# Systemic Inflammatory Response Syndrome

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## Abstract

This short chapter gives a quick overview of the systemic inflammatory response syndrome, what it is, the causes, the consequences and its treatment.

It is of relevance to all severly ill surgical patients and helps the physician understand the patients response to injury and sepsis.

A fuller account of this topic may be found in your favourite surgical textbook.

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# 1 Abbreviations

## 1.1 ARDS

Adult respiratory distress syndrome.

## 1.2 SIRS

The systemic inflammatory response syndrome.

## 1.3 MOF

Multiorgan dysfunction syndrome.

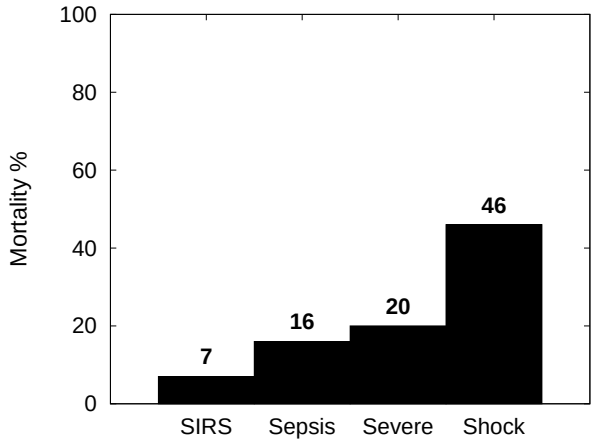


Figure 1: Mortality associated with increasing SIRS [RFPC$^+$95].

Table 1: SIRS[Ame92] is defined as the presence of two or more of the variables below

| Variable | Abnormality |
|---|---|
| Temperature | $> 38^\circ$C or $< 36^\circ$C |
| Heart rate | $> 90$ beats per minute |
| Respiratory rate | $> 20$ breaths per minute or $PaCO_2 < 32$ mmHg ($< 4.3$ kPa) |
| White cell count | $> 12 \times 10^9$ or $< 4 \times 10^9$ or $> 10\%$ immature (band) cells |

# 2 Introduction

The five cardinal signs of inflammation taught in pathology are *calor, rubor, dolor, tumor and function lasso* or heat, redness, swelling and loss of function. It is prudent to remember that inflammation is the bodies response to many different situations. The young physician often confuses inflammation with infection and may wonder why his antibiotics have no effect in superficial thrombo-phlebitis, or similarly scratch her chin when they lack effect in the acute red foot of gout.

The systemic inflammtory response syndrome (SIRS) is a relatively new concept in surgery. It comes from work on sepsis, adult respiratory distress syndrome (ARDS) and multi organ dysfunction syndrome (MOF). Several different stimuli including, trauma (surgery), lipopolysaccharide and ischaemia-reperfusion (trauma, or vascular surgery) will result in an inflammatory response. This response or these responses are aimed at being beneficial but when exaggerated are actually lethal.

The importance of SIRS and MOF is that they are the leading cause of death in the patient in the surgical intensive therapy unit. A patient who survives a nasty trauma, burns or complicated extensive surgery may subsequently succumb to MOF. If the prognosis for these patients is to be improved then SIRS must be understood and novel therapies developed.

↗ **Rangel-Frausto et al, have nicely demonstrated a step-wise progression in mortality in patients based upon the severity of surgical sepsis [RFPC$^+$95].**

## 2.1 SIRS

↗ **The definition of SIRS has varied and most accepted definition is that given by the American College of Chest Physicians (ACCP)/ Society of Critical Care Medicine (SCCM)[Ame92], outlined in table 1.**

In simple terms SIRS is defined as a syndrome characterised by the presence of two or more variables. This definition is fairly wide and all encompassing, indeed any sick surgical patient almost automatically has SIRS.

## 2.2 Infection

Infection is defined as a microbial process characterised by an inflammatory response to the presence of microorganisms or the invasion of normally sterile host tissue by these organisms.

## 2.3 Sepsis

Sepsis is defined as SIRS with a microbial origin.

