CHAPTER 82

Innate Immunity and the Kidney

Steven M. Opal

OBJECTIVES

This chapter will:

1. Briefly review essential features of innate immunity as the first line of defense against microbial invaders.
2. Describe the mechanisms by which the innate immune system rapidly recognizes danger to host viability induced by either infectious or noninfectious tissue injury.
3. Detail the role of innate immune functions that enable specific elements of the adaptive immune system to become activated and begin the process of pathogen clearance without collateral damage to the host.
4. Enumerate the mechanisms by which the innate immune cell activation is held in check and begins to undergo the active process of resolution of inflammation and tissue repair.
5. Review some unique innate immune functions found in the kidney and urinary collecting system.

A BRIEF HISTORY OF DISCOVERIES OF THE INNATE AND ADAPTIVE HOST IMMUNE RESPONSES

The initial recognition of active host defense against microbial pathogens is credited to Élie Ilyich Metchnikoff, who realized amebocytes from starfish would migrate toward foreign invaders and ingest them. He reasoned that such a beneficial host defense would be passed on to higher organisms by Darwinian evolution and would likely be found in humans. He was first to confirm this fact by finding specialized cells in human blood engaged in what he coined to be "phagocytosis." Metchnikoff gained the support of Louis Pasteur, and he spent the rest of his career at the Pasteur Institute in Paris investigating the role of phagocytic cells in innate host defense. He observed two major types of phagocytic cells: large tissue-based macrophages and

smaller circulating "microphages," now known as neutrophils. Metchnikoff's observations were first scoffed at by his peers and public. George Bernard Shaw's play "The Doctor's Dilemma" was written as a humorous satire of Metchnikoff's ideas.[1]

While the cellular contribution of professional phagocytes to host defense was under study in Paris, a competing school of humoral immunity was championed in Germany. Behring and Kitasato of the Koch Institute in Berlin showed that immune serum from animals that survived an infection, in the absence of cells, could passively protect nonimmune animals. The protective factor, serum antibodies, were discovered in immune serum, along with series of proteins that amplified antibody activity, now recognized as the complement system. This work was conducted primarily by Paul Ehrlich et al. in Germany. Subsequent studies clearly established that humoral immune elements, myeloid cells of the innate immune system, and lymphoid cells of the adaptive immune system collaborate to defend patients from microbial invaders. Appropriately, Metchnikoff and Ehrlich shared the Nobel Prize in medicine in 1908, as recognition of their critical discoveries and as an acknowledgment of the equal importance of humoral and cellular immunity in host defense.

Recently, the Nobel Prize in Physiology and Medicine was shared by three scientists for unraveling the mysteries of innate immune sensing, pattern recognition, and Toll-like receptors (TLRs). In 2011 the Nobel Prize went to Bruce Beutler for discovering TLR4, the long sought-after cellular receptor that recognizes bacterial endotoxin. He shared the prize with Ralph Steinman for his discovery of dendritic cells and Jules Hoffman for developing the basic concepts and identity of the pattern recognition receptors in innate immune activation.[2]

INTRODUCTION INTO INNATE IMMUNITY

The innate immune response can be likened to the rapid response team found in most large hospital systems. This team consists of a specified group of healthcare workers, with a clear set of identifiable skills, and has the responsibility to immediately respond and rescue suddenly ill patients within the hospital. The code team does not know what they will find when they arrive at the patient's room. It may be a plugged endotracheal tube in a patient needing ventilator support, chest pain with a dysrhythmia, someone fallen out of bed with a bumped head, a massive gastrointestinal hemorrhage, an adverse drug reaction with anaphylactic shock, or a myriad of other crises that require immediate attention. The team members must make a rapid assessment of the problem, begin appropriate care, and call in other specialists to assist if necessary. Our innate immune system serves similar functions when our physical barriers to infection have been breached by a traumatic injury to the integument.

