PV1: Gatekeeper of Endothelial Permeability

Endothelium is a monolayer of endothelial cells that lines the inner surface of blood vessels and plays a critical role in the maintenance of blood-vessel function and homeostasis. The endothelium is a semipermeable barrier that regulates the passage of molecules between the blood and the underlying interstitial space to meet the metabolic needs of the tissue cells. Caveolae are small (50–80 nm) invaginations of the plasma membrane in many vertebrate cell types, especially abundant in endothelial cells, adipocytes, and muscle cells (1). Caveolae are important vesicle carriers responsible for endocytosis and transcellular transport (transcytosis) in endothelial cells and regulate endothelial cell permeability, angiogenesis, mechanoprotection from shear stress, and signal transduction (2). Accumulating evidence indicates that caveolae participate in physiological processes and the pathogenesis of numerous human diseases, including cancer, atherosclerosis, diabetes, diabetic retinopathy, interstitial lung disease, and muscular dystrophy (3–8). A spoke-like diaphragmatic structure spans the neck region of endothelial caveolae and controls caveolae-mediated uptake. PV1 (plasmalemmal vesicle associated protein-1) is an endothelial cell–specific membrane protein that is involved in the formation of caveolar diaphragms (9, 10). PV1 is a key mediator in the transcytosis of albumin across endothelial cells, inflammation-induced permeability, and leukocyte migration (11, 12). Therefore, PV1 is considered an interesting novel therapeutic target for treating cancer and vasogenic edema.

Endothelial dysfunction and injury are the pathological hallmarks for inflammatory diseases, such as acute lung injury and sepsis. In pathological states of increased endothelial permeability, transcytosis of fluid and macromolecules crossing the endothelium exceeds the clearance capacity of lymphatic vessels, resulting in pulmonary edema, impairment of gas exchange, and hypoxemic respiratory failure. Caveolae-mediated transcytosis plays an important role in LPS-induced transendothelial albumin permeability (13). The role of PV1 in caveolae-mediated transcytosis and permeability remains poorly understood. Evidence from previous studies indicates that the expression of PV1 is regulated by VEGF (vascular endothelial growth factor) signaling (14, 15). VEGF-induced vascular permeability and angiogenesis is regulated by caveolae-mediated transcytosis (15). Caveolins and cavins are major structural coat proteins around the bulb of caveolae (16). Loss of caveolin-1 or cavin-1 by genetic deletion or mutations results in reduction of caveolae and PV1 protein levels (17, 18). As a major component of the caveolar diaphragm, PV1 is believed to also contribute to caveolae-mediated transcytosis and endothelial permeability. In this issue of the *Journal*, the study by Jones and colleagues (pp. 531–539) describes investigation into the role of PV1 in caveolae-mediated uptake and transport endothelial-barrier function (19).

Germline deletion of PV1 leads to premature mortality due to severe noninflammatory protein-losing enteropathy and disruption of vascular homeostasis (11). Inducible endothelial cell–specific PV1 knockout ($PV1^{i\Delta EC}$) mice were used in the study to investigate the role of PV1 in regulation of permeability properties of the continuous endothelium and changes in the structure of caveolae. Ascites and hemorrhaging in lungs were observed in $PV1^{i\Delta EC}$ mice. The extravascular water content in lungs of $PV1^{i\Delta EC}$ mice was also elevated compared with that of control animals. However, $PV1^{i\Delta EC}$ mice did not exhibit any changes in blood cell counts compared with wild-type mice. Furthermore, increased transendothelial permeability to fluid and albumin were observed in $PV1^{i\Delta EC}$ mouse lungs by measuring the lung capillary filtration coefficient and albumin uptake and transportation. Endothelial cell–specific deletion of PV1 failed to increase brain transendothelial albumin permeability because of fewer caveolae and a higher expression of tight-junction proteins in brain microvessel endothelial cells (20, 21). Varying degrees of endothelial permeability were also observed in multiple fenestrated organs after endothelial cell–specific deletion of PV1. The increase in endothelial permeability in lungs and other organs, secondary to endothelial PV1 deletion, resulted in progressive loss of plasma protein and reduction in arterial pressure. Electron morphological analysis of caveolae in lung tissue revealed that endothelial PV1 deletion prevented caveolar diaphragm formation, as well as increasing the diameters of the caveolar neck, bulb, and depth. Accompanied by caveolar shape changes, increased gold-albumin particles were found in the bulb region of caveolae in $PV1^{i\Delta EC}$ mice, suggesting that loss of the PV1 diaphragm in caveolae caused albumin uptake and tissue edema. PV1 deletion, however, did not affect the total caveolae number, which is consist with the findings of another study (10).

The study provides visible evidence in support of PV1 functioning in caveolae-mediated transcytosis and endothelial permeability. Loss of the diaphragm in PV1-depleted endothelial cells changed caveolar shape and enhanced caveolae-mediated endocytosis and intracellular albumin transport from the blood plasma to the subendothelial or interstitial space. Over the past several years, multiple studies have considerably enhanced our understanding of PV1's structure and possible functions under normal physiological conditions and in pathophysiological processes using gene-knockout approaches. However, there are still many open questions that need to be addressed to advance our understanding of the role of PV1 in regulating endothelial permeability. Besides functioning as a barrier-like structure (diaphragm) to control plasma protein uptake into caveolae and caveolae-mediated transcytosis, PV1 is also a structural component of endothelial fenestrae and transendothelial channels (9). Thus, it is still not clear what degree of endothelial hyperpermeability is

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caused by dysfunctional caveolae-mediated transcytosis. Evidence from previous studies suggested that PV1 is upregulated in various pathophysiological processes associated with tumorigenesis, proangiogenic, or proinflammatory responses (13, 22–24). As PV1 is upregulated by VEGF stimulation, it is believed to be a modulator of VEGF-induced vascular permeability and angiogenesis. PV1 upregulation and diaphragm formation by VEGF is dependent on the activation of PI3K and p38MAPK (p38 mitogen-activated protein kinase) signaling pathways (14). The upregulated PV1 expression and caveolae formation may be a compensatory mechanism to increase transendothelial permeability.

The study adds significantly to our understanding of PV1's function in formation of diaphragms, as well as in its biogenesis and regulation. The findings serve as a basis for future investigations that will translate this understanding into the context of diseases and possible therapeutic implications. ■

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