The Immunopathology of Sepsis: Pathogen Recognition, Systemic Inflammation, the Compensatory Anti-Inflammatory Response, and Regulatory T Cells

D.H. Lewis, D.L. Chan, D. Pinheiro, E. Armitage-Chan, and O.A. Garden

Sepsis, defined as the systemic inflammatory response to infection (Box 1), remains the major cause of death in critically ill human patients.1–6 Recent human studies estimate the annual incidence of sepsis to be 240–300 cases per 100,000 population, with associated costs of nearly $17 billion in the United States7,8; the rate of occurrence is also increasing at around 9% each year.8 Although large-scale veterinary epidemiological studies are uncommon, a substantial proportion of the critically ill veterinary population is estimated to be septic.9,10 The case fatality rate associated with sepsis in a variety of veterinary species is reported to approach 50%, emphasizing the need for a greater understanding of the pathophysiology of sepsis to improve therapeutic practices.11–13

The pathophysiology of sepsis remains incompletely understood. A multitude of cell types, inflammatory mediators, and coagulation factors are involved and recent research has focused on the contributions of the innate immune system and T cells in this complex syndrome.14–20 This review will present an account of the current understanding of the functioning of the immune system in sepsis, with emphasis on the interaction of pathogens with innate components of the immune system and the key role of the endothelium in triggering and propagating a pro-inflammatory state.

Abbreviations:

| Abbreviation | Description |
|--------------|-------------|
| AGE          | advanced glycation end product |
| AP-1         | activator protein 1 |
| aPC          | activated protein C |
| BK           | bradykinin |
| CARD         | common caspase activation and recruitment domain |
| CLARP        | caspase-like apoptosis regulatory protein |
| DAMP         | danger-associated molecular pattern |
| gp           | glycoprotein |
| HMBG         | high mobility group box protein |
| HSP          | heat shock protein |
| Ig           | immunoglobulin |
| IKK          | inhibitor of κB kinase |
| IPAF         | interleukin-1 converting enzyme (ICE) protease-activating factor |
| IPS-1        | interferon-β promoter stimulator-1 (also known as CARD adaptor inducing interferon-β [Cardif]) |
| IRAK         | IL-1R associated kinase |
| IRE          | interferon regulatory factor |
| KK           | kallikrein |
| LDL          | low density lipoprotein |
| LGP2         | laboratory of genetics and physiology 2 |
| MAPK         | mitogen-activated protein kinase |
| MDA5         | melanoma differentiation-associated gene 5 |
| MODS         | multiple organ dysfunction syndrome |
| MyD88        | myeloid differentiation primary response gene 88 |
| NAIP         | neuronal inhibitor of apoptosis |
| NEMO         | NF-κB essential modulator |
| NF-κB        | nuclear factor kappa B |
| NLR          | nucleotide-binding domain, leucine-rich repeat containing protein |
| NLRB         | NLR family, baculovirus inhibitor of apoptosis protein repeat domain containing |
| NLRC         | NLR family, CARD containing |
| NLRP         | NLR family, pyrin domain containing |
| NOD          | nucleotide-binding oligomerization domain |
| PAMP         | pathogen-associated molecular pattern |
| PAR          | protease-activated receptor |
| PMN          | polymorphonuclear cell |
| PRR          | pattern-recognition receptor |
| RAGE         | receptor for advanced glycation end products |
| RICK         | RIP-like interacting CLARP kinase |
| RIG-1        | retinoic acid-inducible gene-1 |
In addition, the complex interplay between pro- and anti-inflammatory cytokines and the spectrum of the host defense response will be discussed. Finally, the importance of regulatory T cells (Tregs) in maintaining the balance of host inflammatory mechanisms will be described.

**Immunopathology of Sepsis**

Living organisms face a constant barrage of potentially pathogenic microorganisms. Survival depends upon physical barriers to resist entry of pathogens, as well as the presence of a constitutive, or innate, immune system that can rapidly induce a defensive inflammatory response. Such a system can be found in virtually all species, suggesting that it is evolutionarily ancient and highly successful.²¹,²² The innate immune system further interacts with the adaptive immune system, based on a system of T and B lymphocytes that respond to specific epitopes.

**Surface Barrier Mechanisms and Antimicrobial Peptides**

Whereas the keratinized epithelium of the dermis and the mucous lining of the body cavities discourage pathogenic colonization, various other innate defenses are employed to minimize penetration of the body wall by microorganisms, including antimicrobial peptides (AMPs; otherwise known as host-defense peptides) on mucosal surfaces.²³,²⁴ A comprehensive review of AMPs in veterinary species has recently been published.²⁵ In summary, AMPs comprise 3 main groups; digestive enzymes and peptides that disrupt the microbial cell membrane, peptides that bind essential elements, and peptides that act as decoys for microbial attachment. The 2 major classes of bacterial AMPs in the mammalian immune system are the defensins and cathelicidins. Compared to eukaryotic cells, bacterial cell walls lack cholesterol—which acts to stabilize cell membranes—and negatively charged phospholipids. Instead, bacterial cell walls are rich in anions (basic amino acids) and thus attract the cationic defensins, which carpet the microbial membrane and institute channel formation.²⁶ These channels then lead to transmembrane pore formation, membrane destabilization, and microbial cell death, although recent studies suggest that their function could go beyond that of lipid bilayer perturbation.²⁷ Whereas a large number of AMPs form part of the non-oxidative killing mechanism within phagolysosomes in cells such as neutrophils and macrophages, a growing number are thought to be actively secreted onto epithelial surfaces of the gastrointestinal, respiratory and urinary tracts²⁸–³¹—including the ovine gastrointestinal tract³² and the canine testis.³³ Recent work has revealed that important amounts of these AMPs are secreted not only by immune cells such as neutrophils and alveolar macrophages but also by atypical defense cells such as type II pneumocytes.³⁴ This theme is universal throughout the processes of the innate immune system and challenges the traditional view of a “standing army” of immune defense cells, replacing it with the concept of a body-wide, integrated community of cells contributing to pathogen vigilance.³⁵

**Recognition of Pathogens: PAMPs, MAMPs and DAMPs**

Two factors are vital to the rapid ability of the innate immune system to respond to pathogen incursion: the presence of receptors against pathogen markers and the ubiquitous nature of these receptors in the body.³⁵ Individual receptors are genetically encoded and display strong homology within and between species.²² These receptors are not only expressed on many effector cells of the immune system—including macrophages, neutrophils, dendritic cells, and lymphocytes—but are also found on epithelial cells, endothelial cells, and myocytes³⁶,³⁷; expression has also been detected in the bovine endometrium.³⁸ The major targets of these pattern recognition receptors (PRRs) are known as pathogen-associated molecular patterns (PAMPs), although the presence of these molecules in nonpathogenic and commensal bacteria has led to the suggestion that the term “microbial-associated molecular
patterns” (MAMPs) is more accurate. These molecules share certain core characteristics:

- **PAMPs** are produced only by microbial pathogens, not by the host (eg, peptidoglycans are produced by bacteria but not by eukaryotic cells); this confers automatic self/nonself discriminatory ability.

- **PAMPs** are generally invariant molecules shared by entire classes of microorganisms (eg, lipoparabinomannan is found on the cell wall of all Mycobacteria); this allows the evolutionary retention of a relatively small number of PRRs recognizing vast numbers of potential pathogens.

- **PAMPs** are usually structures vital to the survival or pathogenicity of the microorganism (eg, lipopolysaccharide [LPS] in the outer membrane of Gram-negative bacteria): this allows targeting of highly conserved molecules and obviates the need for variability in host PRRs.

Although bacterial cell surface components such as LPS in the outer cell membrane of Gram-negative bacteria and lipotechoic acid (LTA) in the cell membrane of Gram-positive bacteria represent classic examples of PAMPs, recognition of “altered self” secondary to host cell colonization by viral pathogens is also likely. The innate immune system has thus developed the ability to detect markers of endogenous cell damage called “alarmins” or “danger-associated molecular patterns” (DAMPs)

Table 1 shows key PAMPs and their corresponding receptors, whereas Table 2 shows some of the confirmed interactions of DAMPs. Nonrodent sepsis models, genetic approaches, and immunological studies have demonstrated the presence of a large number of PRRs in clinical veterinary species (Table 3).

Initiation of an inflammatory reaction to necrotic, rather than apoptotic, cell death would appear to be useful in host defense; however, the interaction of DAMPs and PAMPs with their receptors leads to increased case fatality rates in sepsis. Owing to PRR cross-reactivity for both PAMPs and DAMPs, multiple positive feedback systems become established, leading to rapid progression of a global inflammatory response with consequent clinical signs (Fig 1). Functional interactions between PRRs, including synergy and cross-tolerance, also occur.

If an inflammatory state persists, the very defensive mechanisms of the innate immune system designed to protect the host can lead to further tissue damage, as well as diminished antimicrobial activity that allows opportunistic secondary infections. Whereas the existence of such feedback systems in veterinary species can at present only be inferred, experimental data suggest PRR reactivity to both pathogen- and host-derived ligands in cattle, pigs, horses, and dogs. Furthermore, the blunted PAMP-induced TNF, IL-6 and IL-10 response of whole blood in dogs with lymphoma is thought to underlie their higher risk of sepsis.

### Table 1. Key pathogen-associated molecular pattern ligands of the pattern recognition receptors implicated in sepsis.

| PRR         | Location | Cell Type | PAMP Recognized                                      |
|-------------|----------|-----------|-------------------------------------------------------|
| TLR 1/CD281 | pm       | pbmc, np, u | triacyl lipopeptide                                    |
| TLR 2/CD282 | pm, el   | pbmc, dc, mc, nkc | peptidoglycan, lipoprotein, lipopolysaccharide, glycosylphosphatidylinositol, mannan |
| TLR 3/CD283 | pm, el   | dc, epi, fb, blc, nkc | ssRNA, dsRNA, dsDNA                                    |
| TLR 4/CD284 | pm       | pbmc, mc, np, epi | lipopolysaccharide, glycosylphosphatidylinositol, viral envelope proteins, mannan |
| TLR 5       | pm       | pbmc, dc, epi, nkc | flagellin                                             |
| TLR 6/CD286 | pm       | mc, ble    | diacyl lipopeptide                                    |
| TLR 7       | el       | pbmc, dc, blc | ssRNA                                                 |
| TLR 8/CD288 | el       | nkc        | ssRNA                                                 |
| TLR 9/CD289 | el       | dc, blc, nkc, epi | ssDNA, dsDNA                                         |
| TLR 10/CD290| pm       | pbmc, dc, blc | Unknown                                               |
| TLR 11      | pm       | epi, dc, pbmc | profilin                                              |
| NOD1/NLRC1  | cyt, pm  | epi, dc, pbmc | peptidoglycan                                        |
| NOD2/NLRC2/CARD15 | cyt | epi, dc, pbmc, Paneth cells | muramyl dipeptide                                    |
| NLR4/IPAF   | cyt      | unknown    | flagellin                                             |
| NLRP1       | cyt      | unknown    | muramyl dipeptide, Bacillus anthracis lethal toxin    |
| NLRP3       | cyt      | unknown    | bacterial & viral RNA, lipopolysaccharide, lipotechoic acid, muramyl dipeptide |
| NLRB1/NAIP5 | cyt      | unknown    | flagellin                                             |

PRR, pattern-recognition receptor; PAMP, pathogen-associated molecular pattern; TLR, Toll-like receptor; NOD, nucleotide-binding oligomerization domain; NLR, nucleotide-binding domain, leucine-rich repeat containing protein; NLRC, NLR family, CARD containing; IPAF, interleukin-1 converting enzyme (ICE) protease-activating factor; NLRP, NLR family, pyrin domain containing; NLRB, NLR family, baculovirus inhibitor of apoptosis protein repeat domain containing; NAIP, neuronal inhibitor of apoptosis; pm, plasma membrane; el, endolysosomes; cyt, cytoplasm; np, neutrophils; u, ubiquitous; dc, dendritic cells; mc, mast cells; nkc, natural killer cells; epi, epithelial cells; fb, fibroblasts; blc, B lymphocytes; ss, single-stranded; ds, double-stranded.
Evolutionary pressure has resulted in the encoding of a number of different host proteins within three distinct families—the Toll-like receptors (TLRs), the nucleotide-binding domain, leucine-rich repeat containing proteins (NLRs; previously designated as the nucleotide-binding oligomerization domain [NOD]-like receptors) and the retinoic acid-inducible gene-1 (RIG-1)-like receptors (RLRs). Cooperation between the RLRs—intracellular viral nucleic acid sensors—and the endosomal TLRs appears likely, although little is currently known about the involvement of RLRs in the pathogenesis of sepsis. This family of PRRs is therefore not considered further in this review.

Whereas PRRs form a heterogeneous group of proteins, certain characteristics—such as leucine-rich repeat domains, scavenger receptor cysteine-rich domains, and C-type lectin domains—can be commonly recognized.

The Toll-Like Receptors

The Toll gene was first identified in *Drosophila melanogaster* as encoding a transmembrane glycoprotein with a role in determination of dorsoventral polarity in the embryo. Additional investigation revealed a large extracellular component with leucine-rich repeats, with a cytoplasmic portion described as the TIR (Toll/interleukin-1 receptor [L-1R]) domain owing to its similarity to the intracellular region of the mammalian IL-1R. To date, 10 human and 12 murine functional TLRs have been identified by a combination of immunological techniques and examination of genetic databases.

The cytoplasmic TIR domain of the TLRs interacts with a variety of TIR-domain-containing adaptors with a role in determination of dorsoventral polarity in the embryo. PAMP/DAMP ligation of the respective PRR activates a chain of kinases in the IL-1R associated kinase (IRAK) family, ultimately resulting in activation of the inhibitor of κB kinase (IKK) enzyme complex and the mitogen-activated protein kinase (MAPK) pathway. Whereas the connection of TLR activation to the NF-κB and MAPK signaling pathways is well recognized, a number of other pathways are also triggered by TLR stimulation, including Protein Kinase R/eukaryotic translation initiation factor 2α kinase (eIF2α kinase) and the Janus-activated kinase (JAK) pathway.

**Table 2.** Key danger-associated molecular pattern ligands of the pattern recognition receptors.

| PRR     | Location | Cell Type | DAMP Recognized         |
|---------|----------|-----------|-------------------------|
| TLR 2/CD282 | pm, el   | pbmc, dc, mc, nkc | HMGB 1, necrotic cells, HSP-60, HSP-70, gp-96, biglycan, defensins endogenous mRNA |
| TLR 3/CD283 | pm, el   | dc, epi, fb, blc, nkc | HSP-22, HSP-70, HSP-90, fibronectin, fibrinogen, heparan fragments, hyaluronate fragments, β-defensin 2, oxidized LDL, surfactant protein A, neutrophil elastase, HMGB 1, biglycan |
| TLR 4/CD284 | pm       | pbmc, mc, np, epi | chromatin-IgG complex |
| TLR 9/CD289 | el          | dc, blc, nkc, epi | uric acid crystals |
| NLRP3   | cyt        | unknown               | AGEs, HMGB 1, amyloid peptide, S100s |
| RAGE    | pm       | u?                    |                         |

New abbreviations (for previous abbreviations see Table 1): DAMP, danger-associated molecular pattern; RAGE, receptor for advanced glycation end products; HMGB, high mobility group box protein; HSP, heat shock protein; gp, glycoprotein; LDL, low density lipoprotein; Ig, immunoglobulin; AGEs, advanced glycation end products; S100s, S100 proteins (calgranulins).

**Table 3.** Pattern-recognition receptors of those families implicated in sepsis identified in veterinary species to date.

| Species | TLRs | NLRs              |
|---------|------|-------------------|
| Dog     | 1–7, 9 | NOD1, NOD2, NLRC4 |
|         |       | NLRP 1–3, 5, 6, 8–10, 12–14 |
| Cat     | 2–5, 7–9 | NLRC4 |
| Horse   | 2–4, 6, 9 | NOD1, NOD2, NLRC4 |
| Cow     | 1–10 | NLRP 1, 3, 5, 6, 8–10, 12–14 |
| Sheep   | 1–10 | NOD2 |
| Goat    | 1–10 | NOD1, NOD2 |
| Pig     | 1–10 | NOD1, NOD2 |

For abbreviations, see footnote to Table 1.

