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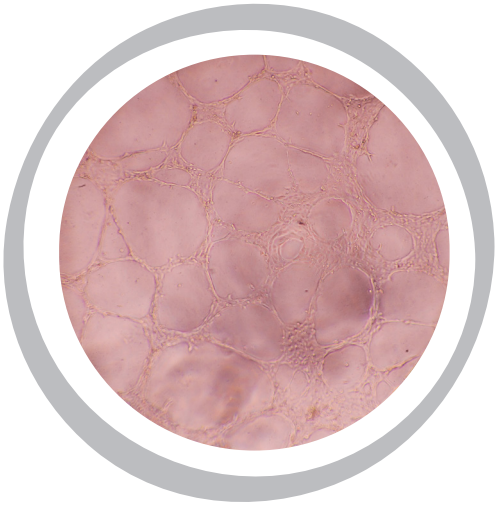
IMMORTALIZED CELL LINES

IMMORTALIZED HUMAN RENAL GLOMERULAR ENDOTHELIAL

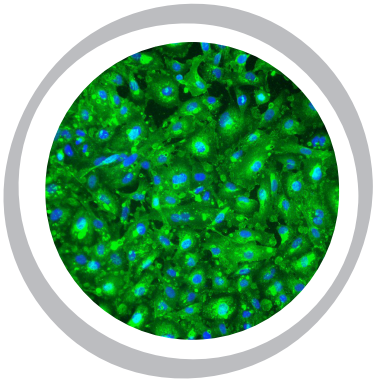
P10665-IM

Human renal glomerular endothelial cells are highly specialized cells that form the inner lining of glomerular capillaries within the kidney, playing a crucial role in maintaining the integrity of the glomerular filtration barrier. These cells are essential for regulating selective permeability, fluid balance, and the exchange of solutes and metabolic waste. They contribute to vascular homeostasis and are actively involved in cross-talk with podocytes and mesangial cells, ensuring the proper function of the glomerulus. In pathological states, such as diabetic nephropathy, glomerulonephritis, or hypertensive nephrosclerosis, glomerular endothelial dysfunction is a key contributor to the disruption of filtration and the progression of kidney injury. Moreover, their response to inflammatory and hemodynamic stress is central to the development of chronic kidney disease and associated vascular complications.

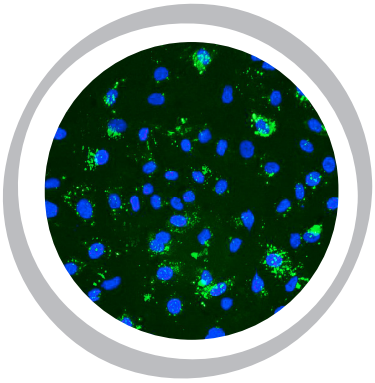
Product Type: Immortalized Human Renal Glomerular Endothelial Cells
Catalog Number: P10665-IM
Immortalization: SV40 Large T Antigen. G418 resistant.
Number of cells: >1x10^6 cells (cryopreserved vials)
Storage: Liquid Nitrogen
Recommended Medium: Endothelial Cell Medium Kit (Ref: P60104)
Product Characterization: Positive for vWF, CD31 and tubule formation.



Tubule formation



CD31



VWF

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About Immortalized Human Renal Glomerular Endothelial Cells

The immortalized human renal glomerular endothelial cell line has been developed through genetic modification of primary culture of human renal glomerular endothelial 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.

Culturing conditions

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 Fibronectin coating step (optional): Prepare a fibronectin coated flask (2 µg/cm2, T-75 flask) is recommended depending on the type of flasks you are using (i.e. no pre-treated flasks for endothelial cells). Add 10 ml of sterile Dulbecco's phosphate buffered saline (DPBS) to a T-75 flask and then add 150 µl of fibronectin stock solution (1 mg/ml, Innoprot cat. no. P8248). Leave the flask in incubator overnight. Aspirate fibronectin solution and add 15 ml of complete medium to the culture vessel. The fibronectin solution can be used twice. Leave the vessel in the sterile field and proceed to thaw the cryopreserved cells.

2.2 Prepare "Thawing medium" by combining 500 ml of basal medium, 25 ml of fetal bovine serum, 5 ml of Growth supplement and 5 ml of penicillin/streptomycin solution.

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

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

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

2.6 Dispense the contents into a 25 cm^2 culture flask with warm complete media (FBS percentage can be increased up to 10% for better culture establishment).

2.7 Place the flask in the incubator.

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Culturing conditions

2.8 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.

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:

Remove medium, rinse with 0.05% trypsin-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.

Recommended subcultivation ratio of 1:2 to 1:6.

Medium Renewal: 2 to 3 times per week.

Reagents for cryopreservation: Cryostor S10.

Quality Control / Biosafety

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

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