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Review

The endothelin system in septic and endotoxin shock

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Abstract

The view of the endothelium as a passive barrier has gradually changed as a number of endothelium-derived substances have been discovered. Substances like nitric oxide, prostaglandins and endothelins have potent and important properties, involving not only the circulation as such but also the response to stimuli like inflammation and trauma. The endothelin system, discovered in 1988, has not only strong vasoconstrictor properties, but also immunomodulating, endocrinological and neurological effects exerted through at least two types of receptors. Septic shock, a condition with high mortality, is associated with vast cardiovascular changes, organ dysfunction with microcirculatory disturbances and dysoxia. In the experimental setting, endotoxaemia resembles these changes and is, as well as septic shock, accompanied by a pronounced increase in plasma endothelin levels. The pathophysiology in septic and endotoxin shock remains to be fully elucidated, but several studies indicate that endothelial dysfunction is one contributing mechanism. Activation of the endothelin system is associated with several pathological conditions complicating septic shock, such as acute respiratory distress syndrome, cardiac dysfunction, splanchnic hypoperfusion and disseminated intravascular coagulation. Through the development of both selective and nonselective endothelin receptor antagonists, the endothelin system has been the object of a large number of studies during the last decade. This review highlights systematically the findings of previous studies in the area. It provides strong indications that the endothelin system, apart from being a marker of vascular injury, is directly involved in the pathophysiology of septic and endotoxin shock. Interventions with endothelin receptor antagonists during septic and endotoxin shock have so far only been done in animal studies but the results are interesting and promising.

Keywords: Endothelin-1; Endothelin receptor antagonism; Septic shock; Endotoxin

1. The endothelin system

Endothelin was first isolated by Yanagisawa et al. (1988). They identified the endothelium-derived contracting factor as a 21-amino acid peptide from porcine aortic endothelium cells. The peptide possessed strong vasoconstrictive properties, at least ten times as potent as those of angiotensin II. This peptide was named endothelin-1 and belongs together with endothelin-2 and endothelin-3, which were identified later, to the endothelin isopeptide family. All three peptides have been cloned from the human genome although the latter two peptides are less well studied (Inoue et al., 1989). The endothelin system is found in most mammals and also in fish and invertebrates, indicating its importance in evolutionary history. Endothelin-1 is mainly produced by the vascular endothelium and is secreted constitutively thereby participating in the regulation of vascular tone (Haynes and Webb, 1994; Weitzberg, 1994). Endothelin-2 may be found in the intestine and endothelin-3 in the lung, central nervous system and intestine (Rubanyi and Polokoff, 1994). Endothelin-1 synthesis is not limited to the endothelium but it is also produced in a broad variety of other cells, including vascular smooth muscle cells (Resink et al., 1990), mucosal epithelium (Takahashi et al., 1990), macrophages (Ehrenreich, 1990), mast cells (Ehrenreich et al., 1992), cardiomyocytes (Suzuki et al., 1993), neurones (Giaid et al., 1989), tracheal epithelium (Endo et al., 1992), renal medulla (Endo et al., 1992), hepatic sinusoids and Kupffer cells (Liu et al., 1997).

Endothelin-1, the predominant form in human plasma and tissue (Opgenorth, 1995), is synthesised as a prepro peptide, containing 212 (203, porcine) amino acids. The prepro peptide is cleaved by endopeptidases to big endothelin-1 (pro endothelin-1), a 38-amino acid peptide (Rubanyi and Polokoff, 1994) (see Fig. 1). Big endothelin-1

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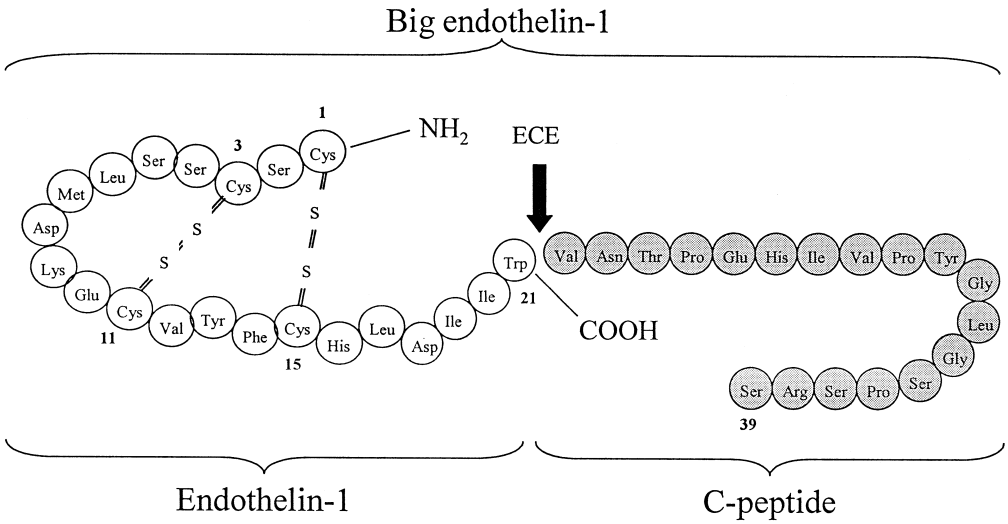

Fig. 1. Amino acid sequence of Big endothelin-1: Big endothelin-1 (pro endothelin-1) containing 39 amino acids (porcine) is converted by endothelin converting enzyme (ECE) to the 21-amino acid peptide, endothelin-1, and the inactive C-peptide (big endothelin-1 22-39), containing 18 amino acids.

has vasoconstrictive properties, although two orders of magnitude weaker than those of endothelin-1 (Kimura et al., 1989). A zinc metalloprotease, endothelin-converting enzyme, trims big endothelin-1 to form endothelin-1 and an inactive C-peptide (Ottoson Seeberger et al., 1998). Three isoforms of ECE with different selectivity for the propeptides are known (Xu et al., 1994; Emoto and Yanagisawa, 1995). Secretion of endothelin-1 occurs in a polar fashion, with approximately 80% towards the abluminal side of the vessel (Wagner et al., 1992). Factors known to stimulate endothelial cells to release endothelin-1 include low shear stress (Yoshizumi et al., 1989), hypoxia (Kourembanas et al., 1991), endotoxin (Sugiura et al., 1989), tumour necrosis factor α (TNF-α) (Marsdeen and Brenner, 1992), interleukin-1 (Maemura et al., 1992), transforming growth factor-β (Kurihara et al., 1989), adrenalin (Yanagisawa et al., 1988), thrombin and angiotensin II (Emori et al., 1989). Inhibition of endothelin-1 release is seen in response to prostacyclin (Razandi et al., 1996), nitric oxide (Boulanger and Luscher, 1990), atrial natriuretic peptide (Hanehira et al., 1997), and heparin (Hanehira et al., 1997) among others.

Endothelin-1 has a plasma half-life of 1–2 min in both human and pig (Pernow et al., 1989b; Hemsen, 1991; Weitzberg et al., 1993). In addition, a second half-life of plasma disappearance has been calculated at about 35 min in pig and human (Weitzberg et al., 1993, 1995b). The pulmonary vascular bed is an important location for plasma clearance of endothelin-1 (Änggård et al., 1989), but both the kidney and the liver have been shown to participate (Weitzberg et al., 1991a).

Under normal conditions, endothelin-1 is considered to act as a paracrine mediator, and effects are mediated by at least two types of receptors, the endothelin ET_A and the endothelin ET_B receptors. Both of these G-protein-coupled receptors have been cloned and characterised (Arai et al., 1990; Sakurai et al., 1990), displaying differences in agonist rank order of potency. The endothelin ET_A receptor has higher affinity for endothelin-1 and endothelin-2 than for endothelin-3 whereas the endothelin ET_B receptor has equal affinity for all three isopeptides. The endothelin ET_B receptor exists as at least two subtypes, the endothelin ET_{B1} receptor, located on the endothelium and the endothelin ET_{B2} receptor, expressed on smooth muscle cells. The endothelin ET_A receptor, located on the smooth muscle cell, together with the endothelin ET_{B2} receptor mediates contraction and endothelin ET_{B1} receptor activation causes relaxation through the release of nitric oxide and prostacyclin (De Nucci et al., 1988). Other functions attributed to the endothelin ET_{B1} receptor include autoinduction of prepro endothelin-1 transcription and plasma endothelin clearance (Iwasaki et al., 1995; Dupuis et al., 1996), suggested to be mediated by a splice variant of this receptor (Elshourbagy et al., 1996). Furthermore, subdivision of the endothelin receptors according to different sensitivity to various antagonists/agonists has been suggested (MacLean et al., 1998; Miasiro et al., 1998). In addition, a cross-talk between the endothelin ET_B receptors has been described, involving increased endothelin ET_B receptor sensitivity during endothelin ET_A receptor antagonism (Ozaki et al., 1997). It is, therefore, obvious that the physiological response to endothelin receptor activation is complex and varies depending on the local receptor population.

Endothelin receptor stimulation results in activation of several intracellular second messengers (see Fig. 2). Activated endothelin receptor–ligand complexes are rapidly internalised by endocytosis and remain in complex for

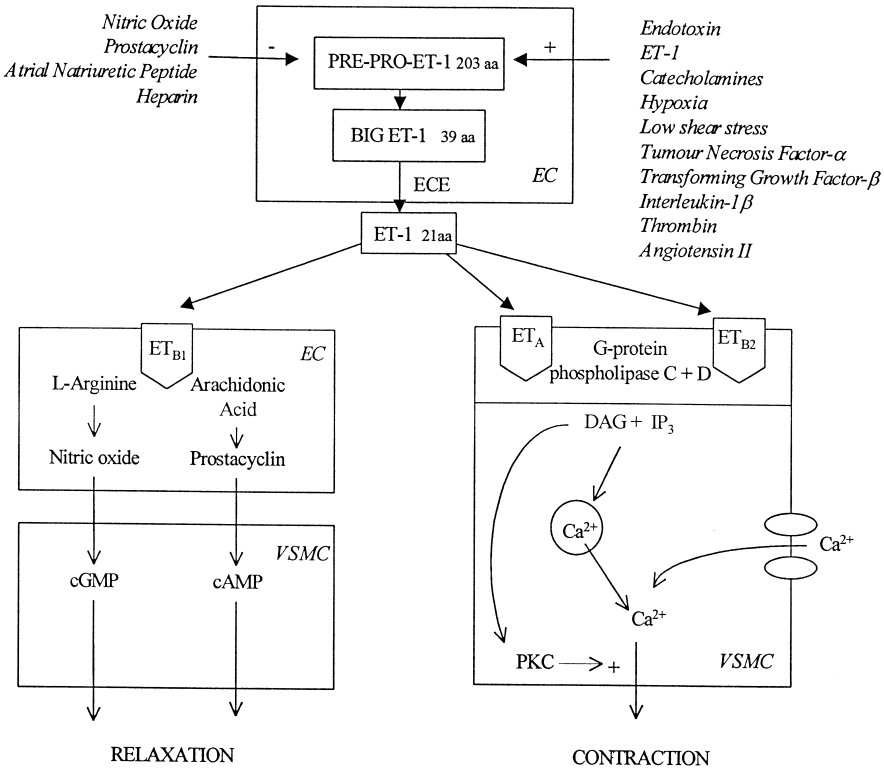

Fig. 2. Brief schematic overview of the endothelin system: aa = amino acid, cAMP = cyclic adenosine monophosphate, cGMP = cyclic guanosine monophosphate, DAG = diacylglycerol, EC = endothelial cell, ECE = endothelin converting enzyme, IP_3 = inositol triphosphate, PKC = protein kinase C, VSMC = vascular smooth muscle cell.

hours, which could explain the prolonged vasoconstriction caused by endothelin-1 (Chun et al., 1995). After internalisation, the receptors have been shown to be recycled as they are externalised (Marsault et al., 1993). In vitro, endothelin degradation is mediated by the metalloendopeptidase, neutral endopeptidase 24.11, but in vivo findings do not unambiguously support the involvement of this peptidase in endothelin degradation (Scili et al., 1989; Asaad et al., 1993).

