Disrupted wall shear stress alters autocrine signaling of brain microvascular endothelial cells at the Blood-Brain Barrier

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BACKGROUND

Flow induced wall shear stress (SS) is known to contribute significantly on the phenotype of microvascular endothelial cells in blood-brain barrier (BBB) formation. Together with the presence of astrocytes, dynamic in vitro models provide more realistic physiological properties of the neurovascular unit[1]. Yet, both direct and indirect relationships have been proposed between brain microvascular damage and abnormal flow patterns caused by upstream stiffened vascular beds[2]. We hypothesize that higher SS rates are responsible of BBB malfunction via autocrine signaling.

METHODS AND RESULTS

In our study, we cultured primary human brain microvascular endothelial cells (HBMVEC) with astrocyte conditioned media (ACM) under different levels of SS on a flow-perfusion bioreactor. Tight junction formation in our dynamic model was evaluated by immunocytochemical analysis of the tight junction marker Zonula Occludens 1 (ZO-1) (Figure 1). Expression levels of Claudin 5, P-glycoprotein (Pgp) and Glucose Transporter 1 (GLUT1) were measured by Western Blot. Results showed an up-regulation of the expression in dynamic cultures compared to static cases (Figure 2). Then, conditioned media was collected from HBMVEC cultured under different SS (SSECM), from 0 to 20 dyn/cm² and used to culture static co-culture models of the BBB. Functional analysis (dextran permeability) showed that SSECM has a positive effect on the BBB (Figure 3).

Immunocytochemical analysis, Western Blot analysis and permeability studies support that shear stress induces BBB properties, but no damage is detected in any case. However, endothelial conditioned media from dynamic cultures alter permeability in static cultures, indicating endothelial secretome alterations induced by shear.

REFERENCES