IMMORTALIZED CELL LINES

P10962-IM

Vascular endothelial cells play a pivotal role in maintaining vascular homeostasis by orchestrating the synthesis and secretion of activators and inhibitors for both the coagulation and fibrinolysis systems. Additionally, these cells release mediators that impact the adhesion and aggregation of blood platelets, and further regulate cell proliferation while modulating vessel wall tone.

In vitro studies of endothelial processes, such as tube formation, are integral to understanding vascular biology. Human umbilical vein endothelial cells (HUVEC) stand as the preeminent cell type for such investigations. Specifically, the IM-HUVEC cell line serves as a suitable model for studying tube formation in vitro, adhering to established and standardized protocols.

Should you require further elucidation or have specific inquiries, please do not hesitate to express them.

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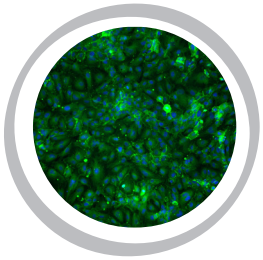
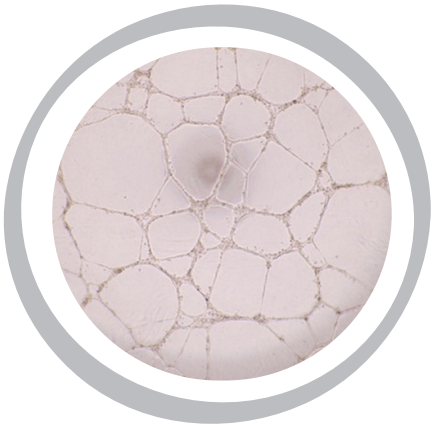
IMMORTALIZED HUVEC



Product Type: Immortalized HUVEC
Catalog Number: P10962-IM
Immortalization: SV40 Large T Antigen
Number of cells: >1x10^6 cells (cryopreserved vials)
Storage: Liquid Nitrogen
Recommended Medium: Endothelial Medium (Reference: P60104)
Product Characterization: Immunofluorescent staining (vWF/Factor VIII and CD31/P-CAM) and uptake of DiI-Ac-LDL.

About Immortalized HUVEC Cells

The immortalized HUVEC cell line (IM-HUVEC) has been developed through genetic modification of primary HUVEC cells, employing the SV40LT protein as the immortalization method. The SV40LT protein, derived from the Simian Virus 40 Large T-antigen, has been introduced to confer immortality to the cells, allowing for an extended lifespan compared to primary cells that typically undergo senescence after a limited number of passages. The use of SV40LT for immortalization is a common technique in cell biology and allows for the establishment of cell lines with a more stable and prolonged growth capacity. This modification enables researchers to conduct long-term experiments and studies that require consistent cellular behavior over an extended period. Primary cells exhibit senescence following the 5th passage, whereas the SV40LT-transduced cells demonstrate a prolonged viability, extending beyond 20 passages.



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Tube formation assay

The assay measures the ability of endothelial cells, plated at subconfluent densities with the appropriate extracellular matrix support, to form capillary-like structures (a.k.a tubes).

Quality Control / Biosafety

The cells test negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast and fungi.

Culturing conditions

The provided information outlines a set of procedures for the immediate handling, thawing, culturing, and maintenance of Immortalized HUVEC cell line. Here is a more structured and concise representation:

1 IMMEDIATELY UPON DELIVERY

1.1 Remove the vial from the shipping container to check for freezing.

1.2 Transfer the frozen vial to liquid nitrogen until ready to thaw.

2 THAWING CELLS:

2.1 Prepare "Thawing medium" by combining 500 ml of Endothelial basal medium, 25 ml of fetal bovine serum, 5 ml of EGS growth supplement, and 5 ml of penicillin/streptomycin solution.

2.2 Thaw cells rapidly in a 37°C water bath; avoid allowing the sample to warm to 37°C. Cryovials should be cool to the touch when removed.

2.3 Remove the vial, wipe it dry, and transfer it to a sterile field.

2.4 Rinse the vial with 70% ethanol, then wipe to remove excess. Open the vial and resuspend its contents using a 1 ml Eppendorf pipette.

2.5 Dispense the contents into a 25 cm2 culture flask with warm complete media (FBS percentage can be increased up to 10% for better culture establishment).

2.6 Place the flask in the incubator.

2.7 For optimal results, avoid disturbing the culture for 16 hours after initiation. Change the growth medium the next day to remove unattached cells, then every other day thereafter.

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Culturing conditions

3 MAINTENANCE OF THE CULTURE:

3.1 Change medium 48 hours after establishing a subculture.

3.2 Subculture when cells are over 90% confluent.

4 SUBCULTURING:

4.1 Remove medium, rinse with 0.05% trypsin, 0.05% EDTA solution. Add 1 to 2 mL of trypsin-EDTA solution and allow the flask to sit until cells detach. Add fresh culture medium, aspirate, and dispense into new culture flasks.

Subcultivation Ratio: Recommended ratio of 1:2 to 1:6.

Medium Renewal: 2 to 3 times per week.

Reagents for cryopreservation: Cryostor S10.
These guidelines provide a systematic approach to handling, thawing, and maintaining Immortalized HUVEC cell line, ensuring optimal culture conditions and reproducibility.

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