Beyond Tissue Injury—Damage-Associated Molecular Patterns, Toll-Like Receptors, and Inflammasomes Also Drive Regeneration and Fibrosis

Hans-Joachim Anders* and Liliana Schaefer†

*Nephrological Center, Medizinische Klinik und Poliklinik IV, University of Munich, Munich, Germany; and †Pharmazentrum Frankfurt, Institute of General Pharmacology and Toxicology, Goethe-University of Frankfurt/Main, Frankfurt/Main, Germany

ABSTRACT

Tissue injury initiates an inflammatory response through the actions of immunostimulatory molecules referred to as damage-associated molecular patterns (DAMPs). DAMPs encompass a group of heterogeneous molecules, including intracellular molecules released during cell necrosis and molecules involved in extracellular matrix remodeling such as hyaluronan, biglycan, and fibronectin. Kidney-specific DAMPs include crystals and uromodulin released by renal tubular damage. DAMPs trigger innate immunity by activating Toll-like receptors, purinergic receptors, or the NLRP3 inflammasome. However, recent evidence revealed that DAMPs also trigger re-epithelialization upon kidney injury and contribute to epithelial-mesenchymal transition and, potentially, to myofibroblast differentiation and proliferation. Thus, these discoveries suggest that DAMPs drive not only immune injury but also kidney regeneration and renal scarring. Here, we review the data from these studies and discuss the increasingly complex connection between DAMPs and kidney diseases.


DAMP GENERATION INSIDE THE KIDNEY

DAMP Release from Dying Cells

Cell death may or may not activate immunity.12 Apoptosis is a silent cell death because apoptosis maintains membrane integrity and DAMP release (Figure 1). Apoptosis is important for homeostatic cell clearance (e.g., of autoreactive lymphocytes during negative selection in the thymus or of senescent blood cells).12 Apoptosis involves a complex series of signaling events to induce surface expression of find-me and eat-me signals that foster phagocytic clearance.13 Uptake of apoptotic cells settles the phagocyte’s host defense modus and rather induces autoimmune or autoinflammatory disorders.3 Nephrologists should have been at the forefront of this debate because sterile inflammation drives the majority of kidney disorders.4–6 But how do sterile injuries trigger kidney inflammation?

The last decade revealed that injured cells release intracellular molecules that activate innate immunity just like pathogen-associated molecular patterns (PAMPs).7 Accordingly, such molecules were named damage-associated molecular patterns (DAMPs). PAMPs and DAMPs activate identical pattern recognition receptors including Toll-like receptors (TLRs) and inflammasomes, a process that induces kidney inflammation and immunopathology.9–11 This review provides an update on the different modes of DAMP generation and how this contributes to tissue remodeling in kidney disease.


Toxins, ischemia, and trauma trigger inflammation just like pathogens, but why? Inflammation was first defined by rubor, calor, dolor, tumor, and function laesa, which all represent responses of the body to injury.1 The discovery of pathogenic bacteria as a cause of inflammation 150 years ago led to the assumption that pathogens trigger most forms of inflammation. The fields of immunology and microbiology evolved and generated the popular concept that the immune system developed from the everlasting competition between host and pathogens, with inflammation being the battlefield.2 However, this concept did not cover numerous clinical observations, including toxin-, ischemia-, or trauma-related inflammation as well as autoimmune or autoinflammatory disorders.3 Nephrologists should have been at the forefront of this debate because sterile inflammation drives the majority of kidney disorders.4–6 But how do sterile injuries trigger kidney inflammation?

