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MINI-REVIEW

Early Growth Response-1, an Integrative Sensor in Cardiovascular and Inflammatory Disease

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ABSTRACT: Early growth response-1 (Egr-1) is a master regulator and transcriptional sensor in vascular dysfunction and disease. This article reviews recent developments in our understanding of the regulatory roles this zinc finger protein and product of an immediate-early gene plays in a range of cardiovascular and inflammatory disorders. Egr-1 can amplify pathologic signals from the extracellular environment by serving as a molecular conduit in the inducible expression of proliferative, migratory and proinflammatory genes driving disease progression. Strategies targeting Egr-1 may provide therapeutic benefit in cardiovascular and inflammatory disorders.

Key Words: cardiovascular disease ■ early growth response-1 ■ inflammation ■ transcription factors ■ vascular dysfunction

Early growth response-1 (Egr-1) is an immediate-early gene that resides on human chromosome 5q23-q31 and encodes a Cys_2-His_2 type zinc finger transcription factor comprising 543 residues. The gene (also known as TIS8,[1] NGFI-A,[2] Krox-24,[3] ZIF-268,[4] and Egr1/EGR1[5]) was cDNA cloned in the mid-1980s from cultured human lymphocytes activated with lectin and cycloheximide, given the name "G0S30" (G0 to G1 Switch Gene 30) and a partial sequence deposited in GenBank.[6] Rapid and transient induction typifies Egr-1 activation in diverse human cell types responding to a range of agonists and conditions.[7] Egr-1 is activated by serum, cytokines, growth factors, hormones, mechanical injury, endotoxin, hypoxia and shear stress, among other stimuli. The Egr-1 promoter contains multiple serum response elements that support serum response factor interactions with ternary complex factors (such as Elk-1 and SAP-1) that undergo phosphorylation by mitogen-activated protein kinases (Figure). Inducible Egr-1 expression is reliant upon the activity of extracellular signal-regulated kinase (ERK), c-Jun N-terminal kinase, p38 kinase, protein kinase A, protein kinase B and protein kinase C. Egr-1 interacts with recognition elements in promoter/enhancer regions of responsive genes, including platelet-derived growth factor (PDGF)-A, PDGF-B, PDGF-C, tissue factor, collagen, suppressor of cytokine signaling-1, colony-stimulating factor 1 receptor and Egr-1 itself.[8] Trizzino et al[9] linked Egr-1 with the transcriptional control of ≈1600 genes during macrophage development.

Egr-1 physically interacts with multiple transcription and regulatory factors such as CREB-binding protein/p300, nuclear factor-κB(NFκB) p65, p53, and YAP-1 and can modulate gene expression by displacing or competing with Sp1 at overlapping binding sites.[10] Egr-1 is also able to suppress inflammatory genes by indirect recruitment and association with the nucleosome remodeling and deacetylation corepressor complex causing deacetylation, thereby reducing chromatin accessibility.[9] Egr-1 undergoes negative regulation by two corepressors, NGFI-A binding protein (NAB) 1, and NAB2. Egr-1 and other family members, Egr-2 and Egr-3, can induce the expression of NAB2, creating a negative autoregulatory feedback loop. Egr-1 is subject to post-translational modification through acetylation,

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Nonstandard Abbreviations and Acronyms

AGE	advanced glycation end products
ALI	acute lung injury
DKD	diabetic kidney disease
EGF	epidermal growth factor
Egr-1	early growth response-1
ERK	extracellular signal-regulated kinase
FGF	fibroblast growth factor
ICAM-1	intercellular adhesion molecule-1
IL-1β	interleukin-1β
IL-6	interleukin-6
LPS	lipopolysaccharide
MCP-1	monocyte chemoattractant protein-1
NAB	NGFI-A binding protein
NFκB	nuclear factor-κB
PAH	pulmonary arterial hypertension
PDGF	platelet-derived growth factor
PKC-ε	protein kinase C-ε
PPAR	peroxisome proliferator-activated receptors
RIPC	remote ischemic preconditioning
S1P	sphingosine-1-phosphate
SMC	smooth muscle cell
TGF-β	transforming growth factor-β

SUMOylation, ubiquitination and phosphorylation. For example, Vedantham et al[11] found Egr-1 hyperacetylation in hyperglycemia and induction of proinflammatory and prothrombotic signals by aldose reductase. Manente et al[12] reported that Egr-1 is SUMOylated and ubiquitinated in cells exposed to epidermal growth factor (EGF), and that this, together with proteasome mediated degradation, is enhanced by SUMO1/Ubc9 overexpression. Egr-1 also interacts with PRC8, a subunit of the proteasome core complex and undergoes multiubiquitination, destining it for proteolysis through the ubiquitin-dependent proteasome pathway.[13] While some of this work has been performed in nonvascular cell types, such processes may provide opportunities for therapeutic intervention in cardiovascular and inflammatory disease.