Despite the relatively short time that has elapsed since the discovery of the endothelin system, there has been an impressive number of studies concerning endothelin. This has largely been a result of the development of specific endothelin receptor agonists and antagonists. Today, more than 80 different antagonists are known and some of them are currently being introduced in clinical trials (Battistini and Dussault, 1998).

2. Endothelin in sepsis

Endothelin has been postulated to participate in the pathogenesis of a number of diseases, such as myocardial infarction (Tomoda, 1993), Raynaud's disease (Cimminiello et al., 1991), bronchial asthma (Mattoli et al., 1991), pulmonary hypertension (Giaid et al., 1993), renal failure (Benigni et al., 1993), gastric ulceration (Matsumaru et al., 1997) and cerebral vasospasm following subarachnoidal haemorrhage (Clozel and Watanabe, 1993). Moreover, in clinical trails, endothelin antagonism proved to be effective and safe in the treatment of hypertension (Krum et al., 1998) and severe heart failure (Sütsch et al., 1998).

Among the pathophysiological conditions known to involve the endothelin system, sepsis presents with the highest plasma levels of endothelin (Battistini et al., 1996). The possible involvement of the endothelin system in human septic shock is further supported by a clear correlation between endothelin plasma levels and morbidity and mortality in septic patients (Pittet et al., 1991; Weitzberg et al., 1991b). In the experimental setting, endotoxin induces the expression of prepro endothelin-1 mRNA in the lung and heart (Hemsen, 1991; Kaddoura et al., 1996), and increased plasma endothelin-1 levels are seen in various species during endotoxaemia (Pernow et al., 1989b; Chou et al., 1995; Kaszaki et al., 1997). Infusion of endothelin-1 to humans causes cardiovascular changes in part resembling those seen during sepsis i.e. decreased cardiac output and vasoconstriction in the pulmonary, renal and splanchnic circulation (Weitzberg et al., 1991a, 1993). Endothelin-1 has also been suggested to contribute to dysfunction of several vital organ systems in septic shock (Pittet et al.,

1991; Ruetten and Thiemermann, 1996; Oldner, 1999; Wanecek, 1999).

2.1. Cardiovascular manifestations

In the heart, the predominant endothelin isopeptide is endotheli-1 (Plumpton et al., 1996) and both endothelin ET_A and endothelin ET_B receptor mRNA are found in human atrial and ventricular myocardium, the conducting system, endocardial cells and in porcine coronary arteries (Molenaar et al., 1993; Awane Igata et al., 1997). In vivo effects of endothelin-1 on the heart include massive coronary vascular constriction (Ezra et al., 1989), decreased cardiac output (Lerman et al., 1991), and arrhythmias (Horkay et al., 1998). In vitro, a positive chronotropic effect has been observed (Ishikawa et al., 1988b). Furthermore, a positive inotropic effect (Ishikawa et al., 1988a), suggested to be mediated by the endothelin ET_B receptor (Beyer et al., 1995), has been described. However, these results, obtained mainly in vitro, have not been uniformly confirmed in in vivo studies (Kiely et al., 1997). Also, in human sepsis the evaluation of plasma endothelin-1 levels correlates inversely with the cardiac index (Pittet et al., 1991).

Exogenous endothelin-1 causes a rapid, transient, vasodilation followed by a sustained vasoconstriction (Yanagisawa et al., 1988), the former mediated by endothelin ET_{B1} and the latter by endothelin ET_{B2} and endothelin ET_A receptors. The vasoconstrictor effects are more pronounced in veins than in arteries (Cocks et al., 1989) and vary between different vascular beds depending on the endothelin-receptor population (Lodge et al., 1995). Endothelin-1 is also a potent modulator of regional perfusion and may well contribute to the maldistribution of blood flow seen in septic shock. As compared to more vital organs, skeletal muscle perfusion seem to be less affected by endothelin-1 (Gasic et al., 1992; Weitzberg et al., 1993).

The mechanisms behind the cardiac dysfunction during endotoxaemia and sepsis have been the object of a vast number of studies, but they still remain to be fully elucidated. Earlier reports from animal studies implicating coronary vasoconstriction as the main mechanism have later been questioned (Peyton et al., 1976; Dhainaut et al., 1987; Cunnion and Parillo, 1989). Despite this, there are indications of a depressed subendocardial blood flow during endotoxaemia (Goldfarb et al., 1986). Recent experimental studies indicate that hypoxic regions in the myocardium (Ince and Sinaasappel, 1999) and troponin I levels are increased in human septic shock, indicating myocardial damage (Turner et al., 1999). Endothelin-1 is a most potent coronary vasocostrictor (Ezra et al., 1989), an effect that is mediated by both endothelin ET_A and endothelin ET_B receptors in the pig (Bonggwan et al., 1994; Awane Igata et al., 1997). The mixed endothelin receptor antagonist, bosentan, has been shown to increase coronary blood flow in a porcine model of endotoxin shock.

Nitric oxide and cytokines, such as TNF-α and interleukin-1, are released during sepsis and endotoxaemia and are suggested exert cardiodepressant effects (Brady, 1995; Kumar et al., 1996). Endotoxin-induced coronary vasoconstriction in rats depends on TNF-α-mediated endothelin-1 release (Hohlfeld et al., 1995) and antagonising endothelin receptors might, therefore, influence the effects of cytokines, such as TNF-α, modulating their cardiodepressant effects. Nevertheless, clinical studies aiming to prevent or oppose the effects of TNF-α, interleukin-1 and nitric oxide have so far not shown favourable effects on cardiac performance (Opal, 1995; Grover et al., 1999). Another suggested mechanism behind endotoxin and septic cardiac dysfunction is that leukocytes, when activated by contact with microbes, might exercise cardiodepressant effects (Granton et al., 1997). Leukocyte transit time increases in the coronary microcirculation during porcine endotoxaemia and endothelin-1 promotes leukocyte adhesion, an effect that could be counteracted by endothelin antibodies (Lopez Farre et al., 1993; Goddard et al., 1995). Endothelin ET_A receptor antagonism has been shown to protect against leukocyte-induced injury during ischemia/reperfusion (Gonon et al., 1998).

Direct actions of endothelin-1 on myocardial function have also been suggested, but these data are contradictory. Positive inotropic effects of endothelin-1 and, therefore, a possible negative inotropic effect of endothelin receptor antagonism, have been described in vitro (Ishikawa et al., 1988a; Mebazaa et al., 1993). The positive inotropic effect has been suggested to depend on an increase in myosin light chain-2 phosphorylation (Rossmanith et al., 1997). De Keulenaer et al. (1995) proposed this positive inotropic effect was endothelin ET_A receptor-mediated, whereas Beyer et al. (1998) in in vivo studies on rats, have suggested that this effect was mediated by the endothelin ET_B receptor.

However, in strong contrast to these studies, negative inotropic actions of endothelin-1 have been observed in vitro in rabbit atria and in vivo in both pig and human (Kaszaki et al., 1997; Kiely et al., 1997; Zhu et al., 1997). In line with these results, several studies have shown that mixed endothelin receptor antagonism induces a clear increase in cardiac output during endotoxaemia, suggesting that the net effect of endothelin during sepsis is severely cardiodepressive (Wanecek et al., 1997a, 1999b). Interestingly, diverse effects of mixed as compared to selective endothelin receptor antagonism have been demonstrated. In one study, selective endothelin ET_A receptor antagonism was without positive effects on cardiac performance and selective endothelin ET_B receptor antagonism was deleterious (Oldner et al., 1999). This may be explained by the fact that selective antagonism leaves an unblocked endothelin receptor population exposed to its agonist, the presence of cross-talk between the two types of receptors

(Mickley et al., 1997; Ozaki et al., 1997), various effects on afterload or coronary blood flow due to the existence of both endothelin ET_A and endothelin ET_B receptors with constrictor properties (Lodge et al., 1995; Zhang et al., 1998).

2.2. Pulmonary manifestations

Endothelin-1 is believed to play an important role in the regulation of pulmonary vascular tone, and pulmonary tissue is rich in endothelin-1-like immunoreactivity, localised to the vascular endothelium (Pernow et al., 1989a). In both pig and man, there are indications that the endothelin system participates in the regulation of pulmonary vascular tone under normal conditions (Weitzberg, 1994). Other cells within the lung that produce endothelin-1 include endocrine cells (Giaid et al., 1991), airway epithelial cells (Black et al., 1989; Mattoli et al., 1990) and macrophages (Ehrenreich, 1990). MRNA encoding both endothelin ET_A and endothelin ET_B receptors has been found in human pulmonary arteries. Although the endothelin ET_A receptors dominates (Davenport et al., 1995), constrictor endothelin ET_B receptors have been shown in pulmonary resistance arteries (Fukuroda et al., 1994; MacLean et al., 1994). The receptor population in peripheral lung tissue appears to vary between species and is also heterogeneously distributed throughout the vascular system (Goldie et al., 1996a; Higashi et al., 1997). Infusion of endothelin-1 in animal models affects pulmonary vessels differently, depending on the existing vascular tone in the pulmonary circulation, but the main vascular effects of endothelin-1 during pathological conditions within the lung are believed to be constrictor (Cassin et al., 1991; Goldie et al., 1996b). Endothelin-1 has also been shown to increase pulmonary microvascular permeability in experimental models (Helset et al., 1993). The effects of endothelin on pulmonary gas exchange are less studied and still need to be clarified. The pulmonary vascular bed clears the blood of circulating endothelin-1 under normal circumstances but, under certain circumstances, such as primary pulmonary hypertension, may become a net producer (Änggård et al., 1989; Stewart et al., 1991).

Since the lungs receive all returning venous blood, possibly containing toxins, inflammatory cells and inflammatory mediators, they are often involved in inflammatory processes. The pulmonary dysfunction during sepsis includes pulmonary hypertension, hypoxemia and low lung compliance and may progress to acute distress syndrome (Bigatello and Zapol, 1996). In this state with a mortality of around 50% (Krafft et al., 1996), the lung may become a net endothelin-1 producer contributing to increased plasma endothelin-1 (Druml et al., 1993; Langleben et al., 1993).

Endotoxin-induced pulmonary hypertension is a reproducible phenomenon seen in several animal models (Leeper Woodford et al., 1991; Booke et al., 1995). The pathophysiology includes a characteristics biphasic increase in mean pulmonary artery pressure and pulmonary vascular resistance index and is thought to involve different mediators, including cytokines, which lead to increased expression of adhesion molecules, leukocyte activation and endothelial damage resulting in endothelial oedema, vascular obliteration and vasoconstriction (Bigatello and Zapol, 1996). The involvement of both the cyclooxygenase pathway in the early phase, and the endothelin system in the late phase of endotoxin-induced pulmonary hypertension, has been

