

**Cardiac Bovine Microvascular Endothelial Cells**

Catalog Number	B1013
Product Name	Cardiac bovine microvascular endothelial cells
Storage	Liquid Nitrogen
Product Format	Frozen vial
Cells Number	>90% confluent in Frozen Vial

**\*Caution:** Although primary cells are tested pathogen-free, investigators should handle these cells with caution and treat all animal cells as potential pathogens, since no test procedure can completely guarantee the absence of infectious agents. Proper precautions must be taken to avoid exposure. Always wear proper protective equipment (Gloves, safety glasses, etc.) when handling these materials. We recommend following the universal procedures for handling products of animal origin as the minimum precaution against contamination.

**GENERAL INFORMATION**

Cardiac bovine microvascular endothelial cells were isolated from young healthy bovine heart tissues. The cells are shipped in proliferating culture or frozen vial with a confluence of > 90% (the cells are provided at passage 2). ENDO-Growth medium (EGM-2102) containing 5% serum and growth supplement is recommended for the expansion of these can be propagated to passage 6 and beyond without losing their morphologic and phenotypic characteristics. When you receive the cells, leave the flask in 37°C CO<sub>2</sub> incubator for 1 hour first and then replace the transport medium with fresh ENDO-Growth medium (EGM-2102). Let the cells grow for 24 hours before subculture.

**CELL CHARACTERIZATION**

PECAM1	>95% positive by immunofluorescence
VE-Cadherin	>95% positive by immunofluorescence
Cardiac bovine microvascular endothelial cells	Negative for mycoplasma

**PRODUCT USE AND SHIPPING STATUS**

Product Use	Cardiac bovine microvascular endothelial cells are for research only
Shipping Status	Frozen vial

### Frozen

- 1) Coating T25 flasks. Add 2 ml AlphaBioCoat (AC001) into a T25 flask and ensure entire interior surface is coated with the solution. After 30 minutes, dispose of AlphaBioCoat (AC001) by aspiration. Gently rinse and aspirate flask with Phosphate Buffer Solution (1XPBS-001). The flask is now ready for use (no need for overnight incubation when coated with AC001)
- 2) If you are using the coated flask the same day, add about 4 ml of Endo-Growth media (EGM-2102) to the coated flask. If the media changes color from pink to yellow, aspirate and discard the media. Add 4ml of fresh media to the coated flask.
- 3) Thaw the cells in a 37°C water bath. Once you see a small amount of ice left in the vial, spray the vial with 70% Ethanol and wipe it down.
- 4) Transfer the vial into your Biosafety cabinet.
- 5) Using a 2 or 5ml pipet, pipet the cells out of the vial.
- 6) Transfer your cell suspension in to your coated plate that have the 4 ml media in it.
- 7) You should have a total working volume of 5ml of cell suspension in the flask; close the cap. Make sure cells are evenly distributed in the flask by moving the flask left and right five times. Move it up and down for an additional five times.
- 8) Place flask in a 37°C incubator with 5% CO<sub>2</sub>. If flask is not vented, please loosen cap.
- 9) Change media after 48 hours.