Any injury can pose a threat to host survival on two fronts: infection by entry of pathogens from the external environment or exsanguination with loss of the internal milieu from bleeding. The innate immune response system is called into action within minutes by danger signals sent out by sentinel macrophages and soluble pattern recognition receptors such as the mannose-binding lectin and alternative complement pathway. Local generation of chemical signals in the form of chemokines and other chemoattractants is detected by adjacent endothelial cells and circulating neutrophils and platelets to begin the delivery of phagocytic cells and antimicrobial peptides to engage and eliminate any microbial pathogens invading the tissues. Concomitantly, the clotting system is activated when exposed collagen binds to circulating von Willebrand factor (VWF), thereby creating long multimers of VWF to which platelets bind and aggregate. Simultaneously, tissue factor exposed by endothelial membrane disruption initiates the coagulation cascade with fibrin deposition. The innate immune response and coagulation systems are coactivated and coregulated to protect against excessive bleeding and infection.[3] Innate immune sensing of danger is a highly evolved and complex network of cellular and soluble receptor molecules that recognize highly conserved molecular patterns found in essential structures for microorganisms, but not for human cells.[4] These exogenous ligands are referred to collectively as PAMPs (pathogen-associated molecular patterns). However, innate immune signaling is not simply initiated by "self" versus "non-self" recognition. Innate immune receptor molecules also recognize endogenous human pattern molecules leaked from dead or dying cells and elements of ground substances that make up the intercellular matrix. The endogenous pattern molecules released during tissue injury are danger signals detectable by innate immune cells by the same type of pattern-recognition receptors that detect PAMPs. The danger signals from tissue injury are DAMPs, or damage-associated molecular patterns. The most widely recognized pattern recognition receptors of innate immunity are the Toll-like receptors (TLRs) but are not the only receptors that participate in the host response to pathogens.[3–7] A brief summary of major ligands that make up the PAMPs, DAMPs, and their cellular receptors expressed on immune cells is found in Table 82.1 and detailed, along with the major intracellular signaling pathways, in Fig. 82.1.[5]

CELLULAR AND HUMORAL COMPONENTS OF HOST INNATE IMMUNE DEFENSES

Cells of the Innate Immune System

Immune cells of the myeloid cell line constitute the major, but not only, cellular components of innate immunity. These cells are called "professional phagocytes," because this is their primary function, but other epithelial and somatic cells can ingest potential microbial pathogens, along with damaged or dying cells. Myeloid cells encompass circulating progenitor monocytes and tissue resident macrophage cells, including hepatic Kupffer cells, lymph-associated macrophages in spleen and lymph nodes, Langerhans cells in the skin, pulmonary alveolar macrophages, and highly specialized dendritic cells found primarily along mucosal surfaces. Polymorphonuclear cells, or neutrophils, constitute the other myeloid cell line of innate immunity.

Other specialized cells such as M cells differentiate from the epithelial cells of the gastrointestinal tract in response to signals from lymphocytes and make up the specialized gut-associated lymphoid tissues called Peyer's patches.[7,8] These cells continuously sample the gut luminal environment through active endocytosis. Like professional phagocytes, M cells take up antigens into endosomes, which then fuse with lysosomes to digest the ingested material and present short peptide sequences (epitopes) bound to major histocompatibility complex (MHC) class 2 molecules on their cell surface. Specific clones of T lymphocytes expressing the requisite T cell receptor recognize and

TABLE 82.1

Common PAMPs, DAMPs, and Their Human Innate Immune Receptors

PAMPs	PAMP RECEPTORS	DAMPs	DAMP RECEPTORS
Lipopolysaccharide (LPS) or endotoxin	CD14:MD2:TLR4	HMGB-1	TLR2, TLR4, RAGE
Triacyl lipopeptides	TLR1/TLR2	Heat shock proteins	TLR2, TLR4
Diacyl lipopeptides	TLR6/TLR1	S100 proteins	TLR4, RAGE
Muranyl dipeptide from bacteria	NOD2	Fibrinogen, fibronectin	TLR4
Diaminopimelic acid (gram-negative bacteria)	NOD1	Hyaluronan	TLR4
RNA viral genomes	RLH	Biglycans	TLR2, TLR4
Lipoteichoic acid (gram-positive bacteria)	TLR2	Modified LDL	MD2:TLR4
Bacterial flagellin	TLR5	Heme	TLR4
Single-stranded RNA viruses	TLR7/8	Histones	TLR4
DNA viruses or bacterial DNA	TLR9	Mitochondrial DNA	TLR9
Double-stranded RNA viruses	TLR3	Nucleosomes	TLR4
Fungal mannans	TLR4, CLR	Neutrophil extracellular traps	TLR4
Malarial hemozoin pigment	TLR9, NLRs	Apoptotic cells	C-reactive protein, pentraxins

CLR, C-type lectin receptor; *DAMPs,* damage-associated molecular patterns; *HMGB-1,* high-mobility group box 1; *LDL,* low-density lipoprotein; *MD2,* myeloid derived 2; *NOD2,* nucleotide binding oligomerization domain; *PAMPs,* pathogen-associated molecular patterns; *RAGE,* receptor for advanced glycated end products; *RLH,* retinoic acid inducible gene 1–like helicase; *TLR,* Toll-like receptor.
(Modified from Opal SM. Immunologic alterations and the pathogenesis of organ failure in the ICU. *Seminars Respir Crit Care.* 2011;32[5]:569–580; Beutler B. TLRs and innate immunity. *Blood.* 2009;113[7]:1399–1407; Hotchkiss RS, Moldawer LL, Opal SM, Reinhart K, Turnbull, Vincent JL. Sepsis and septic shock. *Nat Rev Disease Primers.* 2016; 2:1–21.)

