



B-Bridge International, Inc. B-Bridge

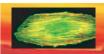


# STREX

**Mechanically Active Cell Strain Instruments** 

## What is STREX? STREX instruments simulate mechanical stress in cell culture. Seeded cells grow in strain chambers which are stretched or compressed by the instrument. The mechanical stress applied to the cells better simulates natural dynamic physiological environments. STREX instruments are automated, computer controlled systems that enables precise control of strain levels on Why is STREX important? Living cells exists in a natural physiological environment that is mechanically active. Cells use receptors to communicate with their extra-cellular environment. This communication is important for cellular growth, degradation, regeneration, development and immunological response. Additionally, disease processes such as osteoporosis, cancer metastasis, skin disorders, and muscle degeneration share a loss of tissue structure, integrity and mechano-transduction. Mechanobiology – Study how mechanical stimuli regulates biological processes Mechanotransduction – Conversion of mechanical signals to a chemical response STREX instruments stretch or compress cells inducing them to respond in a physiological manner similar to what is observed in nature. Unlike classic cell culture conditions that are static with cells immobilized on synthetic plastic dishes. www.b-bridge.com

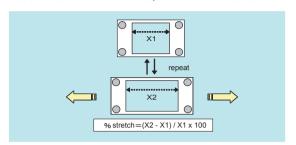




# TECHNICAL BACKGROUND

#### How does STREX work?

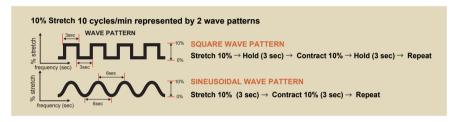
The stretch instrument uses a computer controlled step motor to drive two metal frames closer or further apart. A flexible silicone chamber is attached to both of the frames; the chamber is stretched or compressed depending on the movement of the frames.



The flexible silicone chamber consists of thick outer walls for strength and a thin transparent membrane on the bottom. The thin membrane bottom is first coated with an extra-cellular matrix like collagen or fibronectin to facilitate cell attachment via integrins, an integral plasma membrane protein. Attachment by integrin is a specific cellular function unlike attachment to plastic or glass dishes that is non-specific binding. The transparency of the chamber bottom allows for fluorescent and light microscopic observation of the cultured cells during experimentation.

The amount of strain experienced by the cells is measured as the percent change in stretch or compression from the starting position. For example, a 20% stretch means a 20% increase in the length of the silicone chamber.

Strain programs commonly used on STREX instruments range between 1 - 20% stretch at frequencies between 1 - 60 cycles/minute. Each frequency can be designed to run a unique wave pattern, for example a sine wave or square wave pattern.

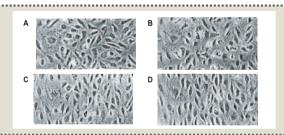


Selection of strain programs is based on cell line and experimental process. For example, a 20%, 60 cycles/min sinusoidal stretch may be appropriate for morphological analysis of cardiomyocytes or signal transduction in endothelial cells lining blood vessels. A smaller stretch and lower frequency would be better suited for studying bone cell regeneration or capturing calcium influx images.

#### **Application Notes**

Involvement of SA channels in orienting response of cultured endothelial cells to cyclic stretch
Naruse et al., Am J Physiol 1998; May;274(5 Pt 2):H1532-8

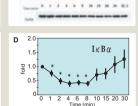
Endothelial cells from human umbilical cord vein were stretched to determine the role of calcium and stretch-activated (SA) channels in the orienting response in cells. Stretch-induced morphological changes were observed after subjecting cells to sinusoidal cyclic stretch (20% stretch, 1 Hz). The cells begun to orient perpendicular to the stretch axis 15 minutes after the onset of stretch and 90% of the cells aligned almost perfectly perpendicular after 120 minutes. A. 0 minutes, B. 30 minutes, C. 60 minutes, and D. 120 minutes.



