

Using three-dimensional imaging to capture the development of the kidney lymphatics

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ABSTRACT

Organotypic roles of lymphatic vessels partly arise from organ-specific mechanisms of lymphatic development, but there is a paucity of information regarding how the lymphatics of the kidney form. We surmounted the challenges of imaging renal lymphatics by combining wholemount immunofluorescence for multiple lymphatic markers, refractive index matching and confocal microscopy; generating a three-dimensional timeline of lymphatic development in mouse embryonic kidneys down to cellular resolution. During normal development, kidney lymphatics first appear as a plexus beneath the nascent renal pelvis at embryonic day 14.5. Thereafter, kidney lymphatics assemble in distinct stages of maturation and extension towards the renal cortex but are largely restricted to the renal hilum. We identified a population of cellular clusters expressing lymphatic markers and anatomically distinct from the main lymphatic tree. Finally, we applied our imaging pipeline to demonstrate anomalies in lymphatic architecture preceding cyst formation in a mouse model of polycystic kidney disease, the most common genetic cause of chronic kidney disease and associated with oedema and inflammation. In conclusion, we have characterised kidney lymphatic development in health and disease. We also provide evidence to suggest that kidney lymphatics form by a combination of extension of pre-existing vessels and *de novo* formation of lymphatic endothelium.