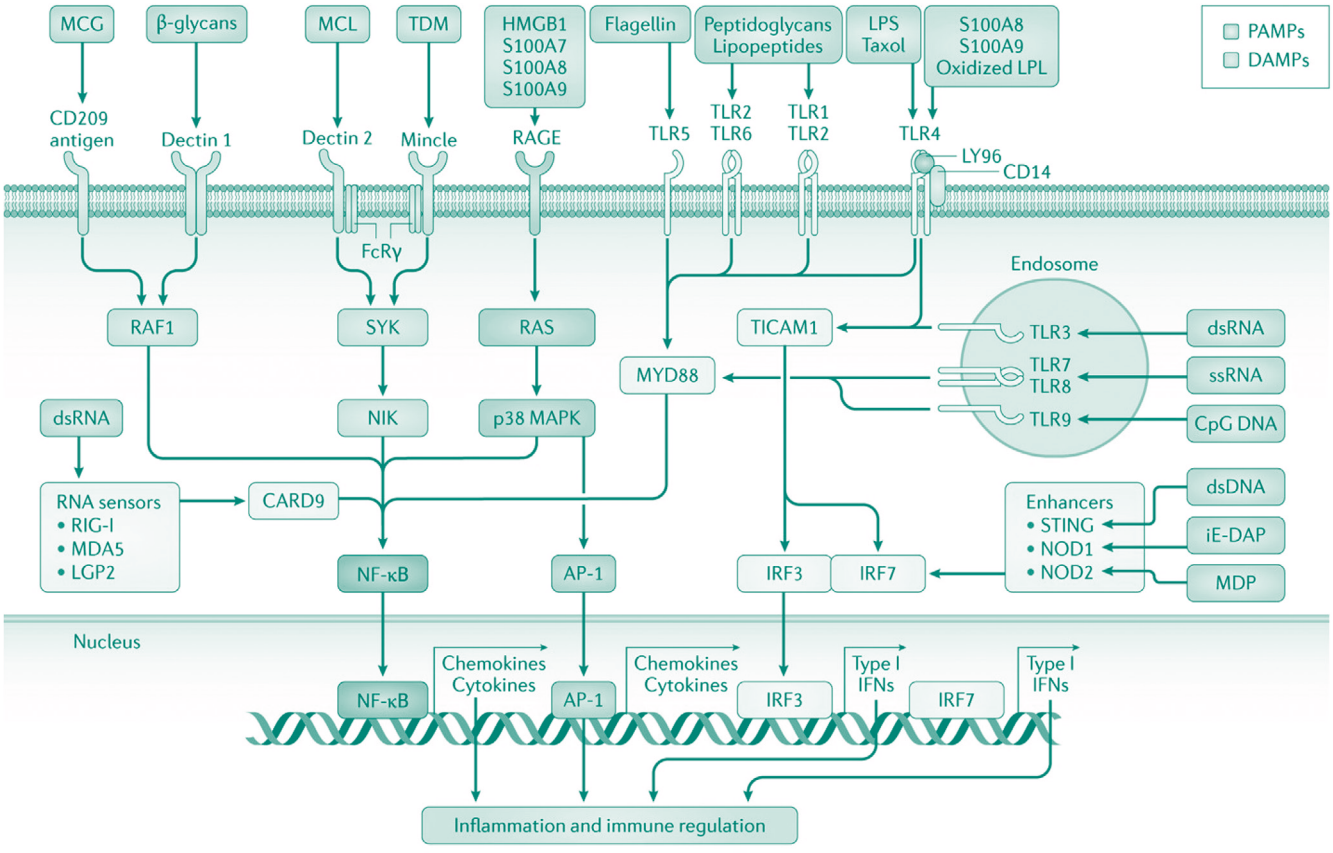

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FIGURE 82.1 Pattern recognition receptors of the human innate immune system and their common pathogen-associated and damage associated ligands. *AP-1,* Activator protein 1; *CARD9,* caspase-associated receptor domain containing protein 9;*CD,* cluster determinant; *CpG,* cytosine phosphate quanidine motif in DNA; *DAMPs,* damage-associated molecular patterns; *ds,* double-stranded; *FcγR,* crystallizable component of immunoglobulin gamma receptor; *HMGB1,* high-mobility group box 1; *iE-DAP,* glutamyl-meso-diaminopimelic acid; *IFN,* interferon; *IRF,* interferon response factor; *LGP2,* laboratory of genetics and physiology 2; *LPL,* lipoprotein lipase; *LPS,* lipopolysaccharide; *Ly96,* lymphocyte antigen 96; *MAPK,* mitogen-activated protein kinase; *MCG,* mannose-containing glycoprotein; *MCL,* mannose-capped lipoarabinomannan; *MDA5,* melanoma differentiation-associated protein 5; *MDP,* muramyl dipeptide; *Mincle,* macrophage inducible C-type lectin; *myD88,* myeloid differentiation primary response protein 88; *NF-κB,* nuclear factor in kappa B cells; *NIK,* NF-κB-induced kinase; *NOD,* nucleotide binding oligomerization domain; *PAMPs,* pathogen-associated molecular patterns; *RAF1,* proto-oncogene serine/threonine protein kinase; *RAGE,* receptor for advanced glycosylation end products; *RIG,* retinoic acid–inducible gene 1protein; *STING,* stimulator of interferon genes; *ss,* single-stranded; *SYK,* spleen tyrosine kinase; *TDM,* trehalose 6,6'dimycolate; *TICAM,* Toll interleukin-1 receptor domain containing adaptor molecule 1; *TLR,* Toll-like receptor. (From Hotchkiss RS, Moldawer LL, Opal SM, Reinhart K, Turnbull, Vincent JL. Sepsis and septic shock. *Nat Rev Disease Primers.* 2016; 2:1–21, with permission.)

respond to the presented epitopes as part of the tightly orchestrated acquired (adaptive) cellular immune response to that antigen. Even classic humoral effector cells of adaptive immunity such as B lymphocytes can phagocytize antibody or complement opsonized microbes, and they possess mechanisms that make them efficient antigen-processing and