



ABSTRACT

Introduction: The mechanisms of molecular and cellular interactions at the blood-nerve barrier (BNB) are incompletely understood.

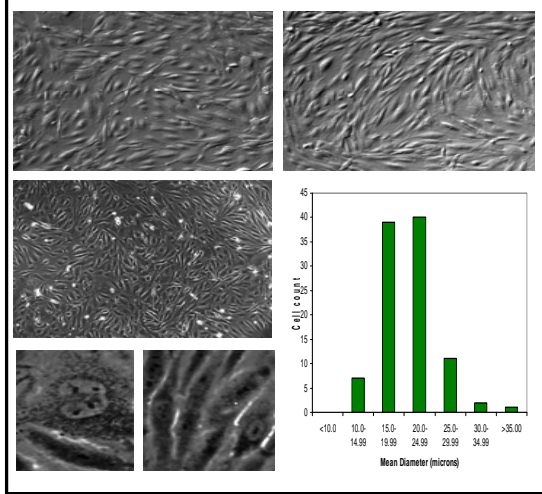
Objectives: To develop a human *in vitro* blood-nerve barrier (IVBNB) model that retains essential BNB characteristics.

Methods: Primary human endoneurial endothelial cells (pHEndECs) were isolated and purified from decedent human sciatic nerves via endoneurial stripping, enzymatic digestion, and density centrifugation. The IVBNB model was developed by plating pHEndECs on collagen-coated transwell inserts. pHEndECs were characterized using contrast and electron microscopy, immunocytochemistry, flow cytometry and polymerase chain reaction. IVBNB barrier properties were measured. Preliminary studies of leukocyte trafficking on cytokine-activated pHEndEC monolayers were also performed.

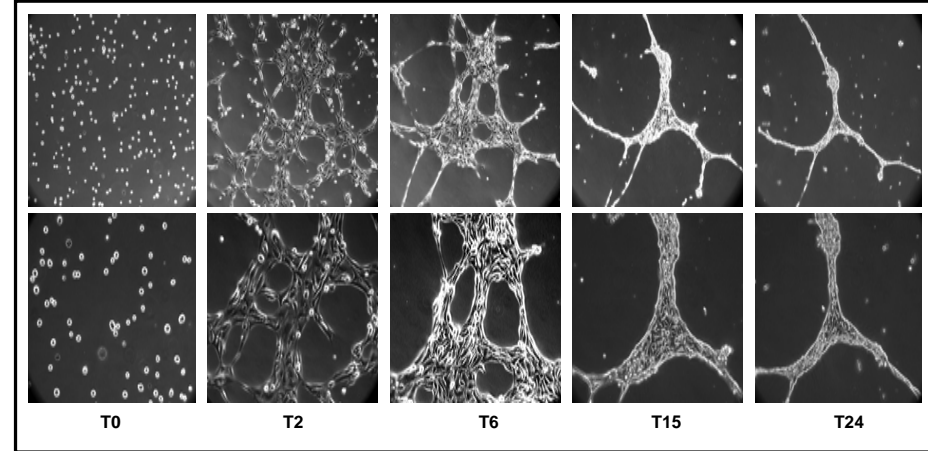
Results: pHEndECs are spindle-shaped, contact inhibited and differentiate to form capillary-like tubes *in vitro*. They are Ulex Europaeus Agglutinin-1, von Willebrand factor-positive and endocytose acetylated low density lipoprotein. The IVBNB model demonstrated high transendothelial electrical resistances (~160 $\Omega \cdot \text{cm}^2$), low solute permeability to fluoresceinated high molecular weight (70 kDa) dextran (~0.7 %) and histological evidence for intercellular tight junctions. Leukocyte rolling, firm arrest/ adhesion, post-arrest locomotion and transmigration were also observed in real-time.

Conclusions: The human IVBNB model provides an avenue to study physiological and pathologic processes at the BNB *ex vivo*.

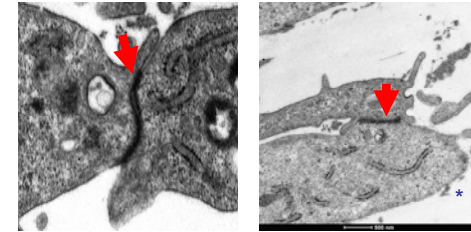
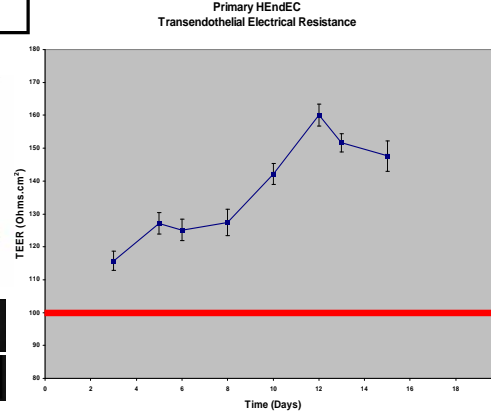
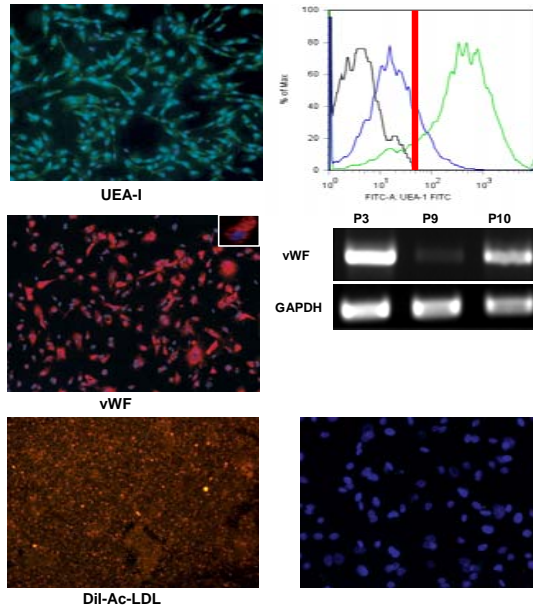
Primary Human Endoneurial Endothelial Cells (pHEndECs)



Capillary-like network formation and angiogenesis



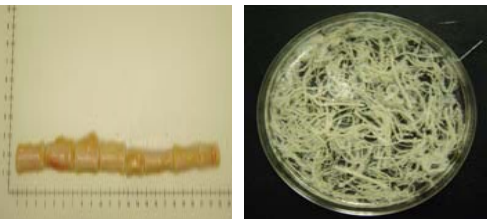
Phenotypic Characterization of pHEndECs



TRANSMISSION ELECTRON MICROSCOPY SHOWING INTERCELLULAR TIGHT JUNCTIONS AND PINOCYTOTIC VESICLES *IN VITRO*.

POTENTIAL APPLICATIONS OF THE IVBNB

- Endogenous solute/ macromolecular transport studies
- Drug transport studies
- Toxin/ xenobiotic influx/ efflux studies
- Mechanisms of leukocyte transmigration
- Microbial invasion processes
- Chemokine translocation/ presentation mechanisms



Human Sciatic Nerve and Endoneurial bundles

