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The role of Yes-associated protein on cell fate decisions of human periodontal ligament cells under compressive force loading

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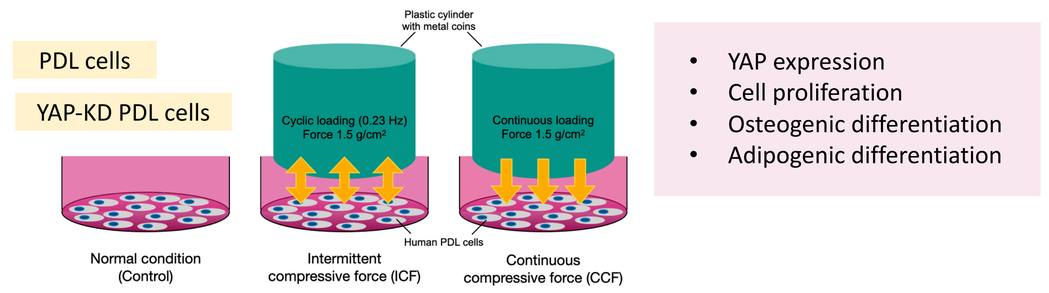
Background and Objective

During tooth movement, mechanical stimuli has been known to influence self-renewal and differentiation fates of periodontal ligament (PDL) cells[1]. In response to mechanical stimuli, the downstream regulator of Hippo pathway, Yes-Associated Protein (YAP) has been identified as a mechanosensitive transcriptional activator that control cellular activities, including cell proliferation and differentiation[2]. However, the role of YAP under compressive force (CF) on the cell fate decision of human PDL cells has not yet been examined. In this study, we aim to identify the role of YAP in PDL cells under CF treatment.

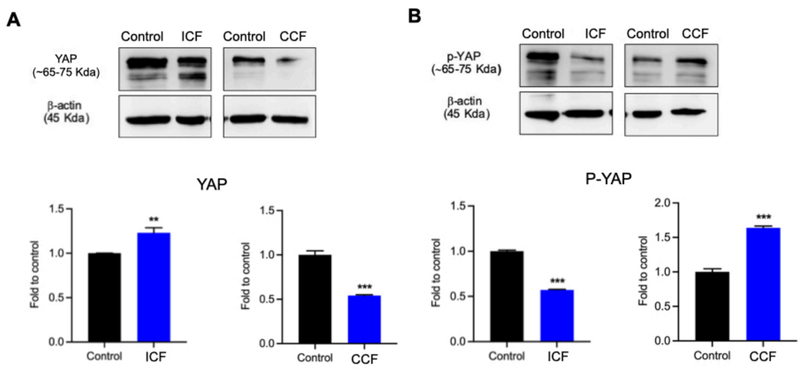
Materials and Methods

- Human PDL cells were derived form PDL tissues of healthy extracted third molars and were cultured in vitro for stem cell characterization.
- YAP expression was determined in human PDL cells treated with intermittent compressive force (ICF) and continuous compressive force (CCF).
- YAP expression was silenced by shRNA. The effects of YAP on cell proliferation, adipogenesis and osteogenesis of PDL cells under ICF loading were determined.