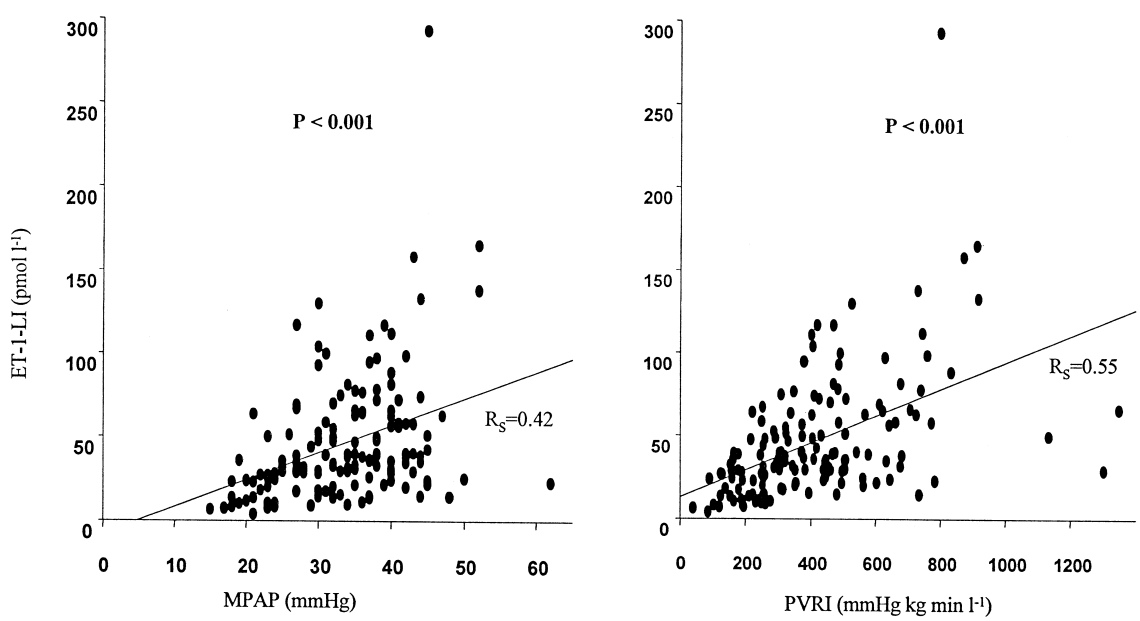

Fig. 3. Relationship between arterial plasma endothelin-1-like immunoreactivity (ET-1-LI) and mean pulmonary artery pressure (MPAP) and pulmonary vascular resistance index (PVRI): Endotoxaemic pigs ($n = 29$).

shown, and mixed as well as selective endothelin endothelin ET_A receptor antagonism can counteract the late changes in the experimental setting (Svartholm et al., 1989; Curzen et al., 1996; Wanecek et al., 1997a, b, 1999a). Also, a positive correlation between mean pulmonary artery pressure, pulmonary vascular resistance index and plasma endothelin-1 levels has been found (see Fig. 3) supporting the involvement of endothelin receptor stimulation in the late phase of endotoxin-induced pulmonary hypertension (Wanecek, 1999). Moreover, endothelin has been advocated as a major mediator of pulmonary hypertension in nonseptic conditions (Cody et al., 1992).

The similarities in effects on pulmonary vascular tone with selective endothelin ET_A receptor antagonism compared to those with nonselective endothelin receptor antagonism indicate that it is mainly endothelin ET_A receptor-mediated mechanisms that are involved in edotoxin-induced pulmonary hypertension. This is consistent with other results obtained under both nonendotoxic and endotoxic conditions, (Curzen et al., 1996; Holm, 1997). In spite of this, some authors have found endothelin ET_B-mediated pulmonary vasoconstriction in the rabbit and rat (Fukuroda et al., 1994; Sato et al., 1995). Actually, when comparing the relative reduction in mean pulmonary artery pressure in response to either nonselective endothelin-receptor antagonism or selective endothelin ET_A receptor antagonism seen in the studies from Wanecek (1999) (see Fig. 4), a possible involvement of the endothelin endothelin ET_{B2} receptor during endotoxin-induced pulmonary hypertension may be anticipated.

In addition to vascular effects, endothelin-1 has oedemagnic effects in the lung (Helset et al., 1994). Moreover, endothelin-1 also can increase the expression of neutrphil adhesive molecules (McCarron et al., 1993; Ishizuka et al., 1999) and promotes leukocyte migration into the alveoli (Filep et al., 1993; Helset et al., 1994). Significantly lower levels of protein and number of white blood cells in broncheo-alveolar lavage fluid after endotoxaemia are seen with mixed endothelin-receptor antagonism (Wanecek et al., 1997a), indicating a lower degree of permeability disturbance in the lungs.

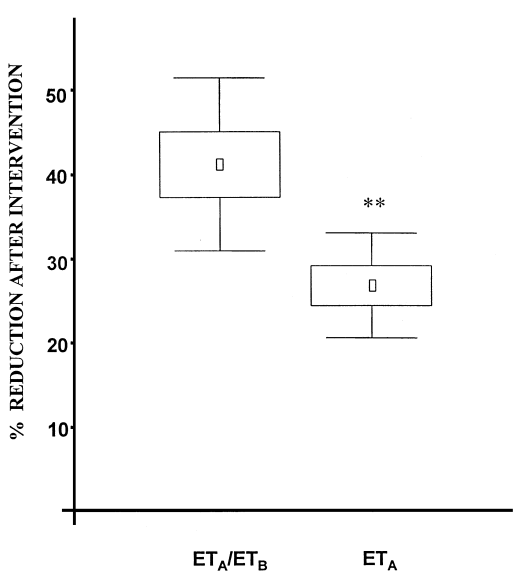

Fig. 4. Effect of selective and nonselective ET receptor antagonism on pulmonary artery pressure: Percent reduction of mean pulmonary artery pressure (MPAP) after intervention with either combined ET_A /ET_B receptor antagonism ($n = 21$) or selective ET_A receptor antagonism ($n = 8$). Data are presented as means (□), ± S.E.M. (box) and S.D. (bars).

2.3. Intestinal manifestations

Disturbances in intestinal perfusion are common findings in states of sepsis and systemic inflammation (Humer et al., 1996). The vulnerability of the gut and, in particular, the intestinal mucosa, is partly due to the vascular anatomy of the intestinal villi, which are perfused by a central arteriole in close contact with the returning capillary network, allowing a counter-current exchange of oxygen, making the tip of the villus hypoxic in state of altered gut perfusion. The pathophysiology of splanchnic perfusion disturbances and mucosal acidosis in the setting of sepsis and inflammation is also complicated by regional hypermetabolism, cytopathic hypoxia and microcirculatory disturbances that may further aggravate the situation.

Interestingly, infusion of endothelin-1 will induce splanchnic vasoconstriction (Weitzberg et al., 1991a) and there is a growing body of data reflecting the involvement of endothelin in the pathogenesis of mucosal lesions with various aetiologies. Local administration of endothelin-1 induces mucosal ulcerations (Lazaratos et al., 1993; Whittle and Lopez-Belmonte, 1993) and endothelin antagonism has been demonstrated to protect against mucosal ulceration induced by HCl and indomethacin, ischaemia/reperfusion and haemorrhagic shock (Michida et al., 1994; Kitajima et al., 1995; Matsumaru et al., 1997). Dikranian et al. (1994) noted a significant increase of colonic endothelial and epithelial endothelin-1 levels in response to hypoxia. Moreover, mucosal levels of endothelin-1 have been shown to be elevated in critically ill patients, in particular, in those exposed to hypoxia (Michida et al., 1997). In a rat model of stress-induced mucosal haemorrhage, the increase in endothelin-1 levels were more pronounced in mucosal tissue than in plasma (Said and El-Mowafy, 1998). In the setting of sepsis and endotoxaemia, with profound increases in endothelin levels in plasma (Pittet et al., 1991; Weitzberg et al., 1991b; Wanecek et al., 1997a) and the intestinal mucosa (Wilson et al., 1993; Miura et al., 1996a), endothelin antagonism has proven to counteract endotoxin-induced intestinal vasoconstriction and to increase red blood cell velocity (Wilson et al., 1993). In 1993, Wilson et al. (1993) were able to restore intestinal blood flow and improve microcirculation by treating septic rats with endothelin antiserum. These findings suggested an important role for endothelin in the pathophysiology of splanchnic perfusion disturbances dur-

ing sepsi. Lately, Oldner et al. (1998) were able to restore both an endotoxin-induced reduction in gut oxygen delivery and intestinal mucosal pHi, as measured by intestinal tonometry, in a porcine model. As opposed to result of several studies using catecholamines, the increase in both systemic and gut oxygen delivery was followed by an improved mucosal pHi. Such data suggests that endothelin may be of importance not only for splanchnic circulation but also for mucosal homeostasis. This possibility is further supported by a clear correlation between arterial plasma levels of endothelin and mucosal pHi as seen in Fig. 5 (Oldner, 1999). The effects of endothelin ET_A, as opposed to endothelin ET_B, receptor stimulation in the gut have not been fully elucidated. Massberg et al. (1998) showed that only endothelin ET_A receptor antagonism was efficient when counteracting endothelin-1-induced gut injury. The importance of endothelin ET_A receptors was also demonstrated in a study by Miura et al. (1996b) in which reducing endotoxin-induced mucosal injury in rats was reduced. However, in a study from our laboratory, only nonselective endothelin receptor antagonism was able to improve splanchnic blood flow and reduce mucosal acidosis in endotoxaemic pigs (Oldner et al., 1999). In that study, selective endothelin ET_A receptor antagonism was without effect in the splanchnic region despite clear effects on pulmonary hypertension in the same experimental model (Wanecek et al., 1999a), whereas selective endothelin ET_B receptor antagonism proved fatal. Possible explanations for these findings have been discussed above. The relative importance of the endothelin ET_B receptor in the regulation of gut blood flow has, however, been indicated in humans, where infusion of endothelin-3, an endothelin ET_B receptor stimulator, resulted in reduced blood flow.

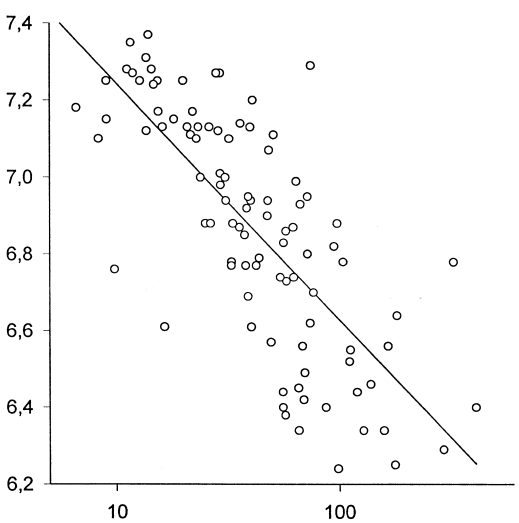

Fig. 5. Relation between ileal mucosal pHi and arterial plasma levels of endothelin-1-like immunoreactivity: The graph illustrates a significant correlation (Spearman rank order correlation) $p < 0.001$ and $R_s = -0.77$ in endotoxaemic pigs. Number of observations = 99. Arterial plasma levels of *of* endothelin-1-like immunoreactivity (ET-1-LI) on the *x*-axis (logarithmic scale) and ileal mucosal pHi on the *y*-axis.

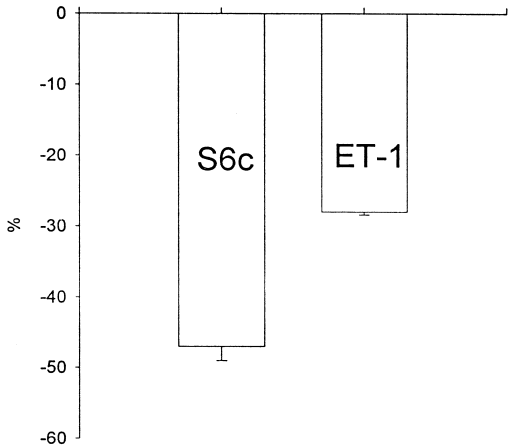

Fig. 6. Relative decrease in portal venous blood flow in sham animals in response to: 200 pmol·kg^{-1} of the selective ET_B receptor agonist, sarafotoxin (S6c, Sigma, Stockholm, Sweden, $n = 5$) or endothelin-1(ET-1) ($n = 3$). Data presented as mean (±S.E.M.).

Moreover, a more pronounced reduction in portal blood flow was seen upon administration of sarafotoxin 6c, a selective endothelin ET_B receptor agonist as opposed to that of endothelin-1, an agonist with higher endothelin ET_A receptor affinity (see Fig. 6) (Oldner, 1999).

How can endothelin mediate mucosal deterioration? Endothelin is a most potent endogenous vasoconstrictor. By means of intravital videomicroscopy, endothelin antagonists have been demonstrated to directly counteract sepsis-induced intestinal vasoconstriction in both large and small arterioli as well as venulae with subsequent increases in red blood cell velocity (Wilson et al., 1993; Miura et al., 1996b). Thus, endothelin seems to be involved in both macro and microcirculatory disturbances. Furthermore, endothelin antagonism has been shown to counteract endotoxin-induced leukocyte sticking in intestinal venulae in rats (Miura et al., 1996b). Leukocyte-mediated tissue damage could contribute to mucosal injury and acidosis.

