How to Cross the Lymphatic Fence

Lessons From Solute Transport

Joanna Kalucka, Laure-Anne Teuwen, Vincent Geldhof, Peter Carmeliet

Mammals contain 2 vascular systems: the blood and the lymphatic system. Because the heart pumps the blood under pressure, fluid leaks out from distal capillaries into the interstitium. Lymph vessels then take up this leaked interstitial fluid and transport it back into blood vessels, thereby ensuring fluid homeostasis.1 Malfunctioning of lymph formation leads to deforming lymphedema, for which no curative therapy exists. Therefore, understanding how lymph is formed is a fundamental unresolved question in vascular biology. Because lymphatic capillaries have a mainly junction-free architecture, it was assumed that paracellular (and not transcellular) transport is the predominant mechanism of lymph formation. However, Triacca et al2 report in this issue of Circulation Research that transcellular and paracellular pathways are equally important for lymphatic solute transport and that both transports contribute to the increased lymphatic permeability on a rise in transmural flow and in case of inflammation. Triacca et al2 thus offer a long-awaited answer to the conundrum whether transcellular transport in lymphatic endothelial cells (LECs) contributes to lymph formation. These insights might pave the way for future treatments of lymphedema.

Furthermore, the lymphatic system is important for immune surveillance in mammals, serving as the main conduit of antigens and antigen-presenting cells from the periphery to lymph nodes. Finally, lymphatics are crucial for the absorption of dietary fats. These processes all depend on the organized movement of fluid, solutes, and cells across the lymphatic vessel wall (Figure 1).

Traditional dogma postulates that increased interstitial pressure pulls apart the anchoring filaments of the LECs, thereby allowing fluid passage into the lymphatic capillary—the so-called paracellular route. For long, this passive lymph formation obeying the Starling law was thought to be the sole lymph-generating mechanism, although it was already postulated decades ago that substances could cross the lymphatic endothelium through vesicular transport (the transcellular route).3 However, only now, in this issue of Circulation Research, Triacca et al2 describe the directional vesicular transport in LECs and further elucidate the mechanism of active lymph formation.

By using albumin uptake assays, the Swartz laboratory showed that in addition to the known paracellular routes, LECs also transport solutes transcellularly2 (Figure 2A). Furthermore, they studied the effect of transmural flow on the different transport routes. Previously, the authors demonstrated that transmural flow increases paracellular lymphatic permeability by downregulating and delocalizing VE (vascular endothelial)-cadherin and PECAM-1 (platelet and endothelial cell adhesion molecule 1).4 However, in this study, they prove that after applying transmural shear stress, the transcellular transport is also greatly enhanced (Figure 2B). A similar mechanism is seen in blood vessel endothelial cells (BECs), where shear stress increases transcellular transport by upregulating caveolin, a crucial component of transcellular transport.5 In line, Triacca et al2 report that the flow-mediated increase in transcellular transport in LECs correlates with an increase in clathrin-positive and caveolin-1-positive vesicles, which both require dynamin for their formation. However, the question remains why LECs use transcellular transport because this active process requires GTP and ATP, instead of solely relying on passive paracellular transport. It also remains intriguing which metabolic pathway LECs use to generate energy for vesicle transport, given that the levels of glycolysis (the major source of ATP in BECs) are lower in LECs than BECs.6 Also, given that laminar flow decreases glycolysis in BECs,7 it remains to be unraveled whether transmural flow also affects energy production for transcellular transport in LECs. For now, these questions remain unanswered and await further characterization of LEC metabolism.

The lymphatic network contains initial lymphatics and collecting ducts.1 Initial lymphatics consist of overlapping LECs, which have loose button-like adhesion junctions that...
open on increasing interstitial pressure and allow lymphatic fluid to enter the vessel. Collecting lymphatics contain a tighter layer of LECs with zipper-like intercellular junctions and are enclosed by a continuous basement membrane and perivascular cells. The collecting lymphatics are therefore less permeable than initial lymphatics and provide transport rather than uptake of lymph. However, LECs are plastic and can differentiate from a button-like to zipper-like phenotype in inflammatory conditions. This process might impair interstitial fluid clearance and promote swelling of inflamed tissues. However, the Swartz group now reports that on treatment with proinflammatory stimuli (eg, tumor necrosis factor-α), LECs increase solute transport by upregulating both the paracellular and transcellular transport routes. It will be interesting to unravel whether these changes in transport are accompanied by changes in zipper-like versus button-like phenotypes.

Lymphatic vessels contribute to the resolution of inflammation by draining excessive interstitial fluid. In models of skin inflammation and Crohn’s disease, lymphangiogenesis therefore reduces the progression of inflammation. However, functional activation of these vessels is also important. With the findings of Triacca et al in mind, upregulation of the transcellular pathway in LECs, which would improve fluid uptake, could thus enhance fluid drainage. However, more study is necessary to assess whether this would be pharmacologically achievable and therapeutically beneficial.

The findings of Triacca et al might perhaps also offer new therapeutic opportunities. For instance, in the brain, failure of lymphatic drainage of the waste product amyloid β (Aβ)
seems to contribute to the pathogenesis of Alzheimer’s disease. Therefore, facilitation of Aβ elimination might help to prevent cerebral amyloid angiopathy and counteract cognitive decline. An elegant—though speculative and as yet untested—means to eliminate Aβ might involve promoting transcellular lymphatic uptake of interstitial Aβ. This process could be further facilitated by administering cell-penetrating peptides that use transcytosis to cross endothelial barriers, at least if these peptides were modified to bind Aβ. While such therapeutic strategy remains hypothetical at this stage, it highlights how the insights of Triacca et al might create novel testable therapeutic opportunities.

As is often the case with an article that provides a leap forward in our understanding, it also invites to raise questions. First, would other large molecules like immunoglobulins also use transcellular transport, leading to potentially enhanced antigen delivery to the lymph nodes during inflammation? Second, would large molecules require specific receptor-mediated transcytosis, as described in BECs for transferrin, insulin, immunoglobulins, and cytokines? Third, is there a cross-talk between the paracellular and transcellular routes to coordinate lymph formation or cellular transport? Fourth, why do LECs prefer to upregulate active transcellular transport over gratuit passive paracellular transport? Is this because the relative surface for paracellular convection is too small, resulting in a satiable passive paracellular transport? Is it because active transport permits tight control of lymphatic endothelial cells regulate changes in solute transport by fluid stress. As is often the case with an article that provides a leap forward in our understanding, it also invites to raise questions. First, would other large molecules like immunoglobulins also use transcellular transport, leading to potentially enhanced antigen delivery to the lymph nodes during inflammation? Second, would large molecules require specific receptor-mediated transcytosis, as described in BECs for transferrin, insulin, immunoglobulins, and cytokines? Third, is there a cross-talk between the paracellular and transcellular routes to coordinate lymph formation or cellular transport? Fourth, why do LECs prefer to upregulate active transcellular transport over gratuit passive paracellular transport? Is this because the relative surface for paracellular convection is too small, resulting in a satiable passive paracellular transport? Is it because active transport permits tight control of lymph flow, as transcytosis of a single caveola is rapid and does not require >30 s. Last, which metabolic pathways do LECs use to produce energy for transcellular transport? As nutrient availability in the blood differs from that in the interstitium and lymph, it is likely that LECs use different metabolic pathways to produce energy. Finding an answer to these and other outstanding questions promises to become an exciting journey.

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References

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