

Review

The Central Role of Endothelial Dysfunction in Cardiorenal Syndrome

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Angiotensin-converting enzyme inhibitor · Asymmetric dimethylarginine · Cardiorenal syndrome · Endothelial activation · Type 1 diabetes · Endothelial dysfunction · Nitric oxide · Oxidative stress · Statins

Abstract

Background: Endothelial dysfunction (ED) has emerged as a critical process in cardiorenal syndrome (CRS). The concept that ED is closely linked with cardiac and renal dysfunction has become an important target for CRS-related research and clinical practice. **Summary:** The sequence of events leading to ED is initiated by type I endothelial activation (almost immediately) and type II endothelial activation (over hours, days, and even months), followed by endothelial apoptosis and endothelial necrosis. The fact that ED is a continual cellular event divides this process into reversible ED (endothelial activation) and irreversible ED (endothelial apoptosis and necrosis). This basic research-defined concept may have clinical implications. Although most antihypertensive drugs (ACE inhibitors, statins, etc.) are effective in patients with hypertension and diabetes, some of them have proved to be ineffective, which may partly be attributed to irreversible ED. Even though the etiology of ED consists mainly of asymmetric dimethylarginine, nitric oxide, oxidative stress, and anti-endothelial cell antibodies, many other inducers of ED have been identified. In addition, a distinct role of ED has been reported for each type of CRS in humans. **Key Messages:** Further study is warranted to prove whether ED holds promise as a pharmacological target in CRS patients.

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Introduction

Over the past decade, endothelial dysfunction (ED) has been recognized as a contributor to the crosstalk between cardiac dysfunction and renal dysfunction [1–6]. Therefore, ED has emerged as a critical process in cardiorenal syndrome (CRS) [1–6]. The concept of ED as a link between cardiovascular disease and chronic kidney disease in CRS was reinforced by evidence of asymmetric dimethylarginine-mediated ED [7]. More recently, ED has been reported to be associated with type 2 CRS [8], type 1 diabetes mellitus [9], and chronic CRS [10].

In this review, it is our goal to introduce the concept of ED as defined by continuous cellular events from endothelial activation to endothelial apoptosis and necrosis, to propose a concept of reversible versus irreversible ED in relation to contradictory results of drugs for ED in patients, to discuss the etiology of ED associated with heart and kidney diseases, to seek potential inducers of ED in the literature, and to elucidate the roles of ED in the pathogenesis of CRS. Finally, this review provides succinct information about the different pathogeneses of ED in hypertensive and diabetic subjects.

A search for original, peer-reviewed articles written in English from 1993 to December 2015 was performed in MEDLINE and PubMed. The search terms used, alone and in combination, were “endothelial activation,” “endothelial dysfunction,” “endothelial apoptosis,” “endothelial necrosis,” “acute or chronic kidney diseases,” “asymmetric dimethylarginine,” “nitric oxide,” “oxidative stress,” “anti-endothelial cell antibody,” “heart failure,” “kidney failure,” “cardiorenal syndrome,” “statins,” “angiotensin-converting enzyme inhibitors,” and “ β -blockers.”

The Concept of ED

Terms such as “endothelial dysfunction,” “endothelial cell activation,” and “endothelial damage/injury” are currently used interchangeably in the medical literature [11–16]. Although widely used, ED still does not have a well-accepted definition [14]. In terms of arterial stiffness, linking cardiac and renal disease, several authors have referred to ED as a maladapted endothelial phenotype characterized by reduced nitric oxide (NO) bioavailability, increased oxidative stress, elevated expression of proinflammatory and prothrombotic factors, and reduced endothelium-derived vasodilation [11]. On the other hand, in terms of de novo protein synthesis or gene transcription, ED can be defined as a series of cellular alterations of the endothelium [15, 16] (Fig. 1). The sequence of events leading to ED may be described as

- (1) type I endothelial activation, in which the surface of the activated endothelium is capable of shedding prestored proteins such as endothelial adhesion and antithrombotic molecules (P-selectin, thrombin, heparin, von Willebrand factor, antithrombin III, and thrombomodulin), thereby requiring no de novo protein synthesis (in addition, a set of protective genes [NF- κ B inhibitor- α , A20, and *Bcl-2*] are constitutively expressed within the endothelial cell; NF- κ B inhibitor- α is a specific inhibitor of NF- κ B, and A20 and *Bcl-2* are antiapoptotic genes; these genes downregulate the expression of the transcription factor NF- κ B, thereby requiring no gene transcription);
- (2) type II endothelial activation, in which de novo protein synthesis and gene transcription are required; activation of NF- κ B triggers endothelial cell activation and provides the endothelium with new capacities and new functions; as a result, activated endothelial cells release new proteins (E-selectin, intercellular adhesion molecule-1, vascular cell adhesion molecule-1, tissue factor, monocyte chemoattractant protein-1, etc.);
- (3) endothelial apoptosis; and
- (4) endothelial necrosis [16].