The last decade revealed that injured cells release intracellular molecules that activate innate immunity just like pathogen-associated molecular patterns (PAMPs).7 Accordingly, such molecules were named damage-associated molecular patterns (DAMPs). PAMPs and DAMPs activate identical pattern recognition receptors including Toll-like receptors (TLRs) and inflammasomes, a process that induces kidney inflammation and immunopathology.9–11 This review provides an update on the different modes of DAMP generation and how this contributes to tissue remodeling in kidney disease.
an anti-inflammatory phenotype, which enforces homeostasis.\textsuperscript{14,15} It has long been thought that tissue injury also involves apoptotic cell death, often based on terminal deoxynucleotidyl transferase–mediated digoxigenin–deoxyuridine nick-end labeling positivity, but this also identifies DNA breaks during cell necrosis or DNA repair.\textsuperscript{16} In fact, it has now become evident that injury-induced cell death mostly involves regulated necrosis (Figure 1).\textsuperscript{17} For example, necroptosis is a receptor–interacting serine/threonine-protein kinase RIP1/RIP3–mediated form of necrosis that is triggered by genotoxic stress as well as by ligands to TLR3/TLR4 and surface receptors of the TNF receptor superfamily.\textsuperscript{18} Necroptosis was documented to contribute to acute tubular necrosis in several AKI models.\textsuperscript{19–23} Cyclophilin D–mediated disruption of the mitochondrial transmembrane potential is another form of regulated necrosis involved in postischemic AKI.\textsuperscript{20} Remarkably, mice lacking both RIP3 and cyclophilin D no longer develop AKI, even upon extended times of renal ischemia.\textsuperscript{20} Ferroptosis is a glutathione peroxidase 4–mediated form of regulated necrosis specifically triggered by oxidative stress.\textsuperscript{24} Some types of regulated necrosis seem restricted to immune cells such as NETosis, a controlled explosion of activated neutrophils. NETosis supports bacterial entrapment and killing during host defense.\textsuperscript{25} NETosis has not yet been demonstrated in infective pyelonephritis but occurs in renal vasculitis with crescentic GN.\textsuperscript{26} Pyroptosis is a caspase–1–dependent and caspase–11–dependent necrosis of infected macrophages.\textsuperscript{27,28} Currently, no functional in vivo data document pyroptosis contributing to kidney inflammation.\textsuperscript{29,30} All of these forms of necrosis have the potential to release DAMPs from different intracellular compartments into the extracellular space (Figure 1, Table 1).

**DAMP Generation during Extracellular Matrix Remodeling**

The extracellular matrix (ECM) is another source of DAMPs.\textsuperscript{31} The renal ECM consists of collagens and elastic fibers, proteoglycans, hyaluronan (HA), and assorted glycoproteins, which undergo a constant turnover.\textsuperscript{31–36} Enzymatic degradation can turn immunologically quiescent ECM components into fragments that ligate TLRs, purinergic receptors, inflammasomes, or integrins of infiltrating and resident renal cells (Table 1).\textsuperscript{33,37–45} As a second mechanism, macrophages and renal resident cells stimulated by TGF–\(\beta\)
\textsuperscript{46–50} and proinflammatory cytokines\textsuperscript{42,51} de novo synthesize soluble DAMPs\textsuperscript{38,42,52} (Figure 2). For example, HA exists under physiologic conditions as a polymer with a high molecular mass of \(\approx 10^6\) Da. HA accumulates in kidneys during AKI,\textsuperscript{33,54} allograft rejection,\textsuperscript{54,55} interstitial nephritis,\textsuperscript{54,56} and lupus nephritis.\textsuperscript{54,57} During inflammation and fibrosis, HA is depolymerized by hyaluronidases, which generates low molecular weight fragments that interact with TLRs and the NLR family, pyrin domain–containing 3 (NLRP3) inflammasome (Figure 2, Table 1).\textsuperscript{33–35} Inflammation also breaks down the glycosaminoglycan heparan sulfate (HS).\textsuperscript{38} Heparanase-mediated HS degradation is a key process in diabetic nephropathy\textsuperscript{59–61} and CKD\textsuperscript{62} (Table 1). However, the majority of extracellular DAMPs\textsuperscript{63–67} are released by matrix metalloproteinases.\textsuperscript{40,68–72} Among those, the TGF–\(\beta\)–binding small leucine-rich proteoglycans biglycan and decorin,
in their soluble form, can promote sterile as well as pathogen-induced inflammation. Numerous inflammatory and fibrotic kidney disorders are associated with decorin and biglycan induction. #40, #73, #33, #74, #37, #40, #75. Transient overexpression of soluble biglycan in mice demonstrates that biglycan triggers inflammation in healthy kidneys and potentiates renal inflammation and fibrosis (Table 1). #38, #74, #76. Finally, there is overwhelming evidence for an anti-fibrotic activity of soluble decorin directly interacting with another crucial members of the TGF-β superfamily, such as connective tissue growth factor (CTGF), and inhibiting apoptosis of renal tubular epithelial cells (TECs) via the IGF type I receptor/Akt-signaling pathway (Figure 2). #33, #48, #49, #73, #77, #78.

