CHAPTER 83

Adaptive Immunity and Critical Illness

Wibke Schulte, Linda L. Maerz, Gilbert Moeckel, and Richard Bucala

OBJECTIVES

This chapter will:

1. Outline the activation of the innate and adaptive immune system in response to invading pathogens.
2. Define characteristics of the adaptive immune response and its interplay with the innate immune response in septic, ischemic, and nephrotoxic acute kidney injury.
3. Describe mechanisms of resolution of inflammation and progression to chronic kidney disease.

IMMUNE SYSTEM: AN OVERVIEW

The primary force that has driven the evolution of the human immune system has been the need to promptly recognize and appropriately respond to danger threatening the survival of the host organism. Danger, however, can take many forms. It may be the invasion of host tissue by infectious pathogens such as bacteria, viruses, or fungi, but it also may be the disruption of tissue from traumatic injury or the neoplastic transformation of normal tissue into cancer. As such, the immune system has evolved into a complex and sophisticated network of immunologic response mechanisms that orchestrate an effective host defense.

The immune system can be conceptualized as comprising two main components: first, the innate immune system, which encompasses a limited number of germline gene products and therefore responds immediately but unspecifically to a broad variety of threats; and second, the adaptive immune system, which becomes activated more gradually as it relies on gene rearrangement, selection, and clonal expansion to produce a specific, tailored response and long-lasting immunologic memory. As crucial as the immediate activation of the immune response is the timely resolution of this response to prevent further tissue damage and organ dysfunction. Without resolution, persistent inflammation promotes deleterious tissue remodeling and organ dysfunction and can promote transformation into cancerous neoplasia.

RECOGNITION OF FOREIGN MOLECULES

The skin and mucous membranes (e.g., in the airways or in the gastrointestinal tract) are natural physical barriers that act as the first line of defense to prevent invasion of external pathogens. A surface layer of mucus prevents microbial binding to the host cell, and tight junctions between cells preclude simple passage of pathogens into deeper tissue. Numerous pathogens have developed various strategies to overcome this initial most primitive form of defense and subsequently encounter more complex defense mechanisms of the host immune system as they invade the underlying tissue (Fig. 83.1).

Specific groups of invading microorganisms are recognized via pathogen recognition receptors (PRRs), which are expressed on epithelial barriers as well as by cells mainly of the innate immune system such as dendritic cells and macrophages. These PRRs act as sensors of microbes and recognize conserved macromolecular motives from microorganisms, called pathogen-associated molecular patterns (PAMPs). Examples of bacterial PAMPs include lipopolysaccharide (LPS, the main virulence factor of gram-negative bacteria), peptidoglycan, lipoteichoic acid (a cell wall component of gram-positive bacteria), flagellin, and bacterial DNA. Damage-associated molecular patterns (DAMPs) are released by damaged host cells and contribute to the activation of the overall immune response. The stimulation of a specific family of PRRs named Toll-like receptors (TLRs) or of the NOD-like receptor (NLR) family of intracellular PRRs results in the triggering of downstream signaling cascades. Depending on the particular receptor engaged, this process leads to the activation of a transcriptional response program, which includes nuclear factor κB (NF-κB), followed by the production and secretion of cytokines, chemokines, and nitric oxide (NO). Proinflammatory cytokines lead in turn to an ensuing, coordinated activation of the innate and adaptive immune response.

INNATE IMMUNE RESPONSE

Cells of the innate immune system include myeloid cells (e.g., monocytes, macrophages, dendritic cells, granulocytes [eosinophils, basophils, neutrophils]), and innate lymphoid cells (e.g., natural killer [NK] cells). These cells are present at important sites of primary pathogen exposure, such as the airways, intestinal mucosa and skin, and are crucial for the initiation of an inflammatory response. For example, recent lineage- and temporal-specific tracking studies suggest that so-called tissue-resident macrophages, which are derived from embryonic progenitor cells without contributions from bone marrow-derived monocytes, are maintained in the periphery, where they patrol the tissue for invading microbes. Once a potential threat is encountered, these cells become activated and release chemokines (most importantly CCL2, also known as monocyte chemoattractant protein-1, MCP-1), which recruit and activate blood monocyte-derived Ly6C+ macrophages and dendritic cells. Thus tissue-resident macrophages act as sentinels, which recruit and assist in the "licensing" of leukocytes. In turn, the recruited innate immune cells release an array of proinflammatory mediators including cytokines (e.g., tumor necrosis factor-α [TNF-α]