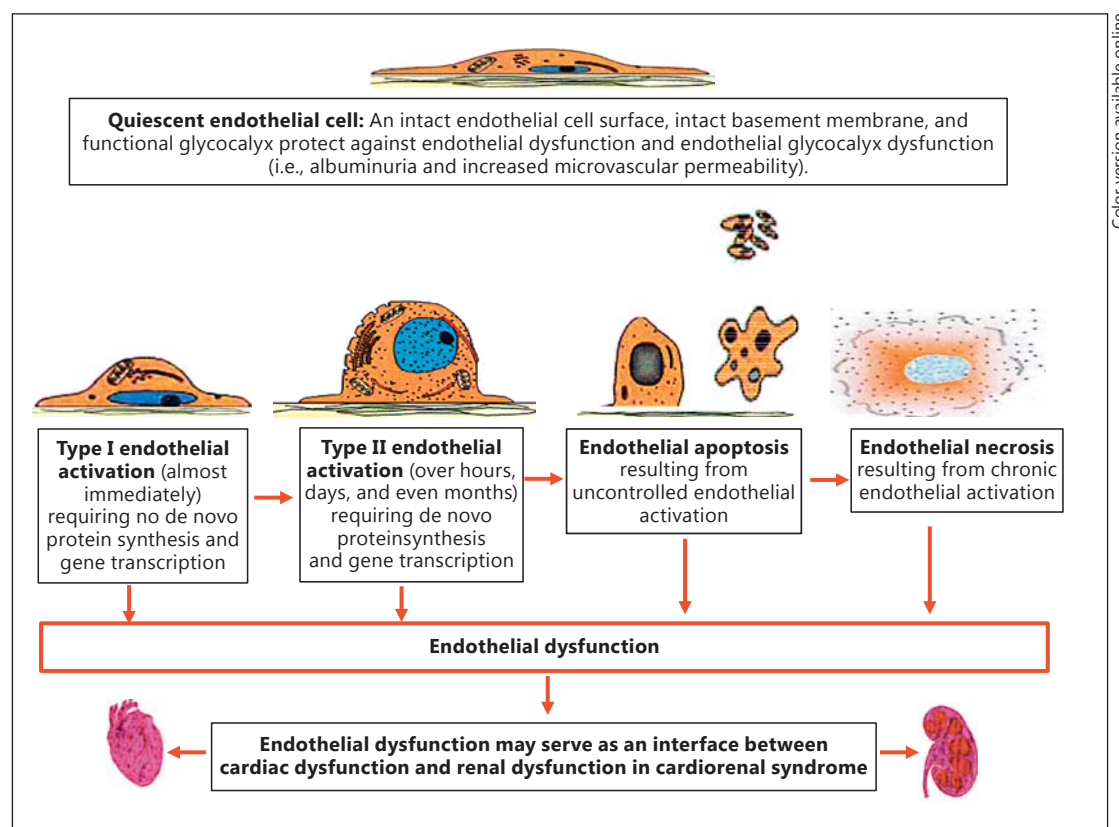
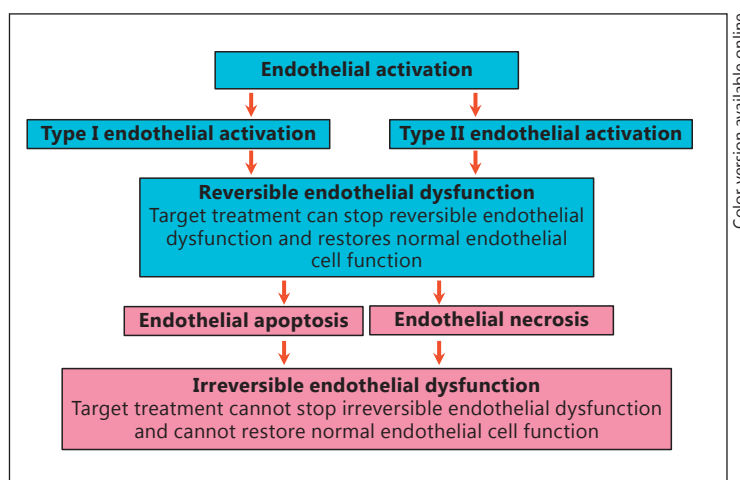


Fig. 1. Schematic representation of the concept of endothelial dysfunction. The sequence of cellular events leading to endothelial dysfunction begins as early as immediately for type I endothelial cell (EC) activation, followed by type II EC activation, EC apoptosis, and EC necrosis. Type I EC activation does not require de novo protein synthesis by means of immediately releasing prestored proteins (von Willebrand factor, P-selectin, thrombin, and histamine), nor does gene transcription by means of a set of protective genes ($\text{I}\kappa\text{B}\alpha$, A20, Bcl-2). Conversely, type II EC activation requires de novo protein synthesis by means of releasing new proteins (E-selectin, ICAM-1, VCAM-1, tissue factor, etc.) and requires activation of transcription factor NF- κB . The early events of EC activation represent reversible cell injury, while the late events of EC apoptosis and necrosis represent irreversible cell injury. See the section The Concept of Endothelial Dysfunction in the text for details [12, 15, 16]. Reprinted and modified from Zhang et al. [16] with permission.

