**Title:** Laboratory Assessment Of Crispr-Mediated Modulation Of Osteoblastic And Osteoclastic Gene Expression Under Simulated Orthodontic Force

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**Abstract:** Introduction: Orthodontic tooth movement is governed by coordinated bone resorption and formation mediated primarily through the Receptor Activator of Nuclear Factor-κB Ligand – Osteoprotegerin signaling axis. Although mechanical force initiates this process, the biological rate of remodeling remains a limiting factor. CRISPR-based transcriptional activation presents a novel strategy to amplify force-induced molecular responses. Material and Methods: Human periodontal ligament stem cells were exposed to simulated compressive orthodontic force (2 g/cm²) and subjected to CRISPR-dCas9-VPR–mediated activation of the TNFSF11 (RANKL) promoter. Samples were divided into control, force-only, scramble control, and CRISPR-RANKL groups. Cell viability was assessed using CCK-8 assay, while gene and protein expression of RANKL, OPG, and RUNX2 were evaluated using RT-qPCR and ELISA. Results: Cell viability exceeded 90% across all groups, indicating no cytotoxic effects. CRISPR-mediated activation significantly enhanced RANKL expression under compressive force, producing a marked increase in the RANKL/OPG ratio compared with force alone ($p < 0.001$). RUNX2 expression was reduced under compression, consistent with osteoclastic dominance, and was unaffected by CRISPR modulation. Conclusion: CRISPR-dCas9-VPR–based activation of RANKL synergistically augments mechanical force–induced osteoclastic signaling in periodontal ligament stem cells. This proofof-concept study highlights the potential of epigenetic modulation as a precision approach for biologically accelerating orthodontic tooth movement.

**Keywords**: CRISPR-Cas9; Orthodontic tooth movement; RANKL; Osteoclastogenesis; Periodontal ligament stem cells; Gene activation

DOI: https://doi.org/10.56936/18290825-2026.20v.2-11