[Frontiers in Bioscience 14, 2970-2982, January 1, 2009]

Innate immunity, coagulation and surgery

Alexander Koch[1], Paula Zacharowski[1], Olaf Boehm[2], Kai Zacharowski[1]

[1]Molecular Cardioprotection and Inflammation Group, University Hospitals Bristol NHS Foundation Trust, Bristol, BS2 8HW, United Kingdom, [2]Anesthesiology and Intensive Care Medicine, University Hospital Bonn, Bonn, Germany

TABLE OF CONTENTS

1. ABSTRACT

Inflammation is the host's defense mechanism to infection or trauma including surgical procedures. In the clinic, non-infectious inflammation plays an important part in cardiology (e.g. Percutaneous transluminal coronary angioplasty, PTCA), intensive care medicine (e.g. polytrauma), cardiac (e.g. extracorporeal circulation) and vascular surgery (e.g. reperfusion injury). An imbalance of the inflammatory response can cause an acute condition like sepsis or long-term Cardiovascular disease (CVD), both of which are leading killers in the Western world. Alterations in coagulation, innate immunity and endothelial function represent key aspects in the mechanism of inflammation and are the link between the pathogenesis of these two diseases. Studying inflammatory pathways or targeting specific mediators during inflammation may help to develop strategies to improve the clinical outcome of patients undergoing major surgery, where postoperative inflammation plays a crucial role.

2. GENERAL INTRODUCTION

Cardiovascular disease (CVD) is a growing health problem with high economic costs to run cardiac intensive care units (1), while sepsis and septic shock are the most frequent cause of death in non-cardiac intensive care units (2). These conditions have more in common than just leading statistics, cardiovascular disease and sepsis are both a result of inflammation (3-4), where coagulation (5-6), innate immunity as well as endothelial dysfunction are underlying factors in their pathogenesis (4,7) (Figure 1). Inflammation can be classified into acute and chronic responses. Chronic inflammation can cause coronary heart disease, while acute inflammation can result in e.g. bacterial infection leading to sepsis. Acute inflammation is also triggered and cascaded by surgery, burns and tissue injury. It is characterized by vasodilatation of blood vessels, edema due to exudation of plasma proteins, leukocyte infiltration and adherence to the endothelium and the release of chemotactic mediators. Local inflammation often results

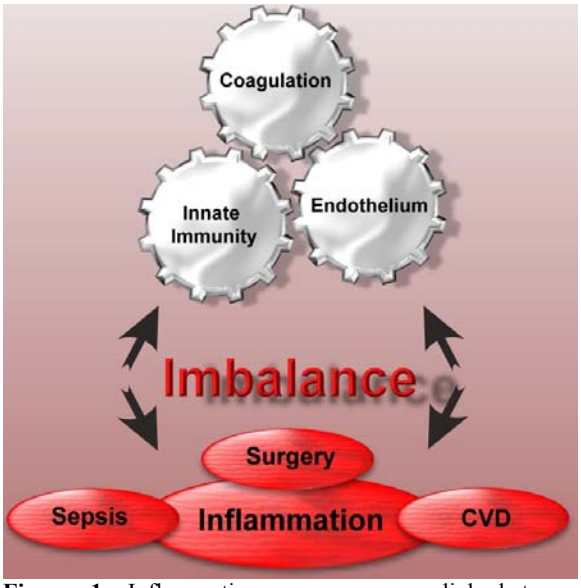

Figure 1. Inflammation – a common link between cardiovascular disease, sepsis and surgery. Under normal physiological conditions, coagulation, the endothelium and innate immunity work in synergy to maintain a functional balance. An imbalance of one system triggers dysfunction of the other systems leading to acute (e.g. sepsis, surgery) or chronic inflammation (e.g. CVD, cardiovascular disease).

in systemic inflammation (acute phase response), where cytokines are synthesized by inflammatory cells, the Hypothalamic-pituitary-adrenal (HPA)-axis is activated (releasing Adrenocorticotropic hormone (ACTH) and glucocorticoids) and acute phase proteins e.g. C-reactive protein are being produced. This review focuses on different aspects of inflammation, i.e. innate immunity and coagulation and their role during surgery.

3. INNATE IMMUNITY

3.1. Toll-like receptor (TLR)-induced signal transduction

In 1997, a human homologue of the *Drosophila* Toll gene and its encoded protein(s) were identified. These proteins were designated as Toll-like receptors (TLRs) and play a significant role in innate immunity. Ten human, and thirteen murine TLRs have been identified, some of which form homodimers or heterodimers (e.g. TLR2/1 or 2/6) (8-9). This family of type I transmembrane receptors is characterized by an extracellular domain with leucine-rich repeats and a cytoplasmatic domain with homology to the type I Interleukin (IL)-1 receptor. Upon activation, the signaling pathway involves several steps, finally resulting in the translocation of Nuclear factor (NF)-κB to the nucleus and the induction of gene transcription for cytokines and effector proteins (10) (Table 1).

3.2. Activation of TLRs by bacterial wall fragments

Inflammatory responses induced by cell wall components and nucleic acids of microorganisms are particularly initiated by the family of TLRs (11-12). Different TLRs are organized as receptor complexes, which sense most of the cell wall components spanning the diverse microbial world (9). TLR2 has been implicated in the signaling process of Gram-positive bacteria (e.g. *Staphylococcus aureus*) (13), while TLR4 is the main receptor for Gram-negative bacteria (e.g. *Escherichia coli*) (14). However, TLR2 detects also LPS and lipoproteins of certain Gram-negative bacteria such as *Helicobacter pylori* and *Francisella tularensis* (15-16). The wall of Gram-negative bacteria contains Lipopolysaccharide (LPS) or endotoxin, which is composed of the lipid A domain, an inner core and outer core oligosaccharide and, the *O*-antigenic polysaccharide domain (17-18). LPS is responsible for the initiation of Gram-negative shock, where pro-inflammatory mediators such as IL-1, IL-6, IL-8 and Tumor necrosis factor (TNF)-α are released by macrophages, monocytes and other cell types (19). In contrast, Gram-positive bacteria such as *Staphylococcus aureus* can cause multiple organ failure and septic shock without causing endotoxemia (20). The cell wall of Gram-positive bacteria contains lipoteichoic acid and peptidoglycan, which is a macroamphiphile (equivalent to LPS in Gram-negative bacteria) containing a substituted poly- (glycero-phosphate) backbone attached to a glycolipid (21). Peptidoglycan is a large polymer, which provides stress resistance and shape determining properties to bacterial cell walls. *In vivo*, lipoteichoic acid and peptidoglycan act in synergy to release cytokines such as TNF-α and Interferon (IFN)-γ and inducing Inducible nitric oxide synthase (iNOS) resulting in shock and multiple organ failure (22).

LPS-induced host cell activation mediates the release of NF-κB, which regulates the expression of diverse genes including cytokines, chemokines and enzymes like Hemeoxygenase (HO)-1 and iNOS, as well as other biological responses (23-25). HO-1 is an enzyme system, catalyzing the rate-limiting conversion of heme to iron, carbon monoxide and biliverdin (converted to bilirubin), whereas iNOS produces Nitric oxide (NO). In stress situations, HO-1 is thought to be protective because of its ability to form the antioxidant agent bilirubin and the vasoactive gas carbon monoxide (26-27). In contrast, copious amounts of NO generated by iNOS have been associated with the pathophysiological features of septic shock, i.e. myocardial dysfunction and multiple organ failure (28-29). Besides extensive formation of NO, endotoxemia causes the generation of large quantities of oxygen free radicals such as hydrogen peroxide and superoxide anions (30). Simultaneously released superoxide anions and NO may yield the production of reactive NO species such as peroxynitrite, which is more potent in causing cell injury than its parent molecules (31). Interestingly, time- and concentration dependent NO can also enfold protective characteristics (32-33), whereas its ability to induce HO-1 might account to this protection (34). In rat lungs, iNOS derived NO induces HO-1 leading to lung protection during a subsequent LPS challenge (35).