## 2.4 Severe Sepsis

Sepsis associated with organ dysfunction, hypoperfusion or hypotension; hypoperfusion and perfusion abnormalities may include, but are not limited to, lactic acidosis, oliguria or an acute alteration in mental status.

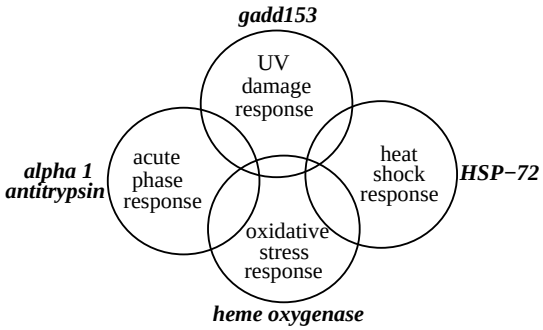


Figure 2: In complex organisms, programs of stress genes expression (circles) overlap with one and another, with the exception of the heat shock and acute-phase responses. Genes characteristic of the four responses are shown in italics. HSP72, heat shock protein-72 kilodalton molecular weight. [Buc94]

## 2.5 MOF

MOF is defined as failure to maintain healthy function without support.

## 2.6 Septic shock

Septic shock, is sepsis with hypotension, despite adequate resuscitation with fluids, along with the presence of perfusion abnormalities

# 3 Basic principles

At the cellular level the cell has only a few basic responses to external stresses[Buc94]. These are the heat shock response, the oxidative stress response and the ultraviolet damage response. A fourth response termed the acute phase response is only seen in multicellular organisms and may have developed to coordinate the response of a variety of cell types to an external threat, see figure 2. Endothelial cells, macrophages (monocytes) and epithelial cells are involved in this complex response. Exaggeration of the acute phase response results in the sepsis-syndrome which is the predecessor of multi-organ failure.

## 3.1 Leucocyte endothelial interactions

One of the responses of the body to injury is to recruite leucocytes to the site of injury. The accumulation of leucocytes in damaged tissues depends upon the interaction of leucocytes with the endothelium in the damaged tissue, see figure 3. There is a continuum between the stages of rolling, adherence and transmigration.

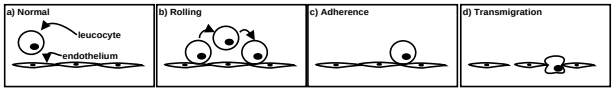


Figure 3: Schematic representation of the stages of migration of leucocytes from the circulation to an area of injury observable on microscopy. a) denotes the normal situation. In b) the leucocytes are seen to roll along the endothelium, they appear ‘sticky’. In c) the leucocytes are tightly adherent to the endothelium and in d) they migrate through the endothelium into the interstium. In the transmigration stage there are gaps formed between the endothelial cells and in the underlying basment membrane.

Most of the manifestations of inflammation in a tissue, (leucocyte accumulation, filtration of fluid and protein) only occur in the *post capillary venule*.

### 3.1.1 Rolling

The first step in the movement of the leucocyte to the injured tissue involves the rolling process. In this the leucocyte is seen to *roll* along the epithelium, this behavior is not seen in healthy tissues.

Rolling may be identified on intra-vital microscopy by looking at the leucocyte velocity. In a 30 $\mu$ post capillary venule, red blood corpuscules travel at about 1–3 mm $s^{-1}$. Leucocytes roll at velocities between 5–300 mm $s^{-1}$ with the most common velocity range of 20–60 mm $s^{-1}$.

The rolling process is believed to be due to the interaction between molecules expressed on leucocytes and the endothelium. E and P selectins are expressed on the endothelium. These interact with L-selectin and the carbohydrate derivative sialyated Lewis on the leucocyte. Both sialyated Lewis and L-selectin are constitutively expressed. It is believed that L-selectin interacts with E and P selectin via sialyated carbohydrate moieties on these molecules.

