar = 50 μ m; T indicates tooth; B, bone; and H, hyalinization.

Histological features and tooth movement

Direct osteoclastic bone resorption was mostly found at the pressure sides of relatively rapid moving teeth, independent of the amount of force. The tension sides of these teeth showed active bone deposition by osteoblastic cells. Focal sites of hyalinized tissue within the PDL were found in slow-moving teeth in all four phases of tooth movement. Sometimes, the PDL of a root of a slow-moving tooth did not show any hyalinization at all. In such a sample, the PDL at the pressure side contained almost no osteoclasts, and only very few osteoblasts were found at the tension side. However, the other root of such a tooth, which was evaluated separately, did always show focal hyalinization areas.

DISCUSSION

Experimental studies on tooth movement are often difficult to compare because of the use of different orthodontic appliances and different magnitudes, types, and duration of

forces.9 The studies of Storey and Smith² and Reitan^{19,20} on force levels and subsequent tissue reaction to orthodontic tooth movement were performed in humans (Table 1). That limited the duration of the experiments; analysis of the exact force levels and a histological evaluation of the changes in the surrounded alveolar bone were of course not possible. At the end of the past century, increasingly more research was done in animals.²¹⁻²⁶ Many previous studies on experimental tooth movement have been performed in rats using relatively high forces.²⁷⁻²⁹ However, the rat as an experimental model has disadvantages. Rodents have continuous eruption of the incisors and physiological distal drift of the molars. Continuous eruption of the incisors may affect the direction of the applied force because incisors are often used as the anchorage unit in these experiments. Distal drift might camouflage the amount of real tooth movement. Furthermore, a rat molar is about 60 times smaller than a human molar; thus, even a force in the range of 5 cN has to be considered a high one. By using the beagle dog model, we could overcome these problems. To avoid



FIGURE 10. Photomicrograph showing focal hyalinization of the PDL and a remaining bone spicula at the pressure side of a second premolar to which a force of 25 cN was applied for 40 days. The section was taken 150 μ m away from the central section lingually of the mesiodistal plane. HE, bar = 50 μ m; T indicates tooth; B, bone; H, hyalinization; BS, bone specula; and OC, osteoclast.

tipping movements caused by the use of elastics or twistflex sectionals,⁶ an appliance was developed that made bodily tooth movement possible. This appliance required the use of an anchorage unit. The results of two studies with this appliance showed that the mean loss of anchorage was about 25%.^{16,17} Therefore, in the present study, the appliance was modified by adding an implant as an anchorage unit.

In the past, many authors have described the formation of cell-free, necrotic areas of the PDL in the arrest phase (second phase of the time-displacement curve) as a result of localized ischemia.^{4,10,19} They reported that after excessive compression of the PDL, the blood supply is cut off, which leads to hyalinized areas and to an arrest of tooth movement. Only removal of the necrotic tissue and bone resorption from the adjacent marrow space allow a resumption of tooth displacement. In this process of undermining resorption, phagocytic cells such as macrophages, foreign body giant cells, and osteoclasts invade from the adjacent undamaged areas and eliminate the hyalinized tissue at the pressure side.^{4,10,30–32} In the absence of necrotic areas, fibroblast- and cementoblast-like cells start the bone



FIGURE 11. Schematic drawing of a horizontal section of a premolar. The horizontal arrow marks the central plane and the direction of tooth movement; the four small arrows indicate the localization of the hyalinization areas, which were mainly found buccally or lingually of the root.

remodeling process at the tension side, and the rate of tooth movement increases.^{11,13}

The outcomes of our experiment are contradictory to this commonly accepted theory. Hyalinization is not only found in the phase of arrest, between four and 20 days of force application but also after 40 and 80 days of tooth movement. This suggests that the development and removal of necrotic tissue is a continuous process during tooth displacement instead of a single event. One of the reasons why research carried out in the past leads to different conclusions could be the short duration of these experiments in which the acceleration phase was not even reached. Furthermore, in our study, the location of hyalinized zones was different from those of earlier reports. Most hyalinized areas were not found in the area of the central plane but lingually and buccally from it (Figure 11). An explanation for this contrasting outcome may be found in the way the sections were cut. In nearly all previous studies, the teeth, which were moved, were sectioned vertically through the central plane, or only a few sections from the entire tooth were evaluated.^{4,16,33} However, in the present study, the teeth were serially sectioned in seven-µm slices from the lingual to the buccal side. The formation of necrotic tissue lingually and buccally from the central plane is probably the consequence of local stress and shear concentrations caused also by local irregularities in bone morphology. Epker and Frost³⁴ suggested a correlation between physical loads and osteoblast or osteoclast activities at the bone surface and further postulated that strain is the major biomechanical factor influencing cell behavior. The amount of bending of the bone could stimulate either formation or resorption.