antigen-presenting cells. They also express the correct antigen-presenting motifs and costimulatory molecules essential to clonally select, activate, and cause clonal proliferation of specific T cells. Activated T lymphocytes also present antigen on class I or II MHC molecules and release proinflammatory cytokines in the process. Thus most cells in the human body possess the capacity to recognize microbes and their toxins at the innate level and initiate and modulate various aspects of the adaptive immune response.

A number of accessory, nonclonal T cell lines, natural killer cells, and even a B cell line are now classified as forming part of the innate immune response found in human immunology (Table 82.2).[3,9–12] Coagulation factors are highly coregulated with innate immune cells and form the essential elements to the initial host response to limit injury, preventing blood loss and blocking invasive pathogens after tissue injury.

Major Distinguishing Features Between Innate Immunity and Adaptive Immunity

Innate immune cells are distinguished from cells of the adaptive (or acquired) immune system for their lack of specificity, in contrast to the exquisite epitope specificity of adaptive immunity. Innate immune cells are nonclonal as opposed to adaptive immune cells, which respond and clonally expand only when the correct T cell receptor or B cell receptor is presented with its specific antigen along with appropriate costimulatory signals. Innate immune cells can attack and kill damaged or virus-infected or tumor cells directly, whereas the adaptive immune system usually requires specific antibodies or activated T cells to kill host cells. Typically innate immune cells are thought to be the fast response system (minutes to hours), whereas adaptive immune responses take days or weeks to respond to a new antigen. Finally, immunologic memory, the faster response to the second exposure to a specific antigen (recall antigens) than the first exposure, is generally a characteristic of the adaptive immunity.[4] Such an anamnestic response to recall antigens is not supposed to occur with the innate immune response. However, recent evidence indicates that some