VASCULAR REMODELING, HYPERTENSION, AND HYPOXIC STRESS

Egr-1 is poorly expressed in the normal blood vessel wall but, when induced, can drive the expression of key growth factor and proinflammatory genes (e.g., PDGF-B, intercellular adhesion molecule-1 [ICAM-1], vascular cell adhesion molecule-1 [VCAM-1]) impacting pathogenesis (Table). For example, Egr-1 regulates atherosclerosis,[14] postangioplasty restenosis,[15] in-stent restenosis,[16] and vein graft stenosis.[17] Mice deficient in Egr-1 and apolipoprotein E have reduced atherosclerosis and vascular inflammation.[18] But how Egr-1 is controlled in the injured artery wall is not well understood. MicroRNAs are noncoding, single-stranded RNAs that can regulate gene expression by reducing levels of mRNA these target. We identified a microRNA targeting Egr-1 (miR-191) that suppresses smooth muscle cell (SMC) growth and intimal thickening after balloon catheter injury of carotid arteries.[19] Other microRNAs targeting Egr-1 include miR-181, miR-183 and miR-150-5p. Vascular injury is able to acutely release endogenous growth factors such as fibroblast growth factor (FGF)-2,[20] which can activate Egr-1 gene expression.[21] A cocktail composed of VEGF-A, VEGF-D, and cyclic Arg-Gly-Asp peptide inhibits both Egr-1 and neointima formation after balloon injury. This was associated with increased endothelial nitric oxide synthase expression and accelerated reendothelialization.[22]

SMC hypertrophy and vascular remodeling underpin hypertension, a process associated with angiotensin II. ERK is phosphorylated and Egr-1 is inducibly expressed in SMC exposed angiotensin II. Recent studies by Troung et al[23] indicate that angiotensin II–inducible Egr-1 expression is dependent upon the

Table. Involvement of Egr-1 in Various Cardiovascular and Inflammatory Disorders (See Text for Detail)

Disease or model of disease	Representative study(s)
Antenatal hypoxia	Xu et al[25]
Gestational diabetes	Rajaraman et al[32]
Diabetic kidney disease	Li et al[26] Ho et al[27] Brennan et al[28]
Angiogenesis	Fahmy et al[49] Lee et al[50] Ye et al[53]
Atherosclerosis	McCaffrey et al[14] Harja et al[18]
Postangioplasty restenosis	Santiago et al[15] Li et al[19]
In-stent restenosis	Lowe et al[16]
Vein graft stenosis	Zhang et al[17]
Ischemia-reperfusion injury	Rayner et al[34] Wang et al[35] Yamamoto et al[48]
Acute lung injury	Chen et al[38] Yan et al[39]
Hypertension	Dickinson et al[41]
Atopic dermatitis	Yeo et al[33]
Sepsis	Chen et al[44]
Graft-versus-host disease	Autieri et al[46] Yamamoto et al[48]

