

CellPlayer™ Angiogenesis StemKit

Essen BioScience Catalog Number: 4507

Presentation, Storage and Stability

The Essen BioScience CellPlayer Angiogenesis StemKit is composed of two boxes containing the reagents required to set up and run the angiogenesis tube formation assay to completion. The contents of the kit, when mixed and treated according to the established protocol, will result in the formation of angiogenic networks over the course of the 4-5 day assay. Kit contents are as follows:

Dry Ice Shipment:

- 1) Adipose Derived Stem Cell (ADSC) Vial
- 2) Endothelial Colony Forming Cell expressing GFP (GFP-ECFC)
- 3) ADSC Growth Media Supplement (1.5 mL total volume)
- 4) ECFC Growth Media Supplement (2.75 mL total volume)
- 5) StemKit Assay Media Supplement (0.85 mL total volume)

Room Temperature Shipment:

- 1) ADSC Growth Basal Media (50 mL total volume)
- 2) ECFC Growth Basal Media (25 mL total volume)
- 3) StemKit Assay Basal Media (100mL total volume)
- 4) 96-well Assay Plate

Storage and stability:

- 1) Upon arrival, the cells in this kit should be **IMMEDIATELY** transferred to liquid nitrogen. When stored in liquid nitrogen, they will remain viable until the expiration date printed on the vial.
- 2) The media supplements contained in this kit should be kept **FROZEN** at -20 °C until use or the expiration date printed on each vial.
- 3) The basal media in this kit should be **REFRIGERATED** at 4 °C until use or the expiration date located on each bottle. The kit contains sufficient culture medium to support development of the assay over the required 4-5 day (neoangiogenic mode) or 10 day (established mode) period.

Background

The CellPlayer Angiogenesis StemKit contains cryopreserved vials of human Endothelial Colony Forming Cells (ECFC) and human Adipose Derived Stem Cells (ADSC). The densities for each cell type have been optimized for performance in a co-culture angiogenesis model. When seeded, grown, and treated according to the Essen BioScience CellPlayer Angiogenesis StemKit protocol, this co-culture model recapitulates the *in vivo* angiogenesis process, involving cell migration,



morphogenesis, and anastomosis. Under basal conditions, these cultures will differentiate into a low level of capillary-like tubules after 4-5 days. Stimulation with a growth factor, such as VEGF or bFGF, results in a 2-3-fold stimulation of tube formation. Tubule development is believed to closely mimic *in vivo* angiogenesis and is enhanced by known angiogenic stimulators. Measuring anti-angiogenic events requires the presence of a stimulating factor, such as VEGF. Both enhancement and suppression occur in a concentration-dependent manner.

Recommended Usage / Quality Control Testing

The CellPlayer Angiogenesis StemKit contains cryogenically preserved ampules of matched cells, medium, and media supplements, all of which are required to complete a successful experiment. Our 3 years of experimental development has repeatedly shown that growth factors purchased from different sources can result in highly variable responses. As such, all growth factor supplement and inhibitor kits available for purchase from Essen BioScience, including VEGF, bFGF, and suramin have been rigorously quality controlled to ensure maximal tube formation and/or inhibition if used as described. When used in conjunction with the IncuCyte-FLR, the resulting effect of growth factors and test agents on tube formation can be measured using the Essen BioScience automated angiogenesis algorithm. The data generated with this algorithm can be used to assess angiogenic potential via tube length, branch point, and tube area metrics.

Extensive testing of the CellPlayer Angiogenesis StemKit is performed to ensure a significant level of growth factor-mediated tube formation compared to the untreated control. Utilizing historical data generated across multiple experiments at Essen BioScience, we typically observe average tube length levels of 13.35 ± 0.67 mm/mm² (Mean \pm SD) when using VEGF at 20ng/ml. Addition of 100 μ M suramin will inhibit VEGF-mediated tube formation by > 90%, resulting in a Z' of 0.80. Careful adherence to the StemKit protocol will minimize unwanted edge effects, enabling the use of the entire 96-well plate for test agent investigation. Our strict criteria for assay performance enable high-quality assessment of a test agents' pro- or anti-angiogenic potential in an *in vitro* setting.

Related Products

- Cat.# 4509 CellPlayer StemKit VEGF/Suramin Supplement Kit
- Cat.# 4510 CellPlayer StemKit bFGF/Suramin Supplement Kit
- Cat.# 4542 CellPlayer Angiogenesis StemKit Optimized Media Kit
- Cat.# 4452 CellPlayer Angiogenesis CryoKit

For additional information including a simplified description of the protocol and an extensive application note, please visit our webpage at http://www.essenbioscience.com/angiogenesis_stemkit.html.

For a detailed protocol including use of the CellPlayer™ Angiogenesis StemKit, as well as additional information, please contact Essen BioScience at: sales@essenbio.com.

This Essen BioScience product contain proprietary nucleic acid(s) coding for proprietary fluorescent protein(s) being, including its derivatives or modifications, the subject of pending patent applications and/or patents owned by Evrogen JSC (hereinafter "Evrogen Fluorescent Proteins"). The purchase of Essen BioScience products incorporating these fluorescent proteins conveys to the buyer the non-transferable right to use Evrogen Fluorescent Proteins only for research conducted by the buyer. No rights are conveyed to modify or clone the gene encoding fluorescent protein contained in this product or to use Evrogen Fluorescent Proteins for commercial purposes. The right to use Evrogen





CellPlayer™ Angiogenesis StemKit

Fluorescent Proteins specifically excludes the right to validate or screen compounds for commercial purposes. For information on commercial licensing, contact Evrogen Licensing Department, email: license@evrogen.com.

For research use only. Not for therapeutic or diagnostic use.