TABLE 82.2

Cellular Components That Contribute to the Innate Immune Response in Humans

CELL TYPE	LIFESPAN IN TISSUES/MAJOR FUNCTIONS	ROLE IN INNATE IMMUNITY
Neutrophils	$T_{1/2}$ 12–48 hr; professional phagocyte, express CD14, CD11/CD18, CD64, TLRs, L-selectin, PSGL-1, form NETs	Mobile phagocytes ingest and kill pathogens, generate ROI and cytokines, naturally apoptotic
Monocytes/ macrophages	$T_{1/2}$: days to weeks; monocytes mature to macrophages, professional phagocytes, APC, express CD14, $MHC_{II,}$ CD80/86	Tissue phagocytes sense and phagocytize DAMPs and PAMPs Produce cytokines and are APCs
Dendritic cells	$T_{1/2}$: months to years/Sense pathogens, APCs Express TLRs, $MHC_{II,}$ CD80/86	Present antigens, express cytokine interferons, mucosal immunity
NK cells	$T_{1/2}$: months to years/no α/β TCR, MHC_{I} restricted, remove virus-infected cells and damaged cells, express CD16, TLRs	Cytotoxic to infected cells, innate viral immunity, produce Th1/Th2 cytokines, immune memory
γδ T cells	$T_{1/2}$: months to years/ no α/β TCR, recognize phosphoantigens, regulate cell-mediated immunity	Recognizes phosphoantigens from bacterial pathogens, regulatory functions in mucosa, granulomas
NKT cells	$T_{1/2}$: years/invariant α/β TCR, CD-1d restricted, recognizes glycolipid antigens, express interferons, IL-4	Contributes to host immune response to viral, mycobacterial, and fungal pathogens
B1 cells and ira B cells	$T_{1/2}$: years/B cells can function in pathogen detection, express TLRs, cytokines, MHC_{II}, CD80/86, ira B cells secrete GM-CSF	Innate response activator (ira) B cells are specialized for antigen detection, GM-CSF production
Treg cells	$T_{1/2}$: years/adaptive immune cells that control and inhibit innate immunity; express CD25 and FoxP3, produce TGFβ and IL-10	Major role in tissue repair and in limiting innate immune response
Innate lymphoid cells (ILCs)	$T_{1/2}$: months to years/ rapidly activated, proinflammatory cells that express IL-17, IL-5, and interferon γ when exposed to microbes	Derived from lymphoid progenitor cells, no TCR or BCR, produce proinflammatory cytokines
Platelets	$T_{1/2}$: 7 to 14 days/express P selectin, CD40L, GP1 to bind VWF, GPIIb/IIIa to bind fibrin	Activate endothelial cells to promote neutrophil binding
Endothelium	$T_{1/2}$: months to years/express P-selectin and E-selectin, CD40, TM, coated with GAGs	Maintains vascular integrity, regulates clotting, directs traffic of myeloid cells to infection sites
Epithelium	$T_{1/2}$: days/mucosal barrier to pathogens, express TLRs, secrete AMPs, APCs and macrophages sense and clear PAMPs	Physical barrier, mucus secretion, AMP secretion, and motility clear potential pathogens

AMP, Antimicrobial proteins; *APC,* antigen-presenting cell; *B cell,* bursa-derived lymphocyte; *BCR,* B cell receptor; *CD,* cluster determinant; *DAMPs,* damage-associated molecular patterns; *E,* endothelial; *FoxP3,* Forkhead box protein 3; *GAGs,* glycosaminoglycans; *GM-CSF,* granulocyte macrophage colony-stimulating factor; *GP,* glycoprotein; *IL,* interleukin; *L,* ligand; *MHC,* major histocompatibility type 2 antigen; *NET,* neutrophil extracellular trap; *P,* platelet; *PAMPs,* pathogen-associated molecular patterns; *PSGL-1,* P selectin glycolipid ligand-1; *ROI,* reactive oxygen intermediates; *T cell,* thymic-derived lymphocyte; *TCR,* T cell receptor; *TGF,* transforming growth factor; *Th1,* T helper cell type 1; *TLR,* Toll-like receptor; *TM,* thrombomodulin; *Treg cells,* T regulatory cells; *VWF,* von Willebrand factor.[3–12]

form of entrained responses can occur in some innate immune responses to repeated antigen responses.[4,13]

NK cells and γδ T cells are typically thought of as innate immune cells that lack specific, clonal, rearranged antigen-generated (RAG) T or B cell receptors of adaptive lymphoid cell immune responses and lack memory to recall antigens. However, these cells appear to a have a form of immunologic memory.[13] Human NK cells can recognize specific antigens on cytomegalovirus (CMV) and respond by expansion and more rapid response to reactivation or reinfection to subsequent exposure to CMV. Similar findings are now found in the NK response to a number of common viruses. These recall features are indicative of immunologic memory. Moreover, a form of recall response that is specifically directed toward *Listeria monocytogenes* has been observed in γδ T cells in mice, conferring greater protection against subsequent challenge to this bacterial pathogen.[13] These findings, in addition to the longstanding evidence that innate immune cells process and present specific antigens for recognition by T and B cells, clearly demonstrate that innate and adaptive immune effector cells collaborate to the mutual benefit and survival of the human host.

Soluble Pattern Recognition Receptors of the Innate Immune Response

Pattern recognition receptors (PRRs) are found not only on the cell surface on the endosomes of innate immune cells. Circulating PRRs and soluble PRRs in the extravascular space are abundant and play an essential role in defending a host from infection.[14] The most well-recognized soluble PRRs make up the second and third arms of the complement (C′) system. The first arm of the C′ system is the classical pathway, which is primarily antibody dependent after recognition and opsonization by specific B cell responses. The alternative C' system is not antibody dependent and allows C3 to directly bind to conserved microbial structures, if accompanied with necessary cofactors that stabilize C3bi molecules on microbial surface structures. Microbial pathogens can be opsonized, followed by assembly of the terminal membrane attack complex (C5-C9), leading to microbial lysis, entirely without the need for antibody. A similar system is found in the third arm of the C' system, consisting of collectin molecules of the mannose-binding lectin pathway. Highly conserved mannosides (such as bacterial cell walls consisting of peptidoglycan) are recognized and bound to the mannose-binding lectin, which activates specific mannose-binding, lectin-associated serine kinases to initiate the complement cascade.[14] This topic is discussed further in the following paragraphs.