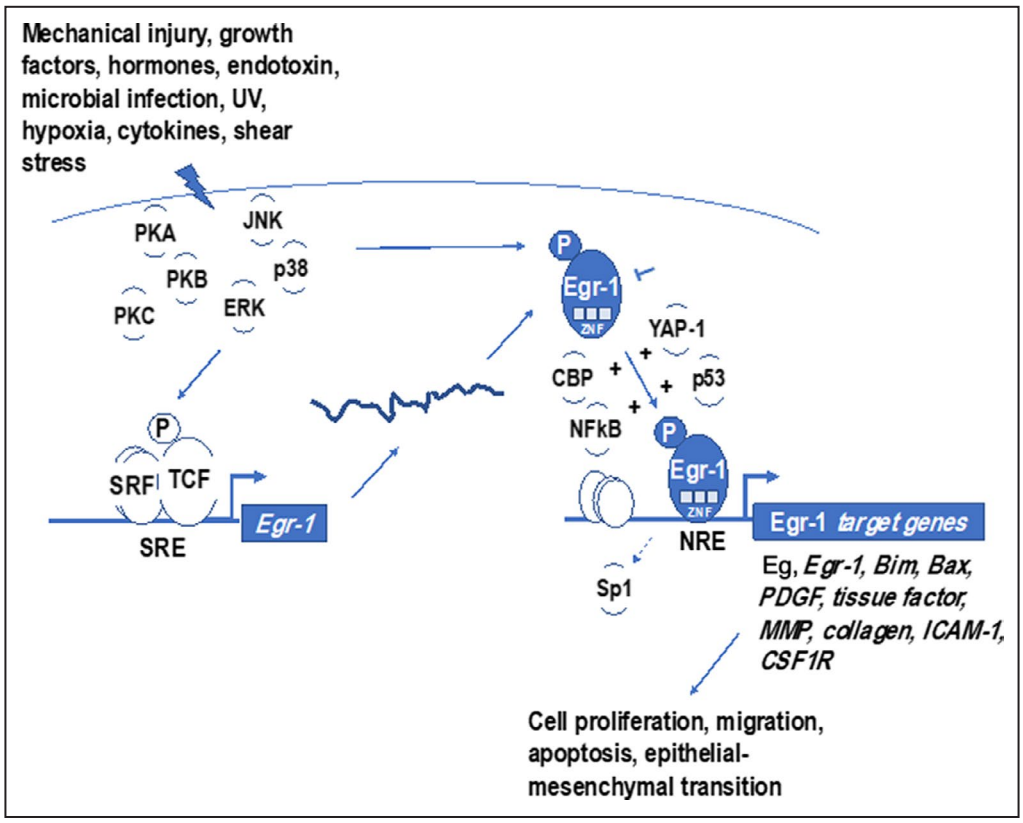

Figure. Simplified representation of induction of early growth response-1 (Egr-1) and Egr-1–dependent genes

Transcription of Egr-1, an immediate-early gene encoding a zinc finger (ZNF) protein, is activated by a range of extracellular stimuli through interactions of serum response factor (SRF) with ternary complex factors (TCF), which undergo phosphorylation, in its promoter region. Egr-1 protein itself undergoes phosphorylation and physical interactions with other transcriptional regulators including NGFI-A binding protein (NAB), CREB-binding protein (CBP), yes-associated protein 1 (YAP-1), p53 and NFκB. Egr-1 binds to recognition elements in the promoter/enhancer regions of target genes and can displace or compete with Sp1. Examples of Egr-1–dependent genes impacting cell phenotype are provided in the figure. ERK denotes extracellular signal-regulated kinase; JNK, c-Jun N-terminal protein kinase; PKA, protein kinase A; PKB, protein kinase B; PKC, protein kinase C; SRE, serum response element; and UV, ultraviolet light.

phosphoinositide 3-kinase/Akt pathway and phosphorylation of histone deacetylase 5. Blockade of phosphoinositide 3-kinase/Akt using wortmannin/SC66 or Akt siRNA inhibited angiotensin II-induced histone deacetylase 5 phosphorylation and its nuclear export. Moreover, depletion of histone deacetylase 5 or Egr-1 reduced SMC hypertrophy in response to angiotensin II.[23] Egr-1 expression activated by angiotensin II is negatively regulated by cAMP, suggesting a mechanism by which cAMP exerts vasculoprotective effects.

The angiotensin II/ERK/Egr-1 axis is also controlled by stromal interaction molecule-1/Orai-1 proteins, which regulate intracellular calcium flux. Simo-Cheyou et al[24] found that pharmacologic inhibition of the inositol-3-phosphate receptor lowers angiotensin II–induced levels of Egr-1 and intracellular calcium in SMC. Moreover, siRNA knockdown of either stromal interaction molecule-1 or Orai-1 reduced angiotensin II induction of Egr-1. Pharmacologic blockade of calmodulin and calmodulin kinase revealed that these effectors of calcium signaling also mediate angiotensin II induction of Egr-1. Egr-1 expression is regulated through Orai-1 channels that facilitate increased intracellular levels of Ca^{2+} that bind calmodulin, which interacts with calcium-calmodulin-dependent protein kinase II and mediates ERK1/2 and CREB activation.