**Table 4.** The spectrum of type I and type II acute phase proteins.

| Acute Phase Protein | Role                                                                 | Type                        |
|---------------------|----------------------------------------------------------------------|-----------------------------|
| Serum amyloid A     | Leukocyte recruitment and activation                                 | I – induced by IL-1 and TNF |
| C-reactive protein  | Enhance microbial phagocytosis and complement binding                |                             |
| C3, C4, C4BP, C1inh| Complement components                                                |                             |
| Haptoglobin (rat)   | Binds free hemoglobin                                                |                             |
| α1-acid glycoprotein| Transport functions                                                  |                             |
| Fibrinogen          | Hemostasis                                                           | II – induced by IL-6 and IL-6-like cytokines |
| Haptoglobin (man)   | Binds free hemoglobin                                                |                             |
| (Apo)Ferritin       | Binds free iron                                                      |                             |
| α1-antitrypsin      | Protease inhibitor                                                   |                             |
| α2-macroglobulin    | Protease inhibitor                                                   |                             |

**Pattern Recognition Receptors: an Overview**

Evolutionary pressure has resulted in the encoding of a number of different host proteins within three distinct families—the Toll-like receptors (TLRs), the nucleotide-binding domain, leucine-rich repeat containing proteins (NLRs; previously designated as the nucleotide-binding oligomerization domain [NOD]-like receptors) and the retinoic acid-inducible gene-1 (RIG-1)-like receptors (RLRs). Cooperation between the RLRs—intracellular viral nucleic acid sensors—and the endosomal TLRs appears likely, although little is currently known about the involvement of RLRs in the pathogenesis of sepsis. This family of PRRs is therefore not considered further in this review.

Whereas PRRs form a heterogenous group of proteins, certain characteristics—such as leucine-rich repeat domains, scavenger receptor cysteine-rich domains, and C-type lectin domains—can be commonly recognized.
factor 2-α kinase 2 (PKR/eIF2α), Notch, phosphoinositide 3-kinases (PI-3K), and small GTPases. 87–92

TLRs are expressed by a number of different cells, including dendritic cells, macrophages, B cells, natural killer (NK) cells, endothelial cells, epithelial cells, and fibroblasts (Tables 1 and 2); their site of expression varies, with TLRs 1, 2, 4, 5, 6, and 11 present on the external plasma membrane and TLRs 3, 7, 8, and 9 in endosomes. 71,93

TLR 2 appears to be of key importance, owing to its ability to recognize PAMPs as diverse as lipoteichoic acid (from Gram-positive bacteria), peptidoglycan (from Gram-positive and Gram-negative bacteria), hemagglutinin (from measles virus), polysaccharides (from yeasts), lipoproteins (from E. coli, Borrelia burgdorferi, Mycoplasma spp. and Mycobacterium tuberculosis), as well as complete pathogens such as Clostridium spp., Chlamydia spp., and herpes simplex virus, and a variety of endogenous ligands. 94–97

Much of its wide-ranging influence stems from the formation of heterodimers with TLRs 1 and 6. 98–100 TLR 4 also has a significant role in triggering the innate immune response as it recognizes molecules such as LPS (from Gram-negative bacteria), various viral protein envelopes, and a large number of endogenous molecules (Table 2). 57,101–103 The vital recognition of LPS by TLR

---

**Fig 1.** The pathophysiology of sepsis—an overview. A primary infectious insult—of bacterial, viral, protozoal or fungal origin—damages host tissues. The ligation of pattern-recognition receptors (TLRs and NLRs) by PAMPs and DAMPs promotes the release of both pro- and anti-inflammatory cytokines, as well as acute phase proteins, with a number of pathophysiological sequelae that ultimately lead to organ hypoperfusion, tissue hypoxia, the generation of ROS and RNS, mitochondrial dysfunction and cell death by necrosis, apoptosis, pyroptosis, and autophagy. Further DAMPs are generated, helping to perpetuate the inflammatory response, and the death of immune cells leaves the organism vulnerable to secondary, or opportunistic, infections. Abbreviations: PAMPs = pathogen-associated molecular patterns; DAMPs = danger-associated molecular patterns; TLRs = Toll-like receptors; NLRs = nucleotide-binding domain, leucine-rich repeat containing protein; RLR = retinoic acid-inducible gene-1 (RIG-1)-like receptor; NFκB = nuclear factor kappa B; PMN = polymorphonuclear cell; ROS = reactive oxygen species; RNS = reactive nitrogen species.
4 appears to be dependent on formation of a complex with other PRRs, myeloid differentiation protein-2 (MD2), membrane-bound CD14 (mCD14), and lipopolysaccharide-binding protein (LBP). Both TLR 2 and 4 have been extensively researched in human septic patients, in whom early upregulation of these TLRs can exacerbate the severity of illness and mortality.

TLR 5 is expressed by epithelial cells of the respiratory and intestinal tract and is a PRR for flagellin, an important component of motile bacteria such as Salmonella spp. Although exactly how the PAMP and PRR interact remains unclear, the intracellular TLRs 3, 7, 8, and 9 all share high sequence homology and recognize nucleotides. TLR 3 is the only TLR yet discovered that does not initiate the MyD88 signaling pathway, interacting solely with TLR-domain-containing adapter molecule 1 (TICAM1, also known as TRIF) (Fig 2). TLR 11, to date only identified in rats and mice, appears to recognize Toxoplasma gondii and certain pathogens of the urinary tract, although a role in sepsis has not so far been described.

The Nucleotide-Binding Domain, Leucine-Rich Repeat Containing Proteins, and Inflammasomes

The realization that TLRs could not account for the full range of PAMP recognition motivated the discovery of additional intracellular PRRs, including the NLRs. Of the NLRs, the 2 cytosolic receptors NOD1 and NOD2 were the first to be discovered; subsequent examination of genomic databases has suggested that there are at least 23 NLRs in humans and 34 in mice. Common to all NLRs is their structure, comprising a leucine-rich repeat domain (thought to be the PAMP receptor region), a central NOD domain, and an N-terminal effector domain responsible for downstream signaling.
IκB, mobilization of NF-κB, and activation of MAPK signaling (Fig 3). NOD1 (NLR family, caspase activation, and recruitment domain [CARD] containing 1; NLRC1) and NOD2 (NLRC2, also known as CARD15) are known to recognize muropeptides derived from peptidoglycan, a major structural component of both Gram-positive and Gram-negative bacterial cell walls (Table 1). Whereas the role of NOD2 is well established in the pathogenesis of Crohn’s disease, little is known about the role of the NOD proteins in sepsis. However, synthetic NOD1 agonists administered to mice stimulate chemokine production, neutrophil recruitment and—in one model—shock and multiple organ dysfunction, raising the possibility of NOD1 involvement in the pathogenesis of sepsis.

A number of studies have been performed on the role of inflammatory cysteinyl aspartate-specific proteinases, or caspases, in murine and human sepsis. Caspase-1 has been the focus of particular attention and is activated by a macromolecular complex of around 700 kDa—one of a number of inflammasomes—comprising pro-caspase-1 together with various adapter proteins. Caspase-1 is important in the synthesis of active IL-1β and IL-18, and induces a type of programmed cell death called pyroptosis.
Pattern Recognition Receptors: Veterinary Species

After mapping of the gene sequences for human and murine PRRs, exploration of the genomeic sequence of other species has allowed the identification of homologs of the majority of TLRs in dogs, 150–154 cats, 49,151,155,156 cattle, 38,50,51,62,157–168 sheep, 48,51,169–173 goats, 52,174,175 horses, 47,168,176–181 and pigs. 169,182–191 Other PRRs have also been identified in these species, although less is known about them than the TLRs (Table 3).

Toll-like receptors 1–10 have been identified in the bovine genome 50,161 with numerous studies documenting their expression in tissues as varied as the endometrium, 38,163 cornea, 158 mammary gland, 167,192 skin, 51 and lung. 168 Whereas most of these studies have utilized PCR techniques to identify TLR expression, 38,51,158,163,167 an increasing number are employing flow cytometry 192,193 or immunohistochemistry. 168 TLR signaling in cattle is similar to that described in mice and humans (Fig 2). 162,192,194 and the PKR/eIF2α pathway appears to be important in bovine viral diarrhea virus (BVDV) and rotavirus infections. 195,196 Comparatively little is known about the NLR family in cattle, although mRNA encoding NOD 1 and 2 has been identified in bovine mammary tissue. 197,198 Because of the financial implications of Johne’s disease and the currently unconfirmed link between Mycobacterium avium subspecies paratuberculosis and Crohn’s disease, a number of studies have examined the potential role of polymorphisms of candidate genes—including TLRs—in susceptibility to paratuberculosis. 199 Recently, TLRs 1–10 have also been cloned and sequenced in sheep 188 and goats. 174 Little is currently known about the implication of TLRs in sepsis of ruminants, although the pathways involved in triggering SIRS in sheep appear to be similar to those reported in other species. 169,200

The impact of infectious agents upon commercial viability of pigs and the contribution of this species to human disease modelling has helped advance the characterization of porcine PRRs. 191 Thus, TLRs 1–10 have been cloned and sequenced 201–208, surface and endosomal TLRs have been detected—both by PCR and immunohistochemistry—in a variety of porcine tissues and enhanced expression demonstrated in response to a number of infectious agents and PAMPs. 168,183,209–211 Members of the NLR family have also been identified 212–216 and current research suggests that porcine PRR signaling pathways are similar to those of other mammalian species. 169,217

Various members of the TLR family have been identified in horses, including TLRs 2, 3, 4, 5, and 9 47,53 Whereas many of these have been characterized by PCR, 176–179 TLRs 4 and 9 have also been localized by immunohistochemistry and immunogold electron microscopy 168,179,181—and a recent study demonstrated TLR 5 expression by equine neutrophils by flow cytometry. 218 Stimulation by PAMPs has increased gene expression of TLRs 2, 3, and 4 in vitro, 177 and TLR 9 in vivo, 181 whereas clinical studies have reported increased TLR 4 gene expression in both foals and adult horses with SIRS/sepsis, but no differences in expression between survivors and nonsurvivors. 219,220 Complementary studies have demonstrated increased plasma endothelin-1 concentrations and decreased long-term survival in horses with severe versus mild-to-moderate endotoxemia. 221 As for cattle and pigs, initial investigations have indicated that the downstream signaling pathways instigated by PAMP stimulation of equine TLRs 2–4 are similar to those identified in other species. 222 To date, there have been no published reports on members of the NLR family in the horse.

Less is currently known about PRRs in small animals. 53–55 Various PCR studies have confirmed the presence of members of the TLR family in both dogs, including TLRs 2, 4, 7, and 9 61,151–154,223 and cats, including TLRs 1–9. 49,151,155,156 Comparative examination of the canine genome has also identified the presence of genes encoding members of the NLRP family, integral to inflammasome and thus caspase activation. 224 A number of canine TLRs have also been detected by immunohistochemical staining and flow cytometry. 168,209,225,226 The use of feline models for investigation of human diseases as diverse as type 2 diabetes mellitus and human immunodeficiency virus infection has yielded additional data on PRRs in cats, including the expression of functional TLRs by the endocrine pancreas 155 and modulation of TLR signaling by retroviral pathogens. 156,227 Enhanced expression of canine TLRs has been observed in clinical cases of osteoarthritis 60 and cystic endometrial hyperplasia/pyometra 61,226, however, the majority of published research in dogs concerns the expression of PRRs in the intestinal tract, particularly in relation to inflammatory bowel disease (IBD). 150,223,228 Ongoing research is attempting to identify whether or not certain single nucleotide polymorphisms (SNPs) of PRRs can be related to the propensity for particular canine breeds to develop IBD and other immune-mediated diseases. 229–231 The analysis of SNPs in PRR-encoding genetic sequences is also an exciting field of research in large animals, 164,213,232 laying the foundation for the breeding of livestock with enhanced disease resistance and...
the design of vaccines better able to target dendritic cells.\textsuperscript{191,233} Finally, recent work has documented the expression of an ortholog of human “triggering receptor expressed on myeloid cells-1” (TREM-1) by canine neutrophils.\textsuperscript{234} Expression of TREM-1 was upregulated by microbial agonists of TLR1/2, TLR2/6, and TLR4/MD2.\textsuperscript{234} This receptor, which has shown promise as a biomarker of sepsis in humans, amplifies pro-inflammatory responses to microbial products.\textsuperscript{59,235}

**Inflammatory Mediators: Cytokines and Chemokines**

PRR ligation triggers signaling cascades that culminate in the activation of NF-κB and AP-1 via MyD88 or TICAM1/TRIF (Fig 2).\textsuperscript{236–238} NF-κB and AP-1 enter the nucleus and activate transcription sites for a variety of genes, including acute phase proteins, inducible nitric oxide synthase (iNOS), coagulation factors, and pro-inflammatory cytokines and chemokines, such as tumor necrosis factor (TNF-α) and IL-1α, 1, 6, 8, and 12. The TICAM1/TRIF pathway results in the phosphorylation of interferon regulatory factors 3 and 7 (IRF3, IRF7), which likewise enter the nucleus and stimulate the transcription of genes encoding interferon (IFN)α, IFNβ, and other type 1 IFN-inducible genes.\textsuperscript{236–238}

Serum concentrations of TNF-α correlate with death in certain types of human sepsis.\textsuperscript{239,240} Studies of naturally occurring sepsis in veterinary patients have yielded similar results, although this pattern appears not to be universal: increased serum concentrations of TNF-α correlate with mortality in canine parvovirus and neonatal septicemia in cattle and horses, but not in septic cats.\textsuperscript{11,241,242} TNF-α is predominantly produced by activated macrophages and T cells—but also by mast cells, B cells, NK cells, neutrophils, endothelial cells, myocytes, osteoblasts, and fibroblasts—as a 26 kDa precursor (pro-TNF) expressed on the plasma membrane; there it is cleaved by TNF-converting enzyme (TACE/ADAM17) to yield a 17 kDa soluble form, both the soluble and membrane-bound forms appearing to be active.\textsuperscript{243} TNF-α exerts its effects by interaction with one of 2 receptors, TNF receptors 1 and 2 (TNFR1, TNFR2).\textsuperscript{244,245} Activation of TNFR1 appears to mediate the proinflammatory and apoptotic pathways associated with inflammation, whereas TNFR2 plays a role in the promotion of tissue repair and angiogenesis.\textsuperscript{245} However, the complexity of signaling networks operating in sepsis is underlined by the observation that NF-κB activity induces molecules that block apoptosis mediated by TNFR1, suggesting that integration of the various signals occurs \textit{in vivo}.\textsuperscript{246} Some “cooperation” between the 2 receptor types, particularly at low TNF-α concentrations, is also likely and stimulation of both TNFRs leads to further NF-κB and AP-1 release.\textsuperscript{244,247}