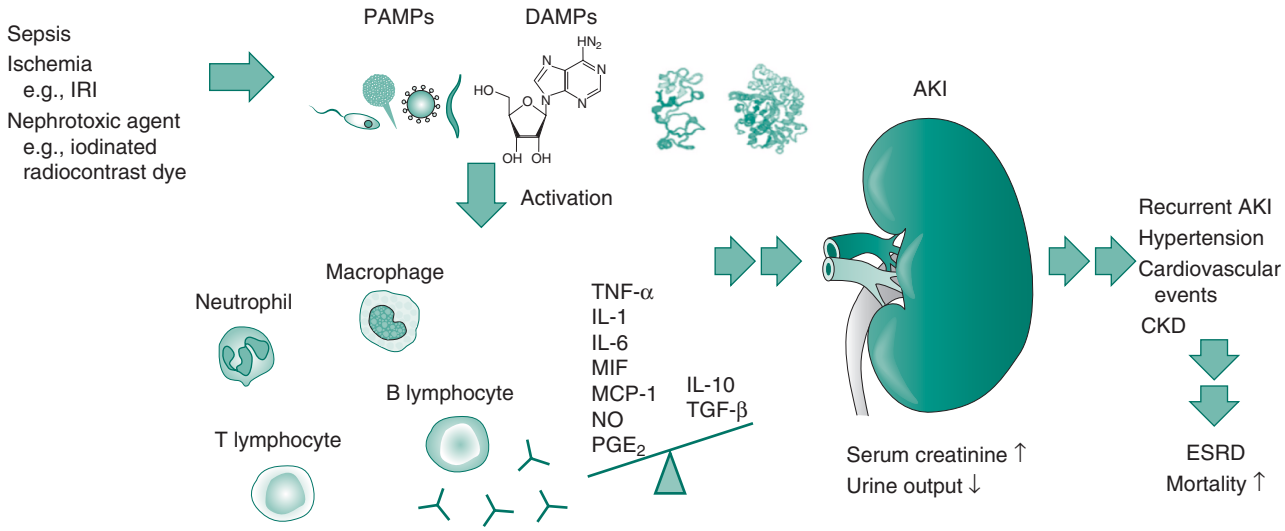

FIGURE 83.1 Schematic figure of sepsis, ischemia, or nephrotoxic agents leading to the activation of the innate and adaptive immune system via pathogen-associated molecular patterns (PAMPs, e.g., LPS) and damage-associated molecular patterns (DAMPs, e.g., adenosine). Depending on the quality and magnitude of the ensuing immune response, kidney dysfunction meeting AKI diagnostic criteria can occur. AKI increases the incidence of recurrent AKI, distant organ dysfunction, and CKD, thus enhancing the susceptibility to ESRD and accelerated mortality. *AKI,* Acute kidney injury; *CKD,* chronic kidney disease; *DAMPs,* damage-associated molecular patterns; *ESRD,* end-stage renal disease; *IL-1,* interleukin-1; *IL-6,* interleukin-6; *IL-10,* interleukin-10; *IRI,* ischemia-reperfusion injury; *MCP-1,* monocyte chemoattractant protein-1; *MIF,* macrophage migration inhibitory factor; *NO,* nitric oxide; *PAMPs,* pathogen-associated molecular patterns; *PGE_2,* prostaglandin E_2; *TGF-β,* transforming growth factor-β; *TNF-α,* tumor necrosis factor-α.

