



Product Sheet

Endothelial Cell Growth Kit-BBE (ATCC® PCS-100-040™)

Please read this FIRST

Storage Temp.

-20°C (or -70°C for long-term storage)

Biosafety Level

1

Description

Product Description: Endothelial Cell Growth Kit-BBE (ATCC® PCS-100-040) and Endothelial Cell Growth Kit-VEGF (ATCC® PCS-100-041) each contain components that when added to Vascular Cell Basal Medium (ATCC® PCS-100-030) create a complete ATCC® Primary Cell Solution™ culture environment for endothelial cells derived from normal human large vessels such as:

- Primary Umbilical Vein Endothelial Cells; Normal, Human, ATCC® PCS-100-010
- Primary Umbilical Vein Endothelial Cells; Normal, Human, Pooled, ATCC® PCS-100-013
- Primary Aortic Endothelial Cells; Normal, Human, ATCC® PCS-100-011
- Primary Coronary Artery Endothelial Cells; Normal, Human, ATCC® PCS-100-020
- Primary Pulmonary Artery Endothelial Cells; Normal, Human, ATCC® PCS-100-022

Your experimental design will dictate which Endothelial Cell Growth Kit should be used. Use of the Endothelial Cell Growth Kit-VEGF (ATCC® PCS-100-041) will support a faster rate of proliferation because of the presence of several purified human recombinant (rh) growth factors (rh VEGF, rh EGF, rh FGF basic and rh IGF-1) combined with heparin and hydrocortisone. Use of the Endothelial Cell Growth Kit-BBE (ATCC® PCS-100-040), which contains Bovine Brain Extract (BBE), is recommended if a less defined cell culture medium is desired.

Volume: 1 kit

Directions for Use

Unpacking and Storage instructions

1. Check all containers for leakage or breakage.
2. Store the growth kit(s) at either -20°C in a freezer that is not self-defrosting or at -70°C for long-term storage. If thawed upon arrival, the Growth Kit can be stored at 2°C to 8°C and added to the basal medium within 72 hours of receipt.

Preparation of Complete Growth Media

1. Obtain one growth kit from the freezer; make sure that the caps of all components are tight.
2. Thaw the components of the growth kit just prior to adding them to the basal medium. It is necessary to warm the L-glutamine component in a 37°C water bath and shake to dissolve any precipitates, prior to adding to the basal medium.
3. Obtain one bottle of Vascular Cell Basal Medium (475 mL) from cold storage.
4. Decontaminate the external surfaces of all growth kit component vials and the basal medium bottle by spraying them with 70% ethanol.
5. Using aseptic technique, and working in a laminar flow hood or biosafety cabinet transfer the volume of each growth kit component, as indicated in Table 1 or 2, to the bottle of basal medium using a separate sterile pipette for each transfer.
6. Tightly cap the bottle of complete growth medium and swirl the contents gently to assure a homogeneous solution. Do not shake forcefully to avoid foaming. Label and date the bottle.
7. Complete media should be stored in the dark at 2°C to 8°C (do not freeze). When stored under these conditions, complete media is stable for 30 days.

Table 1. If using the Endothelial Cell Growth Kit-BBE (ATCC® PCS-100-040), add the indicated volume for each component:

Component	Volume	Final Concentration
Bovine Brain Extract (BBE)	1.0 mL	0.2%
rh EGF	0.5 mL	5 ng/mL
L-glutamine	25.0 mL	10 mM
Heparin sulfate	0.5 mL	0.75 Units/mL
Hydrocortisone hemisuccinate	0.5 mL	1 µg/mL
Fetal Bovine Serum	10.0 mL	2%
Ascorbic acid	0.5 mL	50 µg/mL

Table 2. If using the Endothelial Cell Growth Kit-VEGF (ATCC® PCS-100-041), add the indicated volume for each component:

Component	Volume	Final Concentration
rh VEGF	0.5 mL	5 ng/mL
rh EGF	0.5 mL	5 ng/mL
rh FGF basic	0.5 mL	5 ng/mL
rh IGF-1	0.5 mL	15 ng/mL
L-glutamine	25.0 mL	10 mM
Heparin sulfate	0.5 mL	0.75 Units/mL

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
www.atcc.org

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Fax: 703.365.2750
Email: Tech@atcc.org


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Hydrocortisone hemisuccinate	0.5 mL	1 µg/mL
Fetal Bovine Serum	10.0 mL	2%
Ascorbic acid	0.5 mL	50 µg/mL

Antimicrobials and phenol red are not required for proliferation but may be added if desired. The recommended volume of each optional component to be added to the complete growth media is summarized in Table 3.

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Table 3. Addition of Antimicrobials/Antimycotics and Phenol Red (Optional)

Component	Volume	Final Concentration
Gentamicin- Amphotericin B Solution	0.5 mL	Gentamicin: 10 µg/mL Amphotericin B: 0.25 µg/mL
Penicillin- Streptomycin- Amphotericin B Solution	0.5 mL	Penicillin: 10 Units/mL Streptomycin: 10 µg/mL Amphotericin B: 25 ng/mL
Phenol Red	0.5 mL	33 µM

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This product is intended for laboratory research purposes only. It is not intended for use in humans.

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Additional information on this culture is available on the ATCC web site at www.atcc.org.

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