Many of the classical features of inflammation can be attributed to the actions of TNF-α upon the endothelium, with increased production of iNOS and cyclo-oxygenase 2 (COX-2) leading to vasodilatation and local slowing of blood flow;\textsuperscript{248} TNF-α also stimulates the expression of endothelial adhesion molecules such as E-selectin, intercellular adhesion molecule 1 (ICAM-1), and vascular cell adhesion molecule 1 (VCAM-1).\textsuperscript{249,250} These 3 molecules lead to the tethering of leukocytes to the endothelial wall and their transmigration into the interstitium, accompanied by fluid and plasma macromolecules.\textsuperscript{19} There is also evidence for the upregulation of TNF-α and other proinflammatory cytokines in a canine model of sepsis involving the intravenous infusion of low doses of LPS: increased serum concentrations of TNF-α, IL-1β, and IL-6\textsuperscript{251–253} were accompanied in one of the studies by increased expression of pulmonary E-selectin and ICAM-1 and the influx of neutrophils—although whether the LPS induced the expression of the adhesion molecules directly or via induction of the proinflammatory cytokines, or both, remained unclear.\textsuperscript{251} A similar phenomenon of up-regulation of proinflammatory cytokines, including TNF-α, has also been observed in cats\textsuperscript{254} and horses\textsuperscript{255–257} treated with intravenous LPS, as well as the transcription of neutrophil chemoattractants by equine endothelial cells stimulated by Th2 cytokines.\textsuperscript{258} Furthermore, concentrations of TNF-α, IL-6, and endotoxin were all higher in the blood and peritoneal fluid of horses with colic than in a healthy control group.\textsuperscript{259} The stimulation of feline whole blood with various PAMPs has also elicited the synthesis of proinflammatory cytokines, including TNF, IL-1β, and CXCL-8.\textsuperscript{260} Plasma nitrite and nitrate, oxidation products of NO, have been examined in canine sepsis, in which they were present at higher concentrations than in dogs with SIRS alone.\textsuperscript{261} An additional study showed that the inflammatory response to the intravenous administration of LPS, examined by measuring serum concentrations of TNF-α, IL-1 and IL-6, was mitigated in dogs fed a diet rich in fish oils, suggesting that diet may be an adjunct to conventional anti-inflammatory treatment.\textsuperscript{262} In addition to the upregulation of iNOS, COX-2, and adhesion molecules, TNF-α also induces the expression of procoagulant proteins such as tissue factor (TF)—and down-regulates anti-coagulant factors such as thrombomodulin—leading to activation of the coagulation cascade.\textsuperscript{263} Despite the important role for TNF-α in endothelial activation, experimental evidence suggests that direct stimulation of TLRs expressed by endothelial and vascular smooth muscle cells may provide an alternative pathway for the vascular dysfunction seen in sepsis.\textsuperscript{264–266}

In addition to TNF-α, NF-κB activation results in the transcription of a number of proinflammatory interleukins, such as IL-1, IL-6, CXCL-8 (IL-8), and IL-12 (Fig 1). IL-1 acts in a synergistic manner with TNF-α in the “hyperacute” period after innate immune stimulation in sepsis.\textsuperscript{267,268} Two proinflammatory forms of IL-1 (IL-1α and IL-1β) have been identified and induce the synthesis of adhesion molecules and cytokines by endothelial cells, encouraging leukocyte activation, endothelial tethering, and transmigration into the interstitium.\textsuperscript{269,270} IL-1 also upregulates
iNOS and COX2 production, acts as the major endogenous pyrogen in fever, and increases corticosteroid release via hypothalamic effects.\(^{269,271}\)

Another important proinflammatory cytokine in sepsis is IL-6: as well as stimulatory effects upon leukocyte activation and myeloid progenitor cell proliferation, IL-6 also triggers the acute phase response and is a powerful pyrogen.\(^{272,273}\) Like TNF-\(\alpha\) and IL-1, plasma concentrations of IL-6 are increased in sepsis and may be predictive of progression to multiple organ dysfunction and death.\(^{17,274,275}\) Whereas enhanced gene expression of IL-6 correlates with death in septic foals, the opposite relationship appears to hold for serum IL-6 concentrations.\(^{219,276}\) However, increased serum concentrations of IL-6 and IL-1\(\beta\) do correlate with death in reports of naturally occurring sepsis in dogs and cats.\(^{11,13}\)

Serum concentrations of anti-inflammatory cytokines, most notably IL-10, also increase in sepsis.\(^{277,279}\) This acts not only to inhibit the release of TNF-\(\alpha\), IL-1\(\beta\), and IL-6 from monocytes and macrophages, but also to induce the production of IL-1 receptor antagonist protein (IRAP-1) and soluble TNFR, thus reducing circulating concentrations of these cytokines.\(^{279}\) The critical role of IL-10 in mediating the balance between pro- and anti-inflammatory processes can be seen in experimental models: IL-10 knockout mice are profoundly susceptible to sepsis, whereas IL-10 administration prevents these consequences.\(^{280}\) In clinical situations, however, the pattern appears to be more complex, with increased serum IL-10 concentrations associated with mortality in septic foals and humans,\(^{281,282}\) but a lower prevalence of feline infectious peritonitis in cats infected with feline coronavirus.\(^{283}\)

Two additional cytokines have recently been identified as being critical in sepsis. Macrophage migratory inhibitory factor (MIF), produced by the anterior pituitary gland, is present at increased concentrations in SIRS and sepsis\(^{284,285}\); serum concentrations have not only correlated with mortality, but inhibition of MIF appears to be protective.\(^{286–288}\) Although the trigger for MIF release in vivo is unclear, it is thought to delay apoptosis of activated monocytes and macrophages, thus helping to perpetuate a proinflammatory state.\(^{288}\) High mobility group box protein 1 (HMGB-1) is an endogenous protein involved in nuclear DNA stabilization.\(^{289}\) However, after necrotic or apoptotic cell death, it is released into the circulation where it has direct pro-inflammatory actions.\(^{290}\) In addition, HMGB-1 appears to potentiate the effect of certain PAMPs and DAMPs upon TLR-2, TLR-4, and the receptor for advanced glycation end products (RAGE).\(^{291}\) HMGB-1 was initially reported as a late inflammatory mediator in sepsis,\(^{292}\) although ongoing research has highlighted a number of roles in tissue repair and angiogenesis.\(^{293}\) Circulating concentrations of HMGB-1 appear to correlate with mortality in canine SIRS patients,\(^{294}\) but were unable to predict hospital mortality in 1 study of septic human patients.\(^{295}\)

**The Acute Phase Response**

In addition to the release of cytokines and chemokines from activated immune cells, triggering of PRRs also bring about the release of large quantities of acute phase proteins (APPs) from hepatocytes; these proteins have a variety of functions designed to re-establish homeostasis, assisting in pathogen elimination and subduing inflammation.\(^{296–297}\) The acute phase response is characterized by fever, neutrophilia, activation of the coagulation, and complement cascades (classical, alternative, and mannose-binding lectin pathways), serum iron and zinc binding, enhanced glucocorticoid, increased muscle catabolism, and altered lipid metabolism.\(^{297–299}\)

In general, 2 groups of APPs are recognized: type I, induced by IL-1\(\alpha\), IL-1\(\beta\), and TNF-\(\alpha\), and type II, induced by IL-6.\(^{298}\) As a consequence of the up-regulation of APP production, concentrations of other plasma proteins such as albumin, protein C, protein S, and antithrombin (collectively known as negative APPs) decrease.\(^{299}\) Granulocyte colony-stimulating factor (G-CSF) released from monocytes is also an important component of the acute phase response, thought to mediate a protective role against bacterial infection by virtue of its impact on neutrophils.\(^{301}\) A number of APPs have been characterized in veterinary species, including dogs, cats, cattle, sheep, horses, and chicken,\(^{297,302}\) and they have been used as biomarkers of inflammation in both research and clinical arenas in these species.\(^{303,304}\) Recent studies have identified adiponectin and insulin-like growth factor-1 as negative acute phase proteins in a canine model of endotoxemia.\(^{305}\)

Much research has been conducted to determine whether or not serum levels of APPs are predictive of survival in sepsis, with procalcitonin and C-reactive protein (CRP) the focus of particular attention.\(^{306}\) Plasma procalcitonin concentrations appear to correlate with bacteremia and organ dysfunction in human clinical studies.\(^{306,307}\) Whereas it also appears to be a canine APP, it does not allow the discrimination of inflammatory or infectious from neoplastic disease when measured as a whole blood PCR assay,\(^{308}\) however, extrathyroidal procalcitonin gene expression was documented in dogs with SIRS but not in healthy animals in a preliminary observational study.\(^{309}\) The utility of CRP measurements has been assessed in a number of studies examining infectious,\(^{310,311}\) inflammatory,\(^{312–315}\) neoplastic,\(^{320,321}\) and endocrine\(^{322}\) diseases in the dog, including critically ill dogs.\(^{323}\) In general, serum CRP concentrations provide a sensitive but nonspecific means of measuring inflammation, offering diagnostic and prognostic information in some disorders but not others. A recent study of SIRS and sepsis in dogs demonstrated a correlation between decreasing serum CRP concentration and recovery from disease, suggesting its use as a prognostic biomarker in this context.\(^{324}\)
The Interaction of Inflammation and Coagulation in Sepsis

Despite not classically considered part of the innate immune response, the prevalence of coagulation disorders and disseminated intravascular coagulation (DIC) in sepsis underlines the intimate link between the inflammatory and coagulation pathways. On a local scale, activation of coagulation may act defensively to impede the dispersal of pathogens and inflammatory mediators from the site of insult. Clinical studies have supported experimental models demonstrating increased activation of coagulation, as well as downregulation of anticoagulant mechanisms and reduced fibrinolysis, in human SIRS and sepsis patients. Similar findings have also been reported in dogs and horses. Whereas DIC and pulmonary thromboembolism have both been reported in association with sepsis in cats, experimental models of endotoxin infusion in this species have yielded variable results, a recent study failing to elicit any biologically significant alterations in coagulation parameters, underlining the multifactorial pathogenesis of coagulopathies in clinical patients.

A detailed description of the complexities of the interaction between inflammation and coagulation is beyond the scope of this article; however, several comprehensive reviews of hemostasis in SIRS and sepsis in veterinary species have recently been published and an overview of key interactions is presented in Fig 4. In brief, key to the triggering of the coagulation pathway in sepsis is tissue factor (TF), which initiates coagulation via the contact activation (extrinsic) pathway. In health, lack of TF exposure within the vascular system and the presence of various circulating proteins—such as protein C, antithrombin, and tissue factor plasminogen inhibitor—modulate coagulation by the prevention of TF activation. The expression of TF by monocytes or macrophages and tissue parenchymal cells is activated by various inflammatory cytokines, CRP, and PAMPs such as LPS—a phenomenon that has also been documented in cats and horses (earlier studies in this species citing “procoagulant activity” rather than TF per se). Although findings differ between species, large numbers of TF-expressing microparticles have been identified in blood samples from septic human patients and may correlate with mortality. These microparticles are released from a variety of activated or apoptotic cells—such as platelets, monocytes, erythrocytes, and endothelial cells—and their interaction with endothelial cells and platelets drives the coagulation pathway. Although only an indirect measure, plasma von Willebrand factor concentrations were higher in septic dogs than those in healthy control animals, suggesting that endothelial cell activation also occurs in canine sepsis. Interestingly, platelets enhanced endotoxin-induced equine monocyte TF activity in vitro—although whether microparticles derived from platelets or other sources play a role in the pathogenesis of equine sepsis in vivo remains unknown. Additional work in an equine model of endotoxemia has shown that large volume resuscitation has no impact on coagulation parameters beyond the changes attributed to endotoxemia, providing useful additional data to inform the treatment of sepsis in this species.

Whereas much research has been directed toward the inhibition of coagulation in sepsis, only activated protein C has shown any benefit in human clinical trials. The antithrombotic and anti-inflammatory properties of recombinant human activated protein C led to recommendations for its use in severe sepsis in 2004 and 2008, but a recent meta-analysis found no evidence in support of its administration in the treatment of severe sepsis or septic shock. Indeed, there is still debate about its mechanism of action and, as yet, little experience of its use in veterinary medicine, despite encouraging pharmacological data in experimental models.

In addition to the role of TF in initiating coagulation in the presence of inflammation, activated platelets and endothelial cells—as well as bacterial surfaces—also trigger the contact phase system, leading to the formation of kallikrein and bradykinin. Bradykinin in turn enhances vasodilatation and increases vascular permeability, as well as reducing platelet function; kallikrein accelerates fibrinolysis by conversion of plasminogen to plasmin and causes additional activation of Factor XII, leading to stimulation of the classical complement pathway.

A final connection between coagulation and inflammation in sepsis has become apparent with the recent exploration of the role of “a disintegrin-like and metalloproteinase with thrombospondin type-1 motifs-13” (ADAMTS-13). ADAMTS-13 is produced by the stellate (Ito) cells of the liver and acts to cleave ultra-large von Willebrand’s Factor (vWF) multimers into smaller multimers. These ultra-large vWF multimers are released from endothelial stores after inflammation and lead to platelet activation and aggregation; the ensuing microthrombi further compromise tissue blood flow, leading to additional propagation of the proinflammatory state. Although not yet identified in clinical veterinary species, decreased plasma ADAMTS-13 activity is associated with a poor prognosis in human sepsis patients; decreased activity is attributed to both a diminution of hepatic production and an increase in breakdown by plasma proteases.

The Compensatory Anti-Inflammatory Response Syndrome and Cell Death

Ongoing investigation of the molecular mechanisms of the SIRS response, as well as the notable failure of therapeutic blockade of proinflammatory mediators, led to the realization that the mortality associated with sepsis could not be explained solely by an uncontrollable “cytokine storm”. This resulted in the concept of an opposing “compensatory anti-inflammatory response syndrome” (CARS), thought to be an adaptive response to the excessive proinflammatory process in SIRS and sepsis. Whereas an appropriate balance is struck...
in the vast majority of instances of host-defense challenge, this is lost in sepsis—leading either to an uncontrolled proinflammatory reaction to infection, resulting in organ dysfunction, an undesirable compromise of the immune system permitting opportunistic infection (the “second hit”), or a combination of both.\textsuperscript{367}
Rather than a sequential or compensatory change, as was initially proposed, SIRS and CARS appear to occur simultaneously, and act to balance the host’s need to maintain defense while minimizing self-induced tissue damage. Serum concentrations of both pro- and anti-inflammatory mediators increase early on in sepsis, likewise, concentrations of a variety of both types of mediators (eg, IL-6, IRAP-1) are predictive of septic morbidity or mortality. Thus the changes in cytokine profile are both dynamic and heterogeneous, indicating that prescriptive immunomodulatory therapies are unlikely to meet with success. Furthermore, apoptosis of lymphocytes, hepatocytes, gastrointestinal epithelial cells and endothelial cells is increased, whereas that of neutrophils is decreased. Neutrophil function is also altered: both the migration of neutrophils to infected tissues and their antimicrobial function is diminished; moreover, peritoneal neutrophils are a potential source of IL-10, suppressing inflammatory monocytes in a model of polymicrobial sepsis. Although monocyte survival appears unaltered, sepsis results in the production of molecules such as IL-1 receptor associated kinase M (IRAK-M), MyD88 short variant (MyD88s), and A20-binding inhibitor of NF-kB activation3 (ABIN-3), which reduce activation of the NF-kB signaling pathway and therefore dampen the response to PAMP recognition. Monocyte function is impaired in sepsis, with decreased expression of the MHC class II molecule human leukocyte antigen-DR (HLA-DR) and increased inflammasome expression. Longitudinal observational studies in human patients indicate that a failure to regain >70% normal monocyctic expression of HLA-DR is associated with an increased risk of secondary bacterial infection and decreased survival. In addition to these changes in antigen-presenting function of monocytes, dendritic cells also undergo increased apoptosis in sepsis, further impairing the host’s ability to respond to pathogens.

Although apoptosis, or type 1 programmed cell death (PCD), is responsible for the majority of immune cell death in sepsis and is implicated in immunoparalysis, alternative pathways also play a role. Autophagy is a cellular mechanism that primarily acts as a cytoplasmic “clean-up” process, as well as assisting in delivery of proteins to antigen presentation pathways; however, it may also mediate type II PCD and interact with apoptosis. Much current interest has been directed toward the role of autophagy in trauma and sepsis, although whether it acts in a cytoprotective role or as a mechanism of PCD, or both, remains unclear. A final mechanism of interest in the pathogenesis of sepsis is pyroptosis, a term used to describe the process of caspase-1-mediated PCD, which is distinct from death mediated by the apoptotic caspases 3, 6, and 8. Although some question whether pyroptosis is truly a unique cell death mechanism, or simply a special case of apoptosis or necrosis (oncosis), it is characterized by rapid plasma membrane rupture and release of proinflamma-

A number of studies also suggest an important role for apoptosis in the pathogenesis of SIRS, sepsis, and infectious disease in veterinary species. Apoptotic cells were observed in the liver, kidney, thymus, stomach, and lymphocyte population of endotoxemic pigs and the primary and secondary lymphoid organs of pigs infected with classical swine fever virus. LPS and TNF-α both induced apoptosis of bovine glomerular endothelial cells, modeling a potential pathomechanism of acute renal failure in Gram-negative sepsis; this phenomenon could be potently inhibited by glucocorticoids in vitro. Haemophilus somnus, a Gram-negative pathogen of cattle that causes sepsis and vasculitis, induces caspases 3 and 8, and subsequent apoptosis, of endothelial cells in vitro; furthermore, the bacterium stimulates platelets that are in turn able to induce endothelial cell apoptosis by a contact-dependent mechanism involving the activation of caspases 8 and 9 and the synthesis of reactive oxygen species. The activity of matrix metalloproteinases 2 and 9 and the expression of phosphorylated Paxillin showed positive correlation with cardiomyocyte apoptosis in an ovine model of endotoxemic shock, whereas apoptosis of peripheral blood mononuclear cells and splenocytes of sheep infected with bluetongue virus was thought to contribute to immunosuppression in this disease. Ileal epithelial apoptosis was documented in one feline model of endotoxemia, although a second demonstrated lymphocyte apoptosis in the spleen and Peyer’s patches. Apoptosis of T cells also contributes to the immunosuppression characteristic of feline immunodeficiency virus infection. Finally, apoptosis of intestinal epithelial cells was observed in a canine model of sepsis induced by the intravenous infusion of E. coli.