Uni-axial cyclic stretch induces the activation of transcription factor nuclear factor  $\kappa B$  in human fibroblast cells lnoh et al., FASEB J 2002; 16:405-407

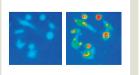
The effects of uniaxial cyclic stretching on translocation of NF-  $\kappa B$  into the nucleus of fibroblasts cells. The figures show the translocation of NF-  $\kappa B$  into the nucleus and the degradation of its inhibitor IκBa in the cytosol. A. Immunoblot stained with RelA mAb and actin mAb. B. After the onset of cyclic stretching, NF-  $\kappa B$  translocation was observed at 2 minutes and peaked at 4 minutes. After 10 minutes, NF-  $\kappa B$  returned to basal level. C. Immunoblot stained with IκBa mAb and actin mAb. D. Significant degradation of IκBa in the cytosol was observed at 1 min and reached its minimum after 4 minutes in response to stretching. For figures B and D the data was normalized by actin content.

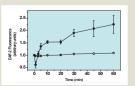
B 5.0 4.0 2.0 1.5 2.0 1.0 0 1 2 4 6 8 10 15 20 30 Time (min)



Bi-phasic activation of eNOS in response to uni-axial cyclic stretch is mediated by differential mechanisms in BAECs Takeda et al., Life Sci 2006; Jun 13;79(3):233-9

The authors investigated the signaling mechanism of stretch-induced nitric oxide (NO) production. When bovine arterial endothelial cells were uni-axial cyclic stretched (20%, 1 Hz), NO production peaked at 5 and 20 minutes after the initiation of stretch. The early peak is mediated by Ca\*2 influx and the later peak is due to activation of Akt.







# STREX INSTRUMENTS



# **Advantages of STREX instruments**

- Precise strain programs
- Consistent stretch/compression at high and low frequencies
- Uniform strain over the entire chamber culture area
- Minimal lateral thinning
- Easy to use no cumbersome parts

#### Bench top stretch instrument and controller

Bench top models are ideal for long term studies including cell morphology, gene and protein expression, and signal transduction. The entire stretching machine can easily be put in an incubator.

# ST-140-04 & ST-140-10

- Uniaxial stretch
- Multi-chamber
- · Fits in laboratory incubator
- 64 unique stretch programs
- · Long term studies

The robust and easy to use design ensures that cells grow over long periods of time with precise cyclic stretching.





#### Microscope mountable stretch instrument and controller

These models are designed for short term studies (20 minutes continuous stretching or compression) and real time observations; typical applications include ion mobilization, Ca<sup>+2</sup> influx, nitric oxide production, and cell morphology.

#### ST-150

- Uniaxial stretch
- Single-chamber
- · Sits on microscope stage
- Real-time observations
- 64 unique stretch programs

The ST-150 is a computer controlled, uniaxial stretching machine that enables real-time observations of a cell culture. The ST-150 fits on most fluorescent microscopes.





- ST-190-XY · Biaxial stretch or compression
  - Single-chamber
  - Sits on microscope stage
  - Real-time observations
  - 64 unique stretch programs

ST-190-XY is an automated instrument that delivers consistent X stretch and compression to a single cell culture to generate reliable results.

#### **ST-195**

- · Uniaxial stretch
- Self-contained in a CO<sub>2</sub> incubator
- · Single-chamber
- Sits on microscope stage
- Real-time observations
- 64 unique stretch programs

Our CO<sub>2</sub> incubator enclosed stretch instrument isolates your test samples from outside contaminants and eliminates the need for a laboratory incubator. The incubator maintains a humidified, 37°C, 5% CO<sub>2</sub> environment.