Antenatal hypoxia, a common form of intrauterine stress, can adversely affect cardiovascular health later in life; however, the underlying mechanisms are incompletely understood. Xu et al[25] studied mechanisms of acquired vascular dysfunction in offspring caused by antenatal hypoxia and identified a role for Egr-1. Antenatal hypoxia upregulated angiotensin II type 1 receptor and hypomethylation-mediated activated transcription of Agtr1a (angiotensin II type 1 receptor-α subtype). Angiotensin II–angiotensin II type 1 receptor interaction increased protein kinase C (PKC)-ε

transcription by stimulating Egr-1 which was enriched at the PKC-ε promoter. Hypomethylation-dependent Agtr1a gene activation with downstream Egr-1 control of PKC-ε therefore may account for vascular hypercontractility in antenatal hypoxic offspring.[25]

DIABETES, KIDNEY FIBROSIS, AND INFLAMMATION

Tubulointerstitial fibrosis in diabetic kidney disease (DKD) can lead to chronic renal failure and involves epithelial-mesenchymal transition. Li et al[26] found that the antiaging factor Klotho can attenuate renal fibrosis in mice by inhibition of ERK1/2 signaling and suppression of Egr-1 activation. Overexpression of Klotho reduced Egr-1 expression whereas silencing Klotho increased Egr-1 levels, which was shut down with an ERK1/2 inhibitor. Hence, Klotho controls Egr-1 through ERK1/2 signaling, a novel mechanism in DKD progression.

Ho et al[27] found that in patients with renal failure Egr-1 expression was increased in renal tubular cells, and used Egr-1 deficient mice to demonstrate that Egr-1 promotes renal inflammation and fibrosis. Egr-1 expression increased in kidneys of mice fed an adenine-rich diet that induced tubulointerstitial nephritis. Mice lacking Egr-1 were protected from renal failure, with reduced cytokine/chemokine expression in kidney, tubular injury, immune cell infiltration and fibrosis. *Egr1*$^{-/-}$ mice with nephritis expressed less tumor necrosis factor-α (TNF-α), macrophage inflammatory protein-2 and monocyte chemoattractant protein-1 (MCP-1) than wild-type mice.[27] Since renal tissue was sourced from individuals with diabetes, it is unclear whether Egr-1 upregulation was attributable to diabetes or renal failure.

Lipoxins are lipid-based mediators that control resolution of inflammatory disorders such as DKD. Brennan et al[28] found that lipoxin A_4 ameliorates diabetes-induced albuminuria, mesangial expansion and collagen deposition in a mouse DKD model. Lipoxin given 10 weeks after disease onset suppressed preexisting DKD and expression of known inflammatory mediators including TNF-α, interleukin-1β (IL-1β) and NFκB. Lipoxin A_4 also reduced levels of Egr-1 in kidney tissue. In cultured human renal epithelial cells, TNF-α induction of Egr-1 was inhibited by lipoxin A_4 while Egr-1 siRNA attenuated VCAM-1 and TNF-α expression in response to TNF-α.[28] The restricted repertoire of genes expressed by these immortalized cells limited the broader interpretation of these experiments. The data nonetheless suggest that lipoxins control transcriptional networks involving Egr-1 and can potentially reverse DKD.

Peroxisome proliferator–activated receptors (PPARs) are ligand-activated transcription factors of the nuclear hormone receptor superfamily that exert anti-inflammatory effects among other impacts on cell metabolism, differentiation, development and immune response. Nemeth et al[29] showed that the PPARγ agonist pioglitazone inhibits transforming growth factor-β (TGF-β)-driven renal fibrosis and its progression, by modulating profibrotic Egr-1 in TGF-β overexpressing transgenic mice. Glomerulosclerosis and tubulointerstitial damage induced by TGF-β were reduced by pioglitazone, which attenuated Egr-1 expression along with signal transducer and activator of transcription 3 and activator protein 1, suggesting that PPARγ agonists may be useful for treatment of chronic kidney disease with effects mediated through Egr-1. Xie et al[30] found that PPARγ expression in pulmonary artery SMC is inhibited by leptin, which stimulates proliferation, and that this involves ERK1/2 phosphorylation and induction of Egr-1. Inhibition of ERK1/2-Egr-1 signaling with pharmacologic inhibitors or siRNA, or activation of PPARγ with pioglitazone, inhibited leptin-inducible cell growth. Furthermore, in a $CaCl_2$-induced model of abdominal aortic aneurysm comparing Egr-1–deficient bone marrow chimeric mice with controls reconstituted with wild-type bone marrow, no aneurysms were observed in mice lacking Egr-1.[31] Pioglitazone inhibited angiotensin II–inducible polycystic kidney disease 1 expression and Egr-1 occupancy of the polycystic kidney disease 1 promoter in cultured macrophages. This suggests the importance of both macrophages and Egr-1 in aneurysm formation and Egr-1 control by PPARγ.