Regulatory T Cells and Sepsis

One of the key features of the adaptive immune system is its ability to generate antigen-specific receptors with an enormous diversity of specificities, some of which may recognize host-derived epitopes. The majority of potentially autoaggressive thymocytes are deleted in the thymic medulla in a process called negative selection. However, this process of central tolerance is imperfect and underlines the importance of a number of peripheral tolerance mechanisms, including clonal deletion, functional inactivation (anergy), and phenotypic skewing. Although a subject of debate for many years, a population of Tregs is now known to play a major role in peripheral tolerance, complementing the preceding (intrinsic) mechanisms. Little is currently known about tolerance mechanisms in veterinary species, but Tregs have been identified in cats, dogs, pigs, cows, sheep, and horses.

Both naturally occurring and peripherally induced Tregs have been characterized—the former the product of a pathway of thymic differentiation called altered
negative selection and the latter the product of peripheral activation of conventional T cells in the context of an environment rich in transforming growth factor \( \beta \) (TGF-\( \beta \)) or IL-10.\textsuperscript{455–457} Naturally occurring Tregs have been identified by their constitutive expression of the IL-2 receptor \( \alpha \) chain (CD25) and Forkhead box P3 (FOXP3), a transcription factor that plays a pivotal role in both their ontogeny and peripheral function. FOXP3 acts to stabilize the Treg transcriptome by repressing a number of pro-inflammatory and growth-promoting genes—for example, IL-2 and IFNG—while activating others encoding key molecules involved in Treg function—for example, CTLA4 and CD25.\textsuperscript{458,459}

Naturally occurring Tregs are known to interact with cells of both the innate and adaptive immune systems—including monocytes, macrophages, natural killer cells, neutrophils, mast cells, dendritic cells, and both T and B cells—generally mediating a suppressive function to prevent the development of autoimmune responses and maintain the population of peripheral CD4\(^+\) T cells, thus contributing to immune system homeostasis.\textsuperscript{456,460,461} Tregs are also known to express a variety of TLRs, stimulation of which has augmented or abolished regulatory function in various studies.\textsuperscript{462–465} The molecular mechanisms of immune suppression mediated by naturally occurring Tregs have not been fully elucidated, but involve cell contact-dependent interactions, induction of cell death, and secretion of immunosuppressive cytokines, including IL-10 and TGF-\( \beta \).\textsuperscript{416,456}

Various studies have documented increased proportions of Tregs in human sepsis during the phase of immunoparalysis,\textsuperscript{466–469} but the role of this change remains unclear because depletion of Tregs in murine models of sepsis has yielded variable conclusions between models, either improving, enhancing, or bearing no influence on mortality.\textsuperscript{470,472} Given the ability of Tregs to induce the alternative activation pathway of macrophages\textsuperscript{473} and to inhibit the LPS-induced survival of monocytes through a proapoptotic mechanism involving the Fas/FasL pathway,\textsuperscript{474} they may make a potentially significant contribution to immune system dysfunction in human septic patients,\textsuperscript{468,469,475,476} although this is still a controversial area.\textsuperscript{477}

Additional research is required to elucidate the role of Tregs in sepsis, as well as to identify their mechanisms of action in this context. Moreover, to the authors' knowledge no veterinary studies have interrogated Treg number or function in septic patients to date. Pharmacological manipulation of Treg activity continues to be explored experimentally and may eventually translate to clinical cases; however, therapeutic interventions to alter the resistance of immune cells to Treg suppression may prove an equally valid alternative approach.\textsuperscript{478,479}

**Conclusions**

As is apparent from Figure 1, a broad concept of the immunopathological mechanisms underlying sepsis is now generally accepted. However, many of the molecular details are both complex and incompletely elucidated. Ongoing research has also indicated the existence of multiple redundant pathways within the innate immune response, potentially explaining the failure of many highly selective therapeutic interventions.\textsuperscript{14,19}

A single “magic bullet” for the treatment of sepsis is highly unlikely to exist: an individually tailored set of therapies, based on point-of-care assessment of the immunopathological status of the patient, is the likely future—albeit a distant one.\textsuperscript{73} Various human studies have resulted in the publication of consensus statements regarding the treatment of sepsis,\textsuperscript{352,353} but current veterinary evidence to substantiate these interventions is thin on the ground. What has become evident is that outside the experimental laboratory, the heterogeneous nature of the septic patient population means that clinical trials must be carefully designed to obtain meaningful data.\textsuperscript{480–482} Ongoing basic research into the immunopathology of sepsis in clinical veterinary species is equally important, to elucidate the underlying molecular mechanisms and thus direct clinical studies to those aspects of disease likely to benefit the greatest number of patients.

**Acknowledgments**

The authors acknowledge funding from the Biotechnology and Biological Sciences Research Council, Novartis Animal Health, the European College of Veterinary Internal Medicine, and the American College of Veterinary Emergency and Critical Care for work on canine regulatory T cells and sepsis.

