



Tech Report 102:

UniFlex™ Culture System

A Method to Apply Uniaxial Mechanical Loading to Monolayer Cell Cultures

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Culturing Cells in a Mechanically Active Environment™
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APPLICATION

UniFlex™ flexible bottomed cell culture plates are designed to apply *uniaxial strain* to cells in monolayer culture.

BACKGROUND

UniFlex™ (Fig. 1) joins the original BioFlex® and Tissue Train® as the newest addition to Flexcell®’s family of patented flexible bottomed culture plates. Based on Flexcell®’s proven mechanical loading technology, UniFlex™’s unique design enables researchers to study the effects of uniaxial strain on their monolayer cell cultures. As with Flexcell®’s BioFlex® and Tissue Train® culture plates, UniFlex™’s application range is broad and extends into numerous areas of molecular cell biology research. A key application distinction between the UniFlex™ culture plates and the BioFlex® culture plates is that UniFlex™ is capable of applying uniaxial strain to monolayer cell cultures whereas BioFlex® applies equibiaxial strain to monolayer cell culture constructs. Similarly, a key application distinction between the UniFlex™ culture plates and the linear Tissue Train® culture plates is that UniFlex™ is capable of applying uniaxial strain to monolayer cell cultures whereas linear Tissue Train® applies uniaxial strain to 3 dimensional tissue culture constructs. These comparisons are summarized in Table 1.

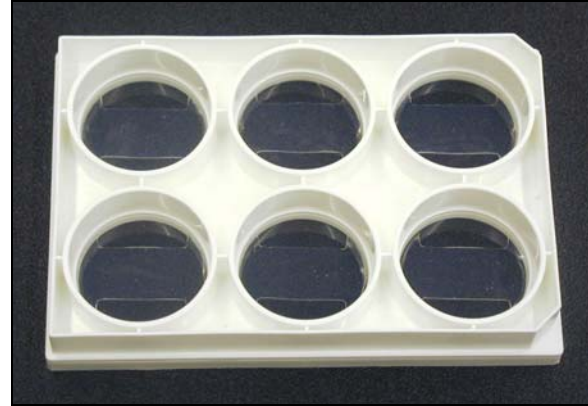


Figure 1. UniFlex™ flexible bottomed cell culture plate.

EQUIPMENT

UniFlex™ culture plates are designed to work with the standard BioFlex® baseplate (Fig. 3) that is included with all FX-5000™ Tension Systems (Fig. 2) and Tissue Train® Tension Systems. UniFlex™ also requires the same Arctangle™ Loading Stations™ (Fig. 4-5) that are used with linear Tissue Train® culture plates. As with BioFlex® and Tissue Train® culture plates, the FX-5000™ Tension System and FlexSoft® software are the hardware and software platforms, respectively, to regulate the uniaxial strain regimen desired with UniFlex™ culture plates. To apply a regulated uniaxial strain to the UniFlex™ culture plates, a regimen must be created with the appropriate strain characteristic information. This procedure and information are available in Flexcell®’s *Culture Plate & Loading Station™ User’s Manual*.

Table 1. Flexcell® culture plate / Loading Station™ application comparison

Culture Plate Style	Culture Type Application	Strain Vector	Loading Stations™
UniFlex™	Monolayer Cell Cultures	Uniaxial	Arctangle™
BioFlex®	Monolayer Cell Cultures	Equibiaxial	Cylindrical
Tissue Train®	3 Dimensional Tissue Culture	Uniaxial	Arctangle™



Figure 2. Flexcell® FX-5000™ Tension System

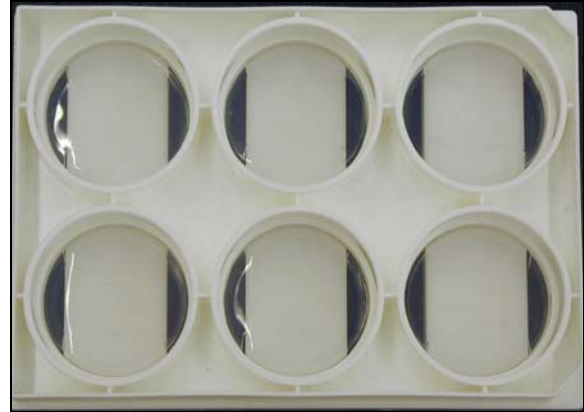


Figure 5. UniFlex™ culture plate on Arcangle™ Loading Stations™



Figure 3. Standard BioFlex® baseplate.