To our knowledge, changes in distribution of blood flow within the intestinal wall in response to endothelin and endothelin antagonists have not yet been studied. Endothelin-mediated redistribution away from the mucosal layer would obviously contribute to mucosal deterioration. Moreover, despite reports of possible endothelin involvement in the pathogenesis of mitochondrial dysfunction (Maxwell et al., 1992; Mino et al., 1992; Yamanaka et al., 1997), there are currently no data available on a potential contribution of endothelin to mitochondrial dysfunction and reduced cellular oxygen utilisation capacity in sepsis and endotoxaemia. All these factors may contribute to the often found discrepancy between beneficial effects on systemic, and even regional, oxygen delivery and the lack of effects on mucosal homeostasis. This was recently reflected in a study by Hayes et al. (1998) in which volume-resuscitated endotoxaemic dogs were treated with catecholamines. Despite increasing systemic oxygen deliv-

ery epinephrine, norepinephrine and dobutamine all decreased mucosal pHi.

Interestingly, endothelin is reported to be more potent as a constrictor of large veins than of arteries (Cocks et al., 1989). This may contribute to blood pooling and oedema formation impairing tissue oxygenation. A disturbed outflow of portal venous blood through the liver may also contribute to splanchnic blood pooling in endotoxaemia (Ayuse et al., 1995).

2.4. Hepatic manifestations

Interestingly, regulation of liver perfusion seems to depend largely on endothelin-receptor activity during endotoxaemia and possibly septicaemia. Infusion of endothelin-1 into the portal vein markedly and dose-dependently reduces sinusoidal blood flow (Bauer et al., 1994). Release of endothelin-1 from sinusoidal endothelial cells acting on Ito cells mediating contraction is one mechanism suggested for flow regulation (Tanikawa, 1995). Moreover, endotoxin has been reported to enhance the portal venous contractile response to endothelin-1. Pannen et al. (1996a) showed that bosentan only slightly reduced hepatic vascular resistance in sham-treated animals, while it had a notable effect during endotoxaemia. In a study from our laboratory, endotoxaemia induced a sixfold increase in portal-hepatic vascular resistance that was effectively counteracted by nonselective endothelin-receptor antagonism (Oldner et al., 1999). These findings support the concept of endothelin being highly important in liver blood flow regulation during endotoxaemia. Both endothelin ET_A and ET_B receptors have been reported to mediate the vasoconstrictor response in the liver (Zhang et al., 1995), but the reports in the literature concerning the relative importance of these receptors are not unanimous. Partly in agreement with the findings above, Iwai et al. (1998) noted that hepatocellular damage and blood flow disturbances in perfused rat livers in response to endothelin-1 infusion were aggravated by both endothelin ET_A and ET_B selective receptor antagonism while their simultaneous administration resulted in improvement of both parameters. Likewise, Zhang et al. (1997) demonstrated that combined endothelin ET_A and ET_B antagonism was necessary to fully antagonise the effect of endothelin in isolated perfused rat livers. However, both endothelin ET_A and endothelin ET_B receptor antagonism was partly effective in that study. In contrast, Ruetten and Thiemermann (1996) observed a reduced hepatocellular injury in endotoxaemic rats treated with endothelin ET_B but not endothelin ET_A receptor antagonists and Nishida et al. (1998) noted aggravated endotoxin-induced hepatic injury in rats treated with selective endothelin ET_A receptor antagonists. Thus, the effects of endothelin on the liver are complex and heterogeneous and may vary between experimental models and species. The liver is, however, a highly important organ under septic conditions. The Kupffer cells constitute the largest population of tissue macrophages in the body and are of great immunological importance partly as they participate in the clearance of bacteria and toxins derived from a leaky septic gut. Endothelin receptor antagonism, by improving liver perfusion and thereby attenuating liver injury, may become an important tool in the treatment of septic shock in the future.

2.5. Renal manifestations

Endothelin exerts multiple effects on renal function. Endothelin-1 has been suggested to have diuretic effects but also negative effects on glomerular filtration rate and renal blood flow (Goetz et al., 1988; Oishi et al., 1991; Clavell, 1994). Infusion of endothelin-1 causes reductions in renal blood flow (Weitzberg et al., 1991a; Cirino et al., 1997), and endothelin-1 constricts afferent arterioles (Nord, 1997). Recently, infusion of endothelin was shown to induce acute renal failure in pigs (Schulz et al., 1995), and increased plasma endothelin-1 levels have been associated with renal failure in several diseases (Nord, 1997). In the porcine kidney, the endothelin ET_B receptor dominates, and endothelin ET_B receptor stimulation may decrease renal blood flow also in humans (Hemsen, 1991; Weitzberg et al., 1995a).

Acute renal failure is a common manifestation in septic shock, Rangel-Frausto et al. (1995) reported an incidence of over 50%. In experimental settings of septic shock decreased renal blood flow as well as renal dysfunction are indeed common findings (Oldner et al., 1998; Mitaka et al., 1999). The effects of endothelin receptor antagonism in septic models are mixed. Mitaka et al. (1999) were recently able to improve renal function in a model of canine endotoxaemia. In that study, pretreatment with a nonselective endothelin receptor antagonist resulted in increased renal blood flow, creatinine clearance and urine volume. Similar results have also been obtained in response to endothelin antibody treatment in endotoxaemic rats (Morise et al., 1994). Other studies, however, have failed to obtain an improved renal blood flow in endotoxin shock models (Oldner et al., 1998).

The somewhat different effects on renal circulation and urinary output during endotoxaemia could depend on the marked endotoxin-induced release of other substances, such as angiotensin, platelet activating factor and catecholamines affecting the kidneys.

Interestingly, endothelin has been suspected as a mediator of radiocontrast-induced as well as myoglobin-induced renal failure, both common problems among septic patients in the intensive care unit (Anarat et al., 1997; Clozel et al., 1999).

2.6. Coagulation

Endothelin-1 has been shown to shorten bleeding and clotting time together with a prolongation of the activated

partial thromboplastin time in a rat model (Pietraszek et al., 1996). In a pig model, Schulz et al. (1995) induced a consumptive coagulopathy by giving endothelin. In sepsis, an early activation of the coagulation system is seen, which ultimately could lead to disseminated intravascular coagulation, involving systemic activation of the coagulation system, leading to microvascular thrombi in various organs, possibly contributing to organ failure. Secretion or leakage of endothelin-1 and big-endothelin from injured endothelial cells may cause vasospasm and aggravate the disseminated intravascular coagulation process by facilitating the formation of intravascular microthrombi, leading to ischemic end-organ dysfunction. Human sepsis complicated by disseminated intravascular coagulation is associated with higher plasma levels of endothelin-1 (Endo et al., 1995). This is also seen during disseminated intravascular coagulation associated with other conditions, such as malignancy (Ishibashi et al., 1994). Interestingly, plasma levels of endothelin-1 have been shown to be higher in patients with disseminated intravascular coagulation complicated by multiple organ failure, endothelin-1 levels were decreased or remained low with clinical improvement in these patients suggesting that endothelin-1 may play an important role in the progression of multiple organ failure (Asakura et al., 1992).

2.7. General aspects

Generally, there are several mechanics by which endothelin may contribute to the pathophysiology of sepsis and endotoxaemia. Apart from its pronounced vascular effects, endothelin-1 also activates neutrophils enhances vascular cell adhesion molecule expression and thereby promotes adhesion of leukocytes to vascular endothelium (Helset et al., 1994; Caramelo et al., 1997; Ishizuka et al., 1999). In line with this, Hogaboam et al. (1996) showed that mixed endothelin-receptor antagonism decreases the degree of leukocyte activation in a rat model of colitis. Furthermore, endothelin-1 has been demonstrated to induce the production of reactive oxygen species, fundamental mediators in the development of septic and endotoxic shock (Cheng et al., 1999). Moreover, endothelin-1 affects the endothelial barrier as it produces a dose-dependent increase in permeability to protein and extravasation of albumin with concomitant haemoconcentration (Filep et al., 1995; Wanecek et al., 1997a; Porter et al., 1999). Endothelins have also been shown to cause mitochondrial damage in several organ systems (Maxwell et al., 1992; Mino et al., 1992; Yamanaka et al., 1997), a possible mechanism underlying the cytopathic hypoxia seen in sepsis (Fink, 1997). Interestingly, in addition to this, endothe-

Table 1
Overview of results of endothelin receptor antagonism during endotoxaemia

Species	Antagonist	Parameters				Ref.
		CI	PVR/PAP	MAP	Regional effects	
Mice	Bosentan (ET A/B antagonist)				↑ mesenteric blood flow ↓ liver and spleenic weight	(Iskit et al., 1999)
Pig	Bosentan (ET A/B antagonist)	↑	↓	↔	↑ portal blood flow ↓ intestinal mucosa acidosis	(Oldner et al., 1998) (Wanecek et al., 1997a)
Rat	Bosetan (ET A/B antagonist)				↓ portal resistance	(Pannen et al., 1996b)
Pig	Bosetan (ET A/B antagonist)	↑	↓	↔	↑ renal blood flow ↑ spleenic blood flow	(Weitzberg et al., 1996)
Pig	Bosetan (ET A/B antagonist) and indometacine (COX I and II inhibitor)	↑	↓		↓ portal resistance ↓ hepatic artery resistance ↔ s. mesenteric artery-portal vein resistance	(Yamamoto et al., 1997)
Rat	SB 209670 (ET A/B antagonist)			↓	↑ renal blood flow ↑ intestinal blood flow	(Gardiner et al., 1995)
Rat	SB 209670 (ET A/B antagonist)			↓	↑ renal dysfunction ↑ metabolic acidosis	(Ruetten et al., 1996)
Dog	TAK-044 (ET A/B antagonist)	↔		↔	↑ renal blood flow ↓ metabolic acidosis	(Mitaka et al., 1998)
Pig	PD 155080 and A 192621 (ET A and B antagonist)	↑	↓	↔	↑ portal blood flow ↓ intestinal mucosa acidosis	(Oldner et al., 1999) (Wanecek et al., 1999b)
Dog	ET-antag. peptide (ETR-P1/fl) (ET A/B antagonist)	↑ and ↑ contractility				(Kaszaki et al., 1997)
Rat	anti-ET-1 antibody (AwET-1N40) (ET A/B antagonist)				↑ urine volume ↑ urinary sodium excretion	(Morise et al., 1994)
Pig	PD 155080 (ET A antagonist)	↔	↓	↓	↔ portal blood flow ↔ intestinal mucosa acidosis	(Oldner et al., 1999) (Wanecek et al., 1999a)
Rat	BQ123 (ET A antagonist) in vitro		↓			(Curzen et al., 1996)
Rat	BQ-788 (ET B antagonist)			↑	↓ degree of hepatocellular injury and dysfunction	(Ruetten and Thiemermann, 1996)
Pig	A 192621 (ET B antagonist)			100% mortality		(Oldner et al., 1999) (Wanecek et al., 1999b)

lin-1 was shown to delay the clearance of *Escherichia coli* bacteria from the circulation and increased bacterial colonization of several vital organs, reflecting a reduction in bacterial killing function (Schmeck et al., 1999). Therefore, the endothelin system may contribute to septic and endotoxin shock not only by vast changes in organ perfusion but also by several other mechanisms.

