혈관염증의 분자생물학적특성 및 제어기술

전병화

Biology of vascular inflammation and therapeutic application

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1. Inflammation and reactive oxygen species

In the past, inflammation has been associated with infections and with the immune system. But more recent evidence suggests that a much broader range of diseases have telltale markers for inflammation. Inflammation is the basic mechanism available for repair of tissue after an injury and consists of a cascade of cellular and microvascular reactions that serve to remove damaged and generate new tissue.

·Reactive oxygen species (ROS) contribute to the pathogenesis of cardiovascular diseases including hypertension, atherosclerosis, cardiac hypertrophy, heart failure and diabetes mellitus. Oxidative stress is resulted from excessive generation of ROS that outstrips the antioxidant system. Various agonists, pathological conditions and therapeutic interventions lead to modulated expression and function of oxidant and antioxidant enzymes, including NAD(P)H oxidase, endothelial nitric oxide synthase, xanthine oxidase, myeloperoxidase, superoxide dismutases, catalase, and glutathione peroxidase. ROS formed in vascular wall target a wide range of signaling molecules and cellular pathways in both endothelium and vascular smooth muscle, such as transcription factors, protein tyrosine phosphatase, protein tyrosine kinase, mitogen-activated protein kinase, Ca(2+) transporting system and protein modification. ROS also have distinct physiological and pathophysiological impacts on vascular cells. ROS contribute to vascular dysfunction and remodeling through oxidative damage by reducing the bioavailability of NO, impairing endothelium-dependent vasodilatation and endothelial cell growth, causing apoptosis, stimulating endothelial cell migration, and activating adhesion molecules and inflammatory reaction, leading to endothelial dysfunction, an initial episode progressing toward hypertension and atherosclerosis.

2. Inflammation and atherosclerosis

Atherosclerosis has traditionally been viewed to simply reflect the deposition of lipids within the vessel wall of medium-sized and large arteries. This concept has changed. It is now assumed that a complex endothelial dysfunction induced by elevated and modified low-density lipoproteins (LDL), free radicals, infectious microorganisms, shear stress, hypertension, toxins after smoking or combinations of these and other factors leads to a compensatory inflammatory response (1).

The development of an atherosclerotic lesion requires a complex interplay between mononuclear cells, endothelia, vascular smooth muscle cells, growth factors, and cytokines. Monocyte rolling and adhesionto the vascular endothelial lining and subsequent diapedesis are not only the first steps, but also seem to be crucial events in the pathological process (2). Expression of cell adhesion molecules, such as vascular cell adhesion molecule-1 (VCAM-1), on endothelial cells represents one of earliest pathological changes in inflammatory diseases, such as atherosclerosis (1). As the

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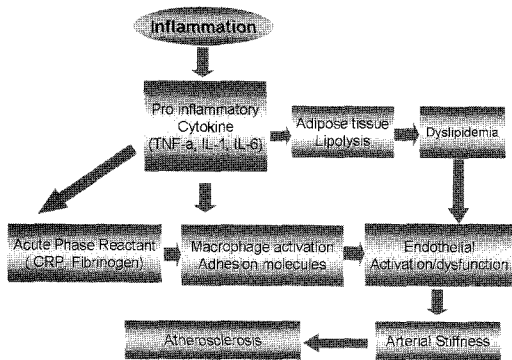
Fig. 1. Schematic concepts of inflammation in the pathogenesis of atherosclerosis

phenotype of endothelial cells is subject to change by oxidative stress, reactive oxygen species (ROS), such as superoxide ions, are implicated in the pathogenesis of cardiovascular disorders (3).

Endothelial dysfunction is characterized by decreased nitric oxide synthesis, local oxidation of circulating lipoproteins and their entry into the vessel wall (4). Intracellular reactive oxygen species similarly induced by the multiple atherosclerosis risk factors lead to enhanced oxidative stress in vascular cells and further activate intracellular signaling molecules involved in gene expression (5) Upregulation of cell adhesion molecules facilitates adherence of leukocytes to the dysfunctional endothelium and their subsequent transmigration into the vessel wall.

3. Therapeutic implication

Although our knowledge on the mechanisms of atherosclerosis and plaque destabilization is still incomplete and largely hypothetical, currently available data provide a conceptual framework that can be used for the design of further pathophysiologically oriented investigations and novel treatment strategies.