3.3. TLRs and the Hypothalamic-pituitary-adrenal(HPA)-axis

The HPA-axis plays an important role in the pathogenesis and course of inflammatory diseases (36). In

Table 1. List of known toll-like receptors (TLR) and examples of agonists

	Agonist	**Species**
TLR1	Triacyl lipopeptides (in association with TLR2)	Bacteria and mycobacteria
	HCV core, NS3 (in association with TLR2)	HCV
	HCMV envelope gp B and gp H (in association with TLR2)	HCMV
	SAA (in association with TLR2)	Host
	hBD-3 (in association with TLR2)	Host
TLR2	Triacyl lipopeptides (in association with TLR1)	Bacteria and mycobacteria
	Diacyl lipopeptides and LTA (in association with TLR6)	Mycoplasma and Gram-positive bacteria
	PG	Gram-positive bacteria
	LCOS 1013	Klebsellia pneumoniae
	Porins	Neisseria
	Lipoarabinomannan	Mycobacteria
	Zymosan (in association with TLR6)	Saccharomyces cerevisiae
	Phospholipomannan	Candida albicans
	Mannan	Candida albicans
	Glucuronoxylomannan	Cryptococcus neoformans
	tGPI-mutin	Trypanosoma
	ND	Encephalitozoon cuniculi
	ND	German cockroach feces
	Yps3p	Histoplasma capsulatum
	Hemagglutinin protein	Measles virus
	HCMV envelope gp B and gp H (in association with TLR1)	HCMV
	ND	HSV1
	HCV core, NS3 (in association with TLR1 and TLR6)	HCV
	SAA (in association with TLR1)	Host
	hBD-3 (in association with TLR1)	Host
	LMW-HA	Host
	amyloid β	Host
TLR3	dsRNA	Viruses
TLR4	LPS	Gram-negative bacteria
	ND	Fusarium oxysporum
	Glucuronoxylomannan	Cryptococcus neoformans
	Glycoinositolphospholipids	Trypanosoma
	Envelope proteins	RSV, MMTV
	Heat-shock protein 60, 70	Host
	Fibrinogen	Host
	LMW-HA	Host
TLR5	Flagellin	Flagellated bacteria
TLR6	Diacyl lipopeptides and LTA (in association with TLR2)	Mycoplasma and Gram-positive bacteria
	HCV core, NS3 (in association with TLR2)	HCV
	Zymosan	Saccharomyces cerevisiae
TLR7	ssRNA	RNA viruses
TLR8	ssRNA	RNA viruses
TLR9	CpG-DNA	Bacteria and mycobacteria
TLR10	ND	ND
TLR11	Uropathogenic bacteria	ND
	Profilin-like molecule	Toxoplasma gondii
TLR12	ND	ND
TLR13	ND	ND

HCV: hepatitis C virus; HCMV: human cytomegalovirus; SAA: serum amyloid A; hBD-3: human beta-defensin-3; LTA: lipoteichoic acid; PG: peptidoglycan; ND: not determined; HSV1: herpes simplex virus 1; LMW-HA: low molecular weight - hyaluronic acid; LPS: lipopolysaccharide; RSV: respiratory syncytial virus; MMTV: mouse mammary tumor virus (9, 119-134).

rats with experimental allergic encephalomyelitis, an adrenalectomy leads to a chronic active disease and, glucocorticoid replacement promotes recovery from this disease (37). Similarly, an adrenalectomy produces severe inflammation in experimental models of acute pancreatitis. Hydrocortisone replacement decreases both the severity and mortality of this disease (38). Peripheral cytokines such as TNF-α, IL-1β and IL-6 modulate the activity of the HPA-axis (39) and are one of the end effectors of this axis. Cortisol and corticosterone, regulate innate and T-cell specific immune responses (40-41). Major inflammatory and infectious diseases are associated with excess glucocorticoid secretion. Several reports highlight the importance of an intact adrenal stress response to infection (36,42-43) and there is good evidence to suggest that impaired innate immunity mediated by TLRs is involved in the pathology of sepsis and cardiovascular disease (44-46). For both, TLR2 and TLR4 knock-out mice we have demonstrated a from Wild-type (WT) mice different morphology of adrenal glands. Basal corticosterone and ACTH plasma concentrations differed also from WT mice. Induction of systemic inflammation resulted in an impaired stress response in both, TLR2 and TLR4 knock-out mice reflected by reduced corticosterone and ACTH concentrations and altered release of pro-inflammatory cytokines (47-48). Recently, we have described the expression of TLR2 and TLR4 in human adrenals (49).

Critically ill patients with sepsis have increased levels of plasma cortisol with relatively normal

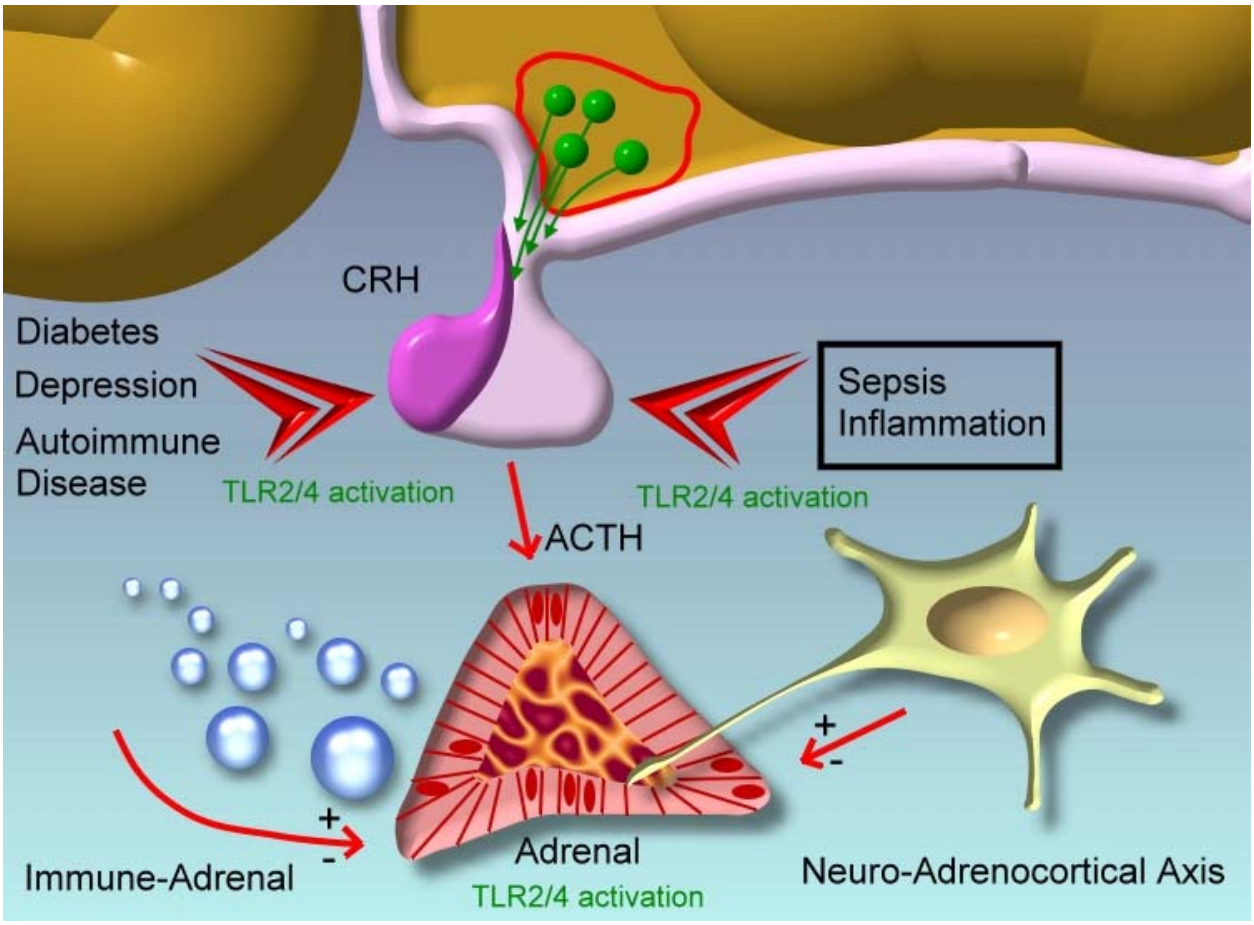

Figure 2. A schematic illustrating the hypothalamic-pituitary-adrenal(HPA)-axis under conditions of stress. Interactions of the immune system involving exogenous and endogenous stimuli at the levels of the hypothalamus, pituitary, and adrenal gland. There is strong evidence that Toll-like receptors (TLR2) and TLR4 are involved in the regulation of the HPA axis. CRH: Corticotropin-Releasing-Hormon; ACTH: Corticotropin-Releasing-Hormon.

concentrations of ACTH (42). Thus, the activation of the adrenal glands in response to inflammation is an important component of the host's anti-inflammatory response. The immune system stimulates the secretion of Corticosteroid-releasing hormone (CRH) and ACTH, and can therefore affect the activity of the HPA-axis. Furthermore, the adrenal gland might be stimulated by inflammatory mediators directly (Figure 2). During chronic stress, high levels of inflammatory cytokines may be required to maintain increased glucocorticoid levels, allowing a shift away from androgen and estrogen synthesis towards increased cortisol production. This shift may be beneficial during sepsis or other severe illnesses; however, it could also contribute to tumor formation in endocrine glands and be a risk factor for patients with arteriosclerosis, diabetes, depression and autoimmune diseases. Relative adrenal insufficiency is a complication in a substantial number of patients with sepsis and is attributable to a high mortality rate. During septic shock or acute respiratory distress syndrome, replacement therapy with low-dose hydrocortisone showed improved survival (50-52). However, a recently published multicenter, randomized, double-blind, placebo-controlled trial suggests that hydrocortisone does not improve survival in patients with septic shock (53). Clinical trials are warranted to study the effects of genetic polymorphisms (e.g. TLRs) on the HPA-axis. This might help us to understand more about the cause of inconsistent effects of systemic inflammation on the HPA-axis in individual patients.