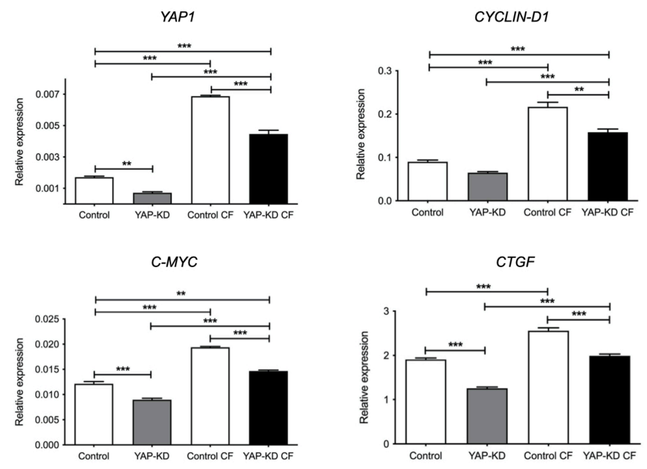
Results

Figure 1



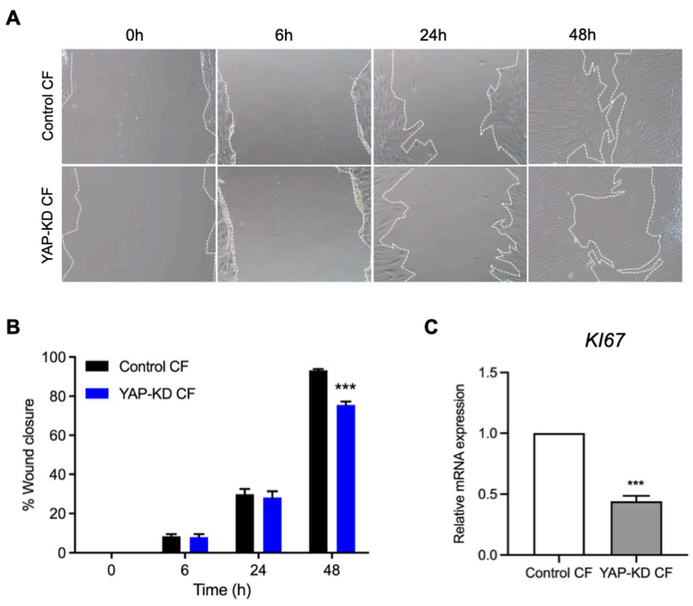
(A) The active form of YAP increased with ICF, whereas it decreased with CCF.
(B) The inactive form of phosphorylated YAP (p-YAP) decreased with ICF, while it increased with CCF.

Figure 2



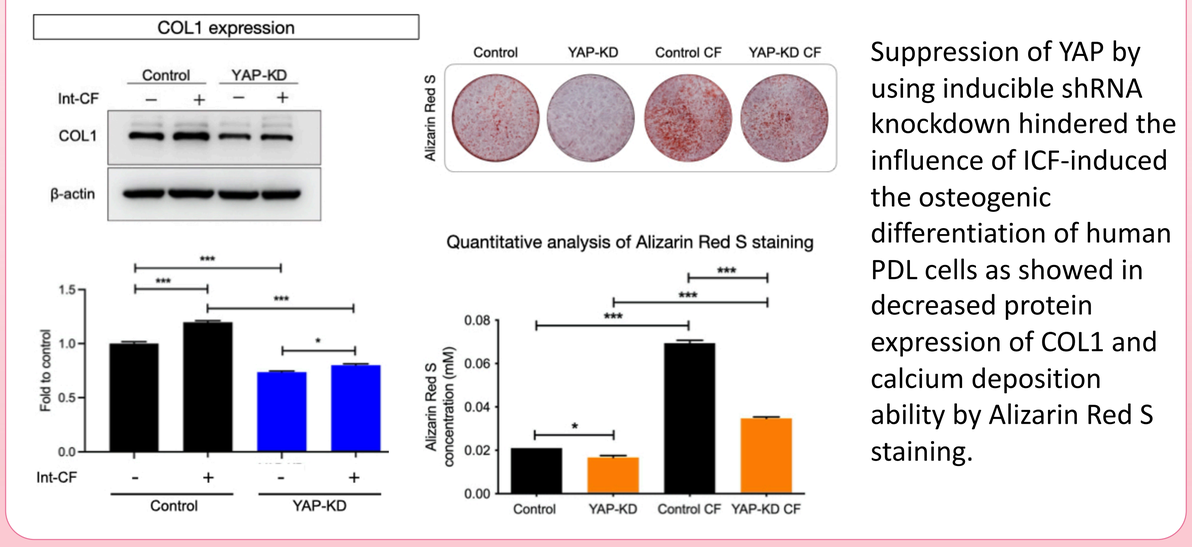
ICF could activate YAP activity and its downstream target genes, which commonly contribute to cell cycle and proliferation.
The mRNA expression levels of *YAP*, *CYCLIN-D1*, *C-MYC*, and *CTGF* were found to be downregulated in both the YAP-KD cells and ICF-treated YAP-KD cells (YAP-KD CF), compared with the control and the ICF-treated control (control CF) groups.