P selectin is released from the Weibel-Palade body in the endothelial cell on stimulation by hypoxia, free radicals, peroxides, histamine and thrombin. Endotoxin, IL-1$\beta$ and TNF-$\alpha$ induce the expression of E-selectin

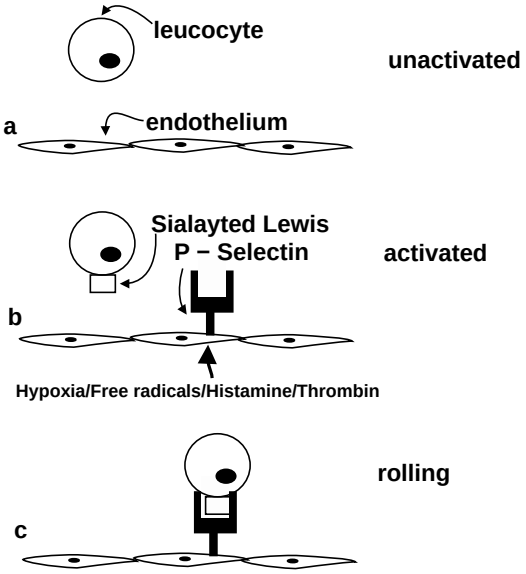


Figure 4: Schematic representation of the molecular mechanism of rolling in leucocytes in post capillary venules. a) shows the unactivated state with no expression of selectins on the leucocyte of the endothelium. b) shows the activated state with P-selectin expressed on the endothelium, P-selectin is released from the Weibel-Palade body in endothelial cells. Expression is induced by hypoxia, free radicals, histamine and thrombin. In c) the leucocyte has grabbed on to the endothelium with binding of sialyated Lewis to P-Selectin. The binding is not very tight as the leucocyte may pop off again with the appearance of rolling on microscopy.

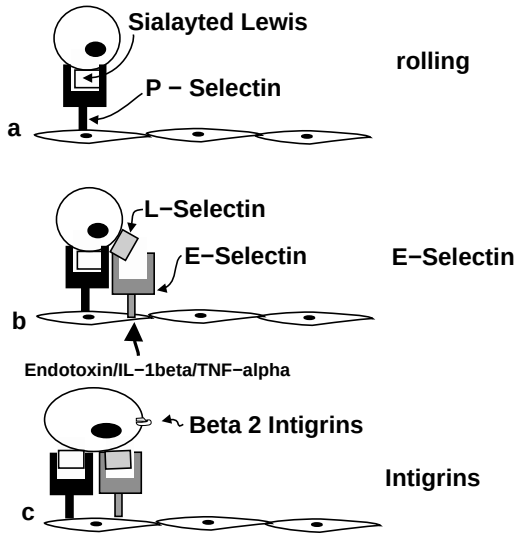


Figure 5: Schematic representation of the molecular mechanism of leucocyte adherence in post capillary venules. a) shows the rolling leucocyte with expression of P-Selectin on the endothelium which interacts with sialyated Lewis. b) shows induced expression of E- Selectin by endotoxin, IL-1$\beta$ and TNF$\alpha$. In c) the leucocyte has grabbed tightly onto the endothelium When E-Selectin on the endothelium is bound to L-Selectin on the leucocytes there is increased affinity of the leucotye for ICAM-1 and like molecules presumabley due to the induction of the expression of $\beta_2$ intigrins on the leucocyte.

on the endothelium. P-selectin interacts with sialyated Lewis and E-selectin interacts with L-selectin. The interaction is visible as *rolling*, see figure 4. It is felt that interaction with E-selectin plays a part in adherence as well as rolling.

L-selectin acts as a ligand for both E and P-selectins. Following capture of the leucocyte on the endothelium L-selectin is rapidly shed from the leucocyte on stimulation by LTB4, C5a, TNF-$\alpha$, IL-1 and PAF.

### 3.1.2 Adhesion

During inflammation leucocytes undergo various morphological changes associated with adherence, firstly they adopt a more spherical shape, then they roll, then they become stationary and flatten. During this process the membrane takes on a ruffelled appearance and there is a change in membrane receptors.

Binding of E-selectin with L-selectin results in very slow rolling and the onset of adherence. Tight adherence of leucocytes is thought to occur only after rolling. Following the interaction of E and L selectin, interaction of $\beta_2$ integrins (LFA-1, Mac-1) on leucocytes with ICAM-1 on the endothelium and the $\beta_1$ integrin (VLA-4) with VCAM-1 on the endothelium are thought to result in adherence of leucocytes onto the endothelium. It has been noted that binding of E to L selectin results in increased binding of $\beta_2$ integrins to ICAM-1.

