Project Title

**Feel the Pressure: Effects of Mechanical Deformation on Collagen Direction and Proliferation**

### Objectives
Examine the effects of periodic, compressive mechanical loading administered via a mechanical pressure device on collagen fiber orientation and fibroblast proliferation.

### Methods
The device used to apply mechanical stress to the collagen was built and designed using a 3D printing program and a 3D printer. Type 1 Collagen gel and NIH3T3 fibroblasts were added to 4 wells in an 8-well cell dish, and only Type 1 Collagen gel was added to the other 4 wells in the dish. Mechanical stress was applied to all eight wells. Reflectance confocal microscopy was used to analyze collagen direction before and after mechanical loading. Bright-field microscopy was used to analyze the number of cells in each well before and after mechanical loading. All imaging was conducted using an Olympus Fluoview 1000 microscope.

### Results
In wells that underwent mechanical stimulation, the average number of cells in the four wells over the course of the experiment were 812, 266, 587, and 621. The average number of cells in the other four wells (the ones that did not receive mechanical stimulation over the course of the experiment) were 18, 90, 348, and 72. The number of cells in wells that underwent mechanical stimulation was approximately 13.6 times greater than the number of cells in wells that did not undergo mechanical stimulation. The orientation of the collagen fibers after mechanical loading changed by approximately 38 degrees in wells with and without fibroblast cells. Collagen direction was determined by the Sobel filter and Gaussian models. The Sobel filter is a method of edge detection conducted by convolving bi-directional kernels with an image. Gaussian models use a non-parametric approach to implement a covariance matrix and a mean function to fit the data as smoothly as possible to a multivariate distribution over possible functions from a bivariate, normal distribution.

### Conclusions
The data collected from this investigation indicates that the change in collagen fiber orientation is not dependent on the presence of fibroblast cells in the wells. In addition, this shows that fibroblast proliferation- and consequently, collagen production- increase significantly after mechanical stress. The number of fibroblasts in wells with and without mechanical stimulation were compared using a one-way ANOVA test. The p-value was less than 0.05 (0.0174), rejecting the null hypothesis that there is no significant difference in cell proliferation after mechanical loading.

### Summary Statement
I found the effects of mechanical stress on collagen fiber orientation and fibroblast proliferation using a 3D-printed mechanical pressure device.

### Help Received
The experiment and data analysis were conducted independently, with the supervision of Dr. Michelle Digman from the University of California, Irvine.