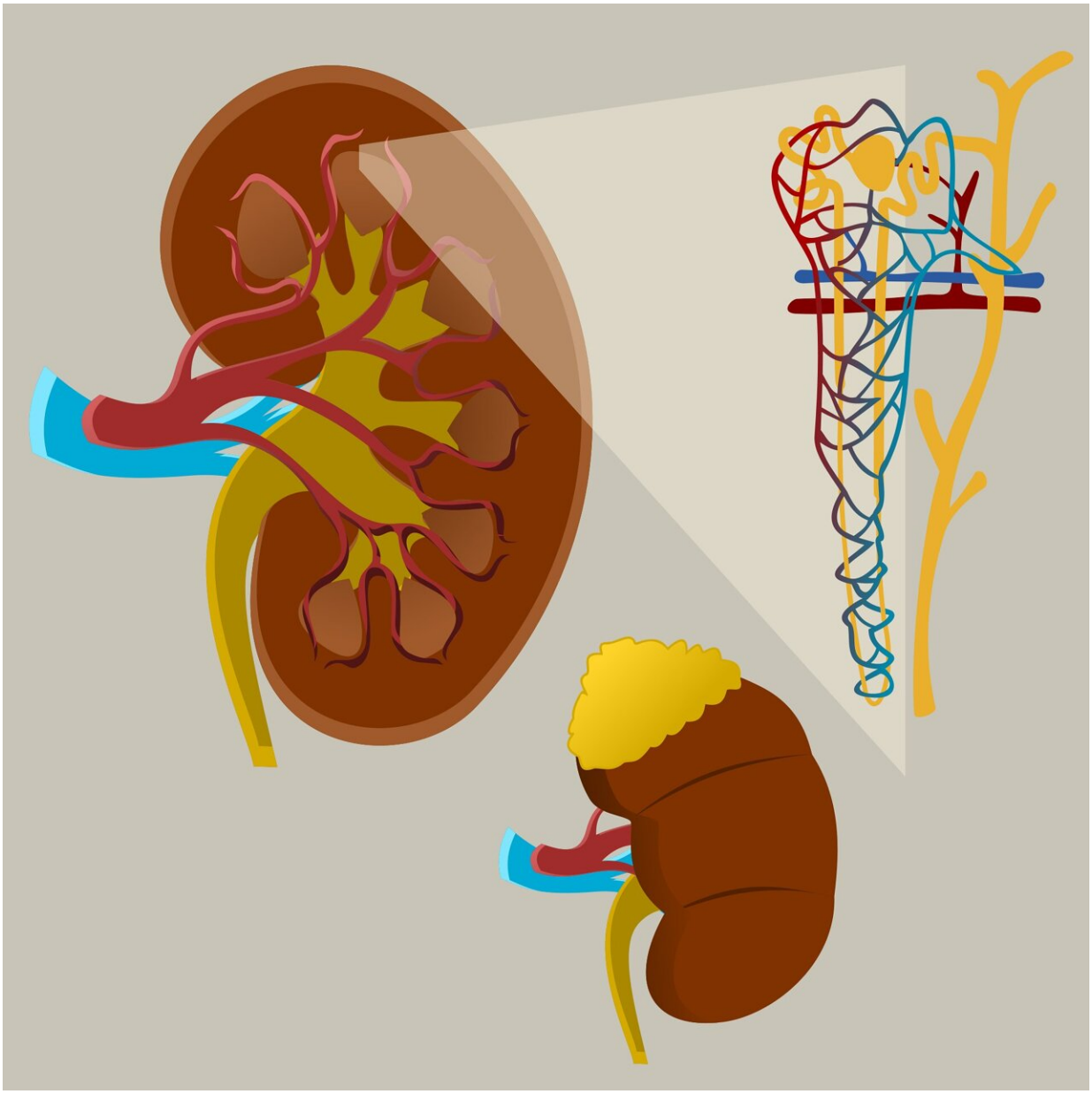
Kidney resident macrophages have distinct subpopulations and occupy distinct microenvironments

September 28 2022, by Jeff Hansen



Credit: Pixabay/CC0 Public Domain

Macrophages are immune cells that engulf and digest pathogens, cancer cells or cellular debris. The kidneys—like other tissues in the body—contain kidney resident macrophages, or KRMs, from the time of birth. These KRMs protect the kidney against infection or injury and help maintain tissue health by phagocytosis of debris or dying kidney cells.