**Kidney-Specific Modes of DAMP Generation**

Tamm–Horsfall protein (renamed uromodulin) is an adhesive, particle-forming protein that is exclusively secreted at the thick ascending limb of the distal tubule. Its adhesive nature coats all particles in the distal tubule such as cells (forming cellular casts) and cell debris (granular casts), crystals (supporting crystal aggregation), bacteria (supporting their clearance), and even serves as a sink for inflammatory cytokines, albumin, and so forth. #79. Uromodulin is immunologically inert inside the tubular lumen. Tubular injury, however, allows uromodulin leakage into the interstitial compartment where it turns into a DAMP that activates interstitial dendritic cells via TLR4. #80. The particle nature of uromodulin fosters phagocytosis and endosomal destabilization in dendritic cells, a process that activates the NLRP3 inflammasome resulting in the release of IL-1β. #81. This mechanism

<table>
<thead>
<tr>
<th>DAMP</th>
<th>Kidney Disease</th>
<th>Receptor and Effects</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intracellular DAMPs</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nucleus</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HMGB1</td>
<td>Sepsis, AKI</td>
<td>TLR2/TLR4-induced inflammation</td>
<td>#142–#145</td>
</tr>
<tr>
<td>Histones</td>
<td>Sepsis, AKI</td>
<td>TLR2/TLR4-induced inflammation</td>
<td>#89, #100</td>
</tr>
<tr>
<td>DNA/RNA/U1 snRNP</td>
<td>IC-GN, inflammation in HD</td>
<td>TLR3-induced mesangial cell activation</td>
<td>#156, #157, #169, #170, #172–#177</td>
</tr>
<tr>
<td>Mitochondrial DNA</td>
<td>Fibrosis, hypertension, metabolic syndrome</td>
<td>TLR9</td>
<td>#178</td>
</tr>
<tr>
<td>ATP</td>
<td>Sepsis</td>
<td>P2X7-mediated renal inflammation</td>
<td>#95, #179–#181</td>
</tr>
<tr>
<td>Extracellular DAMPs</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Biglycan</td>
<td>Renal fibrosis, lupus nephritis, DN, GN</td>
<td>NLRP3-, P2X7-induced inflammation, DC/macrophage activation via TLR2/TLR4</td>
<td>#38, #42, #95, #165</td>
</tr>
<tr>
<td>Decorin</td>
<td>Sepsis</td>
<td>TLR2/TLR4, NLRP3-, P2X7, DC/macrophage activation</td>
<td>#185, #186</td>
</tr>
<tr>
<td>Fibrinogen</td>
<td>FSGS, MPGN, Interstitial fibrosis</td>
<td>Podocyte activation via TLR2/TLR4, Fibroblast activation via TLR2/TLR4</td>
<td>#90, #185, #187</td>
</tr>
<tr>
<td>Fibrinectin</td>
<td></td>
<td>TLR4</td>
<td>#78</td>
</tr>
<tr>
<td>Hyaluronan</td>
<td></td>
<td>TLR2/TLR4-induced inflammation, not confirmed in mesangial cells</td>
<td>#124, #189–#191</td>
</tr>
<tr>
<td>Heparan sulfate</td>
<td>AKI, lupus nephritis, DN</td>
<td>TLR4/CD44</td>
<td>#62, #163, #192</td>
</tr>
<tr>
<td>Versican</td>
<td>DN, CKD</td>
<td>TLR4-induced DC maturation</td>
<td>#62, #193, #194</td>
</tr>
<tr>
<td>Amyloid-β</td>
<td>CKD</td>
<td>TLR2</td>
<td>#195, #196</td>
</tr>
<tr>
<td>HDL, CVD in uremia in absence of uremia</td>
<td>CVD in uremia in absence of uremia</td>
<td>NLRP3-activation in DCs/macrophages</td>
<td>#198–#200</td>
</tr>
<tr>
<td>Crystals</td>
<td>Oxalate nephropathy, Adenine nephropathy</td>
<td>NLRP3-activation in renal DC</td>
<td>#84, #97, #171</td>
</tr>
<tr>
<td>Kidney-specific DAMPs</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uromodulin</td>
<td>AKI</td>
<td>?, cytokine+DAMPs clearance from kidney?</td>
<td>#80, #81, #201</td>
</tr>
</tbody>
</table>