Rajaraman et al[32] investigated the relationship between advanced glycation end products (AGE), Egr-1, and gestational diabetes and found that levels of Egr-1, tissue factor, and soluble ICAM-1 were elevated in umbilical vein endothelial cells isolated from mothers with gestational diabetes. Egr-1 siRNA reduced tissue factor and ICAM-1 expression, confirming Egr-1 reliance by these genes. AGE elevate Egr-1 expression by way of a RAGE–PKC-βII–dependent ERK1/2 pathway, findings demonstrated in part using the macrocyclic bisindolylmaleimide, ruboxistaurin. This implicates the AGE/PKC-βII/ERK/Egr-1 axis in the pathogenesis of gestational diabetes.

Atopic dermatitis is a common, chronic recurrent inflammatory skin disease currently with no cure. Yeo et al[33] studied AB1711, a novel small molecule targeting the DNA-binding domain of Egr-1 in the context of dermal inflammation. Topical application of 2,4-dinitrochlorobenzene to dorsal skin of mice resulted in severe atopic dermatitis, whereas mice lacking Egr-1 developed mild lesions that comprised myeloperoxidase-positive neutrophils, $CD3^+$ T cells, and $F4/80^+$ macrophages. AB1711 suppressed a range of TNF-α–inducible proinflammatory cytokine and chemokines including IL-1ß, interleukin-6 (IL-6), and chemokine (C-C motif) ligand

2 and ligand 5. In a separate group of mice in which 2,4-dinitrochlorobenzene was applied to ears, AB1711 lowered the number of scratching bouts stimulated by 2,4-dinitrochlorobenzene as effectively as dexamethazone. This study did not examine effects of AB1711 on angiogenesis nor investigate its pharmacodynamic profile. The data nonetheless indicate that Egr-1 is involved in the inflammatory microenvironment of skin and indicates the potential therapeutic clinical utility of AB1711 for other inflammatory disorders involving Egr-1.[33]

MYOCARDIAL ISCHEMIA-REPERFUSION INJURY

Ischemia-reperfusion injury after myocardial infarction has adverse cardiovascular outcomes, and effective pharmacotherapeutic interventions are sought. DNAzymes targeting Egr-1 reduce myocardial infarct size in a rat model of ischemia-reperfusion injury accompanied by lower ICAM-1 expression and neutrophil infiltration in myocardium. Rayner et al[34] showed that Egr-1 knockdown following intracoronary delivery of DNAzymes in pigs at the time of reperfusion following acute myocardial ischemia suppressed markers of inflammation (namely, ICAM-1, tissue factor, interleukin-8, complement 3 and complement 3 receptor) and apoptosis. This strategy improved cardiac function through salvaged left ventricular myocardium, ejection fraction, and fractional area change.

In a rat model of ischemia-reperfusion injury involving postconditioning, where multiple cycles of reperfusion/ischemia is applied at the onset of reperfusion, Wang et al[35] found that postconditioning or pretreatment with curcumin reduced Egr-1 expression in myocardial nuclei and microvessels, attenuated inflammation (plasma TNF-α and IL-6 levels) and reduced infarct size. Remote ischemic preconditioning (RIPC) can also reduce myocardial injury following a heart attack, and this again involves Egr-1 modulation. Billah et al[36] used RIPC in a rat model of cardiac ischemia-reperfusion injury involving multiple cycles of hind-limb ligation and release. RIPC reduced Egr-1 expression and myocardial infarct size by activation of the Janus kinase–signal transducer and activator of transcription pathway. The cardioprotective effects of RIPC were abolished by Egr-1 DNAzymes injected in the hind limb.