### 3.1.3 Transmigration

Following a period of tight adherence, the leucocyte is seen to extend pseudopodia and move into the interstitium. Requirements for transmigration include, endothelial cell activation, expression of adhesion molecules, production of inflammatory mediators, cytoskeletal reorganization and alterations in membrane fluidity.

The critial molecular interations appear to be between integrin to ICAM-1 binding and integrin to VCAM-1 binding. Monoclonal antibodies against CD11/CD18 receptors on the leucocyte and ICAM-1 on the endothelium halt transmigration. L and E selectins do not play a role.

If a chemoattractant is present the leucocyte moves from the subendothelial space towards the chemoattractant.

## 3.2 Endothelial permeability

The barrier function of the endothelium is impaired in inflammatory states with the accumulation of protein rich fluid in the interstitium. Changes in permeability in the endothelium are linked to leucocyte adhesion as outlined above.

The three ways by which solutes reach the interstitium are diffusion, vesicular transport and between the endothelial cells. Small solutes pass through the endothelium by simple diffusion. Larger solutes are taken up by the endothelial cell in vesicles which are transported across the cell. There are specialized junctions between the endothelial cells which are altered to allow leucocytes and large solutes through. Stimulation by pro inflammatory mediators such as histamine and thrombin results in retraction and separation of endothelial cells for a period. The result is the accumulation of protein rich fluid in the interstitium and oedema.

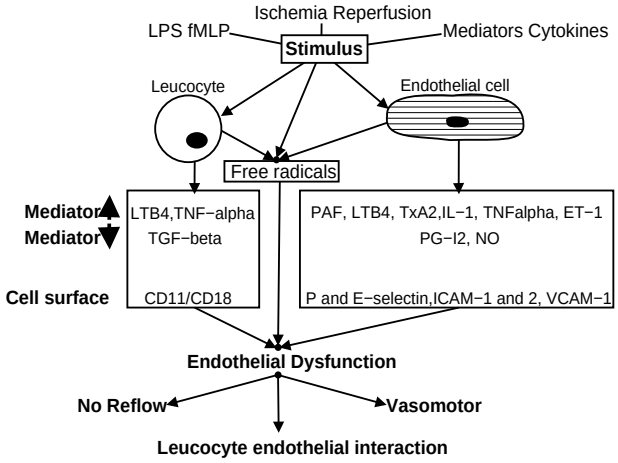


Figure 6: Patho-physiology of microvascular dysfunction in ischemia-reperfusion injury , endotoxemia, diabetes, immunological rejection and dsylipidaemia. External stimuli result in release of free radicals and endothelial and/or leucocyte activation with alterations in inflammatory mediators, cytokines and expression of integrins, selectins and members of the immunoglobulin super-family. This is manifest as endothelial dsyfunction (no-reflow phenomenum, increased leucocyte adhesion and migration and vasomotor dsyfunction).

Separation of the endothelial cells is reversible over a few minutes.

In ischemia reperfusion injury there is a temporal relationship between the migration of leucocytes and the transfer of albumin. The more leucocytes adhering and migrating the greater the amount of albumin transfer and monoclonal antibodies that attenuate leucocyte adhesion and migration also reduce albumin leakage.

The vascular leak syndrome, which occurs clinically in patients administered large doses of IL-2 is due to increased endothelial premeability.

## 3.3 Endothelial dysfunction

Endothelial dysfunction is present when there is an inappropriate alteration with respect to perservation of organ function. Some of the clinical areas where endothelial function and dysfunction are important are ischemia-reperfusion injury, endotoxemia, diabetes, immuno-logical rejection and dyslipidaemia.

The core pathophysiology of microvascular and endothelial dysfunction in these conditions is outlined in figure 6. The pathophysiology is similar despite the stimulus so ischaemia reperfusion injury will be used to describe the process.

## 3.4 Ischaemia-reperfusion injury

While ischemia alone results in tissue damage, reperfusion of ischemic tissue results in further damage both to the tissue and systemically to other organs.