Reversible versus Irreversible ED

The early events of endothelial activation (less than a few hours or days or months) represent reversible ED, whereas late events of endothelial damage/injury (apoptosis and necrosis) represent irreversible ED due to chronic and persistent ED. Endothelial activation is distinct from endothelial cell injury. Endothelial activation represents the alterations resulting in morphological rearrangement, such as an increase in cell size and cytoplasmic organelles and in inducible new functions, but without loss of endothelial integrity [16]. The phenotype of activated endothelial cells may return to the quiescent, nonactivated phenotype after the insult resolves. However, the endothelial activation process, if uncontrolled, can progress to endothelial apoptosis that is characterized by endothelial fragmentation and endothelial separation from the intima [16]. Moreover, endothelial necrosis indicates that the cellular injury is severe and persistent. In endothelial necrosis, mitochondria undergo progressive swelling resulting in cell death [16].

Fig. 2. Schematic representation of the relationship of reversible and irreversible endothelial dysfunction with target treatment [15–21].



Color version available online

In this review, we propose that the concept of reversible versus irreversible ED may have clinical implications (Fig. 2). Whether ED is reversible has long been a subject of concern to researchers in clinical trials [17–21]. Panza et al. [17] concluded that clinically effective antihypertensive therapy does not restore impaired endothelium-dependent vascular relaxation in patients with essential hypertension. Their results indicate that such ED is either primary or becomes irreversible once the hypertensive process has become established. Schmieder and Schobel [18] reported that chronic antihypertensive therapy does not restore ED in patients with essential hypertension; however, they observed a beneficial effect of fluvastatin in patients with hypercholesterolemia. In contrast to the above-mentioned opinions, Celermajer [19] raised the possibility that ED might be reversible with certain interventional strategies. However, this exciting possibility has been challenged by new studies. Hadi and Suwaidi [20] reviewed the role of various modalities of therapy for ED. They found out that many drugs (cerivastatin, atorvastatin, simvastatin, pentoxifylline, vitamin E, and vitamin C) have no effect on ED. Taddei et al. [21] also reviewed the effect of antihypertensive drugs on ED. Although some studies have shown restoration of ED by antihypertensive drugs, this beneficial effect has not been observed in all studies [21]. Table 1 exemplifies the conflicting effects of antihypertensive drugs on ED from selected clinical trials [18, 22–37].

For example, in a patient with type 5 CRS, torsemide had only transient beneficial effects [14]. Despite aggressive treatment with torsemide, hemodialysis, etc., the patient died 9 days later due to end-stage multiple organ failure (cardiac, renal, and pulmonary failure). In this case, from a clinical perspective, improvement in renal function could solely be because of augmentation of cardiac and improved renal perfusion due to inotropic effects. However, from a basic science perspective, hemodialysis-induced ED is likely to play a role (see subsection Inducers of ED below). The loss of sensitivity to changing hemodynamic dysfunction is at least in part related to irreversible ED, which leads to loss of endothelial barrier integrity in systemic vessels and loss of endothelial barrier integrity resulting from endothelial cell membrane damage and endothelial permeability.

Etiology of ED

Asymmetric Dimethylarginine-Mediated ED

Asymmetric dimethylarginine, an inhibitor of nitric oxide synthase, is a potent causal factor for ED in chronic kidney disease; thus, asymmetric dimethylarginine-mediated ED may