interleukin-1 [IL-1], IL-6, macrophage migration inhibitory factor [MIF]), chemokines (e.g., IL-8), prostaglandins, reactive oxygen, and nitrogen species to mount a robust proinflammatory response. Specific cytokines (e.g., IL-1 and TNF-α) and chemokines (e.g., IL-8 and MCP-1) activate the vascular endothelium, increase vascular permeability, and induce site-directed chemotaxis. Cytokines also facilitate the migration of leukocytes from the vasculature to the site of injury and enable interstitial leakage of protein-rich fluid, resulting in the "tumor" of local inflammation. In addition, the upregulation of inducible NO synthase in the adjacent microvasculature leads to increased synthesis and release of NO, which results in local vasodilation. The enhanced generation of prostaglandin E_2 (PGE_2) by cyclooxygenase produces the characteristic "rubor" of local inflammation. IL-1, TNF-α, and IL-6 can act on the hypothalamus to alter the thermoregulatory set point, thereby inducing fever. IL-6 induces acute-phase reactant protein production by hepatocytes. Although all these events promote a strong proinflammatory immune response, other innate immune cell products, such as IL-10 and transforming growth factor-β (TGF-β), are released simultaneously and play an important role in the resolution of the inflammatory response and in the initiation of tissue repair. Taken together, this accumulation of events results either in the elimination of the invasive pathogen and in the subsequent downregulation of the immune response or in a more vigorous immune response by triggering the simultaneous activation of the more specific adaptive immune system.

ACTIVATION OF THE ADAPTIVE IMMUNE RESPONSE

Although some pathogens can be cleared by the innate immune system alone, others survive these defenses and, as a result, trigger the induction of an adaptive immune response. It normally takes many days for the adaptive immune system to become fully activated. However, the resulting response is vigorous against pathogens, specific to the invasive microorganism and establishes long-lasting immunologic memory. The adaptive immune system also has the ability to elicit tolerance to an immunologic stimulus, which differs from the consistently broad robust responsiveness of the innate immune system against a threat.

Adaptive immunity is induced after the processing and presentation of antigens that are not native to the host organism by antigen-presenting cells. Once PAMPs of foreign antigens bind to PRRs, innate immune cells such as macrophages and dendritic cells upregulate major histocompatibility class II (MHC class II) and costimulatory B7 molecules, allowing them to serve as antigen-presenting cells. They take up foreign proteins by phagocytosis, degrade them, and present constituent peptides in association with MHC class II molecules and costimulatory molecules to T cells. The B7 costimulatory molecule on antigen-presenting cells binds to T cell CD28 to promote IL-2 production, which is necessary for clonal expansion of the particular T cell population. Several distinct subpopulations of T cells have been described in the literature. Likewise, it has become evident that certain macrophage cytokines direct the expansion of these subpopulations and thus shape the emerging adaptive immune response. Macrophage IL-12, for instance, activates T helper 1 (T_H1) cells, which express a panel of proinflammatory cytokines, such as interferon-γ (IFN-γ), TNF-α, and IL-2. In turn, these cytokines enhance macrophage antimicrobial activity. In contrast, T_H2 cells are activated by macrophage IL-4 and release predominantly antiinflammatory cytokines such as IL-10, IL-13, and TGF-β, which dampen immune responses and aid in wound repair. Unlike T cells, B cells are effector cells of the adaptive immune system that express specific immunoglobulin as the B cell receptor on their cell surface. Antigen binding to the expressed immunoglobulin leads to clonal expansion

of the particular B cell clone resulting in enhanced secretion of specific antibody against the antigen. The variable region of the antibody represents the antigen-recognition site, whereas the constant region binds dedicated Fc receptors on phagocytic cells. Thus binding of an antigen by immunoglobulin opsonizes the antigen for phagocytosis by cells of the innate immune system. Close communication between innate and adaptive immune cells therefore is crucial for determining the quality and magnitude of the ensuing immune response.