The concept of reperfusion accelerating damage in local tissues may be true in the sense that damage is increased or it may simply represent acceleration in normal patho-physiological processes with restoration of blood flow. For instance, patients with occlusive arterial disease of the lower limbs with critical ischemia, who undergo limb saving arterial reconstructive surgery, will usually demonstrate rapid demarcation of ischaemic tissues that may not have been so obvious prior to surgery. The appearance of the tissues would eventually take on the same appearance if time permitted.

Examination of tissues with ischemia-reperfusion injury show evidence of endothelial cell dysfunction with respect to oedema, and an infiltration of leucocytes.

Ischemic tissues however must, be exposed to molecular oxygen on reperfusion to manifest tissue injury. This suggests that reperfusion with higher than normal (for the tissue in question) oxygen results in generation of reactive oxygen species that cause further tissue injury. Further research suggests that activated leucocytes also contribute to the damage. 'Spilling over' of soluble mediators and activated leucocytes results in remote systemic damage.

## 3.5 Reactive oxygen metabolites

Reactive oxygen metabolites or species refer to molecules containing an oxygen atom with a partially empty outermost electron shell, these oxygen atoms are highly reactive and result in damage to other adjacent molecules. Such metabolites include the superoxide anion radical, hydroperoxyl radical, hurdoxly radical, hydrogen peroxide and hypochlorous acid.

In parynchymal and endothelial cells xanthine oxidase is the main enzyme responsible for the generataion of free oxygen radicals, whilst in leucocytes NADPH oxidase is responsible.

$$Hypoxanthine \xrightarrow{\text{Xanthine Oxidase}} O^{\cdot}$$

$$O_2 \xrightarrow{\text{NADPH Oxidase}} Superoxide^{\cdot}$$

Oxygen free radicals produced from xanthine oxidase have a very short half life (seconds). Thus when the hypoxanthine substrate is exhausted generation of free radicals by xanthine oxidase ceases. In neutrophils production of oxygen fee radicals is unlimited provided $O_2$ is available.

Production of oxygen free radicals in ischemia-reperfusion injury has been shown using electron spin resonance spectroscopy and trapping. By products of the produciton of oxygen free radicals such as peroxidated lipids are indirect evidence of the previous production of oxygen free radicals. Changes similar to those seen in ischemia-reperfusion injury have been seen on exposure of the tissues to oxygen free radicals. Treatment of tissues with agents that limit the production of oxygen free radicals (xanthine oxidase inhibitors such as allopurinol) or that scavange generated oxygen free radicals (superoxide dimutase, catalase, taurine and dimethly sulfoxide) attenuates ischemia-reperfusion injury.

The mechanism of injury of oxygen free radicals is felt to be due to rapid reactions of the free radicals with other molecules resulting in, DNA nicking, lipid membrare peroxidation, crosslinking and degredation of proteins. Radicals also result in attraction and activation of inflammatory cells and increased expression of CD11/CD18 ($\beta_2$ integrins) on the surface of lecocytes, P-selectin and ICAM-1 on endothelial cells, thereby promoting leucoctye adhesion and transmigration.

## 3.6 Activated leucocyte

The three pathways by which neutrophils may cause tissue damage in ischaemia-reperfusion injury are;

1. production of oxygen free radicals
2. release of active agents (lipo-oxygenase products, TNF-$\alpha$, proteolytic enzymes)
3. physical obstruction of capillaries and venules

### 3.6.1 Oxygen free radicals

$$O_2 \xrightarrow{\text{NADPH Oxidase}} Superoxide^{\cdot}$$

$$Superoxide^{\cdot} + H_2O_2 + Cl \xrightarrow{\text{Myeloperoxidase}} HypochlorousAcid$$

NADPH Oxidase is membrane bound and catalyses the formation of Superoxide anion, this reactive anion rapidly reacts with hydrogen peroxide to form the relatively stable hypochlorous acid. This reaction is catalysed by myeloperoxidase (MPO) which is stored in granules in the neutrophil.