2.8. Clinical aspects

Despite enormous progress in intensive care, mortality in sepsis remains high. Even taking into account recent studies, which attempted to modulate the cytokine response seen in sepsis, the results are dissappointing, with no change in, or even an increased, mortality in the clinical setting (Zeni et al., 1997). This emphasises the great complexity of these conditions and the need for new approaches. Several studies using mainly combined endothelin ET_A and endothelin ET_B receptor antagonism in an experimental setting have shown beneficial effects with improvements in central, regional and metabolic parameters (see Table 1), (Curzen et al., 1996; Ruetten et al., 1996; Kaszaki et al., 1997; Wanecek et al., 1997a, 1999b; Oldner et al., 1998, 1999). Hypotension due to vasolidation often accompanies human septic shock and further vasodilatation as a result of endothelin receptor antagonism may be of some concern for the clinician. However, in the experimental setting, the vasodilating properties of endothelin antagonists have resulted in increased perfusion rather than hypotension, even though human data are still lacking. One must also keep in mind that human sepsis has a time course that may be substantially longer then that of most experimental studies, which are conducted in a short-term perspective. Long-term studies of sepsis, utilising endothelin antagonists, are still lacking. So far, endothelin receptor antagonism has not been used in the clinical setting, but the substantial amount of data supporting the involvement of the endothelin system in the pathophysiology of septic and endotoxic shock strongly suggest the potential beneficial effects of such therapy.

References

Anarat, A., Duman, N., Noyan, A., Kibar, M., Anarat, R., 1997. The role of endothelin in radiocontrast nephropathy. Int. Urol. Nephrol. 29, 609–613.

Änggård, E., Galton, S., Rae, G., Thomas, R., McLoughlin, L., de Nucci, G., Vane, J.R., 1989. The fate of radioiodinated endothelin-1 and endothelin-3 in rat. J. Cardiovasc. Pharmacol. 13 (Suppl. 5), S46–S49, discussion S74.

Arai, H., Hori, S., Aramor, I., Ohkubo, H., Nakanishi, S., 1990. Cloning and expression of a cDNA encoding an endothelin receptor. Nature 348, 730–732.

Asaad, M.M., Dorso, C.R., Moreland, S.M., 1993. Effects of neutral endopeptidase inhibition on the clearance of exogeneously administered endothelin in Sprague–Dawley rats. J. Cardiovasc. Pharmacol. 21, 633–636.

Asakura, H., Jokaji, H., Saito, M., Uotani, C., Kumabashiri, I., Morishita, E., Yamazaki, M., Matsuda, T., 1992. Role of endothelin in disseminated intravascular coagulation. Am. J. Hematol. 41, 71–75.

Awane Igata, Y., Ikeda, S., Watanabe, T., 1997. Inhibitory effects of TAK-044 on endothelin induced vasoconstriction in various canine arteries and porcine coronary arteries: a comparison with selective ET_A and ET_B receptor antagonists. Br. J. Pharmacol. 120, 516–522.

Ayuse, T., Brienza, N., Revelly, J.P., Odonell, C.P., Boitnott, J.K., Robotham, J.L., 1995. Alterations in liver hemodynamics in an intact porcine model of endotoxin shock. Am. J. Physiol. 37, H1106–H1114.

Battistini, B., Dussault, P., 1998. Blocking of the endothelin system: the development of receptor antagonists. Pulm. Pharmacol. Ther. 11, 97–112.

Battistini, B., Forget, A.-M., Laight, D., 1996. Potential roles for endothelins in systemic inflammatory response syndrome with a particular relation to cytokines. Shock 5, 167–183.

Bauer, M., Zhang, J.X., Bauer, I., Clemens, M.G., 1994. Endothelin-1 as a regulator of hepatic microcirculation: sublobular distribution of effects and impact on hepatocellular secretory function. Shock 1, 457–465.

Benigni, A., Zoja, C., Corna, D., Orisio, S., Longaretti, L., Bertani, T., Remuzzi, G., 1993. A specific endothelin subtype A receptor antagonist protects against injury in renal disease progression. Kidney Int. 44, 440–444.

Beyer, M.E., Slesak, G., Hoffmeister, H.M., 1995. In vivo hemodynamic and inotropic effects of the endothelin B agonist IRL 1620. J. Cardiovasc. Pharmacol. 26 (Suppl. 3), S190–S192.

Beyer, M.E., Slesak, G., Brehm, B.R., Hoffmeister, H.M., 1998. Hemodynamic and inotropic effects of the endothelin A antagonist BQ-610 in vivo. J. Cardiovasc. Pharmacol. 31 (Suppl. 1), S258–S261.

Bigatello, L., Zapol, M., 1996. New approaches to acute lung injury. Br. J. Anaesth. 77, 99–109.

Black, P.N., Ghatei, M.A., Takahashi, K., Bretherton-Watt, D., Krausz, T., Dollery, C.T., Bloom, S.R., 1989. Formation of endothelin by cultured airway epithelial cells. FEBS Lett. 255, 129–132.

Bonggwan, S., Oemar, B.S., Siebenmann, R., von Segesser, L., Luscher, T.F., 1994. Both ET_A and ET_B receptors mediate contraction to endothelin-1 in human blood vessels. Circulation 89, 1203–1208.

Booke, M., Lingnau, W., Hinder, F., Traber, L.D., Traber, D.L., 1995. Endotoxin versus bacteremia: a comparison focusing on clinical relevance. Prog. Clin. Biol. Res. 392, 393–403.

Boulanger, C., Luscher, T.F., 1990. Release of endothelin from the porcine aorta. Inhibition by endothelium-derived nitric oxide. J. Clin. Invest. 85, 587–590.

Brady, A.J., 1995. Nitric oxide, myocardial failure and septic shock. [Review]. Int. J. Cardiol. 50 (3), 269–272.

Caramelo, C., Lopez Farre, A., Casado, S., 1997. Role of endothelin-1 in the activation of polymorphonuclear leukocytes. Kidney Int. 52, 56–59.

Cassin, S., Kristova, V., Davis, T., Kadowitz, P., Gause, G., 1991. Tone-dependent responses to endothelin in the isolated perfused fetal sheep pulmonary circulation in situ. J. Appl. Physiol. 70, 1228–1234.

Cheng, T.H., Shih, N.L., Chen, S.Y., Wang, D.L., Chen, J.J., 1999. Reactive oxygen species modulate endothelin-I-induced c-*fos* gene expression in cardiomyocytes. Cardiovasc. Res. 41, 654–662.

Chou, M.C., Wilson, M.A., Spain, D.A., Hadjiminas, D., Anderson, G.L., Cheadle, W.G., Garrison, R.N., 1995. Endothelin-1 expression in the small intestine during chronic peritonitis. Shock 4 (6), 411–414.

Chun, M., Lin, H.Y., Henis, Y.I., Lodish, H.F., 1995. Endothelin-induced endocytosis of cell surface ET_A receptors. Endothelin remains intact and bound to the ET_A receptor. J. Biol. Chem. 270, 10855–10860.

Cimminiello, C., Milani, M., Uberti, T., Arpaia, G., Perolini, S., Bonfardeci, G., 1991. Endothelin, vasoconstriction, and endothelial damage in Raynaud's phenomenon. Lancet 337, 114–115.

Cirino, M., Martin, E., Yano, M., Rodger, I.W., 1997. Cardiovascular and

renal actions of the endothelin (B) receptor in pigs. J. Cardiovasc. Pharmacol. 29, 704–712.

Clavell, A., 1994. Physiologic and pathophysiologic roles of endothelin in the kidney. Curr. Opin. Nephrol. Hypertens. 3, 66–72.

Clozel, M., Watanabe, H., 1993. BQ-123, a peptidic endothelin ET_A receptor antagonist, prevents the early cerebral, vasospasm following subarachnoid hemorrhage after intracisternal but not intravenous injection. Life Sci. 52, 825–834.

Clozel, M., Ramuz, H., Clozel, J.P., Breu, V., Hess, P., Loffler, B.M., Coassolo, P., Roux, S., 1999. Pharmacology of tezosentan, new endothelin receptor antagonist designed for parenteral use. J. Pharmacol. Exp. Ther. 290, 840–846.

Cocks, T.M., Faulkner, N.L., Sudhir, K., Angus, J., 1989. Reactivity of endothelin-1 on human and canine large veins compared with large arteries in vitro. Eur. J. Pharmacol. 171, 17–24.

Cody, R.J., Haas, G.J., Binkley, P.F., Capers, Q., Kelley, R., 1992. Plasma endothelin correlates with the extent of pulmonary hypertension in patients with chronic congestive heart failure [published erratum appears in Circulation 1993 Mar; 87 (3) 1064]. Circulation 85, 504–509.

Cunnion, R.E., Parillo, J.E., 1989. Myocardial dysfunction in sepsis. [Review]. Crit. Care Clin. 5 (1), 99–118.

Curzen, N.P., Mitchell, J.A., Jourdan, K.B., Griffiths, M.J., Evans, T.W., 1996. Endothelin-1-induced contraction of pulmonary arteries from endotoxemic rat is attenuated by the endothelin-A receptor antagonist, BQ123. Crit. Care Med. 24, 2007–2013.

Davenport, A.P., O'Reilly, G., Kuc, R.E., 1995. Endothelin ET_A and ET_B mRNA and receptors expressed by smooth muscle in the human vasculature: majority of the ET_A sub-type. Br. J. Pharmacol. 114, 1110–1116.

De Keulenaer, G.W., Andries, L.J., Sys, S.U., Brutsaert, D.L., 1995. Endothelin-mediated positive inotropic effect induced by reactive oxygen species in isolated cardiac muscle. Circ. Res. 76, 878–884.

De Nucci, G., Thomas, R., D'Orleans-Juste, P., Antunes, E., Walder, C., Warner, T.D., Vane, J.R., 1988. Pressor effects of circulating endothelin are limited by its removal in the pulmonary circulation and by the release of prostacyclin and endothelin-derived relaxing factor. Proc. Natl. Acad. Sci. U. S. A. 85, 9797–9800.

Dhainaut, J.F., Huygebaert, M.-F., Monsallier, J.F., Lefevre, G., Dall'Ava-Santucci, J., Brunet, F., Villermant, D., Carli, A., Raichvarg, D., 1987. Coronary hemodynamics and myocardial metabolism of lactate, free fatty acids, glucose and ketones in patients with septic shock. Circulation 75, 533–541.

Dikranian, K., Tomlinson, A., Loesch, A., Winter, R., Burnstock, G., 1994. Increased in immunoreactivity to endothelin-1 in the mucosal vasculature and epithelium of the large intestine during chronic hypoxia. J. Anat. 185, 609–615.

Druml, W., Steltzer, H., Waldhausl, W., Lenz, K., Hammerle, A., Vierhapper, H., Gasic, S., Wagner, O.F., 1993. Endothelin-1 in adult respiratory distress syndrome. Am. Rev. Respir. Dis. 148, 1169–1173.

Dupuis, J., Goresky, C.A., Fournier, A., 1996. Pulmonary clearance of circulating endothelin-1 in dogs in vivo: exclusive role of ET_B receptors. J. Appl. Physiol. 81, 1510–1515.

Ehrenreich, H., 1990. Endothelins, peptides with potent vasoactive properties are produced by human macrophages. J. Exp. Med. 172, 1741–1748.

Ehrenreich, H., Burd, P.R., Rottem, M., Hultner, L., Hylton, J.B., Garfield, M., Coligan, J.E., Metcalfe, D.D., Fauci, A.S., 1992. Endothelins belong to the assortment of mast cell-derived and mast cell-bound cytokines. New. Biol. 4, 147–156.

Elshourbagy, N.A., Adamou, J.E., Gagnon, A.W., Wu, H.L., Pullen, M., Nambi, P., 1996. Molecular characterization of a novel human endothelin receptor splice variant. J. Biol. Chem. 271, 25300–25307.

Emori, T., Hirata, Y., Ohta, K., Shichiri, M., Marumo, F., 1989. Secretory mechanism of immunoreactive endothelin in cultured bovine endothelial cells. Biochem. Biophys. Res. Commun. 160, 93–100.

Emoto, N., Yanagisawa, M., 1995. Endothelin-converting enzyme-2 is a membrane-bound, phosphoramidon-sensitive metalloprotease with acidic pH optimum. J. Biol. Chem. 270, 15262–15268.

Endo, T., Uchida, Y., Matsumoto, H., Suzuki, N., Nomura, A., Hirata, F., Hasegawa, S., 1992. Regulation of endothelin-1 synthesis in cultured guinea pig airway epithelial cells by various cytokines. Biochem. Biophys. Res. Commun. 186, 1594–1599.