3.4. TLRs in surgery

Colorectal surgery is often accompanied by the unwanted effect of anastomotic leakage (see review (54)). This complication leads to post-operative infection increasing morbidity and mortality. On the molecular level, leakage of numerous bacterial species and fungi from a bowel or colon source results in the release of bacterial wall-fragments and the activation of TLRs, leading to local (e.g. peritonitis) or even systemic inflammation (e.g. sepsis) (55). Under experimental settings, the Cecal ligation and puncture (CLP) model consists of ligation of the cecum followed by a small puncture. Feces leaks out of the bowel through the perforation, thereby mimicking anastomotic leakage. In mice within the first 3 hours after CLP surgery, mRNA and protein expression of TLR2 and TLR4 are up-regulated in the liver, lung and spleen (56). In addition,

sham-operated animals (laparotomy only) show also a transient increase in TLR2 and TLR4 mRNA and protein expression, demonstrating that even the stress of anesthesia and sham surgery results in inflammation. Similarly, in the lungs of mice following a *Fumigatus conidia* challenge, an observed increase in TLR2 mRNA expression was still evident some 15 days after CLP surgery (57).

Bacterial translocation is another undesired effect with an occurrence of about 15% in patients undergoing elective surgery (58-59). Enteric organisms translocate from gut across the intact intestinal mucosa to normally sterile conditions like the mesenteric lymph nodes and other internal organs. In addition, this route of passage also allows the transfer of endotoxins (60). Increasingly, reports have inferred a role for bacterial translocation in the morbidity of post-operative sepsis (61). As wall fragments of Gram-positive and Gram-negative bacteria activate TLRs, anastomotic leakage, bacterial translocation and endotoxin release could have implications on their activation and expression. However, the impact of systemic inflammation on TLR expression on leukocytes remains unclear. Up-regulation, no change and down-regulation has been demonstrated and seems to be influenced by time, the cause of systemic inflammation (SIRS, sepsis, septic shock), the type of TLR and leukocyte (62-66). After Coronary artery bypass grafting (CABG) with the use of Cardiopulmonary bypass (CPB), monocytes of patients demonstrate a down-regulation of TLR2 and TLR4 expression, although on day 2 post-surgery, monocytic expression of TLR2 and TLR4 is increased (67). In contrast, Peripheral blood mononuclear cell (PBMC)s of patients who underwent gastrointestinal surgery demonstrated a decrease in the expression of TLR2 and TLR4 initially after surgery. TLR4 expressions returned to preoperative levels within one week after surgery, whilst TLR2 expression took some two weeks to return to preoperative levels (68). The expression status of TLR2 or TLR4 might also have functional consequences. In a study, despite the expression of TLR4 being significantly higher in patients suffering with Systemic inflammatory response syndrome (SIRS) than with normal healthy volunteers, *ex vivo* production of TNF-α by LPS was reduced in blood from patients with SIRS, indicating LPS hyporesponsiveness (69). In another study involving patients with sepsis, severe sepsis and septic shock, no changes in TLR2 or TLR4 expression on PBMCs has been observed. However *ex vivo* stimulation demonstrated a decreased responsiveness of PBMCs to several stimuli (65).

TLR polymorphisms may influence the outcome of surgical procedures. For example renal transplant recipients with TLR4 polymorphism present a lower risk of post-transplant atherosclerotic events and acute allograft rejection, but experience severe infectious episodes more frequently (70). It is a well known fact that whereas some patients succumb to sepsis after surgery, others will recover uneventfully, although having received identical treatment for the same condition. In addition it is unclear why some patients recover from sepsis, whereas others succumb to multi-organ failure and finally death. In these clinical scenarios, it is likely that there are individual differences in the biological responses to SIRS or sepsis. Such differences may include molecules/receptors (e.g. TLRs, CD14, macrophage migration inhibitory factor), various cytokines, CD11b, DNA polymorphisms for genes of the coagulation system, eicosanoids, glucocorticoid secretion, free radicals generation and antioxidant status. The influence of TLR polymorphism on the perioperative phase of patients is still poorly understood. To date there are very few studies with large patient populations to gain further insights into this field. Furthermore, different types of single nucleotide polymorphism for some TLRs are described, which also needs consideration in future clinical trials (71).

4. COAGULATION SYSTEM

4.1. Hemostatis and surgery

The initiation of hemostasis occurs when the integrity of the vasculature is compromised. This process involves the interaction between blood vessel walls, platelets and the activation of a complex cascade of cellular components and soluble factors such coagulation and fibrinolytic factors. Rupture of a blood vessel results in exposure of the sub-endothelium and the initial phase of vasoconstriction to minimize blood loss at the site of injury. Circulating platelets adhere to collagen expressed on endothelial cells via interactions largely mediated with the plasma protein von Willebrand's factor (72). Platelets become activated through the binding of thrombin and they release several mediators such as ADP and thromboxane A2, which promote platelet interactions and aggregation (73). As a result a platelet plug is formed, providing a temporary measure to stop blood loss. Platelets also release a number of phospholipids and lipoproteins, which activate the coagulation cascade, which is sub-divided in to the intrinsic, extrinsic and common pathway. The extrinsic pathway is activated through Tissue factor (TF) expressed which promotes the generation of thrombin and consequently the conversion of fibrinogen to fibrin (74). The platelet plug is strengthened by the polymerization and stabilization of fibrin, forming a mesh or clot. The intrinsic pathway is usually activated in response to abnormalities in the vessel wall, although, this pathway converges with the extrinsic pathway to form a common pathway which results in clot formation. Activation of the fibrinolytic system occurs when injury to the vessel wall is complete and normal blood flow should be resumed. This is achieved by degradation of the fibrin clot. Inactive tissue plasminogen activator becomes activated on binding to fibrin and cleaves plasminogen to plasmin, which lyses clots, yielding soluble fibrin degradation products. The interactions of these fibrinolytic proteins are closely regulated by their respective inhibitors such as plasminogen activator inhibitor and α_1-antiplasmin (75).

During cardiac surgery and CPB, perioperative and postoperative bleeding are a major challenge for surgeons undertaking these procedures. In particular post-operative bleeding has a major influence on morbidity and mortality (76). Therefore patients undergoing such surgery are at considerable risk. Increased postoperative bleeding after CPB has been attributed to impairment of hemostatic system by for e.g. platelet dysfunction (77),

hyperfibrinolysis (78), hemodilution (79) and excessive activation of both the intrinsic and extrinsic pathway by blood contact with extracorporeal surfaces (80-81). Therefore, it is imperative that during such procedures, hemostasis screening and careful monitoring of blood components and coagulation factors are carried out (82-83).

In addition to the risk of excessive bleeding, cardiac surgery and CPB are also associated with major inflammatory responses (67,84-85). Contact activation of the extracorporeal circulation to non-endothelial surfaces and ischemia/reperfusion injury to the heart and lungs triggers inflammation (86-87). Activation of coagulation ties in closely with the activation of the inflammation. Tissue factor is up-regulated by the onset of inflammation leading to hypercoagulability, and increased expression of plasminogen activator inhibitor, which inhibits fibrinolysis. The results are an increase in the tendency of thrombosis (88) and several studies have suggested that this may be an important mechanism in organ injury in conditions like SIRS, sepsis and cardiac surgery (89-93). Therefore, as well as identifying mediators of coagulation, it is also important to identify mediators in inflammation to combat complications in surgery. TF is an ideal candidate as it is a key player in coagulation and a major player in inflammation. In a state of hypercoagulability, TF activates clotting signals which include coagulant mediators, fibrin and its fragments, all of which are pro-inflammatory and contribute to inflammation (for review see (94)).

TF activates clotting factor VII which initiates a cascade of clotting mediators starting with the conversion of X to Xa. The clotting signal is complete with the final conversion of soluble fibrinogen to fibrin and the formation of a clot. The pro-inflammatory signals of these clotting mediators are elicited through the interactions of the protease-activated receptor (95) and such effects include the activation of cytokines, adhesion molecules and growth factors (95-97). Aside to its role in hemostasis, fibrin(ogen) and it fragments mediate a number of pro-inflammatory actions also (98). Fibrin(ogen) and its derivatives are recognized for their role in leukocytes adhesion and transmigration (a key step in acute inflammation) (99). Leukocyte integrins CD11c/CD11b expressed on neutrophils and monocytes bind to gamma region of the fibrinogen, mediating leukocyte-endothelium interactions with intracellular adhesion molecule-1 (100-101). In addition, both macrophage adhesion and cytokine production was suppressed in fibrin(ogen)-deficient mice (102). More recently, the fibrin fragment NDSK-II has been shown to promote leukocyte adhesion and migration via interaction of the $B\beta_{15-42}$ sequence of fibrin with the VE-cadherin (103).