Table 1. Conflicting effects of antihypertensive drugs on ED in patients

Study [Ref.], year	Drug(s)	Beneficial effect on ED	Implications
Mancini et al. [22], 1996	Quinapril (ACE inhibitor)	Yes	In normotensive patients with coronary artery disease, without heart failure and severe hyperlipidemia, the amelioration of the endothelium with quinapril could be attributed in part to reversible ED that still has the capacity for releasing NO to protect against angiotensin II and bradykinin
Clarkson et al. [23], 1996	L-Arginine (substance for NOS)	Yes	In young hypercholesterolemic adults (mean age 29 years) without hypertension, diabetes, and cigarette smoking, dietary supplementation with L-arginine improves endothelium-dependent dilation
Dawes et al. [24], 1999	Nebivolol (β -blocker)	Yes	In patients with uncomplicated essential hypertension, the vasodilator effect of nebivolol is probably related to the ratio of reversible to irreversible ED
Konduracka et al. [27], 2008	Atorvastatin (statin)	Yes	In patients with type 1 diabetes mellitus, without coronary heart disease and arterial hypertension, improvement of ED with atorvastatin is proved by a reduction of plasminogen activator inhibitor-1 and C-reactive protein (biomarkers of endothelial activation)
Lekakis et al. [25], 2002	L-Arginine (substance for NOS)	Yes	In patients with essential hypertension, the beneficial effects of L-arginine may be attributed to NO release
Dogra et al. [26], 2005	Atorvastatin (statin)	Yes	In type 1 diabetes mellitus patients with microalbuminuria, the beneficial effects may be related to reduced oxidative stress and increased NO availability
Zhang et al. [28], 2012 (review article)	Cerivastatin, atorvastatin, simvastatin (statins)	Yes/no	In 845 patients with diabetes mellitus from 10 statin studies, the meta-analysis revealed that statin therapy significantly improved flow-mediated dilatation (a clinical marker of ED), while in a subgroup analysis, patients with a BMI >27.6 did not benefit from statin therapy; the non-effect of statins in subjects with a high BMI may be attributed to irreversible ED and adipocytokines as inducers of ED
Panza et al. [17], 1993	Acetylcholine, sodium nitroprusside	No	In hypertensive patients, ED is either primary or secondarily irreversible with time; under the irreversible condition, the 2 drugs cannot restore endothelium-dependent vascular relaxation
Creager and Roddy [29], 1994	Captopril, enalapril (ACE inhibitors)	No	In hypertensive patients with impaired vasodilative capacity, the 2 ACE inhibitors cannot restore endothelium-dependent vasodilation, which is suggestive of irreversible ED
Schiffrin and Deng [30], 1996	Atenolol (β -blocker)	No	In hypertensive patients, thicker small arteries with abnormal endothelium-dependent relaxation and altered contractility cannot be restored by atenolol, which is suggestive of irreversible ED
Kiowski et al. [31], 1996	Cilazapril (ACE inhibitor)	No	In patients with hypertension, the negative effect of cilazapril on ED does not support the view that cilazapril influences endothelial vasodilator function
Anderson et al. [32], 2000	Quinapril, enalapril (ACE inhibitors); losartan (angiotensin II blockade); amlodipine (calcium channel antagonist)	Yes/no	In patients with coronary disease, quinapril – but not enalapril, losartan, and amlodipine – improves flow-mediated vasodilation, and the effectiveness of quinapril is related to ACE genotype
Taddei et al. [33], 2001	Lacidipine (calcium antagonist); atenolol (β_1 -blocker)	Yes/no	In patients with essential hypertension, lacidipine but not atenolol increases endothelium-dependent vasodilation by restoring NO availability
van de Ree et al. [34], 2001	Simvastatin (statin)	No	In patients with type 2 diabetes, restoration of impaired endothelial function by simvastatin is not proved
van Etten et al. [35], 2002	Atorvastatin (statin)	No	In patients with type 2 diabetes, atorvastatin has no effect on NO availability; except for hyperlipidemia, hyperglycemia is likely responsible for the impaired vasoreactivity
Beishuizen et al. [36], 2005	Cerivastatin (statin)	No	In patients with type 2 diabetes without manifest cardiovascular disease, cerivastatin has no effect of flow-mediated dilation
Sozen et al. [37], 2009	Irbesartan, valsartan (ARB); fosinopril, quinapril (ACE inhibitors)	Yes/no	In patients with essential hypertension, both ACE inhibitors and ARB improve endothelial function at the start of treatment, but they are not maintained long term (1 and 3 years); the results indicate that ED may be resistant or irreversible

This table just exemplifies conflicting effects of drugs from selected reports; we did not intend to comprehensively search for all clinical trials. Readers are referred to review articles for comprehensive information [22, 23, 28]. ED, endothelial dysfunction; ACE inhibitor, angiotensin-converting enzyme inhibitor; ARB, angiotensin receptor blocker; NOS, nitric oxide synthase; NO, nitric oxide; BMI, body mass index.

contribute to the pathogenesis of CRS as a link between cardiovascular disease and chronic kidney disease in CRS patients [7]. An attractive hypothesis was proposed by Ueda et al. [7] to explain a possible role of asymmetric dimethylarginine in CRS. During the bidirectional interaction between cardiac disease and kidney disease in CRS, asymmetric dimethylarginine acts as a major causative factor in the induction of ED via a pathway of increased endogenous nitric oxide synthase. On the one hand, ED induced by asymmetric dimethylarginine contributes to cardiovascular disease, but on the other hand, the ED could increase asymmetric dimethylarginine via 2 pathways within the kidney.