Endo, S., Inada, K., Nakae, H., Takakuwa, T., Kasai, T., Yamada, Y., 1995. Blood levels of endothelin-1 and thrombomodulin in patients with intravascular coagulation and sepsis. Res. Commun. Mol. Pathol. Pharmacol. 90, 277–288.

Ezra, D., Goldstein, R., Czaja, J., Feuerstein, G., 1989. Lethal ischemia due to intracoronary endothelin in pigs. Am. J. Physiol. 257, H339–H343.

Filep, J.G., Sirois, M.G., Foldes-Filep, E., Rousseau, A., Plante, G.E., Fournier, A., Yano, M., Sirois, P., 1993. Enhancement by endothelin-1 of microvascular permeability via the activation of ET_A receptors. Br. J. Pharmacol. 109, 880–886.

Filep, J., Fournier, A., Földes-Filep, E., 1995. Acute pro-inflammatory actions endothelin-1 in the guinea-pig lung: involvement of ET_A and ET_B receptors. Br. J. Pharmacol. 115, 227–236.

Fink, M., 1997. Cytopathic hypoxia in sepsis. Acta Anaesthesiol. Scand. 41, 87–95.

Fukuroda, T., Ozaki, S., Ihara, M., Isçhikawa, K., Yano, M., Nishikibe, M., 1994. Synergetic inhibition by BQ-123/BQ-788 of ET-1 induced contractions of the rabbit pulmonary artery. Br. J. Pharmacol. 113, 336–338.

Gardiner, S.M., Kemp, P.A., March, J.E., Bennett, T., 1995. Enhancement of the hypotensive and vasolidator effects of endotoxemia in conscious rats by the endothelin antagonist, SB 209670. Br. J. Pharmacol. 116, 1718–1719.

Gasic, S., Wagner, O.F., Vierhapper, H., Nowotny, P., Waldhausl, W., 1992. Regional hemodynamic effects and clearance of endotheli-1 in humans: renal and peripheral tissues may contribute to the overall disposal of the peptide. J. Cardiovasc. Pharmacol. 19, 176–180.

Giaid, A., Gibson, S.J., Ibrahim, B.N., Legon, S., Bloom, S.R., Yanagisawa, M., Masaki, T., Varndell, I.M., Polak, J.M., 1989. Endothelin 1, an endothelium-derived peptidem, is expressed in neurons of the human spinal cord and dorsal root ganglia. Proc. Natl. Acad. Sci. U. S. A. 86, 7634–7638.

Giaid, A., Polak, J.M., Gaitonde, V. et al., 1991. Distribution of endothelin-like immunoreactivity and mRNA in the developing and adult human lung. Am. J. Respir. Cell. Mol. Biol. 4, 50–58.

Giaid, A., Yanigisawa, M., Langleben, D. et al., 1993. Expression of endothelin-1 in the lungs of patients with pulmonary hypertension. N. Eng. J. Med. 328, 1732–1739.

Goddard, C.M., Allard, M.F., Hogg, J.C., Herbertson, M.J., Walley, K.R., 1995. Prolonged leukocyte transit time in coronary microcirculation of endotoxemic pigs. Am. J. Physiol. 269, H1389–H1397.

Goetz, K.L., Wang, B.C., Madwed, J.B., Zhu, J.L., Leadley, R.J. Jr., 1988. Cardiovascular, renal, and endocrine responses to intravenous endothelin in conscious dogs. Am. J. Physiol. 255, R1064–R1068.

Goldfarb, R., Nightingale, L., Kish, P., Loegering, D., 1986. Left ventricular function during lethal and sub lethal endotoxemia in swine. Am. J. Physiol. 251, H364–H373.

Goldie, R.G., D'Aprile, A.C., Self, G.J., Rigby, P.J., Henry, P.J., 1996a. The distribution and density of receptor subtypes for endothelin-1 in peripheral lung of the rat, guinea-pig and pig. Br. J. Pharmacol. 117, 729–735.

Goldie, R.G., Knott, P.G., Carr, M.J., Hay, D.W., Henry, P.J., 1996b. The endothelins in the pulmonary system. Pulm. Pharmacol. 9, 69–93.

Gonon, A.T., Wang, Q.D., Pernow, J., 1998. The endothelin A receptor antagonist LU 135252 protects the myocardium from neutrohil injury ischaemia/reperfusion. Cardiovasc. Res. 39, 674–682.

Granton, J.T., Goddard, C.M., Allard, M.F., van Eeden, S., Walley, K.R., 1997. Leukocytes and decreased left-ventricular contractility during endotoxemia in rabbits. Am. J. Respir. Crit. Care Med. 155, 1977–1983.

Grover, R., Zaccardelli, D., Colice, G., Guntupalli, K., Watson, D., Vincent, J.L., 1999. An open-label dose escalation study of the nitric oxide synthase inhibitor, *N*(G)-menthyl-L-arginine hydrochloride (546C88), in patients with septic shock. Glaxo wellcome International Septic Shock Study Group [see comments]. Crit. Care Med. 27, 913–922.

Hanehira, T., Kohno, M., Yoshikawa, J., 1997. Endothelin production in cultured vascular smooth muscle cells-modulation by the atrial, brain, and C-type natriuretic peptide system. Metabolism 46, 487–493.

Hayes, J.K., Luo, X., Wong, K.C., McJames, S., Tseng, C.K., 1998. Effects of dobutamine, norepinephrine and epinephrine on intramucosal pH and hemodynamics of dogs during endotoxic shock. Acta Anaesthesiol. Sinics (Taipei) 36, 113–126.

Haynes, W.G., Webb, D.J., 1994. Contribution of endogenous generation of endothelin-1 to basal vascular tone. Lancet 344, 852–854.

Helset, E., Kjaeve, J., Hauge, A., 1993. Endothelin-1-induced increases in microvascular permeability in isolated, perfused rat lungs requires leukocytes and plasma. Circ. Shock 39, 15–20.

Helset, E., Ytrehus, K., Tveita, T., Kjëve, J., Jörgensen, L., 1994. Et-1 causes accumulation of leukocytes in the pulmonary circulation. Circ. Shock 44, 201–209.

Hemsen, A., 1991. Biochemical and functional characterization of endothelin peptides with special reference to vascular effects. Acta Physiol. Scand., Suppl. 602, 1–61.

Higashi, T., Ishizaki, T., Shigemori, K., Nakai, T., Miyabo, S., Inui, T., Yamamura, T., 1997. Pharmacological heterogeneity of constrictions mediated by endothelin receptors in rat pulmonary arteries. Am. J. Physiol. 272, L287–L293.

Hogaboam, C.M., Muller, M.J., Collins, S.M., Hunt, R.H., 1996. An orally active non-selective endothelin receptor antagonist, bosentan, markedly reduces injury in a rat model of colitis. Eur. J. Pharmacol. 309, 261–269.

Hohlfed, T., Klemm, P., Thiemermann, C., Warner, T.D., Schror, K., Vane, J.R., 1995. The contribution of tumour necrosis factor-alpha and endothelin-1 to the increase of coronary resistance in hearts from rats treated with endotoxin. Br. J. Pharmacol. 116, 3309–3315.

Holm, P., 1997. Endothelin in the pulmonary circulation with special reference to hypoxic pulmonary vasoconstriction. Scand. Cardiovasc. J. Suppl. 46, 1–40.

Horkay, F., Szokodi, I., Merkely, B., 1998. Potential pathophysiologic role of endothelin-1 in canine pericardial fluid. J. Cardiovasc. Pharmacol. 31 (Suppl. 1), S401–S402.

Humer, M.F., Phang, P.T., Friesen, B.P., Allard, M.F., Goddard, C.M., Walley, K.R., 1996. Heterogeneity of gut capillary transit times and impaired gut oxygen extraction in endotoxemic pigs. J. Appl. Physiol. 81, 895–904.

Ince, C., Sinaasappel, M., 1999. Microcirculatory oxygenation and shunting in sepsis and shock [see comments]. Crit. Care Med. 27, 1369–1377.

Inoue, A., Yanagisawa, M., Kimura, S., Kasuya, Y., Miyauchi, T., Goto, K., Masaki, T., 1989. The human endothelin family: three structurally and pharmacologically distinct isopeptides predicted by three separate genes. Proc. Natl. Acad. Sci. U. S. A. 86, 2863–2867.

Ishibashi, M., Ito, N., Fujita, M., Furue, H., Yamaji, T., 1994. Endothelin-1 as an aggravating factor of disseminated intravascular coagulation associated with malignant neoplasms. Cancer 73, 191–195.

Ishikawa, T., Yanagisawa, M., Kimura, S., Goto, K., Masaki, T., 1988a. Positive inotropic actions of novel vasoconstrictor peptide endothelin on guinea pig atria. Am. J. Physiol. 255, H970–H973.

Ishikawa, T., Yanagisawa, M., Kimura, S., Goto, K., Masaki, T., 1988b. Positive chronotropic effects of endothelin, a novel endothelium-derived vasoconstrictor peptide. Pfluegers Arch. 413, 108–110.

Ishizuka, T., Takamizawa Matsumoto, M., Suzuki, K., Kurita, A., 1999. Endothelin-1 enhances vascular cell adhesion molecule-1 expression in tumor necrosis factor alfa-stimulated vascular endothelial cells. Eur. J. Pharmacol. 369, 237–245.

Iskit, A.B., Sungur, A., Gedikoglu, G., Guc, M.O., 1999. The effects of bosentan, aminoguanidine and L-canavanine on mesenteric blood flow, spleen and liver in endotoxaemic mice. Eur. J. Pharmacol. 379, 73–80.

Iwai, M., Yamauchi, T., Shimazu, T., 1998. Endothelin 1 aggravates acute liver injury in perfused livers of rats after treatment with D-galactosamine. Hepatology 28, 503–509.

Iwasaki, S., Homma, T., Matsuda, Y., Kon, V., 1995. Endothelin receptor subtype B mediates autoinduction of endothelin-1 in rat mesangial cells. J. Biol. Chem. 270, 6997–7003.

Kaddoura, S., Curzen, N.P., Evans, T.W., Firth, J.D., Poole-Wilson, P.A., 1996. Tissue expression of endothelin-1 mRNA in endotoxaemia. Biochem. Biophys. Res. Commun. 218 (3), 641–647.

Kaszaki, J., Wolfard, A., Boros, M., Baranyi, L., Okada, H., Nagy, S., 1997. Effects of antiendothelin treatment on the early hemodynamic changes in hyperdynamic endotoxemia. Acta. Chir. Acad. Sci. Hung. 36, 152–153.

Kiely, D.G., Cargill, R.I., Struthers, A.D., Lipworth, B.J., 1997. Cardiopulmonary effects of endothelin-1 in man. Cardiovasc. Res. 33, 378–386.

Kimura, S., Kasuya, Y., Sawamura, T., Shinimi, O., Sugita, Y., Yanagisawa, M., Goto, K., Masaki, T., 1989. Conversion of big endothelin-1 to 21-residue endothelin-1 is essential for expression of full vasoconstrictor active structure-activity relationships of big endothelin-1. J. Cardiovasc. Pharmacol. 13 (Suppl. 5), S5–S7, discussion S18.

Kitajima, T., Tani, K., Yamaguchi, T., Kubota, Y., Okuhira, M., Mizuno, T., Inoue, K., 1995. Role of endogenous endothelin in gastric mucosal injury induced by hemorrhagic shock in rats. Digestion 56, 111–116.

Kourembanas, S., Marsden, P.A., McQuillan, L.P., Faller, D.V., 1991. Hypoxia induces endothelin gene expression and cultured human endothelium. J. Clin. Invest. 88, 1054–1057.

Krafft, P., Fridrich, P., Pernerstofer, T., Fitzgerald, R.D., Koc, D., Schneider, B., Hammerle, A.F., Steltzer, H., 1996. The acute respiratory distress syndrome: definitions, severity and clinical outcome. An analysis of 101 clinical investigations [see comments]. Int. Care Med. 22, 519–529.

