Intra- and inter-organ communication during early pyelonephritis

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Introduction: Tissue microbiological studies have shown that as early as 8 hours after onset of a localized pyelonephritic infection, IFN-γ is secreted by the spleen as a response to the infection. Accumulating evidence shows that the nervous system receives information from the immune system via sensory neurons, and is able to modulate inflammation via the cholinergic anti-inflammatory pathway. In this work we demonstrate that such a well-regulated and rapid signaling system could be responsible for the inter-organ communication between the kidney and the spleen during a pyelonephritic infection.

Methods: *In vivo* an ascending urinary tract infection (UTI) model, as well as a micro-puncture infection model was used in order to induce pyelonephritic infection in rats. Kidneys have been harvested at end point in order to perform immunohistochemical (IHC) analysis of the tissue. The micro-puncture infection model, where GFP^+-expressing UPEC is infused directly into single proximal tubules of exposed kidneys in the living animal, results in a temporally and spatially controlled infection that can be visualized in real-time using multiphoton imaging. Visualization of cell populations and blood flow is enabled by intravenous injections of different sized fluorescent dextran conjugates designed to either stay in the vascular system or enter specific cells.

In vitro renal epithelial cell lines (A498 and CRL4031) and peripheral (THP-1) macrophages were stimulated with pathogen-associated molecular patterns (PAMPs, such as live bacteria or purified LPS) and damage-associated molecular patterns (DAMPs, such as ATP) for 4 hours in order to investigate immune (IL-6, ATP) responses through ELISA analysis of supernatants. Primary mouse dorsal root ganglia cells (DRGs) were stimulated with PAMPs and DAMPs in order to investigate both the immune (IL-6, ATP) and neural (CGRP) responses through ELISA analysis of supernatants.

Results: Through IHC analysis of infected rat kidney tissue we have found that nerves are present in the kidney. Further, some nervous projections reach into the infected foci, where they can be exposed to PAMPs and DAMPs. Both primary mouse DRG cells and renal epithelial cells have immune responses to stimulation with LPS (PAMP). Further, renal epithelial cells secrete extracellular ATP after 4 hours of infection, and ATP (DAMP) stimulation of primary mouse DRG cells result in a neural response with CGRP release. CGRP in its turn, seem to have an attenuating effect on the inflammatory response of infected macrophages (THP-1).

Conclusions: Our work shows possible roles of intra- and inter-organ communication during an early pyelonephritic infection. Nervous projections are present in the kidney, and could potentially sense the presence of an infection in the kidney via PAMP and DAMP stimulation. Such stimulation can give rise to an immune as well as a nervous response. Collected, our data suggests a mean of activation of the cholinergic anti-inflammatory pathway, enabling the inter-organ communication between the kidney and the spleen during an early pyelonephritic infection.