

PRODUCT INFORMATION

GFP Human Umbilical Vein Endothelial Cells (HUVEC) - Angiogenesis Tested, pooled donor, cryopreserved

Product Code: **ZHC-2402**

These cells have been shown to produce tubules in conjunction with Cellworks Human Dermal Fibroblasts – adult Angiogenesis Tested (ZHC-5102), using Cellworks Angiogenesis Seeding and Growth Medium Packages (ZHA-1960 and ZHA-1970, respectively). They have been validated for use in the manufacture of Cellworks GFP V2a Kits (ZHA-4100).

Presentation

Each vial of GFP HUVEC – pooled donor Angiogenesis Tested contains a minimum of 500,000 cells from multiple donors, at passage 2, guaranteed to be 70% viable after thawing. The HUVEC cells contain stably expressed GFP driven off a CMV promoter. Cells are cryopreserved in a cryoprotectant medium.

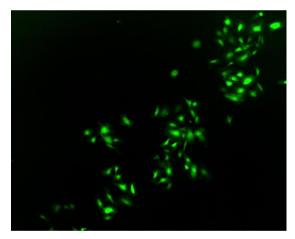


Figure 1. GFP HUVEC (ZHC-2402)

Recommended Media and Supplements

Human Large Vessel Endothelial Cell Growth Medium Package: **ZHM-2953**

Human Large Vessel Endothelial Cell Basal Medium, 500ml (KC1015) Human Large Vessel Endothelial Cell Growth Supplement, 10ml (KC1016) Antibiotic Supplement (Gentamycin/Amphotericin B) 1000x concentrated, 0.5ml

(KC1019)

Contains:

Use of Cells in an Angiogenesis Assay

For tubule formation assays, we recommend the using Cellworks Human Dermal Fibroblasts – adult Angiogenesis Tested (ZHC-5102), using Cellworks Angiogenesis Seeding Medium Package (ZHA-1960) and Angiogenesis Growth Medium Package (ZHA-1970).



Please visit www.cellworks.co.uk/download/GFP_Bespoke_Angiogenesis_Assay_Protocol.pdf for guidance on using the cells in an angiogenesis assay.

Recommended Seeding Density

2,500 viable cells per cm²

Proliferation capacity

Normal human cells have a limited life span *in vitro*. When cells are cultured using our recommended reagents and procedures, these GFP HUVEC can achieve 10 population doublings. We do not recommend passaging these cells past passage 5 prior to using them in an angiogenesis assay.

Storage and maintenance of cells

Cryopreserved cells must be dealt with according to the following guidelines **immediately** on receipt:

GFP HUVEC – pooled donor Angiogenesis Tested will arrive frozen on dry ice and if not seeded immediately must be transferred to the vapour phase of a liquid nitrogen storage facility. **N.B.** Continued storage at -80°C or on dry ice is **NOT** appropriate.

For brief guidelines for the culture of Cellworks cells, see below.

Brief guidelines for culture of Cellworks cells

Initiation of proliferating cultures from cryopreserved cells

- 1. Prepare a bottle of Human Large Vessel Endothelial Cell Growth Medium according to the Cellworks instructions.
- Pre-equilibrate 1x 15ml within a 75cm² culture flask or 3x 5ml into 25cm² flasks in a humidified incubator (37°C, 5% CO₂). Prepare a water bath at 37°C.

N.B. A minimum of 15ml growth medium must be used to dilute out the cryoprotectant.

- 3. Remove the cell vial from liquid nitrogen storage and place in dry ice (or in a dewar of liquid nitrogen).
- 4. Suspend the bottom half of the cryovial in the 37°C water, ensuring that the cap is not submerged.
- 5. Agitate gently for 1-2 minutes until a few ice crystals remain. Wipe the vial with ethanol and immediately transfer into a sterile laminar flow hood.
- 6. Open the vial and gently re-suspend the solution with a pipette to evenly suspend the cells.
- 7. Remove 20 μ l from the vial and dilute the cell suspension in 20 μ l Trypan blue.
- 8. Using a haemocytometer, determine the number of viable cells per ml.
- 9. Inoculate and dilute the cells to a concentration of 1.25 x 10⁴ viable cells/ml into the prepared flasks in step 2 by dispensing the cell suspension in an arc on the surface of the medium, and gently mix for even cell growth.
- 10. Examine the cells microscopically to check for even distribution in the flasks, and transfer to the 37°C, 5% CO₂ humidified incubator. For best results, do not disturb the cell culture for at least 16 hours.
- After 16-24 hours have elapsed after seeding, examine the cultures microscopically to check seeding has been successful, and replace the medium with pre-equilibrated fresh medium i.e. 5ml medium per 25cm².
 N.B. Be careful to dispense the medium over a cell-free surface of the flask and never over the cell layer as this may dislodge the cells.
- 12. Return the cells to the incubator and replenish cells with fresh growth medium every 48 hours according to step 11.
- 13. Double the volume of growth medium when cell cultures are greater than 45% confluent, for culturing over a weekend and subculture when the cells are 60-80% confluent, while they are still actively dividing.



Nelated Floducts	
Human Dermal Fibroblasts (HDF) - Angiogenesis Tested	ZHC-5102
Human Fibroblast Growth Medium Package	ZHM-5923
Human Large Vessel Endothelial Cell Growth Medium Package	ZHM-2953
GFP V2a Kit: Vasculogenesis to Angiogenesis	ZHA-4001
AngioSys 2.0 Image Analysis Software	ZHA-5000
AngioSys 2.0 Image Analysis Software - Demonstration Version	ZHA-5000D
Angiogenesis Growth Medium Package Validated for use with the V2a™ Kit	ZHA-1970
Angiogenesis Seeding Medium Package Validated for use with the V2a™ Kit	ZHA-1960
Angiogenesis Image Analysis Service - Image capture	ZHA-6003
Angiogenesis Service - price per well	ZHA-7001
Angiogenesis Service Set up Fee	ZHA-7000
Angiogenesis Image Analysis Service - First 24 Images	ZHA-6000
Angiogenesis Image Analysis Service - Additional Images	ZHA-6001
Angiogenesis Image Analysis Service Set Up	ZHA-6002
CD31Tubule Staining Kit	ZHA-1225
Angiogenesis Assay Control Kit	ZHA-1300
V2a Kit - Vasculogenesis to Angiogenesis	ZHA-4000
Human Umbilical Vein Endothelial Cells (HUVEC) - Angiogenesis Tested	ZHC-2102
Early Passage Human Umbilical Vein Endothelial Cells (HUVEC)	ZHC-2301
Early Passage HUVEC and Medium Package	ZHP-2353

Caution

Related Products

For research use only, not for therapeutic or diagnostic use.

Treat all human cells as potentially biohazardous and dispose of appropriately.