The first pathway is involved in the sequence of events that is initiated by injury to the renal microvasculature (renal ischemia), followed by interstitial fibrosis and glomerular sclerosis that leads to chronic kidney disease. Oxidative stress and the renin-angiotensin system in chronic kidney disease then result in dysregulation of dimethylarginine dimethylaminohydrolase (DDAH) and protein arginine methyltransferase (PRMT). It is DDAH and PRMT that increase asymmetric dimethylarginine in chronic kidney disease. The second pathway for increased asymmetric dimethylarginine is through proteinuria and resultant dysregulation of DDAH and PRMT [7]. For prevention of CRS in patients with chronic kidney disease, a novel therapeutic strategy may be focused on asymmetric dimethylarginine and DDAH as a drug target, i.e., on reducing levels of asymmetric dimethylarginine by enhancement of DDAH activity or suppression of PRMTs [7]. In addition, asymmetric dimethylarginine appears to be an emerging risk factor for cardiovascular disease and the development of chronic kidney disease [38]. Accumulated asymmetric dimethylarginine in chronic kidney disease is believed to be a missing link between cardiovascular disease and chronic kidney disease [7, 39]. Böger et al. [40] found that simvastatin improves endothelial function in subjects with low levels of asymmetric dimethylarginine, but not in subjects with higher levels of asymmetric dimethylarginine. Plasma levels of asymmetric dimethylarginine were found to be elevated in patients with end-stage renal disease. Such high levels of asymmetric dimethylarginine are considered an important risk factor for chronic vascular disease in end-stage renal disease patients [41]. Moreover, asymmetric dimethylarginine and oxidative stress are suggested to be predisposing factors for ED in chronic kidney disease [42].

Nitric Oxide-Mediated ED

Nitric oxide is an endothelium-derived relaxing factor which opposes the actions of endothelium-derived contracting factors such as angiotensin II and endothelin-1 [43]. A balance between endothelium-derived relaxing and contracting factors is important in maintaining vascular homeostasis [43]. Nitric oxide is synthesized from L-arginine under the influence of the enzyme nitric oxide synthase [43]. On the other hand, nitric oxide can be inactivated by superoxide, thereby generating peroxynitrite and then decreasing nitric oxide activity [43]. Rajapakse et al. [8] published a comprehensive review regarding the role of the L-arginine-nitric oxide pathway. They emphasize that reduced bioavailability of nitric oxide plays a role in the induction of ED, and the impaired L-arginine-nitric oxide pathway participates in the pathogenesis of CRS. In chronic heart failure patients with hemodynamic compromise, inducible nitric oxide synthase levels were significantly higher than those in age-matched, healthy subjects [44]. The higher level of inducible nitric oxide synthase further impairs endothelial function by increasing the local production of reactive oxygen species [44]. Nitric oxide-mediated ED is also suggested to be a predisposing factor for cardiovascular complications in chronic kidney disease [45]. Nitric oxide is a particularly important endothelium-derived mediator, because it has the capacity to possess vasodilative, antiplatelet, antiproliferative, antiadhesive, permeability-decreasing, and anti-inflammatory properties [43, 46]. Stehouwer [46] proposed that decreased nitric oxide availability is related to endothelial activation.

Oxidative Stress-Caused ED

Oxidative stress is defined as an imbalance between prooxidants and antioxidants that is the result of an imbalance between the generation of reactive oxygen species and the antioxidant defense mechanism [45]. In patients with insulin-dependent diabetes mellitus, ED is associated with excessive generation of free radicals and oxidant injury [47]. In hypertensive patients and normotensive subjects, a reduction in nitric oxide availability participates in age-related ED [48]. In patients with end-stage renal disease, a single session of hemodialysis impaired endothelium-dependent vasodilation by an acute increase in oxidative stress [49]. In patients with polycystic kidney disease, oxidative stress and ED are evident in the early stage [50]. In the cardiorenal metabolic syndrome, mitochondrial oxidative stress is believed to contribute to a vicious cycle of enhanced oxidative stress and mitochondrial dysfunction, and alcohol further augments mitochondrial oxidative stress induced by overnutrition associated with the metabolic disease [51]. In addition, oxidative stress in association with endothelial activation and ED is involved in the development of CRS [52].

An elegant review on the role of oxidative stress in the induction of ED was published by Lum and Roebuck [53]. They pointed out that acute (minutes) versus chronic oxidant stress (more than several hours) is a serious causative factor for vascular ED. There is evidence that treatment of human umbilical vein endothelial cells with interleukin-1 and interferon- γ results in dose- and time-dependent increases in superoxide radicals. Superoxide may also react with nitric oxide to form peroxynitrite, which decomposes to form highly reactive hydroxyl radicals and nitrite dioxide [53]. The effects of oxidative stress on the vascular endothelium consist of an increase in vascular endothelial permeability and in endothelial adhesion for leukocytes [53]. The 2 aspects (increased permeability and leukocyte adhesion) are coupled with alterations in endothelial signal transduction and redox-regulated transcription factors such as activator protein-1 and NF- κ B [53].