In other organs, the locations of macrophages affect their functions. Now James George, Ph.D., and colleagues at the University of Alabama at Birmingham (UAB) report for the first time that the mouse kidney contains seven distinct KRM populations located in spatially discrete microenvironments, and that each subpopulation has a unique transcriptomic signature—a measure of which genes are active, which suggests distinct functions.

The UAB study, published in the journal *JCI Insight*, is an application of spatial transcriptomics, which *Nature Methods* crowned as the 2020 Method of the Year.

"Stratification of KRMs into specific zones within the kidney was previously unknown," George said. "The spatial location of macrophages impacts their function in other tissues, such as the lung, spleen and liver, and shapes their response to an immunological challenge. Although many disease states have known connections with KRMs and targeting populations holds great therapeutic promise, successful design and implementation of such strategies are limited by our current understanding of KRM regulation and response to injury as a function of

time."

George, co-corresponding author Anupam Agarwal, M.D., and UAB colleagues traced these KRMs in normal kidneys, and in kidneys after experimental injury caused by restricting the blood flow for 19 minutes. Such acute kidney injury can lead to chronic kidney disease, so knowledge of changes in the KRM subpopulations after injury is an important part of the KRM atlas of the mouse kidney. Such an atlas will serve as a point of reference for future studies into the role of the resident macrophage system in the normal and injured kidney.

The injured kidneys were examined at 12 hours and at one, six and 28 days after injury.

"Following insult, we tracked the subpopulations as they appeared to relocate throughout the tissue, suggesting possible locomotion by these cells in response to injury," George said. Macrophages have the ability to move, similar to amoebas.

At 28 days after injury, three of the macrophage subpopulations largely returned to the locations where they were found before injury, but four subpopulations remained scattered throughout the kidneys. "Thus," George said, "our data support a long-hypothesized dysregulation of the immune system following acute kidney injury that could be a major factor contributing to increased risk for chronic kidney disease following an acute kidney injury event."

Humans have more than 1 million nephrons in each of their two kidneys. A nephron is the tiny, functional unit of the kidney, removing fluid from the blood, and then returning most of that fluid back to the blood while retaining waste urine that will flow through the ureter to the bladder. Different portions of the nephron perform different functions, and the researchers found that the distinct macrophage populations were

associated with distinct portions of the nephron.

The research began with single-cell RNA sequencing of 58,304 KRMs isolated from whole mouse kidneys. Through analysis of 3,000 variable genes, they identified seven major distinct subpopulations that have unique transcriptomic signatures—the messenger RNAs transcribed from active genes.

The differentially expressed genes in six of the clusters indicated at least one specific function. For example, George said, "The most significant gene ontology terms in Clusters 1, 3 and 6 were involved in anti-bacterial, antiviral and anti-fungal responses. Cluster 2 contained terms related to responses to iron, phagocytosis and wound healing, suggesting involvement in homeostatic functions. Clusters 0 and 4 mapped to few terms, but the analysis contained references to tumor necrosis factor and apoptosis."

"These disparate gene ontology mappings suggest that each cluster executes a distinct transcriptional program that could be a function of the location in which each cluster resides."

The locations for the clusters were found by placing a thin slice of the kidney on a Visium Spatial Gene Expression microscope slide that is about one-quarter of an inch square. The technology in the Visium system allowed the researchers to locate where in the kidney anatomy each subpopulation resides based on their transcriptomic signatures.

Two of the clusters in normal kidneys were located in the cortex, the outer region of the kidney. Four were in the medulla, the area below the cortex, and one was in the papilla, or central region of the kidney. One example of the importance of location was the coordinated positioning of three subclusters to protect the kidney from infection. "The transcriptomes and locations of Clusters 1, 3 and 6 depict a strategic

immune barrier from the ureter, the most common origin of kidney infections," George said.

Importantly, the KRM transcriptomic atlas at 28 days after injury—with many KRM subpopulations no longer expressing their original profiles and existing within new locations—was persistently altered. "Given the continued disruption in transcriptional and spatial distribution beyond acute injury, KRMs may influence the transition to chronic kidney disease," George said. "A single acute kidney injury event drastically increases the risk for the development of chronic kidney disease, although the mechanisms that underlie that transition remain unclear."

At UAB, George is a professor in the Department of Surgery, and Agarwal is a professor in the Department of Medicine Division of Nephrology. Co-first authors of the study are Matthew D. Cheung and Elise N. Erman, UAB Department of Surgery.

More information: Matthew D. Cheung et al, Resident macrophage subpopulations occupy distinct microenvironments in the kidney, *JCI Insight* (2022). DOI: 10.1172/jci.insight.161078

Provided by University of Alabama at Birmingham