Region-Specific Mechanical Adaptation of the Periodontal Ligament During Orthodontic Tooth Movement

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INTRODUCTION: The periodontal ligament (PDL) connects the tooth to the jaw, stabilizing the tooth and dissipating loads to the alveolar bone during the high repetitive loads experienced with mastication. Despite its importance for tooth function, the mechanisms of adaptation and associated mechanical properties of the PDL remain poorly understood. We previously observed mechanical heterogeneity throughout the PDL, characterizing the dense collar as critical for tooth stabilization while the softer furcation region resists vertical loads [1]. However, the impact of structural changes, such as during orthodontic tooth movement (OTM), on PDL function has yet to be explored in detail. A comprehensive understanding of the PDL adaptation to altered loads with OTM is required to guide orthodontic treatment design and prevent adverse effects of OTM, such as periodontal inflammation and excessive bone resorption. Thus, the purpose of this study was to investigate the mechanical changes of the PDL in response to OTM. We hypothesized that OTM would destabilize the tooth and therefore cause a softening of the PDL, particularly in the dense collar region.

METHODS: OTM devices were implanted into nine-week-old male and female C57BL/6J mice. Briefly, the mice were anesthetized, and an aluminum wire with a nickel-titanium coil was attached to the lower first molar and lower incisor using a flowable composite resin, creating a tensile force pulling the molar towards the incisor [2]. Mice in the orthodontic movement ('Ortho') groups were sacrificed after 3 or 7 days with the device attached. Additional mice were allowed to recover for 7 days post-removal and then euthanized ('Relapse'). Sectioned hemi-mandible explants were prepared as described for high-frequency nanorheology [1]. Nanoindentation of the PDL at 29 different locations across the furcation and four areas of the dense collar region (mesial-buccal, mesial-lingual, distal-buccal, and distal-lingual) in triplicate. Each indentation consisted of a load-controlled ramp-and-hold followed by a superimposed dynamic protocol (8-12 nm amplitude) ranging from 1 Hz to 10 kHz. The effective indentation modulus was computed from the initial indentation using a Hertzian contact mechanics model. A low-frequency dynamic modulus, a high-frequency dynamic modulus, and a self-stiffening ratio were computed. Each indentation trial was compartmentalized into one of four tissue categories (ligament-like (soft), bone-like (bone), cartilage-like, or "unknown") based on the moduli, phase angle response, and self-stiffening ratios [1]. Kruskal-Wallis tests followed by Dunn's multiple comparisons tests were performed to compare between groups of a given region. A significance level of $p<0.05$ was used for all statistical tests.

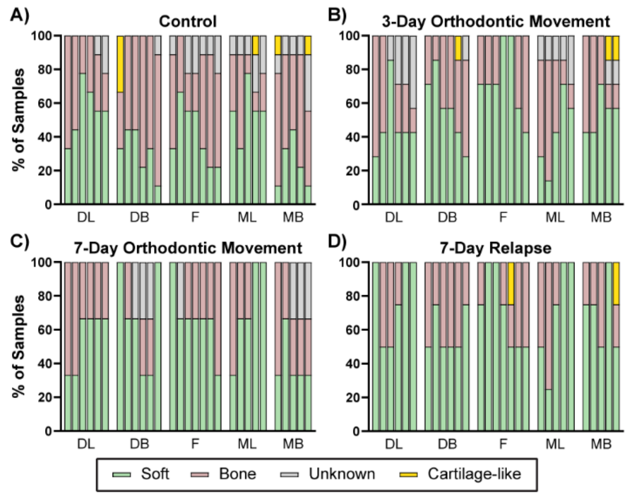

Figure 1. The relative percentage of each tissue type at each indentation location. Each column represents one location indented in the distal-lingual (DL), distal-buccal (DB), furcation (F), mesial-lingual (ML), or mesial-buccal (MB) region of the first molar.

