Supplemental Figure 1. Methodology for fBMVECs isolation from human fetal brain tissue (16-22 weeks). A. Vessels are separated by centrifugation in 17.5% Dextran solution in a 15 mL conical tub. Vessels collect along the wall of the tube while the remnant tissue and myelin collects at the top. B-D. Brightfield images of fBMVEC colonies. B. Images of initial colonies following isolation indicated by yellow arrows. Scale bar at 100 microns C. 10x image of a colony a few days following isolation. Scale bar 200 microns D. Confluent colonies display typical cobblestone endothelial shape and morphology. Scale bar at 100 microns.

Supplemental Figure 2. Sorting of fBMVECs increases purity of EC cultures. f BMVECs were sorted for ICAM-1+/PDGFR β - using a BD Influx Sorter in order to obtain pure EC cultures. 3 fetal donors were stained for the astrocyte marker GFAP (Red) and pericyte marker α–Sma (Green) pre- and post- sorting. After sorting, cultures contain a minimal number of cells positive for astrocyte or pericyte makers (0-1%).

Supplemental Figure 3. Immunofluorescence staining of zona occludens -1 (ZO-1) and von willibrand factor (VWF) in *fBMVEC* cultures A. *fBMVEC*s from 3 donors were stained for ZO-1 (Green) and Dapi (Blue). Expression of the tight junction protein was primarily restricted to the cells borders. Scale bars at 20 microns. B. fBMVECs from 3 donors were stained for VWF (Red) and Dapi (Blue). All cells were positive for this endothelial marker (10x images). Scale bars at 100 microns.

Supplemental Figure 4. Absolute Resistance Values are plotted for *fBMVECs,* *aBMVEC*s, human coronary artery endothelial cells (HCAECs) and Human embryonic kidney cells 293 (HEKs). Absolute resistance is calculated by subtracting resistance values from empty wells. Resistance measurements were taken at 4,000Hz. Cells were plated at 20,000 per well and grown for 3-5 days. Media was exchanged and resistance was measured for 24h (shown is immediately prior and after media exchange). For comparisons with measurements using the EVOM system, which reads at 12.5Hz, resistance measurements for *fBMVECs* were taken at 25Hz (data not shown). Absolute resistance values were ~14,000 Ohms, which converts to approximately 550 ohm·cm^2 when multiplied times 0.03985 cm^2 -the area of the electrode on an ECIS 96W20idf PET 96 well plate: http://www.biophysics.com/cultureware.php).

Supplemental Figure 5. Full Western blot of *fBMVEC*s and aBMVECs. Cell lysates for fetal and adult donors were separated by SDS-page and probed for zona occludens-1 (ZO-1), multidrug resistance-associated protein-5 (MRP-5), platelet endothelial cell adhesion molecule-1 (PECAM-1), transferrin receptor (TfR), occludin, glucose transporter-1 (Glut-1), claudin-5, and β-Actin. Shown are samples from 4 fetal and 4 adult donors.