Stress Induced Ligamentum Flavum Hypertrophy is Inhibited via ROCK Mechanotransduction Modulation

Sami Shaikh[1,2], Eric Esposito[1], Muhamed Hadzipasic[1], Ian Connolly[1], Ryan Burns[1], Joseph Bradley[1], Margaret S. Sten[1], Ganesh M. Shankar[1].

Affiliations:

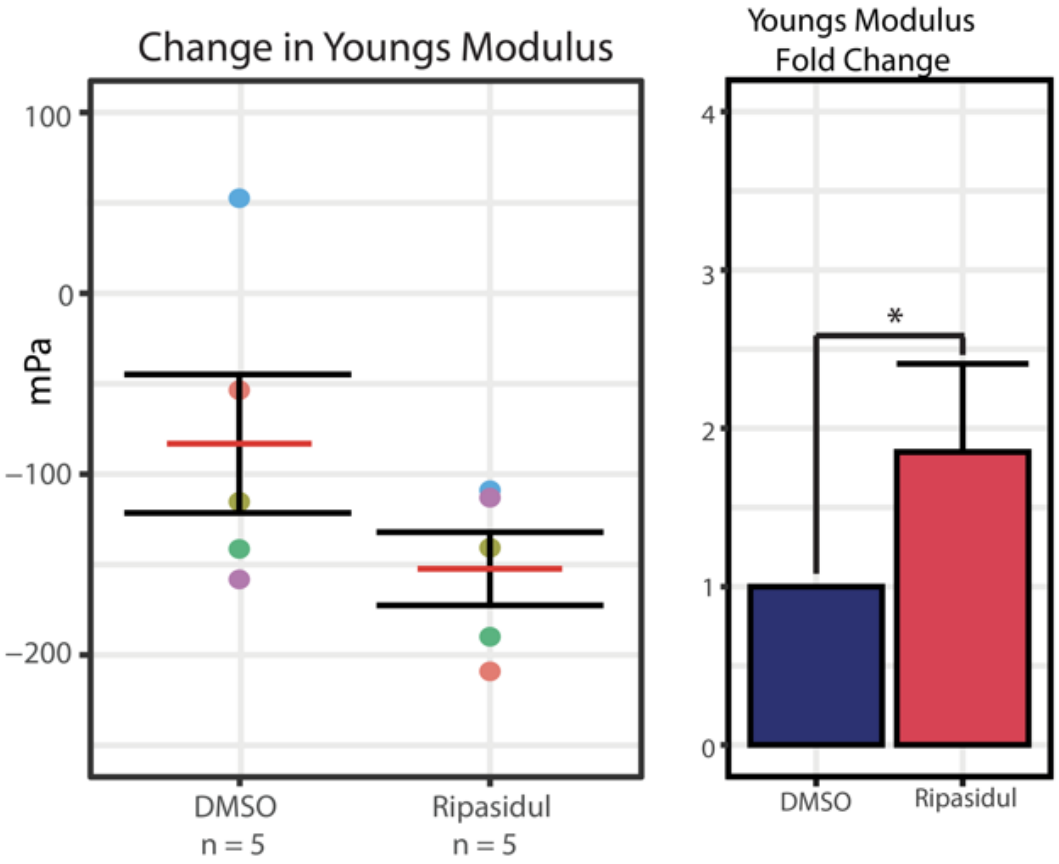
1. Department of Neurosurgery, Massachusetts General Hospital, Harvard Medical School; 2114, Massachusetts, USA
2. Drexel University College of Medicine, 60 N 36th St, Philadelphia, USA

Introduction: Ligamentum Flavum (LF) runs between the lamina of adjacent vertebra extending from the cervical spine to the lumbar region. Mechanical stress due to aging or trauma may result in LF hypertrophy, rigidity, and subsequent radiculopathy via spinal stenosis. Surgical intervention has been the mainstay treatment for decompression; however, there are currently no known medical interventions for ligamentum flavum hypertrophy. Activation of the ROCK kinase pathway promotes macroscale fibrosis, thereby stiffening tissue. Therefore, we propose that inhibition of the ROCK kinase pathway may prevent ligamentum flavum hypertrophy.

Methods: We subjected hypertrophic LF samples from surgical patients to a cyclical strain and measured the change in Young Modulus (YM) as a proxy for tissue stiffness in fascicles incubated with either ripasudil or DMSO. Tissues were then stained for smooth muscle actin (SMA) and cells were visualized under confocal microscopy. Of the five trials, a one-sided Wilcox on-Rank Sum Test was used to evaluate significance between the average fold change in YM between treated and control samples.

Results: LF fascicles treated with Ripasudil had an average change in YM of -152.35mPa ± 20.24 (SE); meanwhile, control fascicles had a lower average change of -83mPa ± 38.31 (SE). Therefore, YM of LF samples treated with Ripasudil were 1.83-fold greater than control, p=0.047 (Table 1). Under confocal microscopy, hypertrophic LF fascicles stretched in DMSO recruited greater SMA myofibroblasts compared to fascicles stretched in Ripasudil. Unstretched samples showed no recruitment of SMA cells.

Conclusion: Inhibition of ROCK mechanotransduction and associated fibrosis showed a marked decrease in LF stiffness, indicated by the greater change in YM. Further workup is essential to explore the use of ROCK inhibitors as an adjunct to the care plan of patients with degenerative spine disease who may seek an alternative and/or may be contraindicated to surgery due to comorbidities.



Mean change in Youngs Modulus for fasicles incubated in either DMSO or Ripasidul. Fold change quantification of change in Youngs Modulus with error bars denoting Standard Error. Statistical significance was evaluated using a one-sided Wilcox on-Rank Sum Test (*p = 0.047)

Table 1.