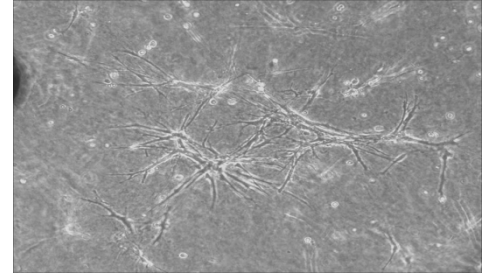


Human Brain Microvascular Angiogenesis Kit

Product Name	Human Brain Microvascular Angiogenesis Kit
Catalog Number	EP003
Product Format	6,12, and 24 well
Storage	37°C



GENERAL INFORMATION

Our Human Brain Microvascular Angiogenesis Kit formed capillary endothelial cells, which are connected by tight junctions with an extremely high electrical resistivity. This model will help overcoming the difficulty of delivering therapeutic agents to specific regions of the brain presents a major challenge to treatment of most brain disorders. The endothelial cells initially form small islands within the culture matrix. They subsequently begin to proliferate and then enter a migratory phase during which they move through the matrix to form threadlike tubule structures with lumens. These gradually join up by (1-2 weeks) to form a network of anastomosing tubules which closely resembles the capillary bed found in vivo.

This model can be used to study the following disease, but not limited to those applications:

- 1) Late-stage neurological trypanosomiasis (sleeping sickness)
- 2) Progressive multifocal leukoencephalopathy (PML)
- 3) Meningitis brain abscess epilepsy multiple sclerosis
- 4) De vivo disease Alzheimer's disease cerebral edema
- 5) Prion and prion-like diseases
- 6) HIV encephalitis
- 7) Rabies
- 8) Neuromyelitis optica

The Human Brain Microvascular Angiogenesis Kit contains all of the materials necessary to perform multiple angiogenesis assays in a 24-well format. The kit is designed that the testing materials, i.e. compounds, conditioned media, or tissue explants, can be added into the system at any time, ranging from the onset of vasculogenesis to advanced angiogenesis. The resulting effect on tubule formation (tubular length, number of branches et al) can be monitored throughout the whole process under inverted fluorescence microscope.

Reagents and Materials Provided:

- (1) 1 x vial of mixture of GFP-tagged Human Brain Microvascular ECs and RFP-tagged supporting cells (-80°C or liquid N₂)
- (2) 1 x 24-well Alpha Coat Solution coated plate (Room temperature, for 2 months)
- (3) 1 x 500ml of Endo-Growth Medium (4°C)

Protocols: Day 1

1. Pre warm Endo-Growth Medium to 37°C in a water bath
2. Accurately pipette 24ml Endo-Growth Medium into a 50ml Falcon tube;
3. Rapidly thaw the vial of cryopreserved cells in a 37°C water bath;
4. Transfer all cells gently into 24ml pre warmed Endo-growth medium;
5. Mix well the cells gently using a serological pipette;
6. Add 1.0ml of cell suspension to each well of the pre coated 24-well plate.
7. Make sure the cells are evenly dispersed in the wells.
8. Place the plate in an incubator (37°C, 5% CO₂ and humidified).

Day 2

9. Take the plate from the incubator and examine cells under inverted fluorescence microscopy (GFP positive Human Brain Microvascular ECs should sparsely and evenly distributed among RFP positive human supporting cells).
10. Wash the cells one with 2 ml of PBS
11. Add 2.0ml of fresh Endo-Growth medium (control) or Experimental media (Endo-Growth medium, plus pro- or anti-angiogenic regents according to customer's needs).
12. Place the plate back into the incubator. Day 4, 6, 8, 10, 12, and 14..... 13. Replace the medium every 2 days until the end of the experiments.