**References**

1. Bone RC, Balk RA, Cerra FB, et al. Definitions for sepsis and organ failure and guidelines for the use of innovative therapies in sepsis. The ACCP/SCCM Consensus Conference Committee. American College of Chest Physicians/Society of Critical Care Medicine. Chest 1992;101:1644–1655.
2. Hotchkiss RS, Karl IE. The pathophysiology and treatment of sepsis. N Engl J Med 2003;348:138–150.
3. Levy MM, Fink MP, Marshall JC, et al. 2001 SCCM/ESICM/ACCP/ATS/SIS International Sepsis Definitions Conference. Crit Care Med 2003;31:1250–1256.
4. Levy MM, Fink MP, Marshall JC, et al. 2001 SCCM/ESICM/ACCP/ATS/SIS International Sepsis Definitions Conference. Intensive Care Med 2003;29:530–538.
5. Vincent JL, Sakr Y, Sprung CL, et al. Sepsis in European intensive care units: Results of the SOAPP study. Crit Care Med 2006;34:344–353.
6. Bone RC. The sepsis syndrome. Definition and general approach to management. Clin Chest Med 1996;17:175–181.
7. Angus DC, Linde-Zwirble WT, Lidicker J, et al. Epidemiology of severe sepsis in the United States: Analysis of incidence, outcome, and associated costs of care. Crit Care Med 2001;29:1303–1310.
8. Martin GS, Mannino DM, Eaton S, et al. The epidemiology of sepsis in the United States from 1979 through 2000. N Engl J Med 2003;348:1546–1554.
9. Brady CA, Otto CM. Systemic inflammatory response syndrome, sepsis, and multiple organ dysfunction. Vet Clin North Am Small Anim Pract 2001;31:1147–1162, v–vi.
10. Hoffman AM, Staempfli HR, Willan A. Prognostic variables for survival of neonatal foals under intensive care. J Vet Intern Med 1992;6:689–95.
11. Declue AE, Delgado C, Chang CH, et al. Clinical and immunologic assessment of sepsis and the systemic inflammatory response syndrome in cats. J Am Vet Med Assoc 2011;238:890–897.
12. Hurcombe SD, Toribio RE, Slovis NM, et al. Calcium regulating hormones and serum calcium and magnesium concentrations in septic and critically ill foals and their association with survival. J Vet Intern Med 2009;23:335–343.
13. Rau S, Kohn B, Richter C, et al. Plasma interleukin-6 response is predictive for severity and mortality in canine systemic inflammatory response syndrome and sepsis. Vet Clin Pathol 2007;36:253–260.
14. Chong DL, Sriskandan S. Pro-inflammatory mechanisms in sepsis. Contrib Microbiol 2011;17:86–107.
15. Cinel I, Opal SM. Molecular biology of inflammation and sepsis: A primer. Crit Care Med 2009;37:291–304.
16. Cohen J. The immunopathogenesis of sepsis. Nature 2002;420:885–891.
17. Ivady B, Beres BJ, Szabo D. Recent advances in sepsis research: Novel biomarkers and therapeutic targets. Curr Med Chem 2011;18:3211–3225.
18. Remick DG. Pathophysiology of sepsis. Am J Pathol 2007;170:1435–1444.
19. Sriskandan S, Altmann DM. The immunology of sepsis. J Pathol 2008;214:211–223.
20. Stearns-Kurosawa DJ, Osuchowski MF, Valentine C, et al. The pathogenesis of sepsis. Annu Rev Pathol 2011;6:19–48.
21. Ganz T, Weiss J. Antimicrobial peptides of phagocytes and epithelia. Semin Hematol 1997;34:343–354.
22. Hoffmann JA, Kafatos FC, Janeway CA, et al. Phylogenetic perspectives in innate immunity. Science 1999;284:1313–1318.
23. Zasloff M. Antimicrobial peptides in health and disease. N Engl J Med 2002;347:1358–1400.
24. Brogden KA, Ackermann M, McCray PB Jr, et al. Antimicrobial peptides in animals and their role in host defences. Int J Antimicrob Agents 2003;22:465–478.
25. Linde A, Ross CR, Davis EG, et al. Innate immunity and host defense peptides in veterinary medicine. J Vet Intern Med 2008;22:247–265.
26. Shai Y. Mechanism of the binding, insertion and destabilization of phospholipid bilayer membranes by alpha-helical antimicrobial and cell non-selective membrane-lytic peptides. Biochim Biophys Acta 1999;1462:55–59.
27. Wilmes M, Carmmue BP, Sahlg HG, et al. Antibiotic activities of host defense peptides: More to it than lipid bilayer perturbation. Nat Prod Rep 2011;28:1350–1358.
28. Ali AS, Townes CL, Hall J, et al. Maintaining a sterile urinary tract: The role of antimicrobial peptides. J Urol 2009;182:21–28.
29. Diamond G, Beckloff N, Ryan LK. Host defense peptides in the oral cavity and the lung: Similarities and differences. J Dent Res 2008;87:915–927.
30. Ham M, Kaunitz JD. Gastroduodenal mucosal defense. Curr Opin Gastroenterol 2008;24:665–673.
31. Laube DM, Yim S, Ryan LK, et al. Antimicrobial peptides in the airway. Curr Top Microbiol Immunol 2006;306:153–182.
32. Huttner KM, Brezinski-Caliguri DJ, Mahoney MM, et al. Antimicrobial peptide expression is developmentally regulated in the ovine gastrointestinal tract. J Nutr 1998;128:2978–2985.
33. Sang Y, Ortega MT, Blecha F, et al. Molecular cloning and characterization of three beta-defensins from canine testes. Infect Immun 2005;73:2611–2620.
34. Salaun B, de Saint-Vis B, Pacheco N, et al. CD208/dendritic cell-lysosomal associated membrane protein is a marker of normal and transformed type II pneumocytes. Am J Pathol 2004;164:861–871.
35. Matzinger P. Friendly and dangerous signals: Is the tissue in control? Nat Immunol 2007;8:11–13.
36. Frantz S, Ertl G, Bauersachs J. Toll-like receptor signaling in the ischemic heart. Front Biosci 2008;13:5772–5779.
37. Rakoff-Nahoum S, Paglino J, Eslami-Varzaneh F, et al. Recognition of commensal microflora by toll-like receptors is required for intestinal homeostasis. Cell 2004;118:229–241.
38. Davies D, Meade KG, Herath S, et al. Toll-like receptor and antimicrobial peptide expression in the bovine endometrium. Reprod Biol Endocrinol 2008;6:53.
39. Bianchi ME. DAMPs, PAMPs and alarms: All we need to know about danger. J Leukoc Biol 2007;81:1–5.
40. Gallucci S, Matzinger P. Danger signals: SOS to the immune system. Curr Opin Immunol 2001;13:114–119.
41. Matzinger P. Tolerance, danger, and the extended family. Annu Rev Immunol 1994;12:991–1045.
42. Akira S, Uematsu S, Takeuchi O. Pathogen recognition and innate immunity. Cell 2006;124:783–801.
43. Miyake K. Innate immune sensing of pathogens and danger signals by cell surface Toll-like receptors. Semin Immunol 2007;19:3–10.
44. Chen GY, Yang J, Zheng P, et al. CD24 and Siglec-10 selectively repress tissue damage-induced immune responses. Science 2009;323:1722–1725.
45. Chun KH, Seong SY, CD14 but not MD2 transmit signals from DAMP. Int Immunopharmacol 2010;10:98–106.
46. Mollen KP, Anand RJ, Tsung A, et al. Emerging paradigm: Toll-like receptor 4-sentinel for the detection of tissue damage. Shock 2006;26:430–437.
47. Astakhova NM, Perelygin AA, Zharkikh AA, et al. Characterization of equine and other vertebrate TLR3, TLR7, and TLR8 genes. Immunogenetics 2009;61:529–539.
48. Chang JS, Russell GC, Jann O, et al. Molecular cloning and characterization of Toll-like receptors 1-10 in sheep. Vet Immunol Immunopathol 2009;127:94–105.
49. Ignacio G, Nordone S, Howard KE, et al. Toll-like receptor expression in feline lymphoid tissues. Vet Immunol Immunopathol 2005;106:229–237.
50. McGuire K, Jones M, Welrion D, et al. Radiation hybrid mapping of all 10 characterized bovine Toll-like receptors. Anim Genet 2006;37:47–50.
51. Menzies M, Ingham A. Identification and expression of Toll-like receptors 1-10 in selected bovine and ovine tissues. Vet Immunol Immunopathol 2006;109:21–30.
52. Tirumurugaan KG, Dhansakaran S, Raj GD, et al. Differential expression of toll-like receptor mRNA in selected tissues of goat (Capra hircus). Vet Immunol Immunopathol 2010;133:296–301.
53. Turin L, Riva F. Toll-like receptor family in domestic animal species. Crit Rev Immunol 2008;28:513–538.
54. Welrion D, Coffey TJ. Pattern recognition receptors in companion and farm animals. The key to unlocking the door to animal disease? Vet J 2007;174:240–251.
55. Welrion D, Jann OC, Oloff V, et al. Variation matters: TLR structure and species-specific pathogen recognition. Trends Immunol 2009;30:124–130.
56. Williams DL, Ha T, Li C, et al. Modulation of tissue Toll-like receptor 2 and 4 during the early phases of polymicrobial sepsis correlates with mortality. Crit Care Med 2003;31:1808–1818.
57. Tsujimoto H, Ono S, Efron PA, et al. Role of Toll-like receptors in the development of sepsis. Shock 2008;29:315–321.
58. Sato S, Nomura F, Kawai T, et al. Synergy and cross-tolerance between Toll-like receptor (TLR) 2- and TLR4-mediated signaling pathways. J Immunol 2000;165:7096–7101.
59. Ford JW, McVicar DW. TREM and TREM-like receptors in inflammation and disease. Curr Opin Immunol 2009;21:38–46.
60. Kuroki K, Stoker AM, Sims HJ, et al. Expression of Toll-like receptors 2 and 4 in stifle joint synovial tissues of dogs with or without osteoarthritis. Am J Vet Res 2010;71:750–754.
61. Silva E, Leitao S, Henrique S, et al. Gene transcription of TLR2, TLR4. LPS ligands and prostaglandin synthesis enzymes are up-regulated in canine uteri with cystic endometrial hyperplasia-pyometra complex. J Reprod Immunol 2010;84:66–74.
62. Taraktsoglou M, Szalabska U, Magee DA, et al. Transcriptional profiling of immune genes in bovine monocye-derived macrophages exposed to bacterial antigens. Vet Immunol Immunopathol 2011;140:130–139.
63. Gundersen Y, Vaagenes P, Thrane I, et al. Early time course of altered leukocyte response to lipopolysaccharide and peptidoglycan in porcine gunshot injury. Acta Anaesthesiol Scand 2008;52:1231–1237.
64. Deitschel SJ, Krel ME, Chang CH, et al. Age-associated changes to pathogen-associated molecular pattern-induced inflammatory mediator production in dogs. J Vet Emerg Crit Care (San Antonio) 2010;20:494–502.
65. Benbarek H, Deby-Dupont G, Caudron I, et al. Interactions between lipopolysaccharides and blood factors on the stimulation of equine polymorphonuclear neutrophils. Vet Immunol Immunopathol 1998;64:313–322.
66. Declue AE, Johnson PJ, Day JL, et al. Pathogen-associated molecular pattern motifs from Gram-positive and Gram-negative bacteria induce different inflammatory mediator profiles in equine blood. Vet J 2011;190:00–00.
67. Fowler BL, Axiak SM, Declue AE. Blunted pathogen-associated molecular pattern motif induced TNF, IL-6 and IL-10 production from whole blood in dogs with lymphoma. Vet Immunol Immunopathol 2011;144:167–171.
68. Ishii KJ, Koyama S, Nakagawa A, et al. Host innate immune receptors and beyond: Making sense of microbial infections. Cell Host Microbe 2008;3:352–363.
69. Medzhitov R, Janeway C Jr. Innate immune recognition: Mechanisms and pathways. Immunol Rev 2000;173:89–97.
70. Zak DE, Aderem A. Systems biology of innate immunity. Immunol Rev 2009;227:264–282.
71. Kawai T, Akira S. Toll-like receptors and their crosstalk with other innate receptors in infection and immunity. Immunity 2011;34:637–650.
72. Koyama S, Ishii KJ, Kumar H, et al. Differential role of TLR- and RLR-signaling in the immune responses to influenza A virus infection and vaccination. J Immunol 2007;179:4711–4720.
73. Takahashi K, Kawai T, Kumar H, et al. Roles of caspase-8 and caspase-10 in innate immune responses to double-stranded RNA. J Immunol 2006;176:4520–4524.
74. Hashimoto C, Hudson KL, Anderson KV. The Toll gene of Drosophila, required for dorsal-ventral embryonic polarity, appears to encode a transmembrane protein. Cell 1988;52:269–279.
75. Gay NJ, Keith FJ. Drosophila Toll and IL-1 receptor. Nature 1991;351:355–356.
76. Brightbill HD, Libraty DH, Krutzik SR, et al. Host defense mechanisms triggered by microbial lipopolysaccharides through Toll-like receptors. Science 1999;285:732–736.
77. Lemaire B, Nicolas E, Michaut L, et al. The dorsoventral regulatory gene cassette zappate/Toll/cactus controls the potent antifungal response in Drosophila adults. Cell 1996;86:973–983.
78. Medzhitov R, Preston-Hurlburt P, Janeway CA Jr. A human homologue of the Drosophila Toll protein signals activation of adaptive immunity. Nature 1997;388:394–397.
79. Takeuchi O, Hoshino K, Kawai T, et al. Differential roles of TLR2 and TLR4 in recognition of gram-negative and gram-positive bacterial cell wall components. Immunity 1999;11:443–451.
80. Horng T, Barton GM, Medzhitov R. TIRAP: An adapter molecule in the Toll signaling pathway. Nat Immunol 2001;2:835–841.
81. Jenkins KA, Mansell A. TIR-containing adaptors in Toll-like receptor signalling. Cytokine 2010;49:237–244.
82. Medzhitov R, Preston-Hurlburt P, Kopp E, et al. MyD88 is an adaptor protein in the hToll/IL-1 receptor family signaling pathways. Mol Cell 1998;2:253–258.
83. O’Neill LA, Bowie AG. The family of five: TIR-domain-containing adaptors in Toll-like receptor signalling. Nat Rev Immunol 2007;7:353–364.
84. West AP, Koblansky AA, Ghosh S. Recognition and signaling by Toll-like receptors. Annu Rev Cell Dev Biol 2006;22:409–437.
85. Kawagoe T, Sato S, Matsushita K, et al. Sequential control of Toll-like receptor-dependent responses by IRAK1 and IRAK2. Nat Immunol 2008;9:684–691.
86. Sato S, Sanjo H, Takeda K, et al. Essential function for the kinase TAK1 in innate and adaptive immune responses. Nat Immunol 2005;6:1087–1095.
87. Aksoy E, Vanden Berge W, Detienne S, et al. Inhibition of phosphoinositide 3-kinase enhances TRIF-dependent NF-kappa B activation and IFN-beta synthesis downstream of Toll-like receptor 3 and 4. Eur J Immunol 2005;35:2200–2209.
88. Liljeroos M, Vuotteenaho R, Morath S, et al. Bruton’s tyrosine kinase together with PI 3-kinase are part of Toll-like receptor 2 multiprotein complex and mediate LTA induced Toll-like receptor 2 responses in macrophages. Cell Signal 2007;19:625–633.
89. Ojanemi M, Glumoff V, Harju K, et al. Phosphatidylinositol 3-kinase is involved in Toll-like receptor 4-mediated cytokine expression in mouse macrophages. Eur J Immunol 2003;33:597–605.
90. Cabanski M, Steinmuller M, Marsh LM, et al. PKR regulates TLR2/TLR4-dependent signaling in murine alveolar macrophages. Am J Respir Cell Mol Biol 2008;38:26–31.
91. Lentschat A, Karahashi H, Michelsen KS, et al. Mastoparan, a G protein agonist peptide, differentially modulates TLR4 and TLR2-mediated signaling in human endothelial cells and murine macrophages. J Immunol 2005;174:4252–4261.
92. Skokos D, Nussenzwieg MC. CD8-DCs induce IL-12-independent Th1 differentiation through Delta 4 Notch-like ligand in response to bacterial LPS. J Exp Med 2007;204:1525–1531.
93. Sabroe I, Parker LC, Dower SK, et al. The role of TLR activation in inflammation. J Pathol 2008;214:126–135.
94. Aldape MJ, Bryant AE, Katahira EJ, et al. Innate immune recognition of, and response to, Clostridium sordellii. Anaerobe 2010;16:125–130.
95. Netea MG, Kullberg BJ, Galama JM, et al. Non-LPS components of Chlamydia pneumoiae stimulate cytokine production through Toll-like receptor 2-dependent pathways. Eur J Immunol 2002;32:1188–1195.
96. Schwannder R, Dziarski R, Wescle H, et al. Peptidoglycan- and lipoteichoic acid-induced cell activation is mediated by toll-like receptor 2. J Biol Chem 1999;274:17406–17409.
97. Zahringer U, Lindner B, Inamura S, et al. TollR2 - promiscuous or specific? A critical re-evaluation of a receptor expressing apparent broad specificity. Immunobiology 2008;213:364–379.
98. Lorne E, Dupont H, Abraham E. Toll-like receptors 2 and 4: Initiators of non-septic inflammation in critical care medicine? Intensive Care Med 2010;36:1826–1835.
99. Takeuchi O, Kawai T, Mulhardt PF, et al. Discrimination of bacterial lipopolysaccharides by Toll-like receptor 6. Int Immunol 2001;13:933–940.
100. Takeuchi O, Sato S, Horiuchi T, et al. Cutting edge: Role of Toll-like receptor 1 in mediating immune response to microbial lipopolysaccharides. J Immunol 2002;169:10–14.
101. Hoshino K, Takeuchi O, Kawai T, et al. Cutting edge: Toll-like receptor 4 (TLR4)-deficient mice are hyporesponsive to lipopolysaccharide: Evidence for TLR4 as the Lps gene product. J Immunol 1999;162:3749–3752.
102. Lu YC, Yeh WC, Ohashi PS. LPS/TLR4 signal transduction pathway. Cytokine 2008;42:145–151.
103. Takeda K, Akira S. Toll-like receptors in innate immunity. Int Immunol 2005;17:1–14.
104. Park BS, Song DH, Kim HM, et al. The structural basis of lipopolysaccharide recognition by the TLR4-MD-2 complex. Nature 2009;451:1191–1195.
105. Andersen-Nissen E, Smith KD, Bonneau R, et al. A conserved surface on Toll-like receptor 5 recognizes bacterial flagellin. J Exp Med 2007;204:393–403.
106. Uematsu S, Fujimoto K, Jang MH, et al. Regulation of humoral and cellular gut immunity by lamina propria dendritic cells expressing Toll-like receptor 5. Nat Immunol 2008;9:769–776.
107. Liaudet L, Szabo C, Evgenov OV, et al. Flagellin from Gram-negative bacteria is a potent mediator of acute pulmonary inflammation in sepsis. Shock 2003;19:131–137.
108. Kumar H, Kawai T, Akira S. Pathogen recognition in the innate immune response. Biochem J 2009;420:1–16.
109. Herskovits AA, Auerbach V, Portnoy DA. Bacterial ligands generated in a phagosome are targets of the cytosolic innate immune system. PLoS Pathog 2007;3:e51.
110. Yamamoto M, Sato S, Hemmi H, et al. Role of adaptor TRIF in the MyD88-independent Toll-like receptor signaling pathway. Science 2003;301:640–643.
111. Yarovsky F, Zhang D, Andersen JF, et al. TLR11 activation of dendritic cells by a protozoan profilin-like protein. Science 2005;308:1626–1629.
112. Zhang D, Zhang G, Hayden MS, et al. A Toll-like receptor that prevents infection by uropathogenic bacteria. Science 2004;303:1522–1526.
113. Carneiro LA, Magalhaes JG, Tattoli I, et al. Nod-like proteins in inflammation and disease. J Pathol 2008;214:136–148.
114. Chamaillard M, Hashimoto M, Horie Y, et al. An essential role for NOD1 in host recognition of bacterial peptidoglycan containing diaminopimelic acid. Nat Immunol 2003;4:702–707.
115. Girardin SE, Boneca IG, Viala J, et al. Nod1 recognizes bacterial flagellin. The plant immune system. Nature 2009;429:751–755.
116. Jones JD, Dangl JL. The plant immune system. Nature 2006;440:237–241.
117. Sutterwala FS, Mijares LA, Li L, et al. Immune recognition of Pseudomonas aeruginosa mediated by the IPAF/NLR4 inflammasome. J Exp Med 2007;204:3245–3253.
118. Franchi L, Amer A, Body-Malapel M, et al. Cytosolic flagellin requires Ipaf for activation of caspase-1 and interleukin 1beta in salmonella-infected macrophages. Nat Immunol 2006;7:576–582.
119. Martino F, Petrilli V, Mayor A, et al. Gout-associated uric acid crystals activate the NALP3 inflammasome. Nature 2006;440:237–241.
120. Rietdijk W, Schrier MJ, Beertema E, et al. Role of NOD1 in monocyte-macrophage viability and oncostatin m production. Metab. J Biol Chem 2005;280:18678–18687.
121. Kawai T, Takeda K, Akira S. NOD-like Toll-like receptors in innate immunity. Curr Top Microbiol Immunol 2004;290:151–173.
122. Yarovinsky F, Zhang D, Andersen JF, et al. TLR11 activation of dendritic cells by a protozoan profilin-like protein. Science 2005;308:1626–1629.
123. Zhang D, Zhang G, Hayden MS, et al. A Toll-like receptor that prevents infection by uropathogenic bacteria. Science 2004;303:1522–1526.
124. Patrignani A, Bussolari M, etc. Cutting edge: Toll-like receptor 4 (TLR4)-deficient mice are hyporesponsive to lipopolysaccharide: Evidence for TLR4 as the Lps gene product. J Immunol 1999;162:3749–3752.
125. Vila J, Chaput C, Boneca IG, et al. Nod1 responds to peptidoglycan delivered by the Helicobacter pylori cag pathogenicity island. Nat Immunol 2004;5:1166–1174.
126. Hasegawa M, Fujimoto Y, Lucas PC, et al. A critical role of RICK/RIP2 polyubiquitination in Nod-induced NF-kappaB activation. EMBO J 2008;27:373–383.
127. Inohara N, Koseki T, del Peso L, et al. Nod1, an Apaf-1-like activator of caspase-9 and nuclear factor-kappaB. J Biol Chem 1999;274:14560–14567.
128. Kim JY, Omori E, Matsumoto K, et al. TAK1 is a central mediator of NOD2 signaling in epithelial cells. J Biol Chem 2008;283:137–144.
129. Ogura Y, Inohara N, Benito A, et al. Nod2, a Nod1/ Apaf-1 family member that is restricted to monocytes and activates NF-kappaB. J Biol Chem 2001;276:4812–4818.
130. Strober W, Watanabe T. NOD2, an intracellular immune sensor involved in host defense and Crohn’s disease. Mucosal Immunol 2011;4:484–495.
131. Masumoto J, Yang K, Varambally S, et al. Nod1 acts as an intracellular receptor to stimulate chemokine production and neutrophil recruitment in vivo. J Exp Med 2006;203:203–213.
132. Cartwright N, Murch O, McMaster SK, et al. Selective NOD1 agonists cause shock and organ injury/dysfunction in vivo. Am J Respir Crit Care Med 2007;175:595–603.
133. Lamkanfi M, Dixit VM. Inflammasomes: Guardians of cytosolic sanctity. Immunol Rev 2006;297:95–105.
134. Lamkanfi M, Dixit VM. The inflammasomes. PLoS Pathog 2009;5:e1000510.
135. Martinon F, Burns K, Tschopp J. The inflammasome: A molecular platform triggering activation of inflammatory caspases and processing of proIL-beta. Mol Cell 2002;10:417–426.
136. Franchi L, Amer A, Body-Malapel M, et al. Cytosolic flagellin requires Ipaf for activation of caspase-1 and interleukin 1beta in salmonella-infected macrophages. Nat Immunol 2006;7:576–582.
137. Martinon F, Petrilli V, Mayor A, et al. Gout-associated uric acid crystals activate the NALP3 inflammasome. Nature 2006;440:237–241.
138. Sutterwala FS, Mijares LA, Li L, et al. Immune recognition of Pseudomonas aeruginosa mediated by the IPAF/NLR4 inflammasome. J Exp Med 2007;204:3245–3245.
139. Franchi L, Amer A, Body-Malapel M, et al. Cytosolic flagellin requires Ipaf for activation of caspase-1 and interleukin 1beta in salmonella-infected macrophages. Nat Immunol 2006;7:576–582.
140. Martinon F, Petrilli V, Mayor A, et al. Gout-associated uric acid crystals activate the NALP3 inflammasome. Nature 2006;440:237–241.
141. Sutterwala FS, Mijares LA, Li L, et al. Immune recognition of Pseudomonas aeruginosa mediated by the IPAF/NLR4 inflammasome. J Exp Med 2007;204:3245–3245.
142. Tsuji NM, Tsutsui H, Seki E, et al. Roles of caspase-1 in Listeria infection in mice. Int Immunol 2004;16:335–343.
143. Mariathasan S, Weiss DS, Dixit VM, et al. Inactive immunity against Francisella tularensis is dependent on the ASC/caspase-1 axis. J Exp Med 2005;202:1043–1049.
144. Sarkar A, Hall MW, Exline M, et al. Caspase-1 regulates Escherichia coli sepsis and splenic B cell apoptosis independently of interleukin-1beta and interleukin-18. Am J Respir Crit Care Med 2006;174:1001–1010.
145. Saleh M, Mathison JC, Wolinski MK, et al. Enhanced bacterial clearance and sepsis resistance in caspase-12-deficient mice. Nature 2006;440:1064–1068.
146. Saleh M, Vaillancourt JP, Graham RK, et al. Differential modulation of endotoxin responsiveness by human caspase-12 polymorphisms. Nature 2004;429:75–79.
147. Li P, Allen H, Banerjee S, et al. Mice deficient in IL-1 beta-converting enzyme are defective in production of mature IL-1 beta and resistant to endotoxic shock. Cell 1995;80:401–411.
148. Mariathasan S, Newton K, Monack DM, et al. Differential activation of the inflammasome by caspase-1 adaptsors ASC and Ipaf. Nature 2004;430:213–218.
Toll-like receptors and impact of biometric molecules. Cells Tissues Organs 2010;192:250–261.