Other soluble PRRs include the ficolins, which are collagen-like proteins that bind conserved carbohydrate molecules in a manner structurally and functionally similar to the mannose-binding lectins. Another related class of soluble PRRs are found in the lungs and are known as surfactant proteins SP-A and SP-D. These proteins bind to an array of oligosaccharides produced in the lung by a large number of gram-positive and gram-negative bacteria and promote clearance of potential pathogens.[14]

The galectin family of lectin proteins are components of the innate immune defense to bacterial and viral pathogens. Some galectins can bind directly to bacterial surfaces, permeabilize cell membranes, and kill bacteria in the absence of antibody or complement.[14,15] In addition, a series of proteins in the pentraxin family are also critical in recognizing PAMPs and assist in pathogen clearance. Best-known members of this family include C-reactive protein (CRP), serum amyloid P, and pentraxin 3. These are all acute phase proteins from the liver that are expressed in acute inflammatory states. CRP is known for its ability to recognize and bind to the C-polysaccharide of *Streptococcus pneumoniae*. CRP then can activate, directly complement, and promote the clearance of this human pathogen. Pentraxin 3 has the capacity to limit ongoing inflammation, because it can block neutrophil attachment to endothelial cells and promote resolution of inflammation.[16] Importantly, the innate immune system also is accompanied by an impressive array of antimicrobial peptides that constantly defend the host from microbial invasion. These peptides generally bind to bacteria and fungi, disrupting their cell wall and inducing cell death. Such peptides are prevalent along mucosal sites and along the lining of the integument, where potential pathogens attempt to gain access to the host. These include defensins, cationic permeabilizing peptides, and the human cathelicidin LL-37. Some of the antimicrobial peptides are expressed in high concentrations in the urinary tract to bolster defense against potential pathogens.[14]

Molecules of the Innate Immune Arsenal That Are Expressed in the Urinary Tract

Defensins

Unlike the gastrointestinal tract and pulmonary airways, the urinary tract lacks a thick unstirred mucous layer and microvillus-rich epithelium. Alternative strategies are used to maintain a sterile environment in the upper urinary tract. The secretions of the renal system are rich with antibacterial substances, including a group of cationic antimicrobial peptides called α- or β-defensins.[17–19] The α-defensins in humans consist of six gene products, including four proteins stored in neutrophil granules and two proteins stored in Paneth cell granules.[19] At least five β-defensins are produced by activated epithelial cells that line the pulmonary, gastrointestinal, and genitourinary tracts. Numerous studies have documented a role for defensins in innate immune defense. α- and β-defensins have been shown to induce degranulation of mast cells.[20,21] Mice lacking α-defensins have decreased resistance to orally administered bacteria compared with wild-type littermates.[22] α-Defensin secretion by Paneth cells is thought to maintain a sterile environment in the intestinal crypt.[23] Mice lacking the *Defb1* β-defensin 1 gene show delayed clearance of *Haemophilus influenzae* from the lung[24] and increased incidence of *Staphylococcus* spp. in the bladder[25] compared with normal littermates. Mice lacking matrilysin, a metalloproteinase that is required for the processing of defensin precursors, are susceptible to microbial invasion of the intestinal mucosa.[22]

Defensins act by binding strongly to the surface of microbes, where they are proposed to perturb lipid ordering in the outer bacterial membrane. This interferes with membrane barrier properties and enzymatic activities of various transport proteins, with subsequent loss of transmembrane potential and eventual death of the cell. Defensins also act as opsonins through interactions with chemokine receptors.[26] β-defensins are produced by epithelial cells in the loop of Henle, distal convoluted tubule, and the collecting duct.[27] β-defensin 1 is produced constitutively at very high levels, and β-defensins 2 and 3 are induced in renal epithelia in response to infection or proinflammatory cytokines.[28]

