Immunomodulator control of Myofibroblast Plasticity & Nuclei Morphology

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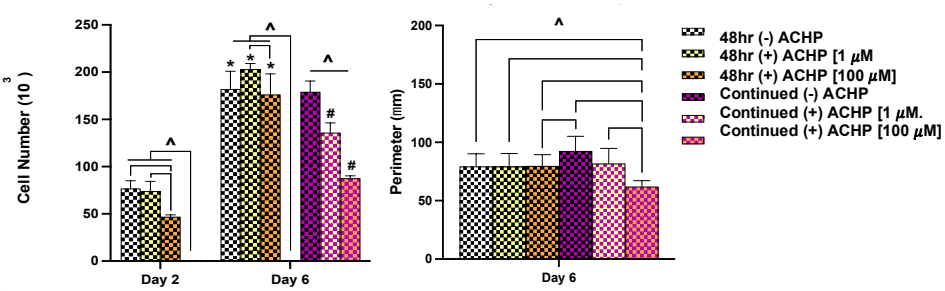
Introduction: Scarring is associated with extra-cellular matrix (ECM) dysregulation[1] and myofibroblast activation and persistence.[2] Myofibroblasts are a contractile pro-fibrotic cell type critical for early remodeling and deposition of predominantly type I collagen after injury in adult healing soft tissues.[1,2,3] The small molecule IKKβ inhibitor, 2-Amino-6-[2-(cyclopropylmethoxy)-6-hydroxyphenyl]-4-(4-piperidinyl)-3 pyridinecarbonitrile (ACHP) blocks IKKβ, which shuts down the inflammatory arm of the NF-κB signaling pathway.[6]

Methods: We did an in vitro experiment and cultured our tgfb-1 stimulated lung fibroblasts over the course of 6 days and assessed the cell behavior through viability and proliferation and their morphology and phenotype through a-sma expression and phospho-p65 immunofluorescence. We quantified DAPI fluorescence to characterize nuclei morphology.

Results: We found between day 2 and 6 cells proliferated to about twice their size and at both timepoints most did not survive the 100um dosage. On day 6 we noticed a the continued tgfb-1 stimulation has also produced an increase in a-sma, NF-kb activation and nuclear size.



Conclusions: We demonstrated that the immunomodulator ACHP successfully targets the NF-**κ**B inflammatory pathway and that 10 μM is the maximum tolerated dose that demonstrates the greatest effect for controlling myofibroblast plasticity. In addition, reduction in myofibroblasts as a result of ACHP treatment was associated with reduced nuclei size. Overall, ACHP Treatment offers a potential anti-fibrotic strategy as it decreases myofibroblast persistence *in vitro*

References:

1. Pakshir, Matrix Biol. 2018;68:81-93.
2. Hinz, EER. 2016;142:56-70.
3. Martin P. Science. 1997;276:75-81
4. Golman et al., Am J Sports Med. 2021;49(3):780-789

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