Anti-Endothelial Cell Antibody-Induced ED

Anti-endothelial cell antibodies can cause ED in patients with granulomatosis with polyangiitis or lupus erythematosus. By now it has become clear that antibodies to kidney endothelial cells contribute to the “leaky” glomerular barrier in chronic kidney disease patients, indicating renal ED-induced alterations in glomerular vascular permeability [54]. The mechanism whereby anti-endothelial cell antibodies increase glomerular vascular permeability is not completely characterized, but it is likely based on decreased expression of both adherens and tight junction proteinases (i.e., vascular endothelial [VE]-cadherin, etc.) [54]. Kidney biopsies from end-stage renal disease patients with anti-endothelial cell antibodies revealed a marked decrease in adherens and tight junctions in the glomerular endothelium [55]. In addition, anti-endothelial cell antibodies in type 2 diabetes mellitus can cause strong endothelial cell contraction (type 1 endothelial activation) and endothelial apoptosis in vitro. It was proposed that anti-endothelial cell antibodies may contribute to the progression of diabetic nephropathy, and that they may be used as markers for this progression [55]. In particular, an interaction between anti-endothelial cell antibodies and albuminuria predicts the composite endpoint of death, end-stage renal disease, or substantial decline in renal function in older adults with type 2 diabetic nephropathy [55].

Inducers of ED

In addition to the above-cited inducers of ED (asymmetric dimethylarginine, nitric oxide, oxidative stress, and anti-endothelial cell antibodies), many other factors can induce ED. Table 2 lists inducers of ED to be found in the literature, though not complete. It categorizes them into transcription factors, major inducers associated with cardiac and renal diseases, cytokines, adipocytokines, drugs, medical interventions, endotoxins, and miscellaneous factors [16, 49,

Table 2. Inducers of endothelial dysfunction

Transcription factors	Nuclear factor- κ B and CD40/CD40 ligand signaling (which is enhanced by activation of nuclear factor- κ B)
Major inducers associated with cardiac and renal diseases	Asymmetric dimethylarginine, nitric oxide, oxidative stress, and anti-endothelial antibody
Cytokines	Tumor necrosis factor- α , interleukin-1, interleukin-18, and interferon
Adipocytokines	Adiponectin, leptin, resistin, apelin, and visfatin
Drugs	Angiotensin II, endothelin-1, phosphodiesterase inhibitors, bleomycin, cisplatin, erythropoietin, cyclosporine, rapamycin, and tacrolimus
Medical interventions	Balloon angiography, stenting, and hemodialysis per se (hemodialysis-induced endothelial dysfunction)
Endotoxins	Lipopolysaccharide
Miscellaneous factors	Histamine, thrombin, hyperhomocysteinemia, hypercholesterolemia, dyslipidemia, C-reactive protein, and high-fat diet with hypoxia
See references 16, 49, and 54.	

54]. However, some factors are merely associated with ED, and thus the association does not necessarily imply “true” inducement. Recently, Malyszko [54] reviewed the relationship of adipocytokines with ED. Possible relations between adipocytokines and markers of ED were identified. Hemodialysis is also included in Table 2, because hemodialysis induces oxidative stress and impairs nitric oxide bioavailability [49, 56]. Zhao et al. [57] revealed in an animal study that in the setting of hypoxia, a high-fat diet leads to earlier and more severe ED than with hypoxia alone. Hypoxia combined with a high-fat diet results in an impaired endothelium-dependent vasorelaxation response to acetylcholine, alters the bioavailability of the nitric oxide synthase substance L-arginine, and blunts increases in endothelial nitric oxide synthase mRNA and protein in aortic endothelial tissue [57]. More importantly, in rats on a high-fat diet up to 32 weeks, once ED is developed, the high-fat diet promotes progressive impairment of coronary and mesenteric ED. Continuous vascular adaptation to a high-fat diet makes the reversibility of ED impossible [58]. The results of this study may have clinical implications with regard to the pathogenesis of impairment of vascular relaxation by obesity in humans.

The Role of ED in the Pathogenesis of CRS

The pathogenesis of CRS is complex and remains unclear. Table 3 summarizes the role of ED in relation to the definition of CRS [52].