185. Auray G, Facci MR, van Kessel J, et al. Differential activation and maturation of two porcine DC populations following TLR ligand stimulation. Mol Immunol 2010;47:2103–2111.

186. Morozumi T, Uenishi H. Polymorphism distribution and structural conservation in RNA-sensing Toll-like receptors 3, 7, and 8 in pigs. Biochim Biophys Acta 2009;1790:267–274.

187. Toka FN, Nfon CK, Dawson H, et al. Accessory-cell-mediated activation of porcine NK cells by Toll-like receptor 7 (TLR7) and TLR8 agonists. Clin Vaccine Immunol 2009;16:866–875.

188. Burkey TE, Skjølaas KA, Dritz SS, et al. Expression of porcine Toll-like receptor 2, 4, and 9 gene transcripts in the presence of lipopolysaccharide and Salmonella enterica serovars Typhimurium and Choleraesuis. Vet Immunol Immunopathol 2011;140:181–190.

189. Liu CH, Chaung HC, Chang HL, et al. Expression of Toll-like receptor mRNA and cytokines in pigs infected with porcine reproductive and respiratory syndrome virus. Vet Microbiol 2009;136:266–276.

190. Chaung HC, Chen CW, Hsieh BL, et al. Toll-like receptor expressions in porcine alveolar macrophages and dendritic cells in responding to poly IC stimulation and porcine reproductive and respiratory syndrome virus. Vet Res 2008;39:11.

191. Uenishi H, Shinkai H. Porcine Toll-like receptors: The front line of pathogen monitoring and possible implications for disease resistance. Dev Comp Immunol 2009;33:353–361.

192. Ibeagha-Awemu EM, Lee JW, Ibeagha AE, et al. Bacterial lipopolysaccharide induces increased expression of Toll-like receptor (TLR) 4 and downstream TLR signaling molecules in bovine mammary epithelial cells. Vet Res 2008;39:11.

193. Worku M, Morris A. Binding of different forms of lipopolysaccharide and gene expression in bovine blood neutrophils. J Dairy Sci 2009;92:3185–3193.

194. Cates EA, Connor EE, Mosser DM, et al. Functional characterization of bovine TIRAP and MyD88 in mediating bacterial lipopolysaccharide-induced endothelial NF-kappaB activation and apoptosis. Comp Immunol Microbiol Infect Dis 2009;32:477–490.

195. Gil LH, van Olphen AL, Mittal SK, et al. Modulation of PKR activity in cells infected by bovine viral diarrhea virus. Virus Res 2006;116:69–77.

196. Sen A, Priausser AJ, Dermody TS, et al. The early interferon response to rotavirus is regulated by PKR and depends on MAVS/IP51, RIG-I, MDA-5, and IRF3. J Virol 2011;85:3717–3732.

197. Bougarn S, Cunha P, Harmache A, et al. Muramyl dipeptide synergizes with Staphylococcus aureus lipoteichoic acid to recruit neutrophils in the mammary gland and to stimulate mammary epithelial cells. Clin Vaccine Immunol 2010;17:1797–1809.

198. Wheleham J, Rundgren M, Forestier J, et al. The early interferon response to rotavirus is regulated by PKR and depends on MAVS/IP51, RIG-I, MDA-5, and IRF3. J Virol 2011;85:3717–3732.

199. Schleifer CJ, Meade KG, Eckersall PD, et al. Experimental Staphylococcus aureus infection of the mammary gland induces region-specific changes in innate immune gene expression. Vet Immunol Immunopathol 2011;140:181–189.

200. Purdie AC, Plain KM, Begg DJ, et al. Candidate gene and genome-wide association studies of Mycobacterium avium subsp. paratuberculosis infection in cattle and sheep: A review. Comp Immunol Microbiol Infect Dis 2011;34:197–208.

201. Fenhhammer J, Rundgren M, Forestier J, et al. Toll-like receptor 4 inhibitor TAK-242 attenuates acute kidney injury in endotoxemic sheep. Anesthesiology 2011;114:1130–1137.

202. Alves MP, Neuhaus V, Guzylack-Priou L, et al. Toll-like receptor 7 and MyD88 knockdown by lentivirus-mediated RNA interference to porcine dendritic cell subsets. Gene Ther 2007;14:836–844.

203. Moue M, Tohno M, Shimazu T, et al. Toll-like receptor 4 and cytokine expression involved in functional immune response in an originally established porcine intestinal epithelial cell line. Biochim Biophys Acta 2008;1780:134–144.

204. Muneta Y, Uenishi H, Kikuma R, et al. Porcine TLR2 and TLR6: Identification and their involvement in Mycobacteria intracellular infection. J Interferon Cytokine Res 2003;23:583–590.

205. Shimosato T, Kitazawa H, Katoh S, et al. Swine Toll-like receptor 9(1) recognizes CpG motifs of human cell stimulant. Biochim Biophys Acta 2003;1627:56–61.

206. Shinkai H, Tanaka M, Morozumi T, et al. Biased distribution of single nucleotide polymorphisms (SNPs) in porcine Toll-like receptor 1 (TLR1), TLR2, TLR4, TLR5, and TLR6 genes. Immunogenetics 2006;58:324–330.

207. Shinkai H, Muneta Y, Suzuki K, et al. Porcine Toll-like receptor 1, 6, and 10 genes: Complete sequencing of genomic region and expression analysis. Mol Immunol 2006;43:1474–1480.

208. Thomas AV, Broers AD, Vandegaert HF, et al. Genomic structure, promoter analysis and expression of the porcine (Sus scrofa) TLR4 gene. Mol Immunol 2006;43:653–659.

209. Wassef A, Janardhan K, Pearce JW, et al. Toll-like receptor 4 in normal and inflamed lungs and other organs of pig, dog and cattle. Histol Histopathol 2004;19:1201–1208.

210. Migue JC, Chen J, Van Alstine WG, et al. Activation of inflammatory cytokines and Toll-like receptors in the brain and respiratory tract of pigs infected with porcine reproductive and respiratory syndrome virus. Vet Immunol Immunopathol 2010;135:314–319.

211. Sang Y, Yang J, Ross CR, et al. Molecular identification and functional expression of porcine Toll-like receptor (TLR) 3 and TLR7. Vet Immunol Immunopathol 2008;125:162–167.

212. Jozaki K, Shinkai H, Tanaka-Matsuda M, et al. Influence of polymorphisms in porcine NOD2 on ligand recognition. Mol Immunol 2009;47:247–252.

213. Kojima-Shibata C, Shinkai H, Morozumi T, et al. Differences in distribution of single nucleotide polymorphisms among intracellular pattern recognition receptors in pigs. Immunogenetics 2009;61:153–160.

214. Luo R, Xiao S, Jiang Y, et al. Porcine reproductive and respiratory syndrome virus (PRRSV) suppresses interferon-beta production by interfering with the RIG-I signaling pathway. Mol Immunol 2008;45:2839–2846.

215. Tohno M, Shimazu T, Aso H, et al. Molecular cloning and functional characterization of porcine nucleotide-binding oligomerization domain-1 (NOD1) recognizing minimum agonists, meso-diaminopimelic acid and meso-lanthionine. Mol Immunol 2008;45:1807–1817.

216. Tohno M, Ueda W, Azuma Y, et al. Molecular cloning and functional characterization of porcine nucleotide-binding oligomerization domain-2 (NOD2). Mol Immunol 2008;45:194–203.

217. Tohno M, Shimazu T, Aso H, et al. Molecular cloning and functional characterization of porcine MyD88 essential for TLR signaling. Cell Mol Immunol 2007;4:369–376.

218. Kwon S, Gewirtz AT, Hurley DJ, et al. Disparities in TLR5 expression and responsiveness to flagellin in equine neutrophils and mononuclear phagocytes. J Immunol 2011;186:6263–6270.

219. Gold JR, Perkins GA, Erb HN, et al. Cytokine profiles of peripheral blood mononuclear cells isolated from septic and healthy neonatal foals. J Vet Intern Med 2007;21:482–488.

220. Lopes MA, Saltor CE, Vandenplas ML, et al. Expression of inflammation-associated genes in circulating leukocytes
collected from horses with gastrointestinal tract disease. Am J Vet Res 2010;71:915–924.

221. Menzies-Gow NJ, Bailey SR, Stevens K, et al. Digital flow blood and plasma endothelin concentration in clinically endotoxemic horses. Am J Vet Res 2005;66:630–636.

222. Figureiro BD, Vandenplas ML, Hurley DJ, et al. Differential induction of MyD88- and TRIF-dependent pathways in equine monocytes by Toll-like receptor agonists. Vet Immunol Immunopathol 2009;127:125–134.

223. Allenspach K, House A, Smith K, et al. Evaluation of mucosal bacteria and histopathology, clinical disease activity and expression of Toll-like receptors in German Shepherd Dogs with chronic enteropathies. Vet Microbiol 2010;146:326–335.

224. Tian X, Pascal G, Monget P. Evolution and functional divergence of NLRP genes in mammalian reproductive systems. BMC Evol Biol 2009;9:202.

225. Burgener IA, Jungi TW. Antibodies specific for human or murine Toll-like receptors detect canine leukocytes by flow cytometry. Vet Immunol Immunopathol 2008;124:184–191.

226. Chotimpanukul S, Sirivaidyaporn S. Differential expression of Toll-like receptor 4 (TLR4) in healthy and infected canine endometrium. Theriogenology 2011;76:1152–1161.

227. Abujamra AL, Spanjaard RA, Akinsheye I, et al. Leukemia virus long terminal repeat activates NF-kappaB pathway by a TLR3-dependent mechanism. Virology 2006;354:390–403.

228. Burgener IA, Konig A, Allenspach K, et al. Upregulation of Toll-like receptors in chronic enteropathies in dogs. J Vet Intern Med 2008;22:553–560.

229. House AK, Binns MM, Gregory SP, et al. Analysis of NOD1, NOD2, TLR1, TLR2, TLR4, TLR5, TLR6 and TLR9 genes in anal furunculosis of German Shepherd Dogs. Tissue Antigens 2009;73:250–254.

230. Kathrani A, House A, Catchpole B, et al. Polymorphisms in the TLR4 and TLR5 gene are significantly associated with inflammatory bowel disease in German Shepherd Dogs. PLoS ONE 2010;5:e15740.

231. Kathrani A, House A, Catchpole B, et al. Breed-independent Toll-like receptor 5 polymorphisms show association with canine inflammatory bowel disease. Tissue Antigens 2011;78:94–101.

232. Mikula I, Bihe M, Pastorekova S. Characterization of ovine TLR7 and TLR8 protein coding regions, detection of mutations and Maedi Visna virus infection. Vet Immunol Immunopathol 2010;138:51–59.

233. McCullough KC, Summerfield A. Targeting the porcine immune system–particulate vaccines in the 21st century. Dev Comp Immunol 2009;33:394–400.

234. Li J, Birkenheuer AJ, Marr HS, et al. Expression and function of triggering receptor expressed on myeloid cells-1 (TREM-1) on canine neutrophils. Dev Comp Immunol 2011;35:872–880.

235. Derive M, Massin F, Gibot S. Triggering receptor expressed on myeloid cells-1 as a new therapeutic target during inflammatory diseases. Self Nonself 2010;1:225–229.

236. Zhu J, Mohan C. Toll-like receptor signaling pathways–therapeutic opportunities. Mediators Inflamm 2010;2010:781235.

237. Yamamoto M, Takeda K. Current views of Toll-like receptor signaling in cats in response to lipopolysaccharide: An in vivo and in vitro study. Vet Immunol Immunopathol 1995;49:183–188.

238. MacKay RJ, Merritt AM, Zertuche JM, et al. Tumor necrosis factor activity in the circulation of horses given endotoxin. Am J Vet Res 1991;52:533–538.

239. Nieto JE, MacDonald MH, Braim AE, et al. Effect of lipopolysaccharide infusion on gene expression of inflammatory cytokines in normal horses in vivo. Equine Vet J 2009;41:717–719.

240. Barton MH, Collatos C. Tumor necrosis factor and interleukin-6 activity and endotoxin concentration in peritoneal fluid and blood of horses with acute abdominal disease. J Vet Intern Med 1999;13:457–464.
260. Stich AN, DeClue AE. Pathogen associated molecular pattern-induced TNF, IL-1beta, IL-6 and CXCL-8 production from feline whole blood culture. Res Vet Sci 2011;90:59–63.

261. Osterbur K, Whitehead Z, Sharp CR, et al. Plasma nitrate/nitrite concentrations in dogs with naturally developing sepsis and non-infectious forms of the systemic inflammatory response syndrome. Vet Rec 2011;169:554.

262. LeBlanc CJ, Horohov DW, Bauer JE, et al. Effects of dietary supplementation with fish oil on in vivo production of inflammatory mediators in clinically normal dogs. Am J Vet Res 2008;69:486–493.

263. Bevilacqua MP, Pober JS, Majeau GR, et al. Recombinant tumor necrosis factor induces procoagulant activity in cultured human endothelium: Characterization and comparison with the actions of interleukin 1. Proc Natl Acad Sci USA 1986;83:453–4537.

264. Cartwright N, McMasters SR, Sorrentino R, et al. Elucidation of Toll-like receptor and adapter protein signaling in vascular dysfunction induced by gram-positive Staphylococcus aureus or gram-negative Escherichia coli. Shock 2007;27:40–47.

265. Jimenez R, Belcher E, Sriskandan S, et al. Role of Toll-like receptors 2 and 4 in the induction of cyclooxygenase-2 in vascular smooth muscle. Proc Natl Acad Sci USA 2005;102:4637–4642.

266. Schouwen M, Wiersinga WJ, Levi M, et al. Inflammation, endothelium, and coagulation in sepsis. J Leukoc Biol 2008;83:536–545.

267. Cavaillon JM, Adib-Conquy M, Fitting C, et al. Cytokine cascade in sepsis. Scand J Infect Dis 2003;35:535–544.

268. Dinarello CA. Proinflammatory and anti-inflammatory cytokines as mediators in the pathogenesis of septic shock. Chest 1997;112:321S–328S.

269. Stylianou E, Saklatvala J. Interleukin-1. Int J Biochem Cell Biol 1998;30:1075–1079.

270. Chaitanya GV, Franks SE, Cromer W, et al. Differential cytokine responses in human and mouse lymphatic endothelial cells to cytokines in vitro. Lymphat Res Biol 2010;8:155–164.

271. Dinarello CA. A clinical perspective of IL-1beta as the gatekeeper of inflammation. Eur J Immunol 2011;41:1203–1217.

272. Jawab RS, Anillo S, Hunton K, et al. Analytic review: Interleukin-6 in surgery, trauma, and critical care: Part I: Basic science. J Intensive Care Med 2011;26:3–12.

273. Scheller J, Chalaris A, Schmidt-Arras D, et al. The pro- and anti-inflammatory properties of the cytokine interleukin-6. Biochim Biophys Acta 2011;1813:878–888.

274. Borrelli E, Roux-Lombard P, Grau GE, et al. Plasma concentrations of cytokines, their soluble receptors, and antioxidan- vaults in patients with severe sepsis or septic shock. J Surg Res 2010;164:e163–171.

275. Mitchell RA, Liao H, Chesney J, et al. Macrophage migration inhibitory factor gene reveals its critical role in sep-sis. J Exp Med 1999;189:341–346.

276. Brenner T, Hofer S, Rosenhagen C, et al. Macrophage migration inhibitory factor (MIF) and manganese superoxide dismutase (MnSOD) as early predictors for survival in patients with severe sepsis or septic shock. J Surg Res 2010;164:e163–171.