Statins

Statins are mainly used in patients as 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) - reductase inhibitors. In patients with coronary artery disease, statins was attributed to the decrease in cholesterol levels. Another mechanism could be reduction of TF and MMP expression. less macrophage and T-cell inflammation, reduced MMP-2 immunoreactivity, increased expression of MMP tissue inhibitor (TIMP), and a higher collagen content. Statins, additionally, exert profound downregulatory effects on systemic markers of inflammation in patients with atherosclerosis such as CRP and serum amyloid A levels.

COX Inhibition

COX exists in 2 isoforms, COX-1 and COX-2. Acetylsalicylic acid (ASS) inhibits both COX isoenzymes. The direct antithrombotic effect of ASS is mediated by inhibition of COX-1 in platelets resulting in a decreased production of thromboxane A2. COX-2 is induced at sites of inflammation and is expressed in human atherosclerotic lesions including carotid artery plaques in conjunction with COX-2 synthase. ASS decreases the adherence of monocytes and T cells to human coronary artery endothelial cells, inhibits TNF-induced nuclear factor-B mobilization, an important transcription factor regulating immune responses, and blocks the expression of immunological cell adhesion molecules.

Inhibition of the Renin-Angiotensin System

In animal models of atherosclerosis, angiotensin-converting enzyme inhibitors exerted consistent beneficial effects on plaque progression. One identified mechanism was the reduction in MCP-1 expression and concomitant macrophage plaque infiltration.

Suppression of Cytokines

In experimental settings blockade of IFN-r and TNF ameliorated the development of atherosclerosis. Pentoxifylline, a TNF-antagonist, significantly decreased plaque formation in apoE -/- mice by shifting T cells toward T-helper-2 differentiation

characterized by increased production of the immunosuppressive cytokine IL-10.

MMP Inhibition

MMP (eg, MMP-1 and MMP-9) contribute to atherosclerosis and destabilization of internal carotid artery plaques. Thus, MMP represent a potential target for therapeutic interventions by restoring the physiological balance between MMP and their tissue inhibitors, TIMP. Doxycycline, a member of the tetracycline family, exhibits a nonselective inhibitory effect on MMP. As mentioned above, statin treatment also profoundly decreased MMP expression in carotid plaques. Similarly, percutaneous delivery of TF pathway inhibitor blocking the extrinsic coagulation cascade concomitantly reduced the neointimal expression of MMP-2 and MMP-9 activity in a rabbit femoral artery model of vascular injury.

4. Therapeutic strategies using redox factor-1

Among the endogenous mechanisms that repair oxidative DNA damage is the ubiquitously expressed apurinic/apyrimidinic endonuclease1/redox factor-1 (APE1/ref-1)(6). APE1/ref-1 is a protein with dual function. It is an essential endonuclease in the base excision repair pathway of oxidatively damaged DNA, as well as having reducing properties that promote the binding of redox-sensitive transcription factors such as activator protein-1 to their cognate DNA sequences. In addition to its nuclear functions, an extra-nuclear role for APE1/ref-1 in the regulation of endothelial oxidative stress has been uncovered. APE1/ref-1 suppresses oxidative stress through modulation of cytoplasmic rac1-regulated ROS generation(7). This action of APE1/ref-1 and its reported effect on endothelial NO levels, suggests a possible protective role for APE1/ref-1 in inflammatory vascular disorders. Recently, it was reported that overexpression of APE1/ref-1 inhibits monocyte adhesion and vascular cell adhesion molecule-1 expression via activation of nitric oxide and inhibiting of ROS generation(8,9). Also MMP-2, MMP-9, and COX-2 induced by lipopolysaccaride was markedly reduced by the overexpression of APE1/ref-1 in the macrophage systems. These kinds of exprimental results in the cellular system yielded promising results. Their therapeutic potential in atherosclerosis, however, awaits further investigation.

5. Application of TAT-fusion protein

Currently, efficient delivery of therapeutic compounds, peptidyl mimetics, and proteins into cells in vivo can be achieved only when the molecules are small- typically less than 600 daltons. Full-length fusion proteins are generated that contain an NH2-terminal 11-amino acid protein transduction domain (PTD) from the human immunodeficiency virus (HIV) TAT protein [first identified in 1988 (10). These proteins are then purified under denaturing conditions. Protein transduction occurs in a rapid, concentration-dependent fashion that appears to be independent of receptors and transporters and instead is thought to target the lipid bilayer component of