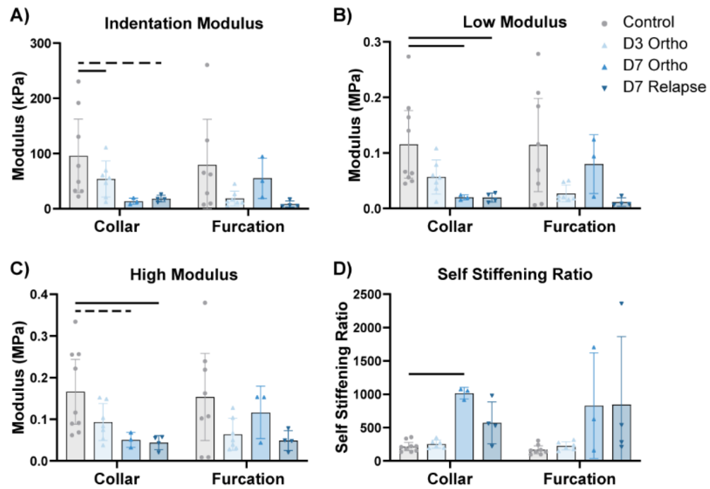

Figure 2. Comparisons of the (A) indentation modulus, (B) low frequency modulus, (C) high frequency modulus, and (D) self stiffening ratio of soft points within the furcation and dense collar regions of the periodontal ligament. Data is plotted as mean +/- 95% CI. Significant differences between experimental groups are indicated with a solid line (--, $p<0.05$) or dotted line (- -, $p<0.1$).

RESULTS: The percentage of indentations characterized as 'soft tissue' was similar to that previously reported in healthy control mice (Fig. 1A) [1]. This percentage increased within the 3 days of OTM and remained elevated after 7 days of relapse (Fig. 1B-D). While significant decreases in the indentation modulus of the dense collar region (Fig. 2A) were observed by the third day of OTM, decreases in the low- (Fig. 2B) and high-frequency dynamic modulus (Fig. 2C) were not observed until 7 days of OTM. Interestingly, a significant increase in the self-stiffening ratio of the dense collar was observed after 7 days of OTM, but this value returned towards the control value after 7 days of relapse (Fig. 2D). No significant differences in mechanical properties were observed in the furcation region of the PDL (Fig. 2A-D).

DISCUSSION: Our data demonstrate that the dense collar region of the PDL, but not the furcation region, softens with OTM. This suggests remodeling of the collar in response to active OTM as well as during the relapse phase, destabilizing the tooth to enable tooth translation. Previous studies have observed elevated matrix metalloproteinases during early phase responses [3], supporting this idea. The continued recovery of the PDL mechanical properties even 7 days after removal of the device suggests a longer timeframe for recovery than the initial movement. This could be due to mechanosensitive feedback circuitry between cells and their ECM; a softer matrix may not activate cells to produce ECM as rapidly [4]. OTM may also increase the susceptibility of the PDL to bacterial penetration and infection, as our previous work demonstrated an association between softer PDL regions and increased permeability [1]. Data were pooled from both sexes to characterize population trends, so it's possible that sex differences in PDL adaptation exist; future studies will explore this. In addition, future studies aim to investigate mechanisms of PDL remodeling following OTM via measurement of matrix turnover, as well as compositional and structural changes.

SIGNIFICANCE/CLINICAL RELEVANCE: Findings from this study provide important insights into tissue-level mechanical changes of the PDL in response to OTM, which may inform the design of orthodontic treatment aimed at optimizing tooth movement while minimizing adverse effects.

REFERENCES: [1] Connizzo+Naveh 2020, *J. Biomech.*, 111:109996 [2] Xu+ 2021, *J. Vis. Exp.*, 170:e62149 [3] Almeida+ 2015, *Angle Orthod.*, 85(6):1009-1014 [4] Tiskratok+ 2023, *Sci. Rep.*, 13(1):1358

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