Bone fragility – the impaired ability of bone to resist fractures – is determined by bone mass and quality, with bone quality shaped by parameters such as porosity, geometry, and the material properties of the ECM. Osteocytes maintain bone quality through perilacunar/canalicular remodeling (PLR), a process by which local matrix is resorbed and replaced and regulated by secreted enzymes and cytokines such as Transforming Growth Factor Beta (TGF-β). TGF-β signaling is partly regulated by a cytoskeletal tension that is calibrated by features of the cellular microenvironment such as cell shape and substrate stiffness. Here, we probe the effects of PLR suppression and aging, a known cause of bone fragility, on osteocyte cytoskeletal tension. Young (4-month) and old (12-month) mice with an osteocyte specific ablation of TGF-β Receptor II (TβRIIocy−/−) were used in this study. Femoral (young) and tibial (old) bones were sectioned to 10µm, fixed and demineralized, and stained for yes-associated protein (YAP), a well-established tension sensitive marker. In the young group, the percentage of YAP expressing cells is lower in TβRIIocy−/− than in WT bone, with not differences detected in old samples. In TβRIIocy−/− sections, the percentage of osteocytes expressing YAP in the old group significantly increased compared to the young. These results suggest the potential involvement of multiple independent mechanisms controlling cytoskeletal tension and YAP levels in osteocytes, providing new insights into the coupling between the mechanical and biological microenvironments in osteocytes and their functional perturbations that contribute to bone fragility.

Figure 1: Osteocytic YAP expression decreases with PLR and aging. (A) YAP expression is largely localized to the cytoplasm in all conditions (white stars in inset marked by dashed white box). (B) the percentage of YAP-positive cells is significantly reduced with PLR suppression in the young samples, with not significant differences detected between conditions in the old group. In the TβRIIocy−/−, there is a large increase in the percentage of YAP-positive cells with age. * indicates significant with respect to WT young, # indicates significant with respect to TβRIIocy−/− young. Blue: DAPI, Red: YAP (Alexa 594) Scale bar 50µm.