New tools for understanding the blood brain barrier

Awardee: Chenghua Gu
Award: Pioneer Award
Awardee Institution: Harvard Medical School

The central nervous system (CNS) requires a tightly controlled environment free of toxins and pathogens to provide the proper chemical conditions for synaptic transmission. This environment is maintained by the 'blood brain barrier' (BBB), which is composed of highly specialized blood vessels whose endothelium display specialized tight junctions and unusually low rates of transcellular vesicular transport (transcytosis). While BBB breakdown has recently been associated with various neurological disorders, an intact BBB also poses a major obstacle for drug delivery to the CNS. Little progress has been made on manipulating the BBB due to a significant knowledge gap in understanding how BBB function is regulated and identifying the essential molecular constituents governing its processes. This limited understanding has also thwarted our ability to therapeutically manipulate the BBB.

The major impediment to understanding the BBB is identifying its essential constituent and unraveling the mechanism by which these key regulators control BBB function. However, the current *in vitro* models rely on fully differentiated endothelial cells, which already contain unique properties that prevent their use in reconstitution studies. Similarly, the main technique to study the BBB has been EM, however its static snapshots do not provide information on active and dynamic vesicular transport, directionality, or their specific routes to allow investigators to interrogate the key molecular mechanisms that regulate BBB integrity. Recently, we mapped the precise timing of BBB formation and then identified molecules with possible roles in BBB function from simple transcriptome comparisons between CNS and peripheral endothelial cells. Surprisingly, we also found that instead of a physical buildup or disruption of structurally important tight junctions as previously thought, transcytosis regulation seems to be the more likely the major mechanism underlying BBB integrity. In characterizing these developmental properties, I realized that these findings are just the tip of the iceberg and that truly fundamental questions remain in identifying the core pathway and understand how they regulate BBB function. New tools thus are needed for understanding the BBB. Here we propose first to develop a new stem cell-based system to allow reconstitution of a functional BBB *in vitro*, and then to develop a genetic-optical system for monitoring the functional integrity of the BBB *in vivo* at subcellular resolution in real time. This integrated approach will address fundamental questions about the regulation of the BBB, which will then lead to more effective therapeutic strategies and specific targets for BBB restoration and manipulation.