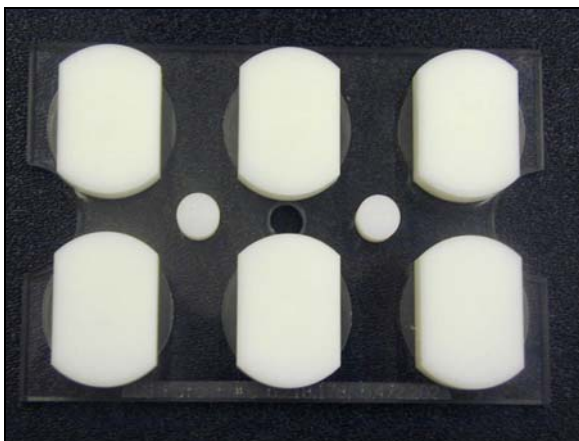


Figure 4. Arcangle™ Loading Stations™

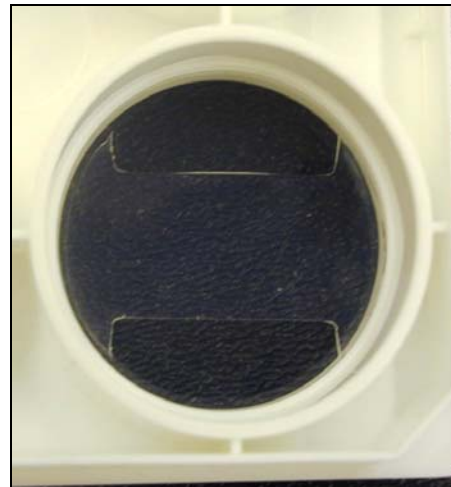


Figure 6. Single UniFlex™ well.

DIMENSIONAL CHARACTERISTICS

UniFlex™ culture plates are a 6 well configuration. The UniFlex™'s well diameter is 35 mm. This is the same overall dimensional configuration as both the BioFlex® and Tissue Train® culture plates. Uniaxial strain is achieved on a centrally located rectangular portion of the UniFlex™ well (Fig. 6-7). The dimensions of that region are 15.25 mm x 24.18 mm (0.600" x 0.952"). The uniaxial strain orientation is along the 24.18 mm axis. This region provides a total uniaxial strain area of 3.68 cm² (0.57 in²).

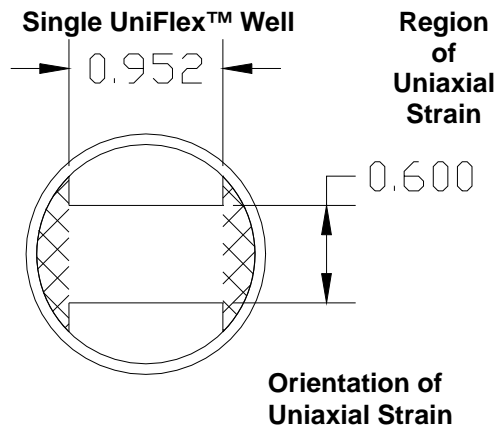


Figure 7. Dimensioned UniFlex™ well showing region and orientation of uniaxial strain. Total strain area = 3.68 cm²

UNIAXIAL STRAIN CHARACTERISTICS:

The strain characteristics for UniFlex™ culture plates have been characterized, and the results are tabulated in Flexcell®'s *Culture Plate & Loading Station™ User's Manual*. These tabulated data reflect the average strain across the specified uniaxial strain region as a function of vacuum. The localized strain can vary $\pm 1.5\%$ of the average strain value over the entire uniaxial strain region. The maximum average strain achieved is 0.122 or 12.2% elongation. The minimum average strain is 0.011 or 1.1% elongation.

EXTRACELLULAR MATRIX (ECM) COATINGS

UniFlex™ culture plates are available in all the standard coatings offered in the BioFlex® family of flexible bottomed culture plates. The various ECM coatings will be applied only to the region of the flexible bottom that will experience the uniaxial strain (see Fig. 7). The standard ECM coatings offered by Flexcell® include: Collagen Type I, Laminin, Elastin, Amino, Pronectin (genetically engineered human Fibronectin), and untreated. ID numbers for

the various coatings are shown below in Table 2.

Table 2. UniFlex™ culture plate with standard ECM coatings

ECM Coating	ID Number
Collagen Type I	UF-4001C
Pronectin	UF-4001P
Laminin	UF-4001L
Elastin	UF-4001E
Amino	UF-4001A
Untreated	UF-4001U

PLATING AND VIEWING CELLS ON THE UNIFLEX™ PLATE

Cells should be plated directly on the silicone UniFlex™ strip, then allowed to attach for 20-30 minutes before filling the rest of the well with medium (this time frame can vary depending on cell type). Cells can be viewed directly through the double silicone membrane using an inverted microscope and a 10x objective lens. Viewing cells with a 40x lens will require a long enough working distance to see through the 0.040" (1 mm) thick double membrane. Cells can also be viewed from above using an upright microscope.

SPECIAL CONFIGURATIONS

In addition to offering standard ECM coatings, Flexcell® can also provide special UniFlex™ configurations to satisfy specific user needs. These configurations can include individual UniFlex™ membranes for Flexcell®'s StageFlexer® single well microscopy tension device. Other examples of special UniFlex™ configurations can also include "aligned" collagen ECM and "troughed" or textured surfaces and geometric surfaces embedded in the flexible bottomed surface. Special applications such as these will be considered and quoted on a "case by case" basis.



REFERENCES FOR UNIFLEX™

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FLEXCELL® PATENTS

Flexcell® International Corp. products are protected by one or more of the following United States or International patents, with other patents pending:

US Patent	4,789,601
US Patent	4,822,741
US Patent	4,839,280
US Patent	5,122,470
US Patent	5,518,909
US Patent	5,593,891
US Patent	6,037,141
US Patent	6,048,723
US Patent	6,218,178
US Patent	6,472,202
US Patent	6,586,235
Japanese Patent	25-28174
German Patent	3855631.6
United Kingdom Patent	0,365,536
Canadian Patent	2,204,862