IRI, ischemia-reperfusion injury; IC-GN, immune complex GN; HD, hemodialysis; RAGE, receptor for advanced glycation end products; MPGN, membranoproliferative GN; EDA, extra domain A; CVD, cardiovascular disease.
is another avenue for activating innate immunity in tubular injury (Figure 3).

The kidney is a preferred site of particle formation (e.g., from anorganic mineral-related crystals or proteins aggregates such as myoglobin and light chains). Crystals or crystalline proteins act as DAMPs by activating the NLRP3 inflammasome in dendritic cells and macrophages. In the distal tubule, uromodulin coats crystals and can trigger immune activation via the aforementioned mechanism. Some disorders, including oxalosis, involve diffuse crystallization in the renal interstitium, in which dendritic cells pick up crystals into lysosomal compartments and again activate the NLRP3 inflammasome (Figure 3). The same mechanism is likely to also contribute to renal inflammation and tubular injury in tumor lysis syndrome and other crystalline nephropathies.

**DAMP EFFECTS ON KIDNEY INJURY AND REPAIR**

**DAMPs Activate Systemic Alloimmunity and Autoimmunity**

Circulating DAMPs activate pattern recognition receptors on immune cells in the circulation or in lymphoid organs. By activating these receptors, DAMPs mimic PAMPs and convert tolerogenic immune responses into immunogenic immune responses. In particular, the activation of antigen-presenting cells such as dendritic cells and B cells enhances antigen presentation, expansion of antigen-specific lymphocyte subsets, and antibody production. This process plays an important role in kidney diseases involving alloimmunity and autoimmunity such as kidney transplantation, immune complex GN, or ANCA vasculitis.

**How DAMPs Activate the Kidney’s Innate Immune System**

**Immunostimulatory “Autoadjuvant” Effects**

DAMPs activate TLRs on renal parenchymal cells as well as on resident and infiltrating immune cells (for a review of the involved signaling pathways, see refs.11,88). TLR-mediated cell activation involves the induction of NF-κB–dependent inflammatory cytokines and chemokines in all cells (Figures 4 and 5). Additional cell type–specific effects include the up-regulation of adhesion molecules, increased vascular permeability in renal endothelial cells, and filtration barrier dysfunction in podocytes. Resident and infiltrating mononuclear phagocytes also host the NLRP3 inflammasome that integrates DAMP signals such as ATP, histones, HA, biglycan, crystals, or uromodulin into the activation of caspase-1,91–97 Caspase-1 and caspase-11 confer proteolytic cleavage of pro-IL-1β and pro-IL-18 into the mature cytokines, which are then secreted and trigger local inflammation inside the kidney (reviewed in ref.10).