Tamm-Horsfall Protein

Tamm-Horsfall protein (THP) is a glycoprotein produced exclusively by renal tubular epithelial cells within the distal loop of Henle, and it is one of the most abundant urine proteins in mammals.[29,30] THP function remained unclear for many years until recent reports that THP binds to type 1[31] and type S[32] fimbriated *Escherichia coli* and impedes microbe interaction with the uroepithelium. THP also appears to activate cells via direct signaling through TLR4.[29,33] THP was shown to induce tumor necrosis factor-α (TNF-α) and tissue factor production via TLR4 in monocytes.[34,35] These findings were supported by reports that intravenous injection with THP rapidly induces systemic TNF-α production in control TLR2$^{-/-}$ mice and TLR9$^{-/-}$ mice but not in TLR4$^{-/-}$ or MyD88$^{-/-}$ mice (MyD88 protein is required for TLR4-dependent signaling).[34] C3H/HeJ mice are homozygous for a nonconservative mutation in the gene encoding TLR4.[36] These mice show decreased responsiveness to gram-negative bacterial endotoxin and decreased recruitment of neutrophils and monocytes in response to gram-negative bacterial infection, two defects that help explain the increased incidence and severity of urinary tract infection in these mice.[33] The relative contribution of lost THP signaling through TLR4 to the development of the urinary tract infection in these mice must be investigated further.

Other Proteins

Bactericidal/permeability-increasing (BPI) protein, lipopolysaccharide (LPS)-binding protein, and a growing number of homologous mammalian proteins[37] play an important role in controlling systemic dissemination with blood-borne bacteria and colonization of epithelial surfaces with microbes.[38–40] Urine from normal volunteers contains BPI at 0.2 to 1.2 ng/mL, in the absence of apparent white blood cells monitored using microscopy.[41] These levels are increased greatly in patients with active infection and renal disease. The majority of urinary BPI is believed to be made locally, and this is most likely the result of production of the bactericidal protein in the uroepithelium. A comparative microarray analysis of mRNA expressed in various organs has detected levels approaching that of myeloid cells in total RNA preparations from kidney, lung, and liver.[42] Thus further studies of the role of BPI in the innate immune defense of the urinary tract seem warranted.

Like the cationic defensins, BPI is directly bactericidal and possesses opsonic properties. Cytotoxicity is due to the high affinity of BPI for the lipid core of endotoxin in the outer leaflet of the gram-negative bacterial outer membrane.[43] The binding receptor for microbe-bound BPI has not been described. Binding of BPI to endotoxins disrupts the membrane leading to bacterial death. The actions of BPI and the defensins are amplified by other factors that are secreted actively from immunostimulated epithelial cells, including phospholipase A2 and complement proteins.[44,45] Although BPI activity usually is associated with its effects on gram-negative bacteria, BPI also has activity against gram-positive bacteria with a compromised cell wall and some fungi.[46]

Even alpha-intercalated cells, known to regulate acid-base balance by the kidney, participate in the innate immune defense against bacteria. These cells, found in the urinary collecting duct, prevent binding of uropathogenic *E. coli* to the epithelial surface by acidification of the urine and secreting the antimicrobial peptide lipocalin 2.[47]

Complement Activation

Complement activation represents another powerful mechanism for controlling microorganisms and their toxins. In humans, there are currently three distinct mechanisms that activate the complement cascade. The classic pathway involves binding of at least one immunoglobulin M or two immunoglobulin G antibodies to a surface-associated antigen. This complex recruits complement protein C1, which cleaves C2 and C4 to C2a and C4b, which assemble to form an active C3 convertase on the surface of the microbe. C3b fragments generated can bind with C2aC4b complexes to form the C5 convertase. The alternative activation pathway is initiated as follows. Serum C3 is activated continuously by reaction with water to form what is referred to as C3 (H_2O). Some microbial carbohydrate structures can activate C3 directly. Once activated, C3 binds factor B, triggering its cleavage by factor D. Factor Bb remains bound to C3 (H_2O) and functions as a soluble C3 convertase. This protein complex can react covalently with any surface in blood or body fluids including urine, where it generates C3b at the surface of target cells and additional convertases are assembled. Binding of an additional C3b fragment to the C3bBb complex forms a functional C5 convertase. The lectin pathway of complement activation is initiated when lectin-like proteins, including mannose-binding protein and ficolins, bind to saccharide units found specifically on the surface of microbes and recruit mannose-binding protein-associated serine proteases (MASPs) to the cell surface. These MASPs cleave C4 to C4b, which cleaves C2 to form C2a, which forms the C3 convertase that culminates in formation of the C5 convertase. Assembly of the C5 convertase initiates the first common step of the complement pathway. At this point, the cells are opsonized for complement receptor 2 on any white blood cells that are nearby. Alternatively, recruitment of complement proteins 5 thru 9 leads to the formation of mature membrane attack complexes that form large pores in the microbial membrane leading to osmotic rupture of the cell. The smaller fragments generated during complement action, particularly the C3a, C4b, and C5a fragments, function as anaphylatoxins, recruiting lymphocytes and leukocytes to the site of infection. Signaling through specific receptors also modulates the immune activity of recruited and resident leukocytes.

