TECH REPORT

103:

StageFlexer®

Quantification of strain at the membrane level

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Culturing Cells in a Mechanically Active Environment™
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INTRODUCTION

The StageFlexer® is designed to allow the user to view cell stretching activity under a microscope (Fig. 1). The cell growth area is part of a treated 42 mm diameter silicone membrane. The membrane is clamped and sealed above a small cylindrical vacuum chamber into which the membrane is pulled to apply strain to the cells growing on the silicone surface. The membrane can be deformed freely in an open chamber or over a Loading Station™ to apply uniform biaxial strain to the cells. The Loading Station™ comes in three different diameters (25 mm, 28 mm, and 31 mm) for varied growth surface area. The StageFlexer® can be run with the Flexcell® Tension System to stretch the cells under specific regimens while they are being viewed under a microscope.

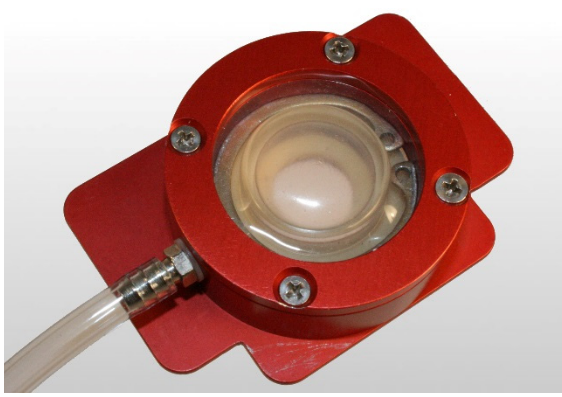

Figure 1. *A StageFlexer® microscopy strain device.*

STAGEFLEXER® LOADING STATIONS™

The StageFlexer® Loading Station™ is designed to provide uniform radial and circumferential strain across the membrane surface along all radii. The uniformity remains along the length of all radii that remain in contact with the top surface of the post during stretching. The Loading Station™ is composed of acrylic plastic. The design centers it within the well of the StageFlexer® body with the top surface of the Loading Station™ just below the StageFlexer® membrane surface. When vacuum is applied to the StageFlexer®, the membrane is pulled over the Loading Station™ creating a single-plane, uniformly stretched circle (Fig. 2). A silicone-based lubricant is used to lubricate the Loading Station™, forming an effective grease boundary layer to minimize friction between the silicone membrane and the acrylic Loading Station™. Given that the part of the membrane stretching over the Loading Station™ (circular loading post) has an infinite number of radii, each cell plated in this area will receive the same strain in the direction of the radius passing through its center.

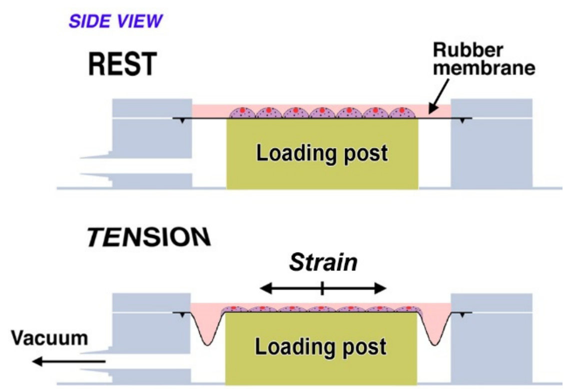

Figure 2. *Schematic of strain application in a StageFlexer®.*

STRAIN QUANTIFICATION

The three different diameters of Loading Stations™ were each placed into the StageFlexer® for testing. The strain was experimentally determined by imprinting the StageFlexer® membrane with a biaxial pattern. Strain was determined by labeling the distance between each pair of dots and measuring their change relative to vacuum levels. All vacuum measurements were made using a digital manometer. Vacuum was applied with a Robinair vacuum pump (model #15101-B). Loctite® lubricant was applied to each Delrin® Loading Station™ to enable frictionless movememt against the rubber membrane. Designated distances were measured using the following method:

A Canon Compact EOS Digital Rebel XTI® camera equipped with a macro lense was leveled and fixed directly above the membrane. The resolution of the image was adjusted to ensure each pair of dots filled the maximum horizontal distance across the digital image, maximizing the number of pixels and measurement accuracy. A FX-5000™ Tension System regimen was designed to step through pressures from 0-90 kPa. At each static step, the image was captured using a Lexar™ memory card. Adobe Photoshop® CS2 image analysis software was used to measure the distances between the dots.

RESULTS AND DISCUSSION

The results showed a nearly linear relationship between vacuum level and strain (Figs. 3-5). Given that two different radii of dots had uniform radial strain, the assumption was made that each radius was increasing uniformly with vacuum level. With this, the change in circumference could be measured from a single point; i.e., the same dots used for radial strain.

The following three figures show the experimental results for the vacuum level vs. strain relationship for each Loading Station™ diameter (25 mm, 28 mm, and 31 mm).

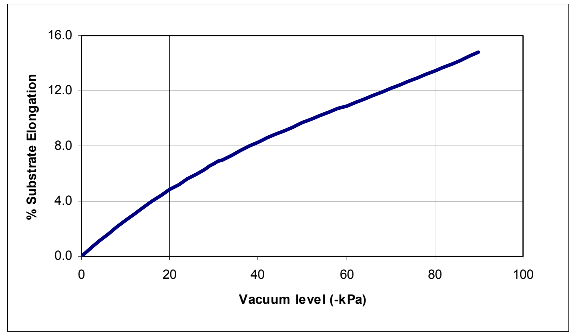

Figure 3. *Average radial strain vs. vacuum level for a StageFlexer® membrane atop a 25 mm BioFlex® Loading Station™.*

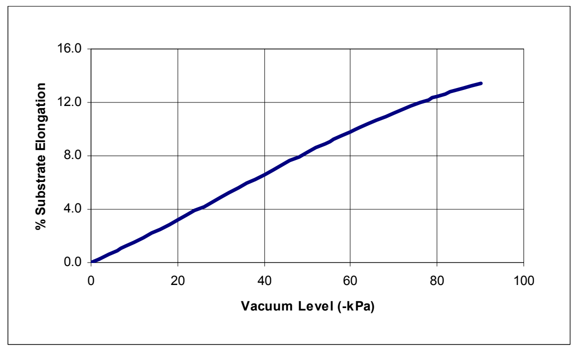

Figure 4. *Average radial strain vs. vacuum level for a StageFlexer® membrane atop a 28 mm BioFlex® Loading Station™.*

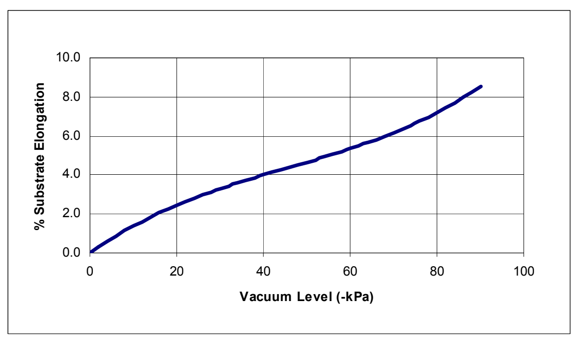

Figure 5. *Average radial strain vs. vacuum level for a StageFlexer® membrane atop a 31 mm BioFlex® Loading Station™.*