Taking this statement as a base for further investigations, Melsen¹³ hypothesized that bone apposition appears as a reaction to bending of the alveolar wall in the tension zone, caused by the stretching of the PDL fibers. She further hypothesized that indirect resorption at the pressure side is not a reaction to force but an attempt to remove ischemic bone lying adjacent to the hyalinized tissue. The subsequent direct resorption could be considered a part of the remodeling process. According to these suggestions, the bending of the alveolar bone lingually and buccally as a reaction to the orthodontic forces could induce localized hyalinization to appear in these areas. Simultaneously, the lowering of the normal stress on the PDL fibers at the central plane of the root leads to direct bone resorption. The findings of the present study support this hypothesis, but more research is still required.

In the past, much research has been performed on the relationship between force magnitude and tooth movement without finding any correlation.^{17,35,36} Owman-Moll et al³⁷ even doubled the applied force but did not find twice as much tooth displacement. Van Leeuwen et al¹⁷ applied different force levels in a split-mouth design. They found large individual differences, but they could not find a correlation between force magnitude and tooth movement. The outcomes of the present study confirm this observation. Teeth on which high forces (300 cN) were applied did not move faster than the ones displaced by low forces (25 cN). However, teeth on which a higher force level was applied showed hyalinization slightly more often, and hyalinization was found in both force groups throughout the whole period. Although the appearance of necrotic tissue might be related to force magnitude, this seems to have no significance for the rate of tooth movement. This means that once tooth movement has started, bone remodeling takes place at a certain rate, independent of force magnitude. Furthermore, the data show that individual variation is large. Differences in bone metabolic capacity could be responsible for this phenomenon. Bone density, morphological differences, and genetic factors could also influence the remodeling process and subsequent tooth movement.¹⁶

CONCLUSIONS

In contrast to earlier research, the present study shows that hyalinization of the PDL can appear at any time during the whole experimental period—from 24 hours up to 80 days of force application. The localization of hyalinization is mostly buccally or lingually of the mesiodistal plane, with a large variation in size. Hyalinization limits tooth movement, but there is no relationship with the force level.

REFERENCES

- Burstone CJ. The biophysics of bone remodeling during orthodontics. In: Norton LA, Burstone CJ, eds. *The Biology of Tooth Movement*. Boca Raton, Fla: CRC Press; 1989:321–334.
- Storey E, Smith R. Force in orthodontics and its relation to tooth movement. Aust J Dent. 1952;56:11–18.
- Jarabak JR, Fizzell JA. *Technique and Treatment with Light-Wire Edgewise Appliances*. Vol 1, 2nd ed. St Louis, Mo: CV Mosby. 1972:353.
- Rygh P. Ultrastructural changes in pressure zones of human periodontium to orthodontic tooth movement. Acta Odontol Scand. 1973;31:109–22.
- Reitan K. Some factors determining the evaluation of forces in orthodontics. Am J Orthod. 1957;43:32–45.
- Boester CH, Johnston LE. A clinical investigation of the concepts of differential and optimal force in canine retraction. *Angle Orthod.* 1975;44:113–119.
- Quinn R, Yoshikawa D. A reassessment of force magnitude in orthodontics. Am J Orthod. 1985;88:252–260.