Pathogenesis in the Setting of CRS Type 1

Cohen [4] reviews the pathophysiology of CRS type 1, providing a possible sequence of ED in the pathogenesis of CRS. ED induced by shear stress, angiotensin II, aldosterone, and inflammatory cytokines leads to decreased glomerular filtration rates and diuretic efficacy, and this in turn results in fluid retention, decreased cardiac output, venous congestion, and decreased renal perfusion [4]. A recent study provided insight into the pathogenesis of CRS type 1, emphasizing the pivotal role of oxidative stress in CRS type 1 [59]. In this study of 23 patients with acute heart failure, 11 patients subsequently developed acute kidney injury due

Table 3. The major roles of ED in each type of CRS

	Definitions of CRS	Major roles of ED
Type 1	Acute CRS: heart failure leading to acute kidney disease	ED induced by shear stress, angiotensin II, aldosterone, inflammatory cytokines leading to renal dysfunction (glomerular filtration rate, diuretic efficacy, fluid retention, etc.)
Type 2	Chronic CRS: chronic heart failure leading to kidney failure	ED induced by abnormal neurohormonal activation leading to cardiac and renal dysfunction via the L-arginine-nitric oxide pathway
Type 3	Acute renocardiac syndrome (acute kidney disease leading to acute heart failure)	ED markers (endothelin-1, von Willebrand factor, C-reactive protein) accelerate progression of kidney disease, with increased permeability of the glomerular membrane, leading to a decreased ejection fraction of the heart
Type 4	Chronic renocardiac syndrome (chronic kidney disease leading to heart failure)	Irreversible ED (depletion of endothelial cells and disturbance of hemodynamics), with resultant endothelium-dependent vasodilation in chronic kidney diseases, leading to heart failure
Type 5	Secondary CRS (systemic disease leading to heart and kidney failure)	Irreversible ED (severely systemic endothelial damage) with coagulopathy in severe sepsis and septic shock, leading to disturbance of systemic hypocoagulopathy
References: definition of CRS [52]; major roles of ED [2, 4, 8, 59–62]. ED, endothelial dysfunction; CRS, cardiorenal syndrome.		

to CRS type 1. The study revealed that levels of oxidative stress markers (myeloperoxidase, nitric oxide, copper/zinc superoxide dismutase, and endogenous peroxidase activity) were significantly higher in CRS type 1 than in acute heart failure without CRS type 1 and in healthy controls. It also highlighted dual oxidative stress pathways via both reactive oxygen species and reactive nitrogen species [59].

Pathogenesis in the Setting of CRS Type 2

It has been proposed that abnormal neurohormonal activation and subsequent impairment of the L-arginine-nitric oxide pathway and resultant ED play a critical role in the pathogenesis of CRS type 2 [8]. In the scenario of CRS type 2 pathogenesis, ED is a key factor for cardiac and renal dysfunction via the L-arginine-nitric oxide pathway, in which low bioavailability of nitric oxide results in increased levels of reactive oxygen species. Therefore, oxidative stress contributes to cardiac and renal failure in CRS type 2 [8].

Pathogenesis in the Setting of CRS Type 3

ED is implicated in the development of CRS in patients with type 1 diabetes mellitus [2]. The clinical characteristics of chronic kidney disease (diabetic nephropathy) with decreased cardiac function (ejection fraction) in these patients are consistent with type 3 CRS. Elevated levels of markers of ED such as endothelin-1, von Willebrand factor, and C-reactive protein were found to be correlated with increased blood pressure and also permeability of the glomerular membrane. Plasma or serum levels of ED markers increased with progression of kidney disease. Left ventricle mass was also noted to be correlated with ED markers and stage of renal disease [2].

Pathogenesis in the Setting of CRS Type 4

Reversible or irreversible ED exerts different effects on endothelium-dependent vasodilation in patients with type 1 diabetes mellitus at different stages of diabetic nephropathy

[60]. Shestakova et al. [60] divided diabetic patients into 4 groups: group 1 (without renal affection), group 2 (with microalbuminuria), group 3 (with proteinuria), and group 4 (with chronic renal failure). Healthy subjects were used as controls. Two parameters, i.e., dilation of the artery resulting from reactive hyperemia and the coefficient of endothelial sensitivity to shift tension, were assessed. Dilation was 9.2% in group 1, 9.6% in group 2, 7.3% in group 3, and 4.4% in group 4. The coefficient of endothelial sensitivity was 0.084 in the control group, 0.083 in group 1, 0.14 in group 2, 0.07 in group 3, and 0.05 in group 4. These results demonstrate that at the earliest stage of diabetic nephropathy with microalbuminuria, endothelium-dependent vasodilation is reversible by early treatment, but at the stage of overt proteinuria and chronic renal failure, endothelium-dependent vasodilation is irreversible. These findings indicate that the irreversible effect is likely due to depletion of endothelial cells and loss of sensitivity to changing hemodynamic conditions [60]. Similar results of endothelium-dependent vasodilation in terms of ED and endothelial apoptosis were reported for chronic kidney disease patients at an early stage [61]. In chronic kidney disease, the reduction of endothelium-dependent vasodilation was 34% in stage I, 52% in stage II, 52% in stage IIIa, and 70% in stage IIIb. Moreover, decrease in endothelium-dependent vasodilation was associated with glomerular filtration rate, homocysteine level, and hemodynamic factors. In chronic kidney disease, an elevation of endothelin-1 levels was seen in stage I (41% of the patients), in stage II (54% of the patients), in stage IIIa (70% of the patients), and in stage IIIb (83% of the patients) [61].