ACUTE LUNG INJURY, LUNG FIBROSIS, AND SEPSIS

Acute lung injury (ALI) is typified by uncontrolled inflammation and macrophage infiltration. Wang and colleagues[37] showed that the allosteric mitogen-activated protein kinase inhibitor trametinib prevents lipopolysaccharide (LPS)-induced ALI in mice by reducing edema and neutrophil infiltration. Trametinib suppressed production of Egr-1 and a range of proinflammatory mediators including IL-1β, MCP-1 and TNF-α, and Egr-1–dependent genes including ICAM-1, tissue factor, and prostaglandin E synthase. Chen et al[38] found that ALI induced by LPS is blocked by trametinib through its inhibition of the mitogen-activated protein kinase–ERK-Egr-1 pathway. These findings suggest the potential clinical use of trametinib for ALI.

Airway deposition of IgG immune complex is associated with ALI and sepsis. Yan et al[39] characterized an inverse relationship between PPARγ and Egr-1 expression. PPARγ was induced using its agonist rosiglitazone or adenoviral vectors, or suppressed with its antagonist GW9662 or shRNA. IgG immune complex stimulated Egr-1 mRNA levels in lung tissue by 13-fold, which was inhibited by airway administration of adenoviral PPARγ. IgG immune complex inducible TNF-α and MCP-1 expression was reduced by Egr-1 shRNA. Conversely, overexpression of Egr-1 increased TNF-α and MCP-1 promoter–dependent gene expression. PPARγ relieved the inflammatory response to IgG immune complex by reducing expression and transcriptional control by Egr-1 of a range of proinflammatory genes, including TNF-α and MCP-1. Treatment of mice with Egr-1 shRNA delivered by lentiviral vector reduced lung permeability and neutrophil recruitment to alveolar compartments. These findings suggest the PPARγ–Egr-1 axis is a targetable strategy for blockade of ALI or acute respiratory distress syndrome.[39]

Akhter et al[40] identified an unknown population of sphingosine-1-phosphate (S1P) receptor 1^+ endothelial cells during ALI critical for reestablishing the endothelial barrier. S1PR1 mediates ERK-dependent expression of Egr-1, which binds to and activates the sphingosine kinase 1 promoter causing S1P generation, vascular repair, and restoration of endothelial barrier function. Intravenous injection of $S1PR1^+$ endothelial cells into conditional endothelial cell–S1PR1 deficient mice with injured lung vasculature resulted in integration of these cells within the intima restoring endothelial integrity. However, mechanisms of homing by these cells remain unresolved.

Patients with pulmonary arterial hypertension (PAH) progressively develop neointimal lesions in small pulmonary arteries, causing elevated pulmonary vascular resistance and pressures, reducing cardiac output and heart failure. Egr-1 levels are elevated in human end-stage PAH. Dickinson et al[41] found a pivotal role for Egr-1 in a flow-associated PAH model in rats in which Egr-1 DNAzymes reduced pulmonary vascular Egr-1 expression and the development of occlusive neointimal lesions. Egr-1 knockdown attenuated pulmonary vascular resistance, right ventricular systolic pressure and hypertrophy, levels of TGF-β, PDGF-B, IL-6, and

p53, and increased apoptosis. Pioglitazone also inhibited neointimal lesion formation.[41] Van der Feen et al[42] examined Egr-1 expression in clinical pulmonary specimens with different forms and stages of PAH and found that Egr-1 expression is increased in PAH specifically with neointimal-type vascular remodeling. Similarly, in a murine model of pulmonary inflammation and fibrosis, Li et al[43] found that SARS coronavirus papain-like protease can stimulate macrophage infiltration and expression of Egr-1 and induction of TGF-β, thrombospondin-1 and profibrotic genes.

Sepsis remains the major cause of mortality in hospital intensive care units and is associated with endothelial permeability and multiorgan dysfunction. Chen et al[44] showed that Egr-1 and ADAM10 levels positively correlate with severe sepsis in patients, with rs653765 GG carriers having higher Egr-1 mRNA levels than those with the AA or GA genotype. Egr-1 binds to and activates the ADAM10 promoter in an allele-dependent manner. Studies in a mouse endotoxemia (LPS) model showed that antisense Egr-1 oligonucleotides reduced host ADAM10 expression, proinflammatory cytokine secretion (TNF-α, IL-6 and IL-1β), adhesion molecule expression (ICAM-1 and VCAM-1), and improved survival.[44] Future studies should further delineate the therapeutic potential of targeting the Egr-1/ADAM10 pathway in sepsis and the prognostic value of rs653765 polymorphisms.