277. Yang H, Tracey KJ. Targeting HMGB1 in inflammation. Biochim Biophys Acta 2010;1799:101–113.

278. van Zoonen MA, Yang H, Florquin S, et al. Role of Toll-like receptors 2 and 4, and the receptor for advanced glycation end products in high-mobility group box 1-induced inflammation in vivo. Shock 2009;31:280–284.

279. Huang W, Tang Y, Li L. HMGB1, a potent proinflammatory cytokine in sepsis. Cytokine 2010;51:119–126.

280. Kuske AM, Rongione AJ, Ashley SW, et al. Interleu-kin-10 prevents death in lethal necrotizing pancreatitis in mice. Surgery 1996;120:284–288; discussion 289.

281. Briassouil G, Venkatakrishnan S, Thompson A. Cytokines and metabolic patterns in pediatric patients with critical illness. Clin Dev Immunol 2010;2010:354047.

282. Pusterla N, Magdesian KG, Mapes S, et al. Expression of molecular markers in blood of neonatal foals with sepsis. Am J Vet Res 2006;67:1045–1049.

283. Kipar A, Mili ML, Failing K, et al. Natural feline coronavirus infection: Differences in cytokine patterns in association with the outcome of infection. Vet Immunol Immunopathol 2006;112:141–155.

284. Gao L, Flores C, Fan-Ma S, et al. Macrophage migration inhibitory factor in acute lung injury: Expression, biomarker, and associations. Transl Res 2007;150:18–29.

285. Calandra T, Roger T. Macrophage migration inhibitory factor: A regulator of innate immunity. Nat Rev Immunol 2003;3:791–800.

286. Bozza M, Satoskar AR, Lin G, et al. Targeted disruption of migration inhibitory factor gene reveals its critical role in sepsis. Blood 2003;98:4471–4481.

287. Brenner T, Hofer S, Rosenhagen C, et al. Macrophage migration inhibitory factor (MIF) and manganese superoxide dismutase (MnSOD) as early predictors for survival in patients with severe sepsis or septic shock. J Surg Res 2010;164:e163–171.

288. Mitchell RA, Liao H, Chesney J, et al. Macrophage migration inhibitory factor (MIF) sustains macrophage proinflammatory function by inhibiting p53: Regulatory role in the innate immune response. Proc Natl Acad Sci USA 2002;99:345–350.

289. Stros M. HMGB proteins: Interactions with DNA and chromatin. Biochim Biophys Acta 2010;1799:101–113.

290. Yang H, Tracey KJ. Targeting HMGB1 in inflammation. Biochim Biophys Acta 2010;1799:149–156.

291. van Zoonen MA, Yang H, Florquin S, et al. Role of Toll-like receptors 2 and 4, and the receptor for advanced glycation end products in high-mobility group box 1-induced inflammation in vivo. Shock 2009;31:280–284.

292. Huang W, Tang Y, Li L. HMGB1, a potent proinflammatory cytokine in sepsis. Cytokine 2010;51:119–126.

293. Mitola S, Belleri M, Urbinati C, et al. Cutting edge: Extracellular high mobility group box-1 protein is a proangiogenic cytokine. J Immunol 2006;176:12–15.

294. Yu DH, Nho DH, Song RH, et al. High-mobility group box 1 as a surrogate prognostic marker in dogs with systemic inflammatory response syndrome. J Vet Emerg Crit Care (San Antonio) 2010;20:298–302.

295. Karlsson S, Pettilla V, Tenhunen J, et al. HMGB1 as a predictor of organ dysfunction and outcome in patients with severe sepsis. Intensive Care Med 2008;34:1046–1053.

296. de Jong HK, van der Poll T, Wiersinga WJ. The systemic pro-inflammatory response in sepsis. J Innate Immun 2010;2:422–430.

297. Cray C, Ziajas J, Altman NH. Acute phase response in animals: A review. Comp Med 2009;59:517–526.

298. Mohmage H. Cytokines and the hepatic acute phase response. J Pathol 1997;181:257–266.

299. Gruys E, Toussaint MJ, Niewold TA, et al. Acute phase reaction and acute phase proteins. J Zhejiang Univ Sci B 2005;6:1045–1056.

300. Cucilli F, Giordano A, Spagnolo V. The systemic reaction during inflammation: The acute-phase proteins. Protein Pept Lett 2002;9:211–223.

301. Noursadeghi M, Pepys MB, Gallimore R, et al. Relationship of granulocyte colony stimulating factor with other acute phase reactants in man. Clin Exp Immunol 2005;140:97–100.
302. Ceron JJ, Eckersall PD, Martynez-Subiela S. Acute phase proteins in dogs and cats: Current knowledge and future perspectives. Vet Clin Pathol 2005;34:85–99.

303. Eckersall PD, Bell R. Acute phase proteins: Biomarkers of infection and inflammation in veterinary medicine. Vet J 2010;185:23–27.

304. Kjelgaard-Hansen M, Jacobsen S. Assay validation and diagnostic applications of major acute-phase protein testing in companion animals. Clin Lab Med 2011;31:51–70.

305. Tvarijonaviciute A, Eraul O, Kocaturk M, et al. Adiponectin and IGF-1 are negative acute phase proteins in a dog model of acute endotoxaemia. Vet Immunol Immunopathol 2011;140:147–151.

306. Castelli GP, Pognani C, Meisner M, et al. Procalcitonin and C-reactive protein during systemic inflammatory response syndrome, sepsis and organ dysfunction. Crit Care 2004;8:R234–R242.

307. Harbarth S, Holeckova K, Froidevaux C, et al. Diagnostic value of procalcitonin, interleukin-6, and interleukin-8 in critically ill patients admitted with suspected sepsis. Am J Respir Crit Care Med 2001;164:396–402.

308. Kuzi S, Aroch I, Peleg K, et al. Canine procalcitonin messenger RNA expression. J Vet Diagn Invest 2008;20:629–633.

309. Giunti M, Peli A, Battilani M, et al. Evaluation of CALC-I gene (CALCA) expression in tissues of dogs with signs of the systemic inflammatory response syndrome. J Vet Emerg Crit Care (San Antonio) 2010;20:523–527.

310. Mukorera V, Dvir E, van der Merwe LL, et al. Serum C-reactive protein concentration in benign and malignant canine spiked cancer. J Vet Intern Med 2011;25:963–966.

311. Koster LS, Van Schoor M, Goddard A, et al. C-reactive protein in canine babesiosis caused by Babesia rossi and its association with outcome. J S Afr Vet Assoc 2009;80:87–91.

312. Sheahan D, Bell R, Mellanby RJ, et al. Acute phase protein concentrations in dogs with nasal disease. Vet Rec 2010;167:895–899.

313. Lowrie M, Penderis J, Eckersall PD, et al. The role of acute phase proteins in diagnosis and management of steroid-responsive meningitis arteritis in dogs. Vet J 2009;189:125–130.

314. Griebsch C, Arndt G, Raila J, et al. C-reactive protein in dogs with primary immune-mediated hemolytic anemia. Vet Clin Pathol 2009;38:421–425.

315. Mitchell KD, Kruth SA, Wood RD, et al. Serum acute phase protein concentrations in dogs with autoimmune hemolytic anemia. J Vet Intern Med 2009;23:585–591.

316. Bathen-Noethen A, Carlson R, Menzel D, et al. Concentrations of acute-phase proteins in dogs with steroid responsive meningitis-arteritis. J Vet Intern Med 2008;22:1149–1156.

317. Nakamura M, Takahashi M, Ohno K, et al. C-reactive protein concentration in dogs with various diseases. J Vet Med Sci 2008;70:127–131.

318. Ohno K, Yokoyama Y, Nakashima K, et al. C-reactive protein concentration in canine idiopathic polycythemia. J Vet Med Sci 2006;68:1275–1279.

319. Dabrowski R, Wawron W, Kostro K. Changes in CRP, SAA and haptoglobin produced in response to ovariohysterectomy in healthy bitches and those with pyometra. Theriogenology 2007;67:321–327.

320. Planellas M, Bassols A, Siracusa C, et al. Evaluation of serum haptoglobin and C-reactive protein in dogs with mammary tumors. Vet Clin Pathol 2009;38:348–352.

321. Nielsen L, Toft N, Eckersall PD, et al. Serum C-reactive protein concentration as an indicator of remission status in dogs with multicentric lymphoma. J Vet Intern Med 2007;21:1231–1236.

322. Caldin M, Tasca S, Carli E, et al. Serum acute phase protein concentrations in dogs with hyperadrenocorticism with and without concurrent inflammatory conditions. Vet Clin Pathol 2009;38:63–68.

323. Chan DL, Rozanski EA, Freeman LM. Relationship among plasma amino acids, C-reactive protein, illness severity, and outcome in critically ill dogs. J Vet Intern Med 2009;23:559–563.

324. Gebhardt C, Hirschberger J, Rau S, et al. Use of C-reactive protein to predict outcome in dogs with systemic inflammatory response syndrome or sepsis. J Vet Emerg Crit Care (San Antonio) 2009;19:450–458.

325. van der Poll T, de Boer JD, Levi M. The effect of inflammation on coagulation and vice versa. Curr Opin Infect Dis 2011;24:273–278.

326. Machado FR, Cesar MS. Sepsis, coagulation and anticoagulants. Endocr Metab Immune Disord Drug Targets 2010;10:204–213.

327. Sun H. The interaction between pathogens and the host coagulation system. Physiology (Bethesda) 2006;21:281–288.

328. Huisse MG, Pease S, Hurtado-Nedelec M, et al. Leukocyte activation: The link between inflammation and coagulation during seastroke. A study of patients during the 2003 heat wave in Paris. Crit Care Med 2008;36:2288–2295.

329. de Laforcade AM, Freeman LM, Shaw SP, et al. Hemostatic changes in dogs with naturally occurring sepsis. J Vet Intern Med 2003;17:674–679.

330. Benzt AI, Palmer JE, Dullap BL, et al. Prospective evaluation of coagulation in critically ill neonatal foals. J Vet Intern Med 2009;23:161–167.

331. Estrin MA, Wehausen CE, Jessen CR, et al. Disseminated intravascular coagulation in cats. J Vet Intern Med 2006;20:1334–1339.

332. Norris CR, Griffey SM, Samii VF. Pulmonary thromboembolism in cats: 29 cases (1987–1997). J Am Vet Med Assoc 1999;215:1650–1654.

333. Lucas WE, Kitzmiller JL. The role of intravascular coagulation in feline endotoxin shock. Surg Gynecol Obstet 1972;134:73–77.

334. DeClue AE, Williams KJ, Sharp C, et al. Systemic response to low-dose endotoxin infusion in cats. Vet Immunol Immunopathol 2009;132:167–174.

335. DelGiudice LA, White GA. The role of tissue factor and tissue factor pathway inhibitor in health and disease states. J Vet Emerg Crit Care (San Antonio) 2009;19:23–29.

336. Dullap Schauer BL, Epstein K. Coagulopathy of the critically ill equine patient. J Vet Emerg Crit Care (San Antonio) 2009;19:53–65.

337. Hopper K, Bateman S. An updated view of hemostasis: Mechanisms of hemostatic dysfunction associated with sepsis. J Vet Emerg Crit Car 2005;15:83–91.

338. Pawlinski R, Mackman N. Cellular sources of tissue factor in endothoxemia and sepsis. Thromb Res 2010;125(Suppl 1):S70–S73.

339. Stokol T, Daddona JL, Choi B. Evaluation of tissue factor procoagulant activity on the surface of feline leukocytes in response to treatment with lipopolysaccharide and heat-inactivated fetal bovine serum. Am J Vet Res 2010;71:623–629.

340. Grunig G, Hulliger C, Winder C, et al. Spontaneous and lipopolysaccharide-induced expression of procoagulant activity by equine lung macrophages in comparison with blood monocytes and blood neutrophils. Vet Immunol Immunopathol 1991;29:295–312.

341. Ouellette AL, Evans RJ, Heath MF. Platelets enhance endotoxin-induced monocyte tissue factor (TF) activity in the horse. Res Vet Sci 2004;76:31–35.

342. Henry MM, Moore JN. Clinical relevance of monocyte procoagulant activity in horses with colic. J Am Vet Med Assoc 1991;198:843–848.
phase of severe sepsis: Longitudinal study of mononuclear histocompatibility leukocyte antigen-DR expression, procollacitin, C-reactive protein, and changes in T-cell subsets in septic and post-operative patients. Crit Care Med 2002;30:1015–1023.

385. Wesche DE, Lomas-Neira JL, Perl M, et al. Leukocyte apoptosis and its significance in sepsis and shock. J Leukoc Biol 2005;78:325–337.

386. Hsieh YC, Athar M, Chaudry IH. When apoptosis meets autophagy: Deciding cell fate after trauma and sepsis. Trends Mol Med 2009;15:129–138.

387. Pinheiro da Silva F, Nizet V. Cell death during sepsis: Integration of disintegration in the inflammatory response to overwhelming infection. Apoptosis 2009;14:509–521.

388. Levine B, Mizushima N, Virgin HW. Autophagy in immunity and inflammation. Nature 2011;469:323–335.

389. Giansanti V, Torriglia A, Scovassi AI. Conversation between apoptosis and autophagy: “Is it your turn or mine?” Apoptosis 2011;16:321–333.

390. Watanabe E, Muenzer JT, Hawkins WG, et al. Sepsis induces extensive autophagic vacuolization in hepatocytes: A clinical and laboratory-based study. Lab Invest 2009;89:549–561.

391. Bergsaken T, Fink SL, Cookson BT. Pyroptosis: Host cell death and inflammation. Nat Rev Microbiol 2009;7:99–109.

392. Kepp O, Galluzzo L, Zitvogel L, et al. Pyroptosis: A cell death modality of its kind? Eur J Immunol 2010;40:627–630.

393. Fink SL, Cookson BT. Apoptosis, pyroptosis, and necrosis: mechanistic description of death and dying eukaryotic cells. Infect Immun 2005;73:1907–1916.

394. Shao W, Yeretsian G, Doirion K, et al. The caspase-1 digestive identifies the glycolysis pathway as a target during infection and septic shock. J Biol Chem 2007;282:36321–36329.

395. Nakajima Y, Mikami O, Yoshioka M, et al. Involvement of apoptosis in the endotoxemic lesions of the liver and kidneys of piglets. J Vet Med Sci 2000;62:621–626.

396. Sato M, Mikami O, Kobayashi M, et al. Apoptosis in the lymphatic organs of piglets inoculated with classical swine fever virus. Vet Microbiol 2000;75:1–9.

397. Messmer UK, Briner VA, Pfieischffer J. Tumor necrosis factor-alpha and lipopolysaccharide induce apoptotic cell death in bovine glomerular endothelial cells. Kidney Int 1999;55:2322–2337.

398. Messmer UK, Winkel G, Briner VA, et al. Glucocorticoids potently block tumour necrosis factor-alpha- and lipopolysaccharide-induced apoptotic cell death in bovine glomerular endothelial cell upstream of caspase 3 activation. Br J Pharmacol 1999;127:1623–1640.

399. Sylte MJ, Leite FP, Kuckleburg CJ, et al. Caspase activation during Haemophilus somnis lipooligosaccharide-mediated apoptosis of bovine endothelial cells. Microb Pathog 2003;35:285–291.

400. Kuckleburg CJ, Tiwari R, Czuprynski CJ. Endothelial cell apoptosis induced by bacteria-activated platelets requires caspase-8 and -9 and generation of reactive oxygen species. Thromb Haemost 2008;99:363–372.

401. Wohlschlaeger J, Stubbe HD, Schmitz KJ, et al. Roles of MMP-2/-9 in cardiac dysfunction during early multiple organ failure in an ovine animal model. Pathol Res Pract 2005;201:809–817.

402. Umeshappa CS, Singh KP, Nanjundappa RH, et al. Roles of Haemophilus somnus in programmable death-1 (PD-1). Vet Immunol Immunopathol 2011;143:307–313.