5. SUREGERY AND CYTOKINES

Surgical procedures alter the expression patterns of cytokines, which relate to postoperative inflammatory responses and/or deterioration of the immune system, leading to increased risk of infection. Open Abdominal aortic aneurysm (AAA) surgery induces a profound inflammatory and coagulative response through influencing cytokine and fibrinogen levels. Preoperatively, IL-6 was elevated in AAA patients versus age-matched controls. During aortic clamping, IL-6, IL-10 and Monocyte chemoattractant protein (MCP)-1 significantly increased, while fibrinogen decreased. After aortic declamping, IL-6, IL-10 and MCP-1 increased further compared with levels during aortic clamping, fibrinogen had a further decrease. After one week, postoperative levels of IL-6, IL-10 and MPC-1 had all decreased (but were still elevated compared with baseline values), while sIl-2R and fibrinogen showed an increase in comparison with baseline (104).

In orthopedic surgery using a LPS-stimulated whole blood assay, one study has revealed that 6 days after surgery, there is suppression in leukocyte capacity to express the cytokines TNF-α and IL-10. In blood taken preoperatively and postoperatively (day 1) and exposed to LPS, no changes were observed in TNF-α and IL-10 levels. However on day 6, a significant reduction in the expression of both TNF-α and IL-10 was observed when compared to preoperative levels, concluding that during this time, patients are susceptible to septic complications (105). A similar method was used to determine cytokine levels in newborns before, 5 and 10 days after cardiac surgery. *Ex vivo* production of the pro-inflammatory cytokines TNF-α, IL-6 and IL-10 was reduced 5 days after surgery. A clear association between the preoperative *ex vivo* production of IL-6 and postoperative respiratory morbidity was also revealed (106). In adult patients undergoing cardiac surgery, suppression of TNF-α after CPB has also been observed (107).

A recent study determined that in cardiac surgery, surgical trauma contributes with a higher degree to the inflammatory response than CPB (108). This is consistent with another study where a number of pro-inflammatory cytokines were profiled during various stages of different cardiac-thoracic procedures. 6 h after CABG, IL-6 levels were maximal in patients who underwent extra-corporeal circulation as well as those who did not (off-pump surgery). IL-6 was also elevated in patients receiving thoracic surgery; however, levels were lower than both cardiac surgery groups. Lipoprotein binding protein was elevated in all three groups from 6 h to 3 days after surgery, although off-pump patient levels were significantly higher than in the other two groups. Procalcitonin was also elevated in all groups, with the extracorporeal circulation patients having the highest levels (109). It was concluded that the impact of extracorporeal circulation on the inflammatory response after cardiac surgery may not be so important as surgical trauma and reperfusion injury. To further elucidate the molecular mechanisms of systemic inflammation in on- and off-pump CABG, the gene expression patterns in leukocytes and plasma proteins was determined. In addition to the usual pro-inflammatory cytokines (IL-6, IL-10 and TNF-α) released, proteomic analysis revealed that IFN-γ, granulocyte colony-stimulating factor, monocyte chemotactic protein-1 and macrophage inflammatory protein-1β also extend the list of mediators released on CPB. Systemic analysis of transcriptional genes in circulating leukocytes revealed that up-regulation of adhesion molecules L-selectin and ICAM-2, signaling

mediators IL-1, IL-8, IL-18 receptors and TLR 4, 5 and 6 also occur after contact with CPB (110). Therefore gene array and muliplex analysis are ideal techniques to profile key players as well as unidentified mediators in the inflammatory responses during various kinds of surgery. This way forward may uncover new potential targets for improving clinical outcome after such procedures.

6. GENETIC POLYMORPHISM AND INFLAMMATION

During surgery, induction of anesthesia and the trauma of surgical procedures induce a localized inflammatory response that stimulates the innate immunity initiating defense mechanisms and wound healing. This is important for patient outcome and recovery, however as discussed there are certain complications that can arise perioperatively and/or postoperatively. Abdominal surgery often results in SIRS and intraperitoneal sepsis (TLR activation) (111-112). Cardiac surgery runs the risk of bleeding (76), thrombosis (93) and inflammatory responses (109). In all cases, continued efforts to reduce such complications are on-going. However whereas in the past clinicians have focused on identifying the mechanisms and pathology of such complications, perhaps a different approach could be to identify and profile genetic variations of crucial genes in innate immunity and coagulation pathways. Genetics may explain the wide variation observed in individual responses' to for e.g. infection or why individuals have different outcomes after surgery. In recent years, the genetic risk for infections has become increasingly recognized, yet still underestimated. For example a Single nucleotide polymorphism (SNP) of TLR1 (TLR1-7202A/G) is associated with worse organ dysfunction, increased gram positive infections and death in sepsis (113). Patients with a TLR4 gene mutation have a higher incidence of Gram-negative infections (44). Specifically patients with TLR4 Asp299Gly allele have higher incidences of Gram-negative infections (114) and this polymorphism is also attributed to the severity of SIRS (115). We have shown that TLR2 (47) and TLR4 (48) knock-out mice have an impaired adrenal stress response to bacterial wall fragments. During surgery, the HPA-axis is activated during the acute phase response (116). Therefore it would be interesting to investigate the outcome and risk of bacterial infection of patients with TLR polymorphism after for e.g. cardiac or colorectal surgery.

Asp299Gly TLR4 polymorphism has been linked to a reduced risk for acute coronary events and atherosclerosis (45,117). In both studies individuals with this polymorphism had also lower levels of fibrinogen, adhesion molecules and in the latter study reduced levels of pro-inflammatory cytokines and acute phase reactants. Again, how would these individuals fair after cardiac surgery, where such mediators are involved in the patho-mechanisms of the inflammatory response (110)? Genetic polymorphisms of TLRs, coagulation factors and cytokines could have a major impact on clinical outcome after surgery. The concept of a patient with a subcutaneous silicon chip carrying a genetic profile has been presented as a theoretical case for the year 2010 (118). Such technology could identify multiple allelic variants resulting in individualized targeted therapy and improving clinical outcome in general and after surgical procedures.

7. PERSPECTIVE

Although current knowledge suggests that numerous factors can influence an individual response to inflammation, little is known about how much each player contributes to a patient's clinical outcome. At present, basic research and large clinical trials (according to www.clinicaltrials.gov, currently 375 trials are underway focusing on sepsis) are needed to separate these factors into positive and negative effectors as well as elucidating their overall importance during inflammation. With such knowledge it would be feasible to screen high risk patients undergoing major surgery, with the aim to optimize their therapy and improve clinical outcome.

8. ACKNOLEGEMENTS

This work was supported by research grants from the Deutsche Forschungsgemeinschaft (Za243/8-1, Za243/8-2 and Za243/9-1 to K.Z.) and the Forschungskommission Duesseldorf (A.K. and K.Z.).

9. REFERENCES

1. N. Garrett and E. M. Martini: The boomers are coming: a total cost of care model of the impact of population aging on the cost of chronic conditions in the United States. *Dis.Manag.* 10, 51-60 (2007)

2. D. C. Angus, W. T. Linde-Zwirble, J. Lidicker, G. Clermont, J. Carcillo and M. R. Pinsky: Epidemiology of severe sepsis in the United States: analysis of incidence, outcome, and associated costs of care. *Crit Care Med.* 29, 1303-1310 (2001)

3. J. Cohen: The immunopathogenesis of sepsis. *Nature* 420, 885-891 (2002)

4. P. Libby: Inflammation in atherosclerosis. *Nature* 420, 868-874 (2002)

5. M. A. Gonzalez and A. P. Selwyn: Endothelial function, inflammation, and prognosis in cardiovascular disease. *Am.J.Med.* 115 Suppl 8A, 99S-106S (2003)

6. M. Levi, E. de Jonge and P. T. van der: New treatment strategies for disseminated intravascular coagulation based on current understanding of the pathophysiology. *Ann.Med.* 36, 41-49 (2004)

7. R. Paoletti, A. M. Gotto, Jr. and D. P. Hajjar: Inflammation in atherosclerosis and implications for therapy. *Circulation* 109, III20-III26 (2004)

8. Y. Kumagai, O. Takeuchi and S. Akira: Pathogen recognition by innate receptors. *J.Infect.Chemother.* 14, 86-92 (2008)

9. S. Akira, S. Uematsu and O. Takeuchi: Pathogen recognition and innate immunity. *Cell* 124, 783-801 (2006)

10. T. Kawai and S. Akira: Signaling to NF-kappaB by Toll-like receptors. *Trends Mol.Med* 13, 460-469 (2007)

11. A. Aderem and R. J. Ulevitch: Toll-like receptors in the induction of the innate immune response. *Nature* 406, 782-787 (2000)