ADAPTIVE IMMUNE RESPONSE IN ACUTE KIDNEY INJURY

Despite remarkable advances in modern medicine, acute kidney injury (AKI) remains a challenging condition that affects approximately 5% of hospitalized patients and 30% of critically ill patients. Sepsis, ischemia-reperfusion injury (IRI) and nephrotoxic agents are among the major causes of AKI. However, burns, trauma, cardiac surgery (especially with cardiopulmonary bypass), and radiocontrast agents also can lead to AKI. AKI worsens the overall clinical course of affected patients by causing uremia, acid-base and electrolyte disturbances, and volume overload with consequential morbidity and mortality. In addition, AKI increases the likelihood of developing chronic kidney disease (CKD) and end-stage renal disease (ESRD). According to the Kidney Disease: Improving Global Outcomes (KDIGO) Foundation clinical practice guidelines, which were released in 2012, AKI is diagnosed by an abrupt decrease in kidney function evident by increased serum creatinine and decreased urine output over either a short (48 hours) or an extended (7 days) time frame. Importantly, most patients are diagnosed with AKI after the injury already has occurred, the disease is well established, and only supportive care with fluid replacement or renal replacement therapy is available. In this chapter we discuss some of the pathophysiologic mechanisms involved in AKI with special focus on adaptive immunity. A more detailed knowledge of the underlying disease processes will assist in the development of more effective diagnostic and therapeutic strategies to improve the clinical outcome of AKI.

SEPTIC ACUTE KIDNEY INJURY

The host response to sepsis is systemic; however, it remains unclear how ongoing immune processes directly affect various organs. For example, as the kidney receives approximately one fifth of the cardiac output, its tubules are continuously exposed to PAMPs, DAMPs, cytokines, reactive nitrogen, and oxygen species, and more, which enter the bloodstream in sepsis. This continual contact with inflammatory mediators either through the blood directly or via its filtrate in the tubular lumen leads to cellular stress and injury to the nephrons. In sepsis, a decreased glomerular filtration rate (GFR) results, which often is followed by transient or complete loss of kidney function, thus meeting diagnostic criteria for AKI. Surprisingly, histologic evidence of cell injury in biopsies performed in patients with septic AKI is remarkably scarce. Septic AKI generally is described as patchy, heterogeneous tubular cell injury with apical vacuolization but without tubular necrosis and without extensive apoptosis. This suggests that adaptive mechanisms are in place to protect the kidney from the acute septic inflammatory process, likely at the expense of kidney function. Recent literature suggests that tubules conserve energy through G1 cell-cycle arrest, that the dislocation of Na^+/K^+ ATPase to the apical or lateral cell segments prevents energy-consuming NaCl reuptake, and that the recruitment of glomerular shunt pathways diverts toxin-rich blood away from the kidney.

Numerous studies of sepsis pathophysiology have revealed a dysregulated immune response, leading to tissue damage, organ failure, and ultimately, death. These studies support the existence of proinflammatory and antiinflammatory responses in sepsis, while they suggest that proinflammatory processes predominate early in the disease process, whereas immunosuppression dominates in later phases of sepsis. As the adaptive immune system normally becomes fully activated in later stages of the inflammatory response, changes in adaptive immunity leading to immunosuppression and impairment in its necessary interactions with the innate immune system have been described mainly in septic patients. In sepsis, defective T and B cell mediated immunity has been reported and quantitative changes at the cellular level include lymphocyte depletion caused by increased apoptosis of T and B lymphocytes. Persistent lymphopenia in septic patients has even been shown to predict early and late mortality. More recent microarray gene expression studies from blood of patients with sepsis support a marked decrease in antigen presentation by innate immune cells to T cells with a resulting depressed T cell function that may be directly associated with the severity of organ failure in sepsis. Likewise, sepsis nonsurvivors show significantly lower expression levels of genes involved in antigen presentation and T cell function when compared with survivors. Sepsis also leads to increased relative numbers of $CD4^+$ $CD25^+$ regulatory T (Treg) cells in the systemic circulation. Treg cells are T cell subsets that control innate and adaptive immune responses by downregulating the proinflammatory effector activities of $CD4^+$ T and $CD8^+$ T cells, B cells, NK cells, and dendritic cells. Thus increased numbers of Treg cells contribute to lymphocyte anergy in sepsis, and particular Treg subgroups have been described as markers of poor prognosis. Alternatively, some studies have suggested that Treg cells play an important role in the termination of inflammatory and cytotoxic immune responses, and are important for achieving immunologic self-tolerance and a homeostatic response to infection. To date, the exact participation of these cells in sepsis pathophysiology remains unknown. Moreover, hypogammaglobulinemia, especially affecting immunoglobulin G (IgG), is a common finding in septic patients. Many different causes for this finding have been proposed, which include limited production or secretion of immunoglobulin by plasma cells, excessive catabolism, changes in IgG clearance by the complement system, vascular leakage secondary to endothelial dysfunction, and redistribution/sequestration in inflamed tissue. In summary, many alterations in the innate and adaptive immune response and in their interaction with each other have been described in sepsis. Future studies that examine both arms of the immune response simultaneously and temporally hold great promise to broaden our understanding of sepsis pathophysiology.