**DAMPs with Direct Killing Effects**

Histones release not only activates TLR2, TLR4, and the NLRP23 inflammasome (Figure 4),89,94,98,99 but extracellular histones also elicit direct toxic effects on vascular endothelial cells. For example, histone injection into the renal artery results in widespread renal necrosis, which is only partially prevented in TLR2/TLR4-deficient mice. Whether histone-induced cell death is a passive or regulated form of necrosis (or both) is unknown to date.

**DAMPs as Autoantigens**

Some DAMPs are important autoantigens that drive intrarenal autoimmune disease. For example, when nucleosomes and double-stranded DNA (dsDNA) are released from glomerular cells, they can promote glomerular binding of lupus autoantibodies that trigger lupus nephritis (Figure 4). Neutrophils undergoing NETosis can also be an important source of the DAMPs in lupus. In lupus, secondary necrosis of apoptotic
cells exposes hypomethylated dsDNA or U1 small nuclear ribonucleoprotein to TLR7 and TLR9 in dendritic cells and B cells, which drives RNA and DNA autoantibody production, systemic inflammation, and lupus nephritis.

**How DAMP Signaling Drives Kidney Regeneration**

Recent data now suggest that DAMPs can accelerate tubule regeneration upon injury either in a direct or indirect manner. Renal progenitor cells are scattered along the thick limbs of the proximal and distal tubule segments of the human kidney. These cells have a higher stress resistance as terminally differentiated TECs and preferentially survive tubule injury. TLR2-agonistic DAMPs enhance the clonal expansion and differentiation of these progenitor cells within the tubule, which accelerates tubule regeneration (Figure 5). As a second mechanism of DAMP-driven kidney regeneration, TLR4-agonistic DAMPs activate interstitial dendritic cells and macrophages to release IL-22, which, in turn, activates the IL-22 receptor, which is exclusively expressed on TECs (Figure 5). IL-22 receptor signaling accelerates tubule re-epithelialization from surviving TECs in the recovery phase of AKI. Whereas TLR4 blockade in the injury phase abrogates AKI by preventing immunopathology, TLR4 blockade in the regeneration phase delays tubule recovery. This dual role of TLR4 agonistic DAMPs in AKI illustrates that DAMPs confer not only immune injury but also wound healing as part of danger control. Thus, DAMPs that ligate TLR2 or TLR4 drive kidney regeneration.

**How DAMP Signaling Affects Kidney Fibrosis/Sclerosis**

Renal fibrosis is considered an aberrant form of injury- or stress-induced wound healing accompanied by excessive ECM deposition. Leukocytes secrete growth factors and matrix metalloproteinases that activate mesangial cells and interstitial fibroblasts to secrete ECM components. ECM-related DAMPs link renal inflammation and fibrogenesis by ligating TLRs, integrins, purinergic receptors, and inflammasomes, a process that also

---

*Figure 3.* Crystals and uromodulin act as DAMPs to induce renal inflammation. Crystals precipitate in the proximal tubule, the distal tubule, and/or in the interstitial compartment of the kidney. Crystals kill TECs. In addition, crystals can be taken up by interstitial dendritic cells via phagocytosis. Lysosomal leakage and potassium efflux (not shown) provide a signal to activate the NLRP3 inflammasome, which cleaves caspase-1 and subsequently pro-IL-1β and pro-IL-18 (not shown). IL-1β ligates the IL-1 receptor (IL-1R) on renal parenchymal cells as well as immune cells, which triggers NF-κB-dependent cytokine and chemokine release. In distal tubule injury, uromodulin leakage into the interstitial compartment activates dendritic cells via TLR4 and the NLRP3 inflammasome. As uromodulin also binds to crystals, crystal precipitation in the distal tubule is likely to involve both mechanisms.
leads to the release of fibrogenic cytokines and chemoattractants (Figure 4). It is of particular interest that although intracellular and extracellular DAMPs are structurally unrelated, they often signal via TLR2/TLR4 as shared receptors (Table 1), thereby resulting in different downstream events depending on the initial trigger. Histo-