12. A. Iwasaki and R. Medzhitov: Toll-like receptor control of the adaptive immune responses. *Nat.Immunol.* 5, 987-995 (2004)

13. R. Schwandner, R. Dziarski, H. Wesche, M. Rothe and C. J. Kirschning: Peptidoglycan- and lipoteichoic acid-induced cell activation is mediated by toll-like receptor 2. *J.Biol.Chem.* 274, 17406-17409 (1999)

14. K. Hoshino, O. Takeuchi, T. Kawai, H. Sanjo, T. Ogawa, Y. Takeda, K. Takeda and S. Akira: Cutting edge: Toll-like receptor 4 (TLR4)-deficient mice are hyporesponsive to lipopolysaccharide: evidence for TLR4 as the Lps gene product. *J.Immunol.* 162, 3749-3752 (1999)

15. S. Yokota, T. Ohnishi, M. Muroi, K. Tanamoto, N. Fujii and K. Amano: Highly-purified Helicobacter pylori LPS preparations induce weak inflammatory reactions and utilize Toll-like receptor 2 complex but not Toll-like receptor 4 complex. *FEMS Immunol.Med Microbiol.* 51, 140-148 (2007)

16. S. Thakran, H. Li, C. L. Lavine, M. A. Miller, J. E. Bina, X. R. Bina and F. Re: Identification of Francisella tularensis lipoproteins that stimulate the toll-like receptor (TLR) 2/TLR1 heterodimer. *J.Biol.Chem.* 283, 3751-3760 (2008)

17. E. T. Rietschel, T. Kirikae, F. U. Schade, U. Mamat, G. Schmidt, H. Loppnow, A. J. Ulmer, U. Zahringer, U. Seydel, F. Di Padova and *et al.*: Bacterial endotoxin: molecular relationships of structure to activity and function. *Faseb J* 8, 217-225 (1994)

18. J. Schletter, H. Heine, A. J. Ulmer and E. T. Rietschel: Molecular mechanisms of endotoxin activity. *Arch Microbiol* 164, 383-389 (1995)

19. M. Guha and N. Mackman: LPS induction of gene expression in human monocytes. *Cell Signal.* 13, 85-94 (2001)

20. G. Wakabayashi, J. A. Gelfand, W. K. Jung, R. J. Connolly, J. F. Burke and C. A. Dinarello: Staphylococcus epidermidis induces complement activation, tumor necrosis factor and interleukin-1, a shock-like state and tissue injury in rabbits without endotoxemia. Comparison to Escherichia coli. *J Clin Invest* 87, 1925-1935 (1991)

21. W. Fischer: Physiology of lipoteichoic acids in bacteria. *Adv Microb Physiol* 29, 233-302 (1988)

22. K. M. Kengatharan, S. De Kimpe, C. Robson, S. J. Foster and C. Thiemermann: Mechanism of gram-positive shock: identification of peptidoglycan and lipoteichoic acid moieties essential in the induction of nitric oxide synthase, shock, and multiple organ failure. *J.Exp.Med.* 188, 305-315 (1998)

23. S. Immenschuh, J. Stritzke, S. Iwahara and G. Ramadori: Up-regulation of heme-binding protein 23 (HBP23) gene expression by lipopolysaccharide is mediated via a nitric oxide-dependent signaling pathway in rat Kupffer cells. *Hepatology* 30, 118-127 (1999)

24. S. Kurata, M. Matsumoto and U. Yamashita: Concomitant transcriptional activation of nitric oxide synthase and heme oxygenase genes during nitric oxide-mediated macrophage cytostasis. *J.Biochem. (Tokyo)* 120, 49-52 (1996)

25. A. M. Vicente, M. I. Guillen and M. J. Alcaraz: Modulation of haem oxygenase-1 expression by nitric oxide and leukotrienes in zymosan-activated macrophages. *Br.J.Pharmacol.* 133, 920-926 (2001)

26. A. M. Choi and J. Alam: Heme oxygenase-1: function, regulation, and implication of a novel stress-inducible protein in oxidant-induced lung injury. *Am.J.Respir.Cell Mol.Biol.* 15, 9-19 (1996)

27. M. D. Maines: Heme oxygenase: function, multiplicity, regulatory mechanisms, and clinical applications. *FASEB J.* 2, 2557-2568 (1988)

28. S. Moncada, R. M. Palmer and E. A. Higgs: Nitric oxide: physiology, pathophysiology, and pharmacology. *Pharmacol Rev* 43, 109-142 (1991)

29. C. Nathan: Nitric oxide as a secretory product of mammalian cells. *FASEB J.* 6, 3051-3064 (1992)

30. K. Tanjoh, A. Shima, M. Aida, R. Tomita and Y. Kurosu: Nitric oxide and active oxygen species in severe sepsis and surgically stressed patients. *Surg.Today* 25, 774-777 (1995)

31. C. Szabo: The pathophysiological role of peroxynitrite in shock, inflammation, and ischemia-reperfusion injury. *Shock* 6, 79-88 (1996)

32. H. Fahmi, D. Charon, M. Mondange and R. Chaby: Endotoxin-induced desensitization of mouse macrophages is mediated in part by nitric oxide production. *Infect.Immun.* 63, 1863-1869 (1995)

33. D. Soszynski: Inhibition of nitric oxide synthase delays the development of tolerance to LPS in rats. *Physiol Behav.* 76, 159-169 (2002)

34. K. Srisook and Y. N. Cha: Biphasic induction of heme oxygenase-1 expression in macrophages stimulated with lipopolysaccharide. *Biochem.Pharmacol.* 68, 1709-1720 (2004)

35. A. Koch, O. Boehm, P. A. Zacharowski, S. A. Loer, J. Weimann, H. Rensing, S. J. Foster, R. Schmidt, R. Berkels, S. Reingruber and K. Zacharowski: Inducible nitric oxide synthase and heme oxygenase-1 in the lung during lipopolysaccharide tolerance and cross tolerance. *Crit Care Med.* (2007)

36. G. P. Chrousos: The hypothalamic-pituitary-adrenal axis and immune-mediated inflammation. *N.Engl.J.Med.* 332, 1351-1362 (1995)

37. I. A. MacPhee, F. A. Antoni and D. W. Mason: Spontaneous recovery of rats from experimental allergic encephalomyelitis is dependent on regulation of the immune system by endogenous adrenal corticosteroids. *J.Exp.Med.* 169, 431-445 (1989)

38. R. Abe, T. Shimosegawa, K. Kimura, T. Abe, J. Kashimura, M. Koizumi and T. Toyota: The role of endogenous glucocorticoids in rat experimental models of acute pancreatitis. *Gastroenterology* 109, 933-943 (1995)

39. H. O. Besedovsky and A. del Rey: Immune-neuro-endocrine interactions: facts and hypotheses. *Endocr.Rev.* 17, 64-102 (1996)

40. T. Wilckens and R. De Rijk: Glucocorticoids and immune function: unknown dimensions and new frontiers. *Immunol.Today* 18, 418-424 (1997)

41. D. T. Boumpas, G. P. Chrousos, R. L. Wilder, T. R. Cupps and J. E. Balow: Glucocorticoid therapy for immune-mediated diseases: basic and clinical correlates. *Ann.Intern.Med.* 119, 1198-1208 (1993)

42. S. R. Bornstein and G. P. Chrousos: Clinical review 104: Adrenocorticotropin (ACTH)- and non-ACTH-mediated regulation of the adrenal cortex: neural and immune inputs. *J.Clin.Endocrinol.Metab* 84, 1729-1736 (1999)

43. J. I. Webster and E. M. Sternberg: Role of the hypothalamic-pituitary-adrenal axis, glucocorticoids and glucocorticoid receptors in toxic sequelae of exposure to bacterial and viral products. *J.Endocrinol.* 181, 207-221 (2004)

44. D. M. Agnese, J. E. Calvano, S. J. Hahm, S. M. Coyle, S. A. Corbett, S. E. Calvano and S. F. Lowry: Human toll-like receptor 4 mutations but not CD14 polymorphisms are associated with an increased risk of gram-negative infections. *J.Infect.Dis.* 186, 1522-1525 (2002)

45. S. Kiechl, E. Lorenz, M. Reindl, C. J. Wiedermann, F. Oberhollenzer, E. Bonora, J. Willeit and D. A. Schwartz: Toll-like receptor 4 polymorphisms and atherogenesis. *N Engl J Med* 347, 185-192 (2002)

46. E. Lorenz, J. P. Mira, K. L. Cornish, N. C. Arbour and D. A. Schwartz: A novel polymorphism in the toll-like receptor 2 gene and its potential association with staphylococcal infection. *Infect.Immun.* 68, 6398-6401 (2000)