- 8. Lee BW. The force requirements for tooth movement. Part 1: tipping and bodily movement. *Aust Orthod J.* 1995;13:238–248.
- Ren Y, Maltha JC, Kuijpers-Jagtman AM. Optimum force magnitude for orthodontic tooth movement: a systematic literature review. *Angle Orthod.* 2003;73:86–92.
- Proffit WR. Biomechanics and mechanics. In: *Contemporary Orthodontics*. 3rd ed. St Louis, Mo: CV Mosby; 2000:298–305.
- Thilander B, Rygh P, Reitan K. Tissue reactions in orthodontics. In: Graber TG, ed. Orthodontics, Current Principles and Techniques. 3rd ed. St. Louis, Mo: CV Mosby; 2000:117–156.
- 12. Middleton J, Jones M, Wilson A. The role of the periodontal ligament in bone remodeling, the initial development of a time dependent finite element model. *Am J Orthod Dentofacial Orthop.* 1996;109:155–162.
- Melsen B. Biological reaction of alveolar bone to orthodontic tooth movement. Angle Orthod. 1999;69:151–158.
- Hujoel PP. A meta-analysis of normal ranges for root surface areas of the permanent dentition. *J Clin Periodont*. 1994;21:225– 229.
- Van Driel WD, Van Leeuwen EJ, Von den Hoff JW, Maltha JC, Kuijpers-Jagtman AM. Time-dependent mechanical behavior of the periodontal ligament. *Proc Inst Mech Eng.* 2000;241:497– 504.
- Pilon JJGM, Kuijpers-Jagtman AM, Maltha JC. Magnitude of orthodontic forces and rate of bodily tooth movement: an experimental study in beagle dogs. *Am J Orthod Dentofacial Orthop*. 1996;110:16–23.
- Van Leeuwen EJ, Maltha JC, Kuijpers-Jagtman AM. Tooth movement with light continuous and discontinuous forces in Beagle dogs. *Eur J Oral Sci.* 1999;107:468–474.
- Van De Wijngaert FP, Burger EH. Demonstration of tartrate-resistent acid phosphatse in undecalcified, glycolmethacrylate embedded mouse bone: a possible marker for (pre)osteoclast identification. J Histochem Cytochem. 1986;34:1317–1323.
- Reitan K. Clinical and histological observations on tooth movement during and after orthodontic treatment. *Am J Orthod.* 1967; 53:721–745.
- Reitan K. Tissue behaviour during orthodontic tooth movement. Am J Orthod. 1960;46:881–900.
- Lee BW. Relationship between tooth-movement rate and estimated pressure applied. J Dent Res. 1964;44:1053.
- Gibson JM, King GJ, Keeling SD. Long-term orthodontic tooth movement response to short-term force in the rat. *Angle Orthod*. 1992;62:211–215.
- Konoo T, Kim YJ, Gu GM, King GJ. Intermittent force in orthodontic tooth movement. J Dent Res. 2001;80:457–60.
- Verna C, Dalstra M, Melsen B. The rate and the type of orthodontic tooth movement is influenced by turnover in a rat model. *Eur J Orthod.* 2000;22:343–352.
- Rodriguez L, Steimetz T, Ubios AM, Cabrini RL. An original orthodontic appliance for experimental mesial movements in rats. *Acta Odontol Latinoam.* 1996;9:45–49.
- Hixon EH, Atiksan H, Callow GE, McDonald HW. Optimal force, differential force, and anchorage. Am J Orthod. 1969;55:437–57.
- Katona TR, Paydar NH, Akay HU, Roberts WE. Stress analysis of bone modeling response to rat molar orthodontics. *J Biomech*. 1995;28:27–38.
- Ohyama N, Yamaguchi S. Effects of phenylephrine and prazosin on axial movement of the rat incisor and arterial blood pressure. *Jpn J Pharmacol.* 1999;80:271–274.
- Row KL, Johnson RB. Distribution of 3H-proline within transseptal fibers of the rat following release of orthodontic forces. *Am J Anat.* 1990;189:179–188.
- Sandstedt C. Einige Beiträge zur Theorie der Zahnregulierung. Nord Tandlæk Tidskr. 1904;5:236–256.
- 31. Hirashita A, Noda K, Kaida K, Nakamura Y, Kuwabara Y. Phago-

cytosis of collagen by fibroblasts incident to experimental tooth movement. *Arch Histol Jpn.* 1985;48:149–158.

- 32. Cooper SM, Sim MR. Evidence of acute inflammation in the periodontal ligament subsequent to orthodontic tooth movement in rats. *Aust Orthod J.* 1989;11:107–109.
- Owman-Moll P. Orthodontic tooth movement and root resorption with special reference to force magnitude and duration. Swed Dent J. 1995; Suppl 105.
- Epker BN, Frost HM. Correlation of bone resorption and formation with the physical behavior of loaded bone. J Dent Res. 1965;44:33–41.
- 35. Iwasaki LR, Haack JE, Nickel JC, Morton J. Human tooth move-

ment in response to continuous stress of low magnitude. Am J Orthod Dentofacial Orthop. 2000;117:175–183.

- 36. Tong Y. Forced tooth movement in rats and its histological changes. *Zhonghua Kou Qiang Za Zhi*. 1990;25:268–270.
- Owman-Moll P, Kurol J, Lundgren D. Effects of a doubled orthodontic force magnitude on tooth movement and root resorption. *Eur J Orthod.* 1996;18:141–150.
- King GJ, Keeling SD, McCoy EA, Ward TH. Measuring dental drift and orthodontic tooth movement in response to various forces in adult rats. *Am J Orthod Dentofacial Orthop.* 1991;99:456– 465.
- 39. Lee BW. The force requirements for tooth movement, part 1: tipping and bodily movement. *Aust Orthod J.* 1995;13:238–248.