Figure 4. DAMP effects in immunopathology. DAMPs activate several classes of pattern recognition receptors, which all induce an immediate activation of innate immunity (i.e., systemic and tissue inflammation). Most DAMPs activate TLR2 and TLR4 at the cell surface. In particular, particles enter the cells via phagocytosis and trigger assembly and activation of the NLRP3 inflammasome. Nucleic acid–related DAMPs activate TLR7 and TLR9 in intracellular endosomes. All pattern recognition receptors finally drive the secretion of proinflammatory cytokines that then activate cytokine receptors on the same cell or on other cells. Extracellular histones also directly kill (endothelial) cells. The molecular mechanism of this process is poorly defined but may involve surface charge. Certain nuclear DAMPs also act as autoantigens in systemic lupus and contribute to lupus nephritis. Their adjuvant-like ability to also activate antigen-presenting cells via TLR7 and TLR9 strongly promotes autoimmunization and the expansion of autoreactive lymphocytes. Local release within the kidney also promotes intrarenal autoantigen recognition (e.g., by circulating autoantibodies and subsequent immune complex GN; not shown). LMW, low molecular weight; HSP, heat shock protein; MAL, MyD88 adaptor–like; IRF, IFN regulatory factor; TRAP, TNF receptor–associated protein; TRAF, TNF receptor–associated factor; TRIF, TIR domain–containing adapter inducing IFN-β; ssRNA, single-stranded RNA; dsDNA, double-stranded RNA; DC, dendritic cell; ASC, apoptotic speck protein; IRAK, IL-1 receptor–associated kinase; TIRAP, Toll IL-1 receptor domain–containing adapter protein; TCR, T-cell receptor; IFNAR, IFN-α receptor; IL-R, IL receptor; CCR, CC-chemokine receptor.
IL-8, and TNF-α, although this could not be confirmed in mesangial cells. HS signals through the TLR4/MyD88 pathway and promotes dendritic cell maturation and production of proinflammatory IL-6 and IL-12. HS also induces the release of IL-1α, IL-1β, IL-6, and TNF-α in macrophages. Biglycan and decorin, endogenous ligands of TLR2/TLR4, activate the mitogen-activated protein kinase p38, extracellular signal-regulated kinase, and NF-κB pathways, leading to the secretion of TNF-α, pro-IL-1β, and a series of chemotactic agents for neutrophils, macrophages, and T/B lymphocytes in either a MyD88- or TIR domain-containing adapter inducing IFN-β-dependent manner. Importantly, both small leucine-rich proteoglycans are orchestrating signaling pathways of diverse receptors probably in a hierarchical manner to sequentially induce signaling pathways. For example, biglycan clusters TLR2/TLR4 with the purinergic P2X7 and P2X4 receptors, which activates NLRP3 (Figure 4, Table 1). The NLRP3 inflammasome triggers secretion of IL-1β and IL-18, which are involved in renal fibrogenesis. NLRP3 activation in TECs also drives epithelial-mesenchymal transition during progressive renal fibrosis, which is associated with progressive renal fibrosis. For example, Nlrp3−/− mice display reduced tubular injury and interstitial fibrosis upon unilateral ureteral ligation compared with wild-type animals. Another study found an effect on early renal vascular permeability, rather than fibrosis, in this model. A role for NLRP3 in fibrosis may not necessarily involve only canonical (caspase/IL-1β/IL-18-dependent) inflammasome activation, because NLRP3 has additional (noncanonical) roles in SMAD2/SMAD3 phosphorylation of the TGF-β1 receptor signaling pathway. Whether this mechanism also operates within fibroblasts (as proposed in Figure 5) remains speculative at this point.

However, DAMP signaling in fibrosis is not only restricted to inflammatory pathways. ECM-related DAMPs also modulate crosslinking of matrix components and cytokine signaling in fibrogenesis. For example, the HS-proteoglycan, syndecan interacts via its HS chains with transglutaminase type 2, an enzyme that promotes ECM crosslinking in fibrosis. Consistently, the lack of syndecan protects from renal fibrosis. In addition, fibronectin can induce fibroblast differentiation via interaction with α5β1 integrin. As another avenue of DAMPs regulating fibrogenesis, decorin sequesters TGF-β in the ECM and competes with TGF-β for receptor binding. Decorin also inhibits CTGF signaling in fibroblasts. In addition, decorin down-regulates microRNA miR-21, which promotes interstitial fibrosis. Together, DAMPs also directly modulate ECM crosslinking and renal fibrogenesis (e.g., by modulating TGF-β and CTGF signaling).