Krum, H., Viskoper, R.J., Lacourciere, Y., Budde, M., Charlon, V., 1998. The effect of an endothelin-receptor antagonist, bosentan, on blood pressure in patients with essential hypertension. Bosentan Hypertension Investigators. N. Engl. J. Med. 338, 784–790.

Kumar, A., Thota, V., Dee, L., Olson, J., Uretz, E., Parillo, J.E., 1996. Tumor necrosis factor alpha and interleukin 1beta are responsible for in vitro myocardial depression induced by human septic shock serum. J. Exp. Med. 183, 949–958.

Kurihara, H., Yoshizumi, M., Sugiyama, T., Takaku, F., Yanagisawa, M., Masaki, T., Hamaoki, M., Kato, H., Yazaki, Y., 1989. Transforming growth factor-beta stimulates the expression of endothelin mRNA by vascular endothelial cells. Biochem. Biophys. Res. Commun. 159, 1435–1440.

Langleben, D., Demarchie, M., Laporta, D., AH, S., Schlesinger, R., DJ, S., 1993. Endothelin-1 in acute lung injury and the adult respiratory distress syndrome. Am. Rev. Respir. Dis. 148, 1646–1650.

Lazaratos, S., Kashimura, H., Nakahara, A., Fukutomi, H., Osuga, T., Urushidani, T., Miyauchi, T., Goto, K., 1993. Gastric ulcer induced by submucosal injection of ET-1: role of potent vasoconstriction and intraluminal acid. Am. J. Physiol. 265, G491–G498.

Leeper Woodford, S., Carey, P., Byrne, K., Fisher, J., Blocher, C., Sugerman, H., Fowler, A., 1991. Ibuprofen attenuates plasma tumor necrosis factor activity during sepsis-induced acute lung injury. J. Appl. Physiol. 71, 915–923.

Lerman, A., Hildebrand, F.L. Jr., Aarhus, L.L., Burnett, J.C. Jr., 1991. Endothelin has biological actions at pathophysiological concentrations. Circulation 83, 1808–1814.

Liu, B., Zhou, J., Chen, H., Wang, D., Hu, D., Wen, Y., Xiao, N., 1997. Expression and cellular location of endothelin-1 mRNA in rat liver following endotoxemia. Chin. Med. J. Engl. 110, 932–935.

Lodge, N.J., Zhang, R., Halaka, N.N., Moreland, S., 1995. Functional role of endothelin ET_A and ET_B receptors in venous and arterial smooth muscle. Eur. J. Pharmacol. 287, 279–285.

Lopez Farre, A., Riesco, A., Espinosa, G. et al., 1993. Effect of endothelin-1 on neutrophil adhesion to endothelial cells and perfused heart. Circulation 88, 1166–1171.

MacLean, M.R., McCulloch, K.M., Baird, M., 1994. Endothelin ET_A- and ET_B-receptor-mediated vasoconstriction in rat pulmonary arteries and arterioles. J. Cardiovasc. Pharmacol. 23, 838–845.

MacLean, M.R., Mackenzie, J.F., Docherty, C.C., 1998. Heterogeneity of endothelin-B receptors in rabbit pulmonary resistance arteries. J. Cardiovasc. Pharmacol. 31 (Suppl. 1), S115–S118.

Maemura, K., Kurihara, H., Morita, T., Oh-hashi, Y., Yazaki, Y., 1992. Production of endothelin-1 in vascular endothelial cells is regulated by factors associated with vascular injury. Gerontology 38 (Suppl. 1), 29–35.

Marsault, R., Feolde, E., Frelin, C., 1993. Receptor externalization determines sustained contractile responses to endothelin-1 in the rat aorta. Am. J. Physiol. 264, C687–C693.

Marsdeen, P., Brenner, B., 1992. Transcriptional regulation of the endothelin-1 gene by TNF alpha. Am. J. Physiol. 262, 854–861.

Massberg, S., Boros, M., Leiderer, R., Baranyi, L., Okada, H., Messmer, K., 1998. Endothelin (ET)-1 induced mucosal damage in the rat small intestine role of ET(A) receptors. Shock 9, 177–183.

Matsumaru, K., Kashimura, H., Hassan, M. et al., 1997. Bosentan, a novel synthetic mixed-type endothelin receptor antagonist, attenuates acute gastric mucosal lesions induced by indomethacin and HCl in the rat: role of endogenous endothelin-1. J. Gastroenterol. 32, 164–170.

Mattoli, S., Mezzetti, M., Riva, G., Allegra, L., Fasoli, A., 1990. Specific binding of endothelin on human bronchial smooth muscle cells in culture and secretion of endothelin-like material from bronchial epithelial cells. Am. J. Respir. Cell. Mol. Biol. 3, 145–151.

Mattoli, S., Soloperto, M., Marini, M., Fasoli, A., 1991. Levels of endothelin in the bronchoalveolar lavage fluid of patients with symptomatic asthma and reversible airflow obstruction. J. Allergy Clin. Immunol. 88, 376–384.

Maxwell, L., Pratt, K., Gavin, J., 1992. Endothelin-3-induced microvascular incompetence and mitochondrial damage in rat myocardium. Clin. Exp. Pharmacol. Physiol. 19, 683–688.

McCarron, R.M., Weang, L., Stanimirovic, D.B., Spatz, M., 1993. Endothelin induction of adhesion molecule expression on human brain microvascular endothelial cells. Neurosci. Lett. 156, 31–34.

Mebazaa, A., Mayoux, E., Maeda, K., Martin, L.D., Lakatta, E.G., Robotham, J.L., Shah, A.M., 1993. Paracrine effects of endocardial endothelial cells on myocyte contraction mediated via endothelin. Am. J. Physiol. 265, H1841–H1846.

Miasiro, N., Karaki, H., Paiva, A.C., 1998. Distinct endothelin-B receptors mediate the effects of sarafotoxin S6c and IRL1620 in the ileum. J. Cardiovasc. Pharmacol. 31 (Suppl. 1), S175–S178.

Michida, T., Kawano, S., Masuda, E. et al., 1994. Role of endothelin-1 in hemorrhagic shock-induced gastric mucosal injury in rats. Gastroenterology 106, 988–993.

Michida, T., Kawano, S., Masuda, E. et al., 1997. Endothelin-1 in the gastric mucosa in stress ulcers of critically ill patients. Am. J. Gastroenterol. 92, 1177–1181.

Mickley, E.J., Gray, G.A., Webb, D.J., 1997. Activation of endothelin ET_A receptors masks the constrictor role of endothelin ET_B receptors in rat isolated small mesenteric arteries. Br. J. Pharmacol. 120, 1376–1382.

Mino, N., Kobayashi, M., Nakajima, A. et al., 1992. Protective effect of a selective endothelin receptor antagonist, BQ-123, in ischmic acute renal failure in rats. Eur. J. Pharmacol. 221, 77–83.

Mitaka, C., Hirata, Y., Yokoyama, K., Nagura, T., Tsunoda, Y., Amaha, K., 1998. Pathologic role of endothelin-1 in septic shock. J. Cardiovasc. Pharmacol. 31, S233–S235.

Mitaka, C., Hirata, Y., Yokoyama, K., Nagura, T., Tsunoda, Y., Amaha, K., 1999. Improvement of renal dysfunction in dogs with endotoxemia by a nonselective endothelin receptor antagonist. Crit. Care Med. 27, 146–153.

Miura, S., Fukumura, D., Kurose, I. et al., 1996a. Roles of ET-1 in endotoxin-induced microcirculatory disturbance in rat small intestine. Am. J. Physiol. 271, G461–G469.

Miura, S., Tsuzuki, Y., Kurose, I. et al., 1996b. Endotoxin stimulates lymphocyte-endothelial interactions in rat intestinal Peyer's patches and villus mucosa. Am. J. Physiol. 271, G282–G292.

Molenaar, P., O'Reilly, G., Sharkey, A., Kuc, R.E., Harding, D.P., Plumpton, C., Gresham, G.A., Davenport, A.P., 1993. Characterization and localization of endothelin receptor subtypes in the human atrioventricular conducting system and myocardium. Circ. Res. 72, 526–538.

Morise, Z., Ueda, M., Aiura, K., Endo, M., Kitajima, M., 1994. Pathophysiologic role of endothelin-1 in renal function in rats with endotoxin shock. Surgery 115 (2), 199–204.

Nishida, T., Huang, T.P., Seiyama, A., Hamada, E., Kamiike, W., Ueshima, S., Kazuo, H., Matsuda, H., 1998. Endothelin A-receptor blockade worsens endotoxin-induced hepatic microcirculatory changes and necrosis. Gastroenterology 115, 412–420.

Nord, E.P., 1997. Role of endothelin in acute renal failure. Blood Purif. 15, 273–285.

Oishi, R., Nonoguchi, H., Tomita, K., Marumo, F., 1991. Endothelin-1 inhibits AVP-stimulated osmotic water permeability in rat inner medullary collecting duct. Am. J. Physiol. 261, F951–F956.

Oldner, A., 1999. Splanchnic organ homeostasis in experimental endotoxin shock: with special reference to the endothelin and renin–angiotensin systems, Dep. Surgical Sciences. Karolinska Institute, Stockholm.

Oldner, A., Wanecek, M., Goiny, M., Weitzberg, E., Rudehill, A., Alving, K., Sollevi, A., 1998. The endothelin receptor antagonist bosentan restores gut oxygen delivery and reverses mucosal acidosis in porcine endotoxin shock. Gut 42, 696–702.

Oldner, A., Wanecek, M., Weitzberg, E., Sundin, P., Sollevi, A., Rubio, C., Hellstrom, P.M., Alving, K., Rudehill, A., 1999. Differentiated effects on splanchnic homeostasis by selective and non-selective endothelin receptor antagonism in porcine endotoxaemia. Br. J. Pharmacol. 127, 1793–1804.

Opal, S., 1995. Clinical trials of novel therapeutic agents: why did they fail? In: Vincent, J. (Ed.), Yearbook of Intensive Care and Emergency Medicine. pp. 425–436.

Opgenorth, T.J., 1995. Endothelin receptor antagonism. Adv. Pharm. 33, 1–65.

Ottoson Seeberger, A., Hemsen, A., Lundberg, J.M., Ahlborg, G., 1998. The C-terminal fragment of big endothelin-1 does not potentiate the vasoactive effects of endothelin-1. Clin. Physiol. 18, 61–68.

Ozaki, S., Ohwaki, K., Ihara, M., Ishikawa, K., Yano, M., 1997. Coexpression studies with endothelin receptor subtypes indicate the existence of intracellular cross-talk between ET(A) and ET(B) receptors. J. Biochem. (Tokyo) 121, 440–447.

Pannen, B.H., Bauer, M., Zhang, J.X., Robotham, J.L., Clemens, M.G., 1996a. Endotoxin pretreatment enhances portal venous contractile response to endothelin-1. Am. J. Physiol. 270, H7–H15.

Pannen, B.H., Bauer, M., Zhang, J.X., Robotham, J.L., Clemens, M.G., 1996b. A time-dependent balance between endothelins and nitric oxide regulating portal resistance after endotoxin. Am. J. Physiol. 271, H1953–H1961.

Pernow, J., Hemsen, A., Lundberg, J., 1989a. Tissue specific distribution, clearance and vascular effects of endothelin in the pig. Biochem. Biophys. Res. Commun. 161, 647–653.

Pernow, J., Hemsen, A., Lundberg, J.M., 1989b. Increased plasma levels of endothelin-like immunoreactivity during endotoxin administration in the pig. Acta Physiol. Scand. 137, 317–318.

Peyton, M.D., Hinshaw, L.B., Greenfield, L.J., Elkins, R.C., 1976. The effects of coronary vasodilation on cardiac performance during endotoxin shock. Surg., Gynecol. Obstet. 143, 533–538.

Pietraszek, M., Chabielska, E., Azzadin, A., Pawlak, R., Buczko, W.,

1996. Effect of endothelin-1 on some hemostatic parameters in normotensive rats. Acta Physiol. Acad. Sci. Hung. 84, 329–330.