TRANSPLANT REJECTION

Egr-1 may also play a role in heart transplant rejection. Pivotal studies by Okada et al[45] demonstrated that Egr-1, together with PDGF-A, ICAM-1 and VCAM-1 are strongly expressed in cardiac allografts after transplantation, while Autieri et al[46] showed that Egr-1 expression may serve as a surrogate marker of cardiac allograft rejection, demonstrating a relationship between Egr-1 expression and rejection grade in endomyocardial biopsies.

The success of allogeneic organ transplantation is limited by T-cell–mediated alloimmune reactions resulting in graft-versus-host disease. Peltier et al[47] recently reported that the long noncoding RNA Linc00402 is a regulator of T-cell alloimmunity following hematopoietic stem cell transplantation. Overexpression of Linc00402 in Jurkat cells increased ERK1/2 phosphorylation and stimulated expression of Egr-1, which can enhance T-cell activation and interleukin-2 production. Linc00402 increased in donor T cells from patients undergoing allogeneic heart transplantation and was reduced in those who developed acute graft-versus-host disease.[47] Future studies should explore the therapeutic use of Linc00402 in allograft rejection or acute graft-versus-host disease and its effects on Egr-1.

Ischemia-reperfusion injury is an important cause of morbidity and mortality after lung transplantation. In an orthotopic vascularized lung transplantation mouse model, Yamamoto et al[48] found that Egr-1 controls this process by regulating neutrophil infiltration. Immunohistochemistry revealed normal lungs showing no staining of Egr-1 whereas transplanted lungs expressed Egr-1 within an hour of reperfusion in pulmonary artery endothelial cells and neutrophils. Egr-1 deficiency improved pulmonary graft function and reduced neutrophil infiltration. Egr-1 may therefore be a useful target for lowering lung ischemia-reperfusion injury. However, the specific Egr-1-dependent factors controlling neutrophil infiltration remain to be elucidated.

ANGIOGENESIS AND RELATED PROCESSES

Egr-1 stimulates angiogenesis in a range of settings including wound healing, corneal neovascularization, and tumor angiogenesis.[49] Adenoviral delivery of a form of Egr-1 unable to be suppressed by NAB can increase expression of FGF-2, PDGF-A, PDGF-B, insulin-like growth factor II, and TGF-ß and tissue perfusion in a mouse model of hindlimb ischemia.[50] Even though Egr-1 was described as a phosphoprotein when initially characterized 3 decades ago, actual residue(s) phosphorylated in Egr-1 remained elusive until we identified Ser26, which is highly conserved in Egr-1 and unique to Egr-1 in the Egr family. Ser26 is phosphorylated by ERK.[51] To establish the functional significance of Ser26 we devised a CRISPR/Cas9 strategy to introduce a homozygous Ser26>Ala mutation into Egr-1 in human vascular endothelial cells.[52] Cells bearing this mutation proliferated and migrated slower than those with wild-type Egr-1. Mutant cells formed fewer networks on solubilized basement membrane preparations and increased expression of vascular endothelial cadherin, which controls endothelial adhesion, permeability, and angiogenesis, and underwent apoptosis. The phenotypic effects of this mutation were comparable to deletion of Egr-1 itself, thereby underlining the importance of Ser26 to the function of Egr-1.

Egr-1 also regulates endothelial progenitor cells, which differentiate into mature endothelial cells and regulate neovascularization in conditions such as myocardial ischemia. Ye and colleagues[53] showed that the androgen dihydrotestosterone can promote the migration and proangiogenic properties of endothelial progenitor cells. Egr-1 knockdown inhibited endothelial progenitor cell migration stimulated by dihydrotestosterone, suggesting that androgen regulates endothelial progenitor cell function through Egr-1 signaling.[53]

CONCLUSIONS

As an immediate-early gene that responds rapidly and transiently to a wide range of extracellular stimuli, Egr-1 integrates changes in the local microenvironment with programs of inducible gene expression. While it is known that Egr-1 undergoes posttranslational modification and interacts with a range of transcription factors, our understanding of mechanisms with which Egr-1 regulates transcription and drives the pathogenesis of disease is far from complete. Future studies should provide a more detailed dissection of the mechanistic roles played by Egr-1 in vascular dysfunction and inflammatory disease and develop new strategies exploiting this knowledge.

ARTICLE INFORMATION

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Disclosures

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