Figure 3



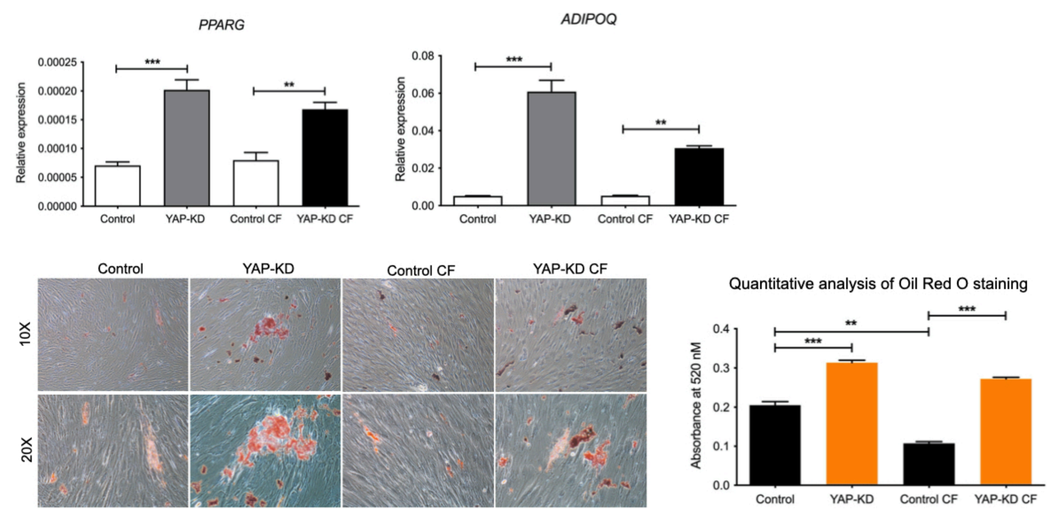
Silencing of YAP decreased cell proliferation of human PDL cells under ICF loading .
(A-B) The YAP-KD CF cells presented a lower percentage of wound reduction than the control CF cells.
(C) A qRT-PCR analysis used to determine the mRNA transcript levels of the proliferation marker KI67 in YAP-KD CF cells revealed that the levels had significantly decreased, relative to those of the control CF.

Figure 4



Suppression of YAP by using inducible shRNA knockdown hindered the influence of ICF-induced the osteogenic differentiation of human PDL cells as showed in decreased protein expression of COL1 and calcium deposition ability by Alizarin Red S staining.

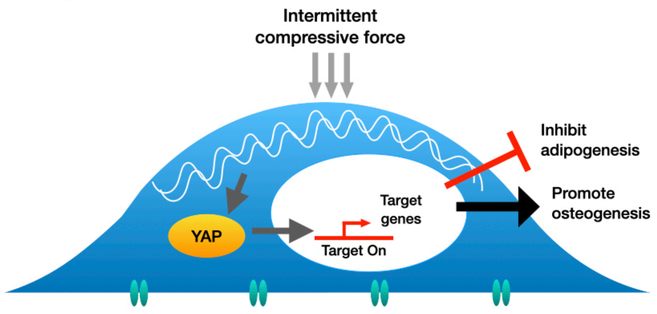
Figure 5



YAP suppression showed the potential to drive PDL cells into adipogenic lineage as showed in upregulation of *PPARG* and *ADIPOQ* gene expression and lipid accumulation by Oil Red O staining.

Discussion and Conclusion

- YAP has also been signified as a key regulator in a mechanotransduction process to control cellular activities in response to mechanical stimuli[3].
- Previous study showed that ICF could enhance cell proliferation and osteogenic differentiation of human PDL cells[4].
- ICF could enhance cell proliferation and osteogenic differentiation of human PDL cells via YAP signaling cascade.
- Suppression of YAP hindered the influence of ICF-induced cell proliferation, and the osteogenic differentiation, while promote adipogenic differentiation in human PDL cells.



"YAP could be a mechanosensing regulator response to ICF that require for cell proliferation and osteogenic differentiation capacities of human PDL cells."

Our findings may provide the possibility to facilitate periodontal tissue regeneration by manipulation of Hippo-YAP signaling pathway.

Acknowledgements

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