ISCHEMIC ACUTE KIDNEY INJURY

Initially, immune mechanisms were not expected to be involved in the pathogenesis of aseptic ischemic renal

injury. However, numerous studies over the past decade have demonstrated a crucial role of innate and adaptive immune responses in ischemic AKI caused by cold and warm ischemia. Cold ischemia is encountered in organ transplantation when the procured organ is immersed in cold perfusion solution. Warm ischemia occurs at physiologic temperatures and ends when perfusion is restored, either after completion of a vascular anastomosis or after resolution of organ hypoxia. Robust inflammatory responses usually begin during the initial ischemic event but accelerate upon reperfusion of the organ, resulting in what has been termed ischemia-reperfusion injury (IRI). In the kidney, IRI causes mechanical interruption of renal vascular endothelial integrity, leading to a rapid influx of immune cells through the disrupted endothelium. The resulting innate response is aggravated by enhanced transcription of genes encoding cytokines and chemokines that are induced by DAMPs and by hypoxia-inducible factors (HIFs), which are released after hypoxic or anoxic cell injury. TLR expression on tubular epithelial cells also contributes to the initiation of tissue-damaging innate responses. Studies have demonstrated that $CD4^+$ T cells contribute significantly to renal tissue damage in the early reperfusion phase, which was supported further by the finding that T cell–targeted medications (such as tacrolimus or mycophenolate mofetil) substantially alleviate early renal injury after IRI. These medications are used frequently to prevent allograft rejection in renal transplant patients. B cells also traffic into postischemic kidneys. They are activated and differentiate into plasma cells in the injury phase and enhance tubular atrophy and suppress tubular regeneration during recovery. In contrast, Treg cells increase in the late injury phase, promote tubular proliferation, and thus aid in renal regeneration and renoprotection. More recently, studies have identified activated T cells and effector memory T cells in the postischemic kidney several weeks after IRI, suggesting that these cells are involved in long-term structural tissue changes and possibly in the transition from AKI to CKD.

NEPHROTOXIC ACUTE KIDNEY INJURY

Large population-based studies have indicated that the combination of more than two nephrotoxic drugs (e.g., diuretics, angiotensin receptor blockers or angiotensin-converting enzyme inhibitors, nonsteroidal antiinflammatory drugs, aminoglycosides) increases the risk of AKI. In addition, iodinated radiocontrast dye-induced AKI (contrast induced nephropathy, CIN) is a well-known disease entity, which is thought to be caused by a combination of hypoxic and toxic damage with endothelial dysfunction. However, significantly less is known about the pathophysiology of nephrotoxic AKI, and the roles of various immune cells have been investigated primarily in mouse models of cisplatin-induced AKI. Injured or necrotic nephrons cause an influx and activation of immune cells, but cisplatin also acts as a sterile inflammatory stimulus. T cell numbers are increased in cisplatin-exposed kidneys and $CD4^+$ T cell–deficient mice are protected from cisplatin-induced mortality and renal dysfunction, indicating an important role of these cells in disease progression. Other studies have linked Fas-mediated apoptosis with cisplatin-induced AKI, which is mediated by Fas ligand (FasL) expressed on renal tubular cells and infiltrating T cells. In accordance with studies of other causes of AKI, renoprotective effects have been attributed to Treg cells in cisplatin-induced AKI, as these cells were demonstrated to attenuate renal injury and decreased macrophage infiltration. However, more detailed studies are needed to delineate the immune mechanisms involved in nephrotoxic acute kidney injury.