**DAMPs in Distinct Kidney Disorders**

There is currently mostly experimental evidence for a role of DAMPs in kidney disease, as listed in detail in Table 1. Data obtained from mice deficient for DAMP receptors provide only indirect evidence on the role of DAMPs; hence, we do not further discuss this here because numerous reviews on this topic exist.

**AKI**

**Tubular Necrosis**

Among kidney disorders, AKI is most frequently associated with cell necrosis, which implies DAMP release in acute tubular necrosis. For example, septic, ischemic, or toxic forms of tubular necrosis...
involves the release of histones and high-mobility group protein B1 (HMGB1), which drive the associated sterile inflammatory and immunopathology that determines organ failure. In addition, lethality in sepsis relates to the release of HMGB1, histones, decorin, or biglycan. This process seems to preferentially involve DAMP-mediated endothelial dysfunction via TLR2/TLR4, which increases vascular permeability, shock, and hypoperfusion. HMGB1 also can facilitate ischemic preconditioning, which implies that HMGB1 exposure protects from subsequent postischemic AKI. This TLR2-mediated process involves the upregulation of Siglec as one of many counterregulatory mediators that limit DAMP-related immunity just like endotoxin tolerance. In addition, NLRP3-deficient mice were protected from postischemic AKI. However, apoptotic speck protein or caspase-1 deficiency as well as IL-1/IL-18 blockade were not protective, largely excluding an inflammasome-related role of NLRP3 in postischemic AKI and arguing for yet unknown noncanonical effects.

**GN**
Necrotizing GN is another form of AKI that involves DAMP release from renal cells. Data on necrotizing GN are currently limited to models induced in TLR2/TLR4-deficient mice or to serum and tissue expression levels of HMGB1 that correlate with disease activity. However, histones are released from necrotizing GN, and contribute to glomerular necrosis in crescentic GN in a TLR2/TLR4-dependent manner. Nuclear DAMPs also contribute to lupus nephritis by activating TLR7 and TLR9 outside and inside the kidney, as well as by activating dendritic cells and B cells, which enhances local as well as systemic autoimmunity.

**CKD**
Diabetic Nephropathy
Inflammatory responses mediated by activation of TLR2, TLR4, and NLRP3 inflammasome play key roles in the progression of diabetic nephropathy (DN). Under diabetic conditions, NLRP3 is activated by intracellular reactive oxygen species or extracellular ATP. Furthermore, hyperglycemia evokes the expression of the ATP receptor P2X4 in renal TECs of patients with type 2 DN, and this correlates with IL-1 cytokine release. The production of another proinflammatory molecule (i.e., HA) is also induced by hyperglycemia through a protein kinase C/TGF-β pathway. Besides HA, several DAMPs, including the glucose-inducible HMGB1, biglycan, and decorin, are overexpressed in diabetic kidneys and may trigger inflammation by activating TLR2/TLR4 receptors and NLRP3 inflammasomes.

Increased renal biglycan levels correlate with enhanced infiltration of macrophages and renal LDL accumulation, and appear to promote kidney injury. By contrast, because of its antiapoptotic effects on TECs and ability to neutralize TGF-β1 and CTGF, decorin is nephroprotective in DN. Whereas HA accumulation is considered as a marker of renal damage during DN and is potentially involved in the development of interstitial fibrosis, high molecular weight-HA reduces diabetes-induced renal injury. Thus, decorin and low molecular weight versus high molecular weight HA, orchestrating signaling of various receptors, appear to act in diabetic kidneys in a scenario more complex than canonical DAMPs.