	Models				
Features	ST-140-04	ST-140-10	ST-150	ST-190-XY	ST-195
Applications	<ul> <li>Cytoskeleton rearrangements</li> <li>Cell morphology</li> <li>Gene or protein expression</li> <li>Signal transduction</li> <li>Long duration studies (hours-days)</li> </ul>		Cytoskeleton rearrangements     Cell morphology     Ion mobilization     Calcium influx     Nitric oxide production     Real-time observation of cultures     Short duration studies (15-20 minutes)		
No. of chambers	6	5	1	1	1
Chamber size - culture surface area	4 cm <sup>2</sup>	10 cm²	4 cm <sup>2</sup>	4 cm <sup>2</sup>	4 cm²
Strain	Uniaxial stretch	Uniaxial stretch	Uniaxial stretch	Biaxial stretch and compression	Uniaxial stretch
Microscope mountable	_	_	Fits Nikon and Olympus. Optional for Zeiss, and Leica	Fits Nikon and Olympus. Optional for Zeiss, and Leica	Fits Nikon and Olympus. Optional for Zeiss, and Leica
Incubator	Fits in standard incubator	Fits in standard incubator	Fits in standard incubator	Fits in standard incubator	Self contained CO <sub>2</sub> incubator
No. strain programs	64	64	64	64	64



## STREX CHAMBERS

### **REFERENCES**



The patented STREX chambers are made of silicone (poly[dimethylsiloxane]; PDMS). The chamber consists of 400  $\mu m$  thick outer walls and a 200  $\mu m$  thick membrane bottom. It is on this thin membrane bottom that cells are cultured and can be observed microscopically. Always a concern when stretching cells is applying equal strain to all cells and minimizing lateral

thinning. Lateral thinning is the detachment of cells around the perimeter of the culture area in the chamber. The thick side walls provide strength and support to prevent narrowing of the chamber bottom when stretched. The thin membrane bottom ensures equal and consistent strain is applied across the entire membrane minimizing lateral thinning. Lateral thinning does not exceed 1% at a 20% stretch (Naruse K; et al, 1998).

#### **Advantages to STREX Chambers**

- · Uniform strain for accurate results
- · Minimal lateral thinning
- · Optically compatible for light and fluorescent microscopy
- Durable



Culture area: 2.0 x 2.0 x 1.0 cm





Corresponding STREX Models

ST-140-04 ST-150 ST-195

ST-CH-10 Culture area 3.2 x 3.2 x 1.0 cm

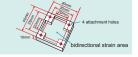




Corresponding STREX Models

ST-140-10

ST-CH-04-XY Culture area 2.0 x 2.0 x 1.0 cm





Corresponding STREX Models

ST-190-XY

ST-CH-10ST

on ST-CH-10



ST-CH-4W Culture area for each well 1.5 x 1.5 x 1.0 cm



Corresponding STREX Models

ST-140-10

#### Automated, bench top:

Model	Description	Chamber size
ST-140-04	Uniaxial stretch, multi-chamber	4 cm <sup>2</sup> chamber
ST-140-10	Uniaxial stretch, multi-chamber	10 cm <sup>2</sup> chamber

#### Automated, microscope-mountable:

Model	Description	Chamber size
ST-150	Uniaxial stretch, single chamber	4 cm <sup>2</sup> chamber
ST-195	Uniaxial stretch, single chamber, CO2 incubator	4 cm <sup>2</sup> chamber
ST-190-XY	Biaxial stretch/compress, single chamber	4 cm <sup>2</sup> XY chamber

#### Stretch Chambers:

Catalog #	Description	Corresponding STREX model
ST-CH-04	4 cm² silicone chamber	ST-140-04, ST-150, ST-195
ST-CH-10	10 cm² chamber	ST-140-10
ST-CH-4W	10 cm <sup>2</sup> chamber with 4-wells	ST-140-10
ST-CH-04-XY	4 cm <sup>2</sup> XY chamber	ST-190-XY

#### **Chamber Stands:**

Catalog #	Description	Corresponding Chambers	
ST-CH-04ST	1 metal stand, holds 1 chamber	ST-CH-04	
ST-CH-10ST	1 metal stand, holds 1 chamber	ST-CH-10	

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