Pittet, J., Morel, D., Hemsen, A., Gunning, K., Lacroix, J.-S., Suter, P., Lundberg, J., 1991. Elevated plasma endothelin-1 concentrations are associated with the severity of illness in patients with sepsis. Ann. Surg. 213, 261–264.

Plumpton, C., Ashby, M.J., Kuc, R.E., O'Reilly, G., Davenport, A.P., 1996. Expression of endothelin peptides and mRNA in the human heart. Clin. Sci. 90, 37–46.

Porter, L.P., McNamee, J.E., Wolf, M.B., 1999. Endothelin-1 induces endothelial barrier failure in the cat hindlimb. Shock 11, 111–114.

Rangel-Frausto, M.S., Pittet, D., Costigan, M., Hwang, T., Davis, C.S., Wenzel, R.P., 1995. The natural history of the systemic inflammatory response syndrome (SIRS). A prospective study [see comments]. JAMA, J. Am. Med. Assoc. 273, 117–123.

Razandi, M., Pedram, A., Rubin, T., Levin, E.R., 1996. PGE2 and PG12 inhibit ET-1 secretion from endothelial cells by stimulating particulate guanylate cyclase. Am. J. Physiol. 270, H1342–H1349.

Resink, T.J., Hahn, A.W., Scott-Burden, T., Powell, J., Weber, E., Buhler, F.R., 1990. Inducible endothelin mRNA expression and peptide secretion in cultured human vascular smooth muscle cells. Biochem. Biophys. Res. Commun. 168, 1303–1310.

Rossmanith, G.H., Hoh, J.F., Turnbull, L., Ludowyke, R.I., 1997. Mechanism of action of endothelin rat cardiac muscle: cross-bridge kinetics and myosin light chain. J. Physiol. (London) 505, 217–227.

Rubanyi, G., Polokoff, M., 1994. Endothelins: molecular biology, biochemistry, pharmacology, physiology and pathophysiology. Pharmacol. Rev. 46, 325–415.

Ruetten, H., Thiemermann, C., 1996. Effect of selective blockade of endothelin ET_B receptors on the liver dysfunction and injury caused by endotoxaemia in the rat. Br. J. Pharmacol. 119, 479–486.

Ruetten, H., Thiemermann, C., Vane, J.R., 1996. Effects of the endothelin receptor antagonist, SB 209670, on circulatory failure and organ injury in endotoxic shock in the anaesthetized rat. Br. J. Pharmacol. 118, 198–204.

Said, S.A., El-Mowafy, A.M., 1998. Role of endogenous endothelin-1 in stress-induced gastric mucosal damage and acid secretion in rats. Regul. Pept. 73, 43–50.

Sakurai, T., Yanagisawa, M., Takuwa, Y., Miyazaki, H., Kimura, S., Goto, K., Masaki, T., 1990. Cloning of a cDNA encoding a non-isopeptide-selective subtype of the endothelin receptor. Nature 348, 732–735.

Sato, K., Oka, M., Hasunuma, K., Ohnishi, M., Sato, K., Kira, S., 1995. Effects of separate and combined ET_A and ET_B blockade on ET-1-induced constriction in perfused rat lungs. Am. J. Physiol. 269, L668–L672.

Schmeck, J., Heller, A., Phan, T.L., Urbschek, R., Koch, T., 1999. Effects of endothelin-1 on bacterial clearance in rabbits. Eur. J. Anaesthesiol. 16, 169–175.

Schulz, E., Ruschitzka, F., Lueders, S., Heydenbluth, R., Schrader, J., Muller, G.A., 1995. Effects of endothelin on hemodynamics, prostaglandins, blood coagulation and renal function. Kidney Int. 47, 795–801.

Scili, A., Vijaaraghavan, J., Hersh, L., Carretero, O., 1989. Neutral endopeptidase 24.11 (NEP) inactivates endothelin. Hypertension 14, 353.

Stewart, D., Levy, R., Cernacek, P., Langleben, D., 1991. Increased plasma endothelin-1 in pulmonary hypertension: marker of mediator of disease? Ann. Intern. Med. 114, 464–469.

Sugiura, M., Inagami, T., Kon, V., 1989. Endotoxin stimulates endothelin-release in vivo and in vitro as determined by radioimmunoassay. Biochem. Biophys. Res. Commun. 161, 1220–1227.

Sütsch, G., Kiowski, W., Yan, X.W., Hunziker, P., Christen, S., Strobel, W., Kim, J.H., Rickenbacher, P., Bertel, O., 1998. Short-term oral endothelin-receptor antagonist therapy in conventionally treated patients with symptomatic severe chronic heart failure. Circulation 98, 2262–2268.

Suzuki, T., Kumazaki, T., Mitsui, Y., 1993. Endothelin-1 is produced and secreted by neonatal rat cardiac myocytes in vitro. Biochem. Biophys. Res. Commun. 191, 823–830.

Svartholm, E., Bregqvist, D., Hedner, U., Ljungberg, J., Haglund, U., 1989. Thrombaxane A2-receptor blockade and prostacyklin in porcine *Escherichia coli* shock. Arch. Surg. 124, 669–672.

Takahashi, K., Jones, P.M., Kanse, S.M., Lam, H.C., Spokes, R.A., Ghatei, M.A., Bloom, S.R., 1990. Endothelin in the gastrointestinal tract. Presence of endothelin-like immunoreactivity, endothelin-1 messenger RNA, endothelin receptors, and pharmacological effect. Gastroenterology 99, 1660–1667.

Tanikawa, K., 1995. Hepatic sinusoidal cells and sinusoidal circulation. J. Gastroenterol. Hepatol. 10, S8–S11.

Tomoda, H., 1993. Plasma endothelin-1 in acute myocardial infarction with heart failure. Am. Heart J. 125, 667–672.

Turner, A., Tsamitros, M., Bellomo, R., 1999. Myocardial cell injury in septic shock [see comments]. Crit. Care Med. 27, 1775–1780.

Wagner, O.F., Christ, G., Wojta, J., Vierhapper, H., Parzer, S., Nowotny, P.J., Schneider, B., Waldhausl, W., Binder, B.R., 1992. Polar secretion of endothelin-1 by cultured endothelial cells. J. Biol. Chem. 267, 16066–16068.

Wanecek, M., 1999. The endothelin system and cardiopulmonary dysfunction in porcine endotoxine shock. Thesis.

Wanecek, M., Oldner, A., Rudehill, A., Sollevi, A., Alving, K., Weitzberg, E., 1997a. Cardiopulmonary dysfunction during porcine endotoxin shock is effectively counteracted by the endothelin receptor antagonist bosentan. Shock 7, 364–370.

Wacenek, M., Rudehill, A., Hemsen, A., Lundberg, J.M., Weitzberg, E., 1997b. The endothelin receptor antagonist, bosentan, in combination with the cyclooxygenase inhibitor, diclofenac, counteracts pulmonary hypertension in porcine endotoxin shock. Crit. Care Med. 25, 848–857.

Wanecek, M., Oldner, A., Rudehill, A., Sollevi, A., Alving, K., Weitzberg, E., 1999a. Endothelin A receptor antagonism attenuates pulmonary hypertension in porcine endotoxin shock. Eur. Respir. J. 13, 145–151.

Wanecek, M., Oldner, A., Sundin, P., Alving, K., Weitzberg, E., Rudehill, A., 1999b. Effects on hemodynamics by selective endothelin ET(B) receptor and combined endothelin ET(A)/ET(B) receptor antagonism during endotoxin shock. Eur. J. Pharmacol. 386, 235–245.

Weitzberg, E., 1994. Porcine basal pulmonary and systemic vascular tone is endothelin-dependent. Acta Anaesthesiol. Scand.

Weitzberg, E., Ahlborg, G., Lundberg, J.M., 1991a. Long-lasting vasoconstriction and efficient regional extraction of endothelin-1 in human splanchnic and renal tissues. Biochem. Biophys. Res. Commun. 180, 1298–1303.

Weitzberg, E., Lundberg, J.M., Rudehill, A., 1991b. Elevated plasma levels of endothelin in patients with sepsis syndrome. Circ. Shock 33, 222–227.

Weitzberg, E., Ahlborg, G., Lundberg, J.M., 1993. Differences in vascular effects and removal of endothelin-1 in human lung, brain, and skeletal muscle. Clin. Physiol. 13, 653–662.

Weitzberg, E., Hemsen, A., Lundberg, J.M., Ahlborg, G., 1995a. ET-3 is extracted by and induces potent vasoconstriction in human splanchnic and renal vasculatures. J. Appl. Physiol. 79, 1255–1259.

Weitzberg, E., Lundberg, J., Rudehill, A., 1995b. Inhibitory effects of diclofenac on the endotoxin shock response in relation to endothelin turnover in the pig. Acta Anaesthesiol. Scand. 39, 50–59.

Weitzberg, E., Hemsén, A., Rudehill, A., Modin, A., Wanecek, M., Lundberg, J.M., 1996. Bosentan-improved cardiopulmonary vascular performance and increased plasma levels of endothelin-1 in porcine endotoxin shock. Br. J. Pharmacol. 118, 617–626.

Whittle, B.J., Lopez-Belmonte, J., 1993. Actions and interactions of endothelins, prostacyclin and nitric oxide in the gastric mucosa. [Review]. J. Physiol. Pharmacol. 44, 91–107.

Wilson, M.A., Steeb, G.D., Garrison, R.N., 1993. Endothelins mediate intestinal hypoperfusion during bacteremia. J. Surg. Res. 55 (2), 168–175.

Xu, D., Emoto, N., Giaid, A., Slaughter, C., Kaw, S., deWit, D., Yanagisawa, M., 1994. ECE-1: a membrane-bound metalloprotease that catalyzes the proteolytic activation of big endothelin-1. Cell 78, 473–485.

Yamamoto, S., Burman, H.P., O'Donnell, C.P., Cahill, P.A., Robotham, J.L., 1997. Endothelin causes portal and pulmonary hypertension in porcine endotoxemic shock. Am. J. Physiol. 272, H1239–H1249.

Yamanaka, N., Takaya, Y., Oriyama, T. et al., 1997. Hepatoprotective effect of a nonselective endothelin receptor antagonist (TAK-044) in the transplanted liver. J. Surg. Res. 70, 156–160.

Yanagisawa, M., Kurihara, H., Kimura, S., Tomobe, Y., Kobayashi, M., Mitsui, Y., Yazaki, Y., Goto, K., Masaki, T., 1988. A novel potent vasoconstrictor peptide produced by vascular endothelial cells. Nature 332, 411–415.

Yoshizumi, M., Kurihara, H., Sugiyama, T., Takaku, F., Yanagisawa, M., Masaki, T., Yazaki, Y., 1989. Hemodynamic shear stress stimulates endothelin production by cultured endothelial cells. Biochem. Biophys. Res. Commun. 161, 859–864.

Zeni, F., Freeman, B., Natason, C., 1997. Anti-inflammatory therapies to treat sepsis and septic shock: a reassessment [editorial; comment]. Crit. Care Med. 25, 1095–1100.

Zhang, J.X., Bauer, M., Clemens, M.G., 1995. Vessel- and target cell-specific actions of endothelin-1 and endotheline-3 in rat liver. Am. J. Physiol. 269, G269–G277.

Zhang, B., Calmus, Y., Wen, L., Sogni, P., Lotersztajn, S., Houssin, D., Weill, B., 1997. Endothelin-1 induces liver vasoconstriction through both ET_A and ET_B receptors. J. Hepatol. 26, 1104–1110.

Zhang, Y., Oliver, J.R., Horowitz, J.D., 1998. Endothelin B receptor-mediated vasoconstriction induced by endothelin A receptor antagonist. Cardiovasc. Res. 39, 665–673.

Zhu, Y., Yang, H.T., Endoh, M., 1997. Negative chronotropic and inotropic effects of endothelin isopeptides in mammalian cardiac muscle. Am. J. Physiol. 273, H119–H127.