### 3.6.2 Release of active agents

The lipooxygenase pathway product of arachadinoic acid leucotriene-B4 (LTB4), is released by activated leucocytes, it is a potent chemoattractant and induces ahesion and trans migration in venules.

Tumor necrosis factor-$\alpha$ (TNF-$\alpha$) is one of the most pro-inflammatory cytokines known. It is produces by the activated leucocyte.

Elastase, collagenase and gelatinase are released, these play a role in making a hole in the basement membrane to permit transmigration of leucocytes. They also function to degrade the interstitial matrix.

### 3.6.3 Physical occlusion of capilleries

Accumulation of activated leucocytes that are sticky, results in blockage of capilleries and venules. This results in the no-reflow phenomenum with impairment of the microcirculation and tissue death.

## 3.7 Inflammatory mediators and cytokines

Cytokines are soluble glycoproteins or small peptides released by living cells such as parenchyma, endothelium and leucocytes. Cytokines having an effect on inflammation may be conveniently subdivided into pro and anti inflammatory.

Inflammatory mediators are larger molecules than cytokines, they are proinflammatory and include platelet activated factor (PAF), leukotriene B4 (LTB4), C5 and thrombin.

### 3.7.1 Inflammatory mediators

**PAF** PAF is a potent pro inflammatory mediator that is formed from a membrane phospholipid in a variety of cells including, platlets, endothelial cells and leucocytes. There are three mechanisms by which PAF augments inflammation.

Firstly, there is a receptor on the surface of leucocytes for PAF. Binding of PAF results in increased expression of CD11/CD18 on the surface of the leucocyte thereby augmenting adhesion and transmigration of leucocytes. Transmigration also increases microvascular permeability.

Secondly, PAF stimulates the formation of oxygen free radicals and the release of myeloperoxidase and lactoferrin.

And finally, hypoxia stimulates endothelial cells to release PAF with further leucocyte adhesion and ICAM-1 expression.

**LTB4** LTB4 is a product of arachadinoic acid via the lipo-oxygenase pathway. It is a potent chomoattractant. It enhances vascular permeability and leucocyte adhesion. It also promotes release of free oxygen radicals and proteolytic enzymes from leucocytes.

### 3.7.2 Cytokines

These soluble glycoproteins and peptides are released from living cells such as mocrophages and lymphocytes in response to noxious stimuli such as ischemia, endotoxin, physical and chemical injury. They may be conviently subdivided into pro and anti- inflammatory cytokines.

Pro-inflammatory cytokines are predominantly released by macrophages and up regulate inflammatory reactions. Anti-inflammatory cytokines are predominantly released by T lymphocytes and are involved with down regulating the inflammatory response. Transforming growth factor $\beta$ has both pro and anti inflammatory effects.

**Pro-inflammatory cytokines** These inlcude IL-1, IL-2, IL-6, IL-8 and TNF-$\alpha$. Administration of antagonists to these agents such as IL-1ra (IL-1 receptor antagonist), or monoclonal antibodies against TNF-$\alpha$ and soluble TNF-receptor block acute and chronic inflammatory responses in animal models of inflammatory disease.

IL-1 and TNF-$\alpha$ are the main mediators involved in endotoxic shock.

Up regulation of the inflammatory response is also effected by IL-11, IF-$\gamma$ and by members of the chemokine superfamily.

Production of pro-inflammatory cytokines is reduced by the anti-inflammatory cytokines (IL-4, IL-10, IL-

13). And, production of IL-1, IL-6 and TNF-$\alpha$ is inhibited by TGF-$\beta$.

**IL-2** The main source of endogenous IL-2 is the T-cell. The targets are lymphocytes with T and B cell activation and NK cell activation. It has been used to treat a number of malignancies such as metastatic melanoma and renal cell carcinoma. However, its clinical usefulness appears to be limited by dose-dependent toxicity manifest as the vascular leak syndrome. It also initially increases the number of T cells which then fall dramatically. The vascular leak syndrome is thought to be due to IL-2 induction of leucocyte adherence and transmigration leading to a loss in endothelial barrier function.