RESOLUTION OF INFLAMMATION

The resolution of inflammation is an active process that limits proinflammatory immune responses and activates tissue regeneration important for the restoration of organ function. Generally, the removal of the injurious insult significantly drives the resolution of inflammation; however, antiinflammatory responses and regenerative processes can begin while the infectious threat is still present. In the current literature, one of the most described mechanisms involved in the resolution of inflammation is the phenotypic switch of proinflammatory M1 macrophages toward alternatively activated M2 macrophages. M2 macrophages secrete antiinflammatory and proregenerative mediators, such as IL-10, IL-22, and TGF-β and thus drive tissue repair. This switch is triggered by various humoral factors secreted from neighboring immune cells into the immunologic milieu (including cytokines, chemokines) and direct cell-cell interaction (e.g., with Treg cells, which actively promote the regeneration process by suppressing innate immunity). In the kidney, the persistence of M1 macrophages is sufficient to induce glomerulosclerosis, tubular atrophy, and progressive CKD, whereas M2 macrophages accelerate tubular reepithelialization. This M1/M2 dichotomy has been defined in controlled experimental studies, and the true pathophysiology may be represented more accurately by a phenotypic continuum. Nevertheless, macrophages with M2 character significantly drive regeneration and enhance nephron survival during the recovery phase of AKI.

CHRONIC KIDNEY DISEASE

Nephron recovery is more likely to occur after short-term injurious triggers that are present in AKI compared with repetitive or persistent harmful insults, such as hypertension, diabetes mellitus, chronic ischemia, and long-term toxin exposure, which can lead to progressive CKD and eventually ESRD. However, numerous clinical observational studies have demonstrated that AKI is associated with subsequent kidney failure, such as recurrent AKI, incidental or progressive CKD (both through persistent structural kidney impairment caused by interstitial inflammation and capillary rarefaction), and distant organ dysfunction, such as elevated blood pressure (presumably caused by impaired sodium handling), and subsequent cardiovascular events (exacerbated by hypertension and CKD). Importantly, the presence of AKI also has been associated with increased mortality either through direct impairment of kidney function or via indirect effects on other organ systems. With an increased incidence of in-hospital AKI and an expanded population of AKI survivors, it is imperative to detect and diagnose AKI early to treat the disease more effectively and to develop optimal care practices to prevent long-term consequences. This is particularly important because AKI is currently no longer viewed as a self-limiting disease, but rather the initiation of a potential pathway to CKD, ESRD, and accelerated mortality.

Key Points

1. An effective host response against invading pathogens is crucial to ensure host survival. However, a tight regulation of the immune response is equally essential to maintain a healthy balance between protective and tissue damaging responses.
2. The innate and adaptive immune systems closely interact to ensure rapid pathogen removal, but they also downregulate each other to prevent tissue-damaging excessive inflammation.
3. In acute kidney injury (AKI), whether because of sepsis, IRI, or nephrotoxins, studies have elucidated various active inflammatory processes that involve both the innate and adaptive immune system. However, the exact interplay between different immune cell subtypes remains unclear.
4. We anticipate that ongoing research will expand our knowledge of AKI pathophysiology and lead to the identification of new therapeutic strategies that may be exploited for the more effective future treatment of not only AKI itself but also the pathologic sequelae of chronic kidney disease and end-stage renal disease.

Key References

1. Marshall JC, Charbonney E, Gonzalez PD. The immune system in critical illness. *Clin Chest Med*. 2008;29(4):605-616, vii.
2. Rivera A, et al. Innate cell communication kick-starts pathogen-specific immunity. *Nat Immunol*. 2016;17(4):356-363.
3. Gomez H, et al. A unified theory of sepsis-induced acute kidney injury: inflammation, microcirculatory dysfunction, bioenergetics, and the tubular cell adaptation to injury. *Shock*. 2014;41(1):3-11.
4. Jang HR, Rabb H. Immune cells in experimental acute kidney injury. *Nat Rev Nephrol*. 2015;11(2):88-101.
5. Anders HJ. Immune system modulation of kidney regeneration–mechanisms and implications. *Nat Rev Nephrol*. 2014;10(6):347-358.