47. S. R. Bornstein, P. Zacharowski, R. R. Schumann, A. Barthel, N. Tran, C. Papewalis, V. Rettori, S. M. McCann, K. Schulze-Osthoff, W. A. Scherbaum, J. Tarnow and K. Zacharowski: Impaired adrenal stress response in Toll-like receptor 2-deficient mice. *Proc.Natl.Acad.Sci.U.S.A* 101, 16695-16700 (2004)

48. K. Zacharowski, P. A. Zacharowski, A. Koch, A. Baban, N. Tran, R. Berkels, C. Papewalis, K. Schulze-Osthoff, P. Knuefermann, U. Zahringer, R. R. Schumann, V. Rettori, S. M. McCann and S. R. Bornstein: Toll-like receptor 4 plays a crucial role in the immune-adrenal response to systemic inflammatory response syndrome. *Proc.Natl.Acad.Sci.U.S.A* 103, 6392-6397 (2006)

49. S. R. Bornstein, R. R. Schumann, V. Rettori, S. M. McCann and K. Zacharowski: Toll-like receptor 2 and Toll-like receptor 4 expression in human adrenals. *Horm.Metab Res.* 36, 470-473 (2004)

50. D. Annane, V. Sebille, C. Charpentier, P. E. Bollaert, B. Francois, J. M. Korach, G. Capellier, Y. Cohen, E. Azoulay, G. Troche, P. Chaumet-Riffaut and E. Bellissant: Effect of treatment with low doses of hydrocortisone and fludrocortisone on mortality in patients with septic shock. *JAMA* 288, 862-871 (2002)

51. S. R. Bornstein and J. Briegel: A new role for glucocorticoids in septic shock: balancing the immune response. *Am.J.Respir.Crit Care Med.* 167, 485-486 (2003)

52. M. S. Cooper and P. M. Stewart: Corticosteroid insufficiency in acutely ill patients. *N.Engl.J.Med.* 348, 727-734 (2003)

53. C. L. Sprung, D. Annane, D. Keh, R. Moreno, M. Singer, K. Freivogel, Y. G. Weiss, J. Benbenishty, A. Kalenka, H. Forst, P. F. Laterre, K. Reinhart, B. H. Cuthbertson, D. Payen and J. Briegel: Hydrocortisone therapy for patients with septic shock. *N.Engl.J.Med* 358, 111-124 (2008)

54. J. Bruce, Z. H. Krukowski, G. Al Khairy, E. M. Russell and K. G. Park: Systematic review of the definition and measurement of anastomotic leak after gastrointestinal surgery. *Br.J.Surg* 88, 1157-1168 (2001)

55. Y. Parc, P. Frileux, G. Schmitt, N. Dehni, J. M. Ollivier and R. Parc: Management of postoperative peritonitis after anterior resection: experience from a referral intensive care unit. *Dis.Colon Rectum* 43, 579-587 (2000)

56. D. L. Williams, T. Ha, C. Li, J. H. Kalbfleisch, J. Schweitzer, W. Vogt and I. W. Browder: Modulation of tissue Toll-like receptor 2 and 4 during the early phases of polymicrobial sepsis correlates with mortality. *Crit Care Med.* 31, 1808-1818 (2003)

57. C. F. Benjamim, S. K. Lundy, N. W. Lukacs, C. M. Hogaboam and S. L. Kunkel: Reversal of long-term sepsis-induced immunosuppression by dendritic cells. *Blood* 105, 3588-3595 (2005)

58. P. C. Sedman, J. MacFie, P. Sagar, C. J. Mitchell, J. May, B. Mancey-Jones and D. Johnstone: The prevalence of gut translocation in humans. *Gastroenterology* 107, 643-649 (1994)

59. J. MacFie: Current status of bacterial translocation as a cause of surgical sepsis. *Br.Med Bull.* 71, 1-11 (2004)

60. E. Bolke, P. M. Jehle, K. Orth, G. Steinbach, A. Hannekum and M. Storck: Changes of gut barrier function during anesthesia and cardiac surgery. *Angiology* 52, 477-482 (2001)

61. J. MacFie: Current status of bacterial translocation as a cause of surgical sepsis. *Br.Med.Bull.* 71, 1-11 (2004)

62. H. Tsujimoto, S. Ono, T. Majima, P. A. Efron, M. Kinoshita, H. Hiraide, L. L. Moldawer and H. Mochizuki: Differential toll-like receptor expression after *ex vivo* lipopolysaccharide exposure in patients with sepsis and following surgical stress. *Clin.Immunol.* 119, 180-187 (2006)

63. L. Harter, L. Mica, R. Stocker, O. Trentz and M. Keel: Increased expression of toll-like receptor-2 and -4 on leukocytes from patients with sepsis. *Shock* 22, 403-409 (2004)

64. J. S. Hadley, J. E. Wang, L. C. Michaels, C. M. Dempsey, S. J. Foster, C. Thiemermann and C. J. Hinds: Alterations in inflammatory capacity and TLR expression on monocytes and neutrophils after cardiopulmonary bypass. *Shock* 27, 466-473 (2007)

65. M. K. Brunialti, P. S. Martins, C. H. Barbosa de, F. R. Machado, L. M. Barbosa and R. Salomao: TLR2, TLR4, CD14, CD11B, and CD11C expressions on monocytes surface and cytokine production in patients with sepsis, severe sepsis, and septic shock. *Shock* 25, 351-357 (2006)

66. D. Bastian, H. Shegarfi, B. Rolstad, C. Naper, S. P. Lyngstadaas and O. Reikeras: Investigation of lipopolysaccharide receptor expression on human monocytes after major orthopaedic surgery. *Eur.Surg.Res.* 40, 239-245 (2008)

67. B. Dybdahl, A. Wahba, E. Lien, T. H. Flo, A. Waage, N. Qureshi, O. F. Sellevold, T. Espevik and A. Sundan: Inflammatory response after open heart surgery: release of heat-shock protein 70 and signaling through toll-like receptor-4. *Circulation* 105, 685-690 (2002)

68. H. Ikushima, T. Nishida, K. Takeda, T. Ito, T. Yasuda, M. Yano, S. Akira and H. Matsuda: Expression of Toll-like receptors 2 and 4 is downregulated after operation. *Surgery* 135, 376-385 (2004)

69. J. E. Calvano, D. M. Agnese, J. Y. Um, M. Goshima, R. Singhal, S. M. Coyle, M. T. Reddell, A. Kumar, S. E. Calvano and S. F. Lowry: Modulation of the lipopolysaccharide receptor complex (CD14, TLR4, MD-2) and toll-like receptor 2 in systemic inflammatory response syndrome-positive patients with and without infection: relationship to tolerance. *Shock* 20, 415-419 (2003)

70. D. Ducloux, M. Deschamps, M. Yannaraki, C. Ferrand, J. Bamoulid, P. Saas, A. Kazory, J. M. Chalopin and P. Tiberghien: Relevance of Toll-like receptor-4 polymorphisms in renal transplantation. *Kidney Int.* 67, 2454-2461 (2005)

71. N. W. Schroder and R. R. Schumann: Single nucleotide polymorphisms of Toll-like receptors and susceptibility to infectious disease. *Lancet Infect.Dis.* 5, 156-164 (2005)

72. K. Tomokiyo, Y. Kamikubo, T. Hanada, T. Araki, Y. Nakatomi, Y. Ogata, S. M. Jung, T. Nakagaki and M. Moroi: Von Willebrand factor accelerates platelet adhesion and thrombus formation on a collagen surface in platelet-reduced blood under flow conditions. *Blood* 105, 1078-1084 (2005)

73. B. Savage, M. Cattaneo and Z. M. Ruggeri: Mechanisms of platelet aggregation. *Curr.Opin.Hematol.* 8, 270-276 (2001)

74. K. G. Mann, C. van't Veer, K. Cawthern and S. Butenas: The role of the tissue factor pathway in initiation of coagulation. *Blood Coagul.Fibrinolysis* 9 Suppl 1, S3-S7 (1998)

75. F. J. Castellino and V. A. Ploplis: Structure and function of the plasminogen/plasmin system. *Thromb.Haemost.* 93, 647-654 (2005)

76. K. Karkouti, D. N. Wijeysundera, T. M. Yau, W. S. Beattie, E. Abdelnaem, S. A. McCluskey, M. Ghannam, E. Yeo, G. Djaiani and J. Karski: The independent association of massive blood loss with mortality in cardiac surgery. *Transfusion* 44, 1453-1462 (2004)

77. N. Tabuchi, I. Tigchelaar and W. van Oeveren: Shear-induced pathway of platelet function in cardiac surgery. *Semin.Thromb.Hemost.* 21 Suppl 2, 66-70 (1995)

