

CellPlayer™ Angiogenesis CryoKit

Essen BioScience Catalog Number: 4452

Presentation, Storage and Stability

The Essen BioScience CellPlayer Angiogenesis CryoKit 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 8-10 day assay. Kit contents are as follows:

Dry Ice Shipment:

- 1) Normal Human Dermal Fibroblast (NHDF) Cell Vial
- 2) Human Umbilical Vein Endothelial Cells expressing GFP (GFP-HUVEC)
- 3) Seeding Media Supplement (2 mL total volume)
- 4) Growth Media Supplement (0.4 mL total volume)
- 5) Assay Media Supplement (2.2 mL total volume)

Room Temperature Shipment:

- 1) Seeding Basal Media (40 mL total volume)
- 2) Growth Basal Media (20 mL total volume)
- 3) Assay Basal Media (110mL 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 8-10 day period.

Background

The CellPlayer Angiogenesis CryoKit contains cryopreserved vials of early passage normal human endothelial cells and early passage normal human interstitial cells. 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 CryoKit protocol, this co-culture model recapitulates all phases of the *in vivo* angiogenesis process,





including cell proliferation, migration, morphogenesis, and anastomosis. Under basal conditions, these cultures exhibit a low level of capillary-like tubules after 8-10 days, unless stimulated with a growth factor such as VEGF, EGF, or bFGF. 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 CryoKit 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, EGF, 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 CryoKit 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 8.0 ± 1.4 mm/mm² (Mean \pm SD) when using VEGF at 4ng/ml. Addition of 100 μ M suramin will inhibit VEGF-mediated tube formation by > 90%. Careful adherence to the CryoKit 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.# 4436 CellPlayer Angiogenesis LiveKit
- Cat.# 4453 CellPlayer Cryo-preserved GFP-HUVEC
- Cat.# 4437 CellPlayer VEGF/Suramin Supplement Kit
- Cat.# 4438 CellPlayer bFGF/Suramin Supplement Kit
- Cat.# 4439 CellPlayer EGF/Suramin Supplement Kit
- Cat.# 4409 CellPlayer Angiogenesis Assay Media 100 mL
- Cat # 4410 CellPlayer Angiogenesis Assay Media 500 mL

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

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

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