**TNF-$\alpha$** TNF-$\alpha$ appears to be very important. It is release systemically early on and is responsible for cachexia, the old description of TNF-$\alpha$ is *cachectin*. Macrophages, Kupfer cells, natural killer cells and lymphocytes produce TNF-$\alpha$. The half life is 16 minutes. Plasma levels normally do not exceed 35 picogrammes/Litre. A slight increase in TNF-$\alpha$ induces cell proliferation and differentiation and regulation of cytokine interaction. Higher levels result in tissue remodelling, inflammation and cytotoxicity. In mice antibodies against TNF-$\alpha$ or knockout of the TNF-$\alpha$ gene confers resistance to the lethal effects of endotoxin.

**Anti-inflammatory cytokines** These include IL-4, IL-10 and IL-13. IL-10 is capable of protecting mice from endotoxic shock.

**TGF-$\beta$** As outlined above TGF-$\beta$ inhibits the production of most monokines and lymphokines. In this respect it is anti-inflammatory by reduction in release of IL-1, IL-6 and TNF-$\alpha$.

TGF-$\beta$ besides its anti-inflammatory effects is pro inflammatory. It acts as a chemoattractant for neutorphils, T cells and unactivated monocytes.

Systemic TGF-$\beta$ is anti-inflammatory while local TGF-$\beta$ is pro-inflammatory. It has an important role to play in scar formation and promotes neovascularisation and proliferation of connective tissue cells.

# 4 Endotoxemia

Endotoxin is lipopolysaccharide (LPS) a component of the other membrane of most gram negative bacteria. Administration of endotoxin results in the systemic inflammatory response syndrome and subsequent multiple organ failure.

LPS directly induces upregulation of $\beta_2$ integrins on leucocytes and expression of P and E selectins and ICAM-1 on endothelial cells. This enhances leucocyte - endothelial cell interactions with adhesion and transmigration. LPS also induces secretion of free oxygen radicals and pro-inflammatory cytokines (IL-1 and TNF-$\alpha$).

Endotoxic shock is a lethal inflammatory state mediated by TNF-$\alpha$ and IL-1. Mice may be protected from endotoxic shock by IL-10.

# 5 Causes of SIRS

SIRS represents a final-common pathway for a variety of insults. It is not just due to infection. The major causes of SIRS are;

- Infection
- Trauma
- Burns
- Pancreatitis
- Hypovolemic or haemmorhagic shock

# 6 Pathophysiology of SIRS

There are three clinical stages of SIRS described from 1–3. Stage one is in the main a local response with increased recruitment of neutrophils and monocytes. In stage two there is a spill over of activated cytokines into the circulation but balance is retained by the dampening effects of anti-inflammatory cytokines. In stage three balance is lost and there is a massive pro-inflammatory swing. There is generalised loss in capillary barrier function and generalised organ dysfunction.

In severe SIRS there is systemic vasodilatation (histamine, prostaglandins) with a fall in systemic vascular resistance and blood pressure. Widespread leaky capillaries lead to extensive third space extra-cellular fluid loss with hypovolemia. Myocardial function is impaired due to leucocyte induced myocardial cell dysfucntion and to the effects of myocardial hypo-perfusion.

The terminal state is characterised by severe hypotension, refractory to fluid and inotropic support. There is severe worsening lactic acidosis due to impaired tissue perfusion.

# 7 Activated Protein C and sepsis/SIRS

It has been noted that abnormalities of coagulation accompany SIRS and sepsis. In many patients there appears to be a disseminated intravascular coagulation and sepsis is one of the main causes of elevations in D-dimers and in fibrinogen degradation products.

Children with meningococcal septicaemia often develop pupura, this is termed purpura fulminans. In these children low levels of activated protein C are associated with a poor prognosis. A recombinant human activated protein C has been developed and has been used in clinical trials. It has been shown to be beneficial in patients with sepsis.

↗ **Human recombinant activated protein C has been shown to reduce mortality in adults with sepsis in a randomised controlled trial. [DLJ+03]**

The downside to administrating activated protein C is that it is a natural anticoagulant and its use is associated with a low but significant risk of serious bleeding. Studies in the United States suggest that the cost of using recombinant activated protein C is about 160 thousand dollars per life saved.

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