Pathogenesis in the Setting of CRS Type 5

Vascular endothelial activation and damage, concurrent with coagulopathy and microcirculatory failure, are usually present in patients with severe sepsis [62]. Therefore, ED plays a critical role with coagulopathy in severe sepsis in balancing hemostasis. ED is independently associated with concurrent coagulopathy in severe sepsis. This attractive conclusion was reached by using viscoelastic hemostatic whole blood tests (thromboelastography) and an enzyme-linked immunosorbent assay measuring plasma/serum markers of endothelial activation and damage in 184 patients with severe sepsis and septic shock. This study indicates that ED (mainly endothelial damage and to a lesser extent endothelial activation) is intimately linked to functional whole blood hypocoagulopathy as assessed by thromboelastography and functional fibrinogen [62]. Moreover, excessive damage to the glycocalyx, as evidenced indirectly by high levels of syndecan-1 (an endothelium-derived marker), is inversely associated with functional hyperfibrinolysis on thromboelastography [62].

Different Pathological Mechanisms in Endothelial Cells of Hypertensive and Diabetic Subjects

Although ED is associated with the pathogenesis of many cardiovascular diseases such as hypertension and diabetes mellitus, the pathogeneses of these 2 diseases are unlikely based on precisely the same mechanism. The spontaneously hypertensive rat strain and blood pressure high mice were used for 2 independent rodent models of human essential hypertension [63]. The animal models showed that spontaneously hypertensive rats and blood pressure high mice have common genetic mechanisms of hypertension across mammalian species that might be pertinent to human hypertension [63]. Transcript pattern evidence was found for involvement of several systems in the pathology of hypertension in the 2 models. The transcript pattern evidence includes the following: (1) adrenal catecholamines and sympathetic function; (2) steroid hormone synthesis; (3) catabolism, and its contribution to enhanced glucocorticoid sensitivity in spontaneously hypertensive rats; (4)

oxidative stress, and its role as a common mechanism of vascular and end-organ injury; and (5) intermediary metabolism with global but mechanistically different perturbation in the 2 models. In addition, the 2 models share approximately 10% of the differentially expressed orthologous genes (overexpressed or underexpressed) [63]. Viridis et al. [64] studied the role of hyperhomocystinemia in normotensive subjects and patients with essential hypertension. They found that hyperhomocystinemia impaired endothelial function by producing oxidative stress that reduces nitric oxide availability, and hyperhomocystinemia can lead to a further reduction in endothelial function by exacerbating the production of oxidative stress [64]. It should be noted that in normotensive subjects, the endothelium modulates vascular tone mainly by the production of nitric oxide, whereas in hypertensive patients, essential hypertension is associated with impaired basal release of nitric oxide, which is likely to be a dysfunction secondary to increases in blood pressure [65].

Impairment of vasodilation due to ED in small-vessel disease is an important factor in diabetes [66]. The hyperglycemic state causes cell damage by promoting advanced glycation end products, by activating protein kinase C, and through polyol pathway activation [66]. Activation of the polyol pathway consumes nicotinamide adenine dinucleotide phosphate, which reduces endothelial nitric oxide synthase activity and decreases nitric oxide production, thereby causing ED [66]. There is also evidence that impairment of vasodilation mediated by endothelium-derived nitric oxide plays a role in animal models of diabetes and in patients with insulin-dependent and non-insulin-dependent diabetes mellitus [67]. It seems likely that the pathogenesis of diabetic vascular disease involves a reduced bioavailability of endothelium-derived nitric oxide [67]. By now it has become clear through human studies that nitric oxide synthase or its bioavailability within the periendothelial environment in type 2 diabetic patients is indeed reduced [68].

Conclusion

The molecular and cellular aspects of asymmetric dimethylarginine, oxidative stress, nitric oxide synthase, and low bioavailability of nitric oxide add to our understanding of the pathophysiology of ED in CRS. Further studies are warranted to investigate whether ED holds promise as a target in the treatment of CRS patients.

Statement of Ethics

This work did not involve human subjects or animals and was exempt from review by the Baylor Institutional Review Board.

Disclosure Statement

There are no conflicts of interest to disclose.

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