**Lupus Nephritis**
Lupus nephritis involves immune activation by nuclear DAMPs that share autobody- and autoantigen features, and also contain adjuvant qualities. Ribonucleoproteins activate TLR7 and hypomethylated dsDNA activates TLR9 (Table 1), which enhances autoantigen presentation and the expansion of autoreactive lymphocytes. In addition, these TLR7- and TLR9-specific DAMPs trigger plasmacytoid dendritic cells to release IFN-α, which initiates antiviral gene transcription accounting for many of the unspecific (viral infection-like) symptoms of LN and IFN-related glomerular pathology. Furthermore, TLR7- and TLR9-specific DAMPs activate intrarenal macrophages and dendritic cells toward an M1 phenotype, a process that accelerates immunopathology in lupus nephritis. In addition, DAMP ligands of TLR2/TLR4 accelerate renal damage in lupus nephritis. Biglycan also aggravates lupus nephritis. Transient overexpression of soluble biglycan induces CCL2, CCL3, and CCL5 and aggravates murine lupus nephritis, whereas its deficiency suppresses disease activity and renal damage.

**Crystalline Nephropathies**
Crystal formation within the kidney creates a DAMP that has the potential to activate renal inflammation and immunopathology via the NLRP3 inflammasome (e.g., in renal dendritic cells). For this to occur, crystals need to reach the interstitial compartment, which happens during diffuse crystallization or upon tubular injury from inside the tubular lumen. NLRP3 activation results in IL-1β and IL-18 secretion, which initiates downstream inflammatory effects by activating their respective IL receptors. This was consistently demonstrated in animal models of acute and chronic oxalate nephropathy as well as adenine overload-induced CKD (Table 1).

**SUMMARY**
Tissue injury emits DAMPs as danger signals to activate danger control (e.g., inflammation for host defense). DAMPs can either be intracellular molecules that signal cell necrosis (HMGB1, histones, ATP), matrix constituents that signal extensive matrix remodeling (decorin, HA, HS, biglycan), or luminal factors that signal barrier destruction (urotholin). DAMPs activate TLRs, purinergic receptors, and inflammasomes in parenchymal cells and leukocytes. DAMP-induced inflammation initiates an autoamplification loop by triggering regulated forms of necrotic cell death, which aggravates immunopathology and further DAMP release. Hence, blocking DAMP signaling in the early injury phase of acute disorders limits immunopathology.
However, DAMPs have additional effects. Certain nuclear DAMPs (RNA/DNA) combine adjuvant and autoantigen qualities and thereby promote systemic lupus and lupus nephritis. TLR2-agonistic DAMPs activate renal progenitor cells to regenerate epithelial defects in injured tubules. TLR4-agonistic DAMPs induce renal dendritic cells to release IL-22, which also accelerates tubule repaphealization in AKI. Finally, DAMPs also promote renal fibrosis by inducing NLRP3, which also promotes TGF-β receptor signaling. It is likely that more exciting discoveries are to be made in this area.

ACKNOWLEDGMENTS
H.J.A. and L.S. are supported by the German Research Foundation (AN372/9-2 and 14-1 to H.J.A.; and Sonderforschungsbereich (SFB) 815, project A5, SFB 1039, project B2, Excellence Cluster Cardio-Pulmonary System and Landes-Offensive zur Entwicklung Wissenschaftlich-ökonomischen Exzellenz program Ub-Net to L.S.).

DISCLOSURES
None.

REFERENCES


DAMPs in Kidney Disease 1395
BRIEF REVIEW

www.jasn.org


Biglycan fragmentation in pathologies associated with extracellular matrix remodeling by matrix metalloproteinases. Fibrogenesis Tissue Repair 6: 9, 2013


91. Khandoga AG, Khandoga A, Anders HJ, Krombach F: Postischemic vascular permeability requires both TLR-2 and TLR-4, but only TLR-2 mediates the transendothelial migration of leukocytes. Shock 31: 592–598, 2009