78. M. J. Ray, N. A. Marsh and G. A. Hawson: Relationship of fibrinolysis and platelet function to bleeding after cardiopulmonary bypass. *Blood Coagul.Fibrinolysis* 5, 679-685 (1994)

79. S. Dial, E. Delabays, M. Albert, A. Gonzalez, J. Camarda, A. Law and D. Menzies: Hemodilution and surgical hemostasis contribute significantly to transfusion requirements in patients undergoing coronary artery bypass. *J.Thorac.Cardiovasc.Surg.* 130, 654-661 (2005)

80. R. L. Heimark, K. Kurachi, K. Fujikawa and E. W. Davie: Surface activation of blood coagulation, fibrinolysis and kinin formation. *Nature* 286, 456-460 (1980)
81. M. D. Boisclair, D. A. Lane, H. Philippou, M. P. Esnouf, S. Sheikh, B. Hunt and K. J. Smith: Mechanisms of thrombin generation during surgery and cardiopulmonary bypass. *Blood* 82, 3350-3357 (1993)

82. H. J. Hertfelder, M. Bos, D. Weber, K. Winkler, P. Hanfland and C. J. Preusse: Perioperative monitoring of primary and secondary hemostasis in coronary artery bypass grafting. *Semin.Thromb.Hemost.* 31, 426-440 (2005)

83. M. D. Boisclair, H. Ireland and D. A. Lane: Assessment of hypercoagulable states by measurement of activation fragments and peptides. *Blood Rev.* 4, 25-40 (1990)

84. P. Li, J. Sanders, E. Hawe, D. Brull, H. Montgomery and S. Humphries: Inflammatory response to coronary artery bypass surgery: does the heme-oxygenase-1 gene microsatellite polymorphism play a role? *Chin Med.J. (Engl.)* 118, 1285-1290 (2005)

85. P. J. Hess, Jr.: Systemic inflammatory response to coronary artery bypass graft surgery. *Am.J.Health Syst.Pharm.* 62, S6-S9 (2005)

86. G. Kalweit, J. Bach, H. Huwer, J. Winning and P. Hellstern: The impact of cardiac ischemia and reperfusion on markers of activated haemostasis and fibrinolysis during cardiopulmonary bypass: comparison of plasma levels in arterial and coronary venous blood. *Thromb.Res.* 116, 33-39 (2005)

87. R. Grossmann, J. Babin-Ebell, M. Misoph, S. Schwender, K. Neukam, T. Hickethier, O. Elert and F. Keller: Changes in coagulation and fibrinolytic parameters caused by extracorporeal circulation. *Heart Vessels* 11, 310-317 (1996)

88. A. J. Chu: Tissue factor upregulation drives a thrombosis-inflammation circuit in relation to cardiovascular complications. *Cell Biochem.Funct.* (2004)

89. M. E. Lissauer, S. B. Johnson, G. Siuzdak, G. Bochicchio, C. Whiteford, B. Nussbaumer, R. Moore and T. M. Scalea: Coagulation and complement protein differences between septic and uninfected systemic inflammatory response syndrome patients. *J.Trauma* 62, 1082-1092 (2007)

90. G. Lissalde-Lavigne, C. Combescure, L. Muller, C. Bengler, A. Raillard, J. Y. Lefrant and J. C. Gris: Simple coagulation tests improve survival prediction in patients with septic shock. *J.Thromb.Haemost.* 6, 645-653 (2008)

91. S. Gando, S. Nanzaki, S. Sasaki and O. Kemmotsu: Significant correlations between tissue factor and thrombin markers in trauma and septic patients with disseminated intravascular coagulation. *Thromb.Haemost.* 79, 1111-1115 (1998)

92. S. Gando, S. Nanzaki and O. Kemmotsu: Disseminated intravascular coagulation and sustained systemic inflammatory response syndrome predict organ dysfunctions after trauma: application of clinical decision analysis. *Ann.Surg.* 229, 121-127 (1999)

93. B. Dixon, J. Santamaria and D. Campbell: Coagulation activation and organ dysfunction following cardiac surgery. *Chest* 128, 229-236 (2005)

94. M. Schouten, W. J. Wiersinga, M. Levi and P. T. van der: Inflammation, endothelium, and coagulation in sepsis. *J.Leukoc.Biol.* 83, 536-545 (2008)

95. M. Riewald, V. V. Kravchenko, R. J. Petrovan, P. J. O'Brien, L. F. Brass, R. J. Ulevitch and W. Ruf: Gene induction by coagulation factor Xa is mediated by activation of protease-activated receptor 1. *Blood* 97, 3109-3116 (2001)

96. G. Busch, I. Seitz, B. Steppich, S. Hess, R. Eckl, A. Schomig and I. Ott: Coagulation factor Xa stimulates interleukin-8 release in endothelial cells and mononuclear leukocytes: implications in acute myocardial infarction. *Arterioscler.Thromb.Vasc.Biol.* 25, 461-466 (2005)

97. A. Rahman, A. L. True, K. N. Anwar, R. D. Ye, T. A. Voyno-Yasenetskaya and A. B. Malik: Galpha (q) and Gbetagamma regulate PAR-1 signaling of thrombin-induced NF-kappaB activation and ICAM-1 transcription in endothelial cells. *Circ.Res.* 91, 398-405 (2002)

98. M. E. Lee, K. J. Rhee and S. U. Nham: Fragment E derived from both fibrin and fibrinogen stimulates interleukin-6 production in rat peritoneal macrophages. *Mol.Cells* 9, 7-13 (1999)

99. L. R. Languino, A. Duperray, K. J. Joganic, M. Fornaro, G. B. Thornton and D. C. Altieri: Regulation of leukocyte-endothelium interaction and leukocyte transendothelial migration by intercellular adhesion molecule 1-fibrinogen recognition. *Proc.Natl.Acad.Sci U.S.A* 92, 1505-1509 (1995)

100. D. C. Altieri, J. Plescia and E. F. Plow: The structural motif glycine 190-valine 202 of the fibrinogen gamma chain interacts with CD11b/CD18 integrin (alpha M beta 2, Mac-1) and promotes leukocyte adhesion. *J Biol.Chem.* 268, 1847-1853 (1993)

101. D. C. Altieri, A. Duperray, J. Plescia, G. B. Thornton and L. R. Languino: Structural recognition of a novel fibrinogen gamma chain sequence (117-133) by intercellular adhesion molecule-1 mediates leukocyte-endothelium interaction. *J Biol.Chem.* 270, 696-699 (1995)

102. F. M. Szaba and S. T. Smiley: Roles for thrombin and fibrin (ogen) in cytokine/chemokine production and macrophage adhesion *in vivo*. *Blood* 99, 1053-1059 (2002)

103. P. Petzelbauer, P. A. Zacharowski, Y. Miyazaki, P. Friedl, G. Wickenhauser, F. J. Castellino, M. Groger, K. Wolff and K. Zacharowski: The fibrin-derived peptide

Bbeta15-42 protects the myocardium against ischemia-reperfusion injury. *Nat.Med.* 11, 298-304 (2005)
104. A. Holmberg, D. Bergqvist, B. Westman and A. Siegbahn: Cytokine and fibrinogen response in patients undergoing open abdominal aortic aneurysm surgery. *Eur.J.Vasc.Endovasc.Surg.* 17, 294-300 (1999)

105. O. Reikeraas, J. Sun, C. D. Krohn, J. E. Wang and A. O. Aasen: Reduced capacity of whole blood leucocytes to express tumour necrosis factor-alpha and interleukin-10 following major orthopaedic surgery. *Eur.Surg Res.* 37, 210-215 (2005)

106. K. Schumacher, S. Korr, J. F. Vazquez-Jimenez, G. von Bernuth, J. Duchateau and M. C. Seghaye: Does cardiac surgery in newborn infants compromise blood cell reactivity to endotoxin? *Crit Care* 9, R549-R555 (2005)

107. J. Borgermann, I. Friedrich, S. Flohe, J. Spillner, M. Majetschak, O. Kuss, A. Sablotzki, T. Feldt, J. C. Reidemeister and F. U. Schade: Tumor necrosis factor-alpha production in whole blood after cardiopulmonary bypass: downregulation caused by circulating cytokine-inhibitory activities. *J.Thorac.Cardiovasc.Surg* 124, 608-617 (2002)

108. R. Prondzinsky, A. Knupfer, H. Loppnow, F. Redling, D. W. Lehmann, I. Stabenow, R. Witthaut, S. Unverzagt, J. Radke, H. R. Zerkowski and K. Werdan: Surgical trauma affects the proinflammatory status after cardiac surgery to a higher degree than cardiopulmonary bypass. *J.Thorac.Cardiovasc.Surg.* 129, 760-766 (2005)

109. A. Franke, W. Lante, V. Fackeldey, H. P. Becker, E. Kurig, L. G. Zoller, C. Weinhold and A. Markewitz: Pro-inflammatory cytokines after different kinds of cardio-thoracic surgical procedures: is what we see what we know? *Eur.J.Cardiothorac.Surg.* 28, 569-575 (2005)