The majority of complement protein production occurs in the liver, but surface epithelia and endothelial cells also have been shown to produce complement proteins.[48] Glomerular epithelia[49] and mesangial cells[50,51] release complement proteins locally. Tubular epithelial cells also synthesize complement proteins in normal and diseased states.[52,53]

Genes encoding complement proteins have been identified in organisms as evolutionarily distant as horseshoe crabs, in which a C3 convertase activity and lectin-like nucleating activity have been identified.[54] Given that antigen-specific antibodies that are generated through somatic recombination of highly polymorphic alleles are not present in the vertebrate lineage until the appearance of the bony jaw fish, the absence of the classic complement activation system in this animal was not surprising. Thus the initial selective advantage of the complement system was to opsonize invading organisms for destruction inside specialized phagocytes. Receptors on dedicated arthropod phagocytes called *hemocytes* bind complement-coated microbes and rapidly trigger their phagocytosis. This is critical because, unlike higher vertebrates, most antimicrobial peptides and enzymes are located within these specialized immune effector cells.

RESOLUTION OF INFLAMMATION AND DEACTIVATION OF INNATE IMMUNE RESPONSES

Once the potential pathogen has been recognized and cleared by innate and/or adaptive immune responses, resolution of the inflammatory process must follow or risk ongoing collateral damage to normal tissues. Repair and remodeling of tissues then occur with a return to normal homeostasis. Recent evidence indicates that resolution of inflammation is not a passive process of absence of continued need, but an active proresolution process directed by specific sets of biochemical signals and sets of cellular responses.[54] Active restraint of the inflammatory response by antiinflammatory signals and cytokines (e.g., IL-10, IL-4, IL-13, TGF-β) is an essential first step to restoring order after infection. However, the expression of antiinflammatory cytokines alone will not return the tissues to their original state without activation of specific proresolving mechanisms. Much of the proresolving process is driven by a set of oxidized lipid mediators derived from prostaglandins and leukotrienes via the lipoxygenase pathway.[55] These lipid mediators generate compounds known as lipoxins, resolvins, protectins, and maresins, which promote clearance of damaged and apoptotic cells. This is accomplished by a process called efferocytosis. Efferocytosis is the phagocytosis of apoptotic neutrophils and cellular debris primarily by macrophages, whereupon they are clear the tissues and migrate to regional lymphatics for disposal.[56] Recently, evidence has emerged that the proinflammatory Th17 cell line responsible for local and mucosal proinflammatory responses has a rather unique control mechanism to limit excessive tissue damage. These activated cells over time within tissue will transdifferentiate into regulatory T cells (Treg).[57] Treg cells have the polar opposite immune effects in tissues compared with Th17 cells. Treg cells are almost exclusively antiinflammatory in their functions and express large amounts of the inhibitory cytokines TGF-β and IL-10. The discovery of the proresolving pathways opens up an opportunity to target these pathways for therapies that could curb inflammation, promote healing, and begin tissue repair.

CONCLUSION

At every stage of the host–pathogen interaction, all the cells in the vicinity of the infection, including T cells and B cells and endothelial cells that are not immediately thought of as *innate* immune cells, can release cytokines, induce production of antimicrobial proteins, and present antigenic substances to other cells. These defensive processes usually are thought to be the purview of myeloid cells and other "dedicated" antigen-presenting cells. Therefore it may be simpler to consider T and B cells as members of the acquired immune cell lineages and assume that all cell types, including uroepithelial cells, are capable of functioning in an innate immune capacity to some extent. The relatively recent advances in the TLR field only recently have been directed at studying the uroepithelium. Evidence now exists for an important role for epithelial TLR-dependent recognition of bacteria within the urinary tract and against uropathogenic bacterial infection.[58] These findings represent some of the earliest insights into the potential importance of the innate functions of the urinary epithelium itself in maintaining immune homeostasis in the urinary tract. These findings provide novel potential options to bolster host defenses and promote resolution of tissue injury. Whether this knowledge will lead to new preventive and therapeutic strategies against urinary tract infection remains to be demonstrated.

Key Points

1. A variety of general and kidney-specific innate immune mechanisms are operating continuously in normal health and disease to control microbial growth in the urinary tract.
2. Early advances in the field of immunology favored research into antibody-dependent responses at the cost of research advances in the field of innate immunity.
3. The identification of pathogen-sensing systems that are present on most mammalian cells has greatly increased the number of cells that can be considered innate immune effector cells.
4. The uroepithelium is well equipped to function in the sensing and elimination of potentially pathogenic microbes.

Key References

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