403. Crouser ED, Julian MW, Weinstein DM, et al. Fluid replacement with hypertonic or isotonic solutions guided by mixed venous oxygen saturation in experimental hypodynamic sepsis. J Trauma 2009;67:1205–1212.

404. Rahal L, Garrido AG, Cruz RJ Jr, et al. Fluid replacement with hypertonic or isotonic solutions guided by mixed venous oxygen saturation in experimental hypodynamic sepsis. J Trauma 2009;67:1205–1212.

405. Collins AM, Sewell WA, Edwards MR. Immunoglobulin gene rearrangement, repertoire diversity, and the allergic response. Pharmacol Ther 2003;100:157–170.

406. Nikoel-Zugich J, Sliata MK, Messaudoi I. The many important facets of T-cell repertoire diversity. Nat Rev Immunol 2004;4:123–132.

407. Hollander GA, Peterson P. Learning to be tolerant: How T cells keep out of trouble. J Intern Med 2009;265:541–561.

408. McCaughtry TM, Hoggquist KA. Central tolerance: What have we learned from mice? Semin Immunopathol 2008;30:399–409.

409. Parish IA, Heath WR. Too dangerous to ignore: Self-tolerance and the control of ignorant autoreactive T cells. Immunol Cell Biol 2008;86:146–152.

410. Waldmann H. Tolerance: An overview and perspectives. Nat Rev Nephrol 2010;6:569–576.

411. Mueller DL. Mechanisms maintaining peripheral tolerance. Nat Immunol 2010;11:21–27.

412. Miyara M, Sakaguchi S. Human FoxP3+CD4 regulatory T cells: their knowns and unknowns. Immunol Cell Biol 2011;89:346–351.

413. Sakaguchi S. Regulatory T cells: History and perspective. Methods Mol Biol 2011;707:3–17.

414. Shevach EM. The resurrection of T cell-mediated suppression. J Immunol 2011;186:3805–3807.

415. Garden OA, Pinheiro D, Cunningham F. All creatures great and small: Regulatory T cells in mice, humans, dogs and other domestic animal species. Int Immunopharmacol 2011;11:576–588.

416. Lankford S, Petty C, LaVoy A, et al. Cloning of feline FOXP3 and detection of expression in CD4+CD25+ regulatory T cells. Vet Immunol Immunopathol 2008;122:159–166.

417. Petty CS, Tompkins MB, Tompkins WA. Transforming growth factor-beta/transforming growth factor-betaRII signaling may regulate CD4+CD25+ T regulatory-cell homeostasis and suppressor function in feline AIDS lentivirus infection. J Acquir Immune Defic Syndr 2008;47:148–160.

418. Smithberg SR, Fogle JE, Mexas AM, et al. In vivo depletion of CD4+CD25+ regulatory T cells in cats. J Immunol Methods 2008;329:81–91.

419. Vahlenkamp TW, Tompkins MB, Tompkins WA. Feline immunodeficiency virus infection phenotypically and functionally activates immunosuppressive CD4+CD25+ T regulatory cells. J Immunol 2004;172:4752–4761.

420. Fogle JE, Tompkins WA, Tompkins WA. Feline immunodeficiency virus infection phenotypically and functionally activates immunosuppressive CD4+CD25+ T regulatory cells. J Immunol 2004;172:4752–4761.

421. Fogle JE, Tompkins WA, Tompkins MB. CD4+CD25+ T regulatory cells from FIV+ cats induce a unique anergic profile in CD8+ lymphocyte targets. Retrovirology 2010;7:97.

422. Fogle JE, Mexas AM, Tompkins WA, et al. CD4+CD25+ T regulatory cells inhibit CDS' IFN-gamma production during acute and chronic FIV infection utilizing a membrane TGF-beta-dependent mechanism. AIDS Res Hum Retroviruses 2010:26:201–216.

423. Achleiter A, Clark ME, Biezdle D. T-regulatory cells infected with feline immunodeficiency virus up-regulate programmed death-1 (PD-1). Vet Immunol Immunopathol 2011;143:307–313.

424. Miller BJ, Elmslie RE, Burnett RC, et al. Use of FoxP3 expression to identify regulatory T cells in healthy dogs and dogs with cancer. Vet Immunol Immunopathol 2007;116:69–78.

425. Keppel KE, Campbell KL, Zuckermand P, et al. Quantitation of canine regulatory T cell populations, serum interleukin-10 and allergen-specific IgE concentrations in healthy control dogs and canine atopic dermatitis patients receiving allergen-specific immunotherapy. Vet Immunol Immunopathol 2008;123:337–344.
426. O’Neill K, Guth A, Biller B, et al. Changes in regulatory T cells in dogs with cancer and associations with tumor type. J Vet Intern Med 2009;23:875–881.
427. Horiiuchi Y, Tominaga M, Ichikawa M, et al. Increase of regulatory T cells in the peripheral blood of dogs with metastatic tumors. Microbiol Immunol 2009;53:468–474.
428. Mizuno T, Suzuki R, Umeki S, et al. Crossreactivity of antibodies to canine CD25 and Foxp3 and identification of canine CD4+CD25+ Foxp3+ cells in canine peripheral blood. J Vet Med Sci 2009;71:1561–1568.
429. Abrams VK, Hwang B, Lesnikova M, et al. A novel monoclonal antibody specific for canine CD25 (PA1A10): Selection and evaluation of canine Tregs. Vet Immunol Immunopathol 2010;135:257–265.
430. Rissetto KC, Rindt H, Selting KA, et al. Cloning and expression of canine CD25 for validation of an anti-human CD25 antibody to compare T regulatory lymphocytes in healthy dogs and dogs with osteosarcoma. Vet Immunol Immunopathol 2010;135:137–145.
431. Horiiuchi Y, Tominaga M, Ichikawa M, et al. Relationship between regulatory and type 1 T cells in dogs with oral malignant melanoma. Microbiol Immunol 2010;54:152–159.
432. Pinheiro D, Singh Y, Grant CR, et al. Phenotypic and functional characterization of a CD4+CD25highFOXP3high regulatory T-cell population in the dog. Immunology 2011;132:111–122.
433. Burton JH, Mitchell L, Thamm DH, et al. Low-dose cyclophosphamide selectively decreases regulatory T cells and inhibits angiogenesis in dogs with soft tissue sarcoma. J Vet Intern Med 2011;25:920–926.
434. Kaser T, Gerner W, Hammer SE, et al. Detection of Foxp3 protein expression in porcine T lymphocytes. Vet Immunol Immunopathol 2008;125:92–101.
435. Kaser T, Gerner W, Hammer SE, et al. Phenotypic and functional characterisation of porcine CD4+CD25high regulatory T cells. Vet Immunol Immunopathol 2008;122:153–158.
436. Bolzer K, Kaser T, Saalmuller A, et al. Molecular characterisation of porcine Forkhead-box p3 (Foxp3). Vet Immunol Immunopathol 2009;132:275–281.
437. Wongyian P, Buranapraditkun S, Chokeshai-Usaha K, et al. Induction of inducible CD4+CD25+ Foxp3+ regulatory T lymphocytes by porcine reproductive and respiratory syndrome virus (PRRSV). Vet Immunol Immunopathol 2010;133:170–182.
438. Silva-Campa E, Flores-Mendoza L, Resendiz M, et al. Induction of T helper 3 regulatory cells by dendritic cells infected with porcine reproductive and respiratory syndrome virus. Virology 2009;387:373–379.
439. Dawson H, Solano-Aguilar G, Beal M, et al. Localized Th1-, Th2-, T regulatory cell-, and inflammation-associated hepatic and pulmonary immune responses in Ascariis suum-infected swine are increased by retinoic acid. Infect Immun 2009;77:2576–2587.
440. Pilon C, Meuners F, Dauba A, et al. Induction of porcine regulatory cells by mycophenolic acid-treated dendritic cells. Transplant Proc 2009;41:700–702.
441. Kaser T, Gerner W, Mair K, et al. Current knowledge on porcine regulatory T cells. Vet Immunol Immunopathol 2011;http://doi.org/10.1016/j.vetimm.2011.05.035 (in press).
442. Kaser T, Gerner W, Saalmuller A. Porcine regulatory T cells: Mechanisms and T-cell targets of suppression. Dev Comp Immunol 2011;35:1166–1172.
443. Gerner W, Stadler M, Hammer SE, et al. Sensitive detection of Foxp3 expression in bovine lymphocytes by flow cytometry. Vet Immunol Immunopathol 2010;138:154–158.
444. Hoek A, Rutten VP, Kool J, et al. Subpopulations of bovine WC1+ gammadelta T cells rather than CD4+CD25high Foxp3+ T cells act as immune regulatory cells ex vivo. Vet Res 2009;40:6.
445. Seo KS, Davis WC, Hamilton MJ, et al. Development of monoclonal antibodies to detect bovine FOXP3 in PBMCs exposed to a staphylococcal superantigen. Vet Immunol Immunopathol 2009;128:30–36.
446. de Almeida DE, Colvin CJ, Coussens PM. Antigen-specific regulatory T cells in bovine paratuberculosis. Vet Immunol Immunopathol 2008;125:234–245.
447. McNelly TN, McIntyre J, Frew D, et al. Infestation of sheep with Psoroptes ovis, the sheep scab mite, results in recruitment of Foxp3+ T cells into the dermis. Parasite Immunol 2010;32:361–369.
448. Rocchi MS, Wattegedera SR, Frew D, et al. Identification of CD4+(+)-CD25(high) Foxp3+(+) T cells in ovine peripheral blood. Vet Immunol Immunopathol 2011;144:172–177.
449. Wagner B, Hillegas JM, Brinker DR, et al. Characterization of monoclonal antibodies to equine interleukin-10 and detection of T regulatory 1 cells in horses. Vet Immunol Immunopathol 2008;122:57–64.
450. Hamza E, Torsteinssottir S, Eydal M, et al. Increased IL-4 and decreased regulatory cytokine production following relocation of Icelandic horses from a high to low endoparasite environment. Vet Immunol Immunopathol 2010;133:40–50.
451. Hamza E, Steinbach F, Marti E. CD4+CD25+ T cells expressing Foxp3 in Icelandic horses affected with insect bite hypersensitivity. Vet Immunol Immunopathol 2011;http://doi.org/10.1016/j.vetimm.2011.05.033 (in press).
452. Wagner B, Burton A, Ainsworth D. Interferon-gamma, interleukin-4 and interleukin-10 production by T helper cells reveals intact Th1 and regulatory TR1 cell activation and a delay of the Th2 cell response in equine neonates and foals. Vet Res 2010;41:47.
453. Heimann M, Janda J, Sigurdardottir OG, et al. Skin-infiltrating T cells and cytokine expression in Icelandic horses affected with insect bite hypersensitivity: A possible role for regulatory T cells. Vet Immunol Immunopathol 2011;140:63–74.
454. Robbin MG, Wagner B, Noronha LE, et al. Subpopulations of equine blood lymphocytes expressing regulatory T cell markers. Vet Immunol Immunopathol 2011;140:90–101.
455. Langier S, Sade K, Kivity S. Regulatory T cells: The suppressor arm of the immune system. Autoimmun Rev 2010;10:112–115.
456. Sakaguchi S, Wing K, Onishi Y, et al. Regulatory T cells: How do they suppress immune responses? Int Immunol 2009;21:1105–1111.
457. Workman CJ, Szymczak-Workman AL, Collison LW, et al. The development and function of regulatory T cells. Cell Mol Life Sci 2009;66:2603–2622.
458. Rudensky AY. Regulatory T cells and Foxp3. Immunol Rev 2011;241:260–268.
459. Campbell DJ, Ziegler SF. FOXP3 modifies the phenotypic and functional properties of regulatory T cells. Nat Rev Immunol 2007;7:305–310.
460. Shalev I, Schmelze M, Robson SC, et al. Making sense of regulatory T cell suppressive function. Semin Immunol 2011;23:282–292.
461. Wan YY. Regulatory T cells: Immune suppression and beyond. Cell Mol Immunol 2010;7:204–210.
462. Caramalho I, Lopes-Carvalho T, Ostler D, et al. Regulatory T cells selectively express Toll-like receptors and are activated by lipopolysaccharide. J Exp Med 2003;197:403–411.
463. Dai J, Liu B, Li Z. Regulatory T cells and Toll-like receptors: What is the missing link? Int Immunopharmacol 2009;9:528–533.
464. Liu H, Komai-Koma M, Xu D, et al. Toll-like receptor 2 signaling modulates the functions of CD4+CD25+ regulatory T cells. Proc Natl Acad Sci USA 2006;103:7048–7053.
465. Nyirenda MH, O’Brien K, Sanvito L, et al. Modulation of regulatory T cells in health and disease: Role of toll-like receptors. Inflamm Allergy Drug Targets 2009;8:124–129.

466. Monneret G, Debard AL, Venet F, et al. Marked elevation of human circulating CD4+CD25+ regulatory T cells in sepsis-induced immunoparalysis. Crit Care Med 2003;31:2068–2071.

467. Saito K, Wagatsuma T, Toyama H, et al. Sepsis is characterized by the increases in percentages of circulating CD4+CD25+ regulatory T cells and plasma levels of soluble CD25. Tohoku J Exp Med 2008;216:61–68.

468. Venet F, Chung CS, Kherouf H, et al. Increased circulating regulatory T cells (CD4+CD25+CD127+) contribute to lymphocyte anergy in septic shock patients. Intensive Care Med 2009;35:678–686.

469. Venet F, Pachot A, Debard AL, et al. Increased percentage of CD4+CD25+ regulatory T cells during septic shock is due to the decrease of CD4+CD25- lymphocytes. Crit Care Med 2004;32:2329–2331.

470. Heuer JG, Zhang T, Zhao J, et al. Adoptive transfer of in vitro-stimulated CD4+CD25+ regulatory T cells increases bacterial clearance and improves survival in polymicrobial sepsis. J Immunol 2005;174:7141–7146.

471. Scumpia PO, Delano MJ, Kelly KM, et al. Increased natural CD4+CD25+ regulatory T cells and their suppressor activity do not contribute to mortality in murine polymicrobial sepsis. J Immunol 2006;177:7943–7949.

472. Wisnosi N, Chung CS, Chen Y, et al. The contribution of CD4+CD25+ T-regulatory cells to immune suppression in sepsis. Shock 2007;27:251–257.

473. Tiemessen MM, Jagger AL, Evans HG, et al. CD4+CD25+Foxp3+ regulatory T cells induce alternative activation of human monocytes/macrophages. Proc Natl Acad Sci USA 2007;104:19446–19451.

474. Venet F, Pachot A, Debard AL, et al. Human CD4+CD25+ regulatory T lymphocytes inhibit lipopolysaccharide-induced monocyte survival through a Fas/Fas ligand-dependent mechanism. J Immunol 2006;177:6540–6547.

475. Hodge G, Scott J, Osborn M, et al. Increased T regulatory cells and decreased Th1 pro-inflammatory cytokines correlate with culture-positive infection in febrile neutropenia childhood oncology patients. Cytokine 2011;53:286–288.

476. Huang LF, Yao YM, Dong N, et al. Association between regulatory T cell activity and sepsis and outcome of severely burned patients: a prospective, observational study. Crit Care 2010;14:R3.

477. Hein F, Massin F, Cravoisy-Popovic A, et al. The relationship between CD4+CD25+CD127- regulatory T cells and inflammatory response and outcome during shock states. Crit Care 2010;14:R19.

478. van Maren WW, Jacobs JF, de Vries IJ, et al. Toll-like receptor signalling on Tregs: To suppress or not to suppress? Immunology 2008;124:445–452.

479. Walker LS. Regulatory T cells overturned: The effectors fight back. Immunology 2009;126:466–474.

480. Marshall JC. Sepsis: Rethinking the approach to clinical research. J Leukoc Biol 2008;83:471–482.

481. Girbes AR, Beishuizen A, Strack van Schijndel RJ. Pharmacological treatment of sepsis. Fundam Clin Pharmacol 2008;22:355–361.

482. Wittebole X, Collienne C, Castanares-Zapatero D, et al. Adjunctive therapies for severe sepsis. Int J Antimicrob Agents 2008;32(Suppl 1):S34–S38.