110. V. Tomic, S. Russwurm, E. Moller, R. A. Claus, M. Blaess, F. Brunkhorst, M. Bruegel, K. Bode, F. Bloos, J. Wippermann, T. Wahlers, H. P. Deigner, J. Thiery, K. Reinhart and M. Bauer: Transcriptomic and proteomic patterns of systemic inflammation in on-pump and off-pump coronary artery bypass grafting. *Circulation* 112, 2912-2920 (2005)

111. R. Sarbinowski, S. Arvidsson, M. Tylman, T. Oresland and A. Bengtsson: Plasma concentration of procalcitonin and systemic inflammatory response syndrome after colorectal surgery. *Acta Anaesthesiol.Scand.* 49, 191-196 (2005)

112. R. R. Hutchins, M. P. Gunning, D. N. Lucas, T. G. Allen-Mersh and N. C. Soni: Relaparotomy for suspected intraperitoneal sepsis after abdominal surgery. *World J.Surg.* 28, 137-141 (2004)

113. M. M. Wurfel, A. C. Gordon, T. D. Holden, F. Radella, J. Strout, O. Kajikawa, J. T. Ruzinski, G. Rona, R. A. Black, S. Stratton, G. P. Jarvik, A. M. Hajjar, D. A. Nickerson, M. Rieder, J. Sevransky, J. P. Maloney, M. Moss, K. C. Barnes, J. A. Russel and T. R. Martin: Toll-like Receptor 1 Polymorphisms Affect Innate Immune Responses and Outcomes in Sepsis. *Am.J.Respir.Crit Care Med* (2008)

114. E. Lorenz, J. P. Mira, K. L. Frees and D. A. Schwartz: Relevance of mutations in the TLR4 receptor in patients with gram-negative septic shock. *Arch.Intern.Med.* 162, 1028-1032 (2002)

115. N. J. Child, I. A. Yang, M. C. Pulletz, K. Courcy-Golder, A. L. Andrews, V. J. Pappachan and J. W. Holloway: Polymorphisms in Toll-like receptor 4 and the systemic inflammatory response syndrome. *Biochem.Soc.Trans.* 31, 652-653 (2003)

116. R. F. De Jongh, K. C. Vissers, L. H. Booij, K. L. De Jongh, P. Vincken and T. F. Meert: Interleukin-6 and perioperative thermoregulation and HPA-axis activation. *Cytokine* 21, 248-256 (2003)

117. N. Ameziane, T. Beillat, P. Verpillat, S. Chollet-Martin, M. C. Aumont, P. Seknadji, M. Lamotte, D. Lebret, V. Ollivier and D. de Prost: Association of the Toll-like receptor 4 gene Asp299Gly polymorphism with acute coronary events. *Arterioscler.Thromb.Vasc.Biol.* 23, e61-e64 (2003)

118. C. L. Holmes, J. A. Russell and K. R. Walley: Genetic polymorphisms in sepsis and septic shock: role in prognosis and potential for therapy. *Chest* 124, 1103-1115 (2003)

119. N. Cheng, R. He, J. Tian, P. P. Ye and R. D. Ye: Cutting edge: TLR2 is a functional receptor for acute-phase serum amyloid A. *J.Immunol.* 181, 22-26 (2008)

120. S. Thakran, H. Li, C. L. Lavine, M. A. Miller, J. E. Bina, X. R. Bina and F. Re: Identification of Francisella tularensis lipoproteins that stimulate the toll-like receptor (TLR) 2/TLR1 heterodimer. *J.Biol.Chem.* 283, 3751-3760 (2008)

121. N. Funderburg, M. M. Lederman, Z. Feng, M. G. Drage, J. Jadlowsky, C. V. Harding, A. Weinberg and S. F. Sieg: Human -defensin-3 activates professional antigen-presenting cells via Toll-like receptors 1 and 2. *Proc.Natl.Acad.Sci.U.S.A* 104, 18631-18635 (2007)

122. S. Chang, A. Dolganiuc and G. Szabo: Toll-like receptors 1 and 6 are involved in TLR2-mediated macrophage activation by hepatitis C virus core and NS3 proteins. *J.Leukoc.Biol.* 82, 479-487 (2007)

123. K. W. Boehme, M. Guerrero and T. Compton: Human cytomegalovirus envelope glycoproteins B and H are necessary for TLR2 activation in permissive cells. *J.Immunol.* 177, 7094-7102 (2006)

124. S. Gariboldi, M. Palazzo, L. Zanobbio, S. Selleri, M. Sommariva, L. Sfondrini, S. Cavicchini, A. Balsari and C. Rumio: Low molecular weight hyaluronic acid increases the self-defense of skin epithelium by induction of beta-

defensin 2 via TLR2 and TLR4. *J.Immunol.* 181, 2103-2110 (2008)
125. S. Gillet-Hladky, K. Duperrier, S. Picandet, V. Mathias, M. C. de Carvalho, J. Bernaud, D. Masseau, J. Bienvenu and D. Rigal: Induction of high expression of CCR7 and high production of IL-12 in human monocyte-derived dendritic cells by a new bacterial component: LCOS 1013. *Int.Immunopharmacol.* 8, 1151-1163 (2008)

126. R. Ahmad, B. S. El, P. Cordeiro and J. Menezes: Requirement of TLR2-mediated signalling for the induction of IL-15 gene expression in human monocytic cells by HSV-1. *Blood* (2008)

127. A. B. Tarabishy, B. Aldabagh, Y. Sun, Y. Imamura, P. K. Mukherjee, J. H. Lass, M. A. Ghannoum and E. Pearlman: MyD88 regulation of Fusarium keratitis is dependent on TLR4 and IL-1R1 but not TLR2. *J.Immunol.* 181, 593-600 (2008)

128. K. L. Richard, M. Filali, P. Prefontaine and S. Rivest: Toll-like receptor 2 acts as a natural innate immune receptor to clear amyloid beta 1-42 and delay the cognitive decline in a mouse model of Alzheimer's disease. *J.Neurosci.* 28, 5784-5793 (2008)

129. N. J. Nilsen, S. Deininger, U. Nonstad, F. Skjeldal, H. Husebye, D. Rodionov, A. S. von, T. Hartung, E. Lien, O. Bakke and T. Espevik: Cellular trafficking of lipoteichoic acid and Toll-like receptor 2 in relation to signaling; role of CD14 and CD36. *J.Leukoc.Biol.* 84, 280-291 (2008)

130. N. J. Nilsen, S. Deininger, U. Nonstad, F. Skjeldal, H. Husebye, D. Rodionov, A. S. von, T. Hartung, E. Lien, O. Bakke and T. Espevik: Cellular trafficking of lipoteichoic acid and Toll-like receptor 2 in relation to signaling; role of CD14 and CD36. *J.Leukoc.Biol.* 84, 280-291 (2008)

131. K. Page, K. M. Lierl, V. S. Hughes, P. Zhou, J. R. Ledford and M. Wills-Karp: TLR2-mediated activation of neutrophils in response to German cockroach frass. *J.Immunol.* 180, 6317-6324 (2008)

132. P. S. Coelho, A. Klein, A. Talvani, S. F. Coutinho, O. Takeuchi, S. Akira, J. S. Silva, H. Canizzaro, R. T. Gazzinelli and M. M. Teixeira: Glycosylphosphatidylinositol-anchored mucin-like glycoproteins isolated from Trypanosoma cruzi trypomastigotes induce *in vivo* leukocyte recruitment dependent on MCP-1 production by IFN-gamma-primed-macrophages. *J.Leukoc.Biol.* 71, 837-844 (2002)

133. J. Fischer, C. Suire and H. Hale-Donze: TLR2 recognition of the microsporidia Encephalitozoon spp. induces nuclear translocation of NF-{kappa}B and subsequent inflammatory responses. *Infect.Immun.* (2008)

134. K. Bieback, E. Lien, I. M. Klagge, E. Avota, J. Schneider-Schaulies, W. P. Duprex, H. Wagner, C. J. Kirschning, M. Ter, V and S. Schneider-Schaulies: Hemagglutinin protein of wild-type measles virus activates toll-like receptor 2 signaling. *J.Virol.* 76, 8729-8736 (2002)

Key Words: Toll-like receptors, Innate Immunity, Coagulation, Fibrin Fragments, Inflammation, Surgery, Review

Send correspondence to: Kai Zacharowski, Molecular Cardioprotection and Inflammation Group, University Hospitals Bristol NHS Foundation Trust, Bristol, BS2 8HW, United Kingdom, Tel: 44-0-1179283152, Fax: 44-0-1179268674, E-mail: kai.zacharowski@bristol.ac.uk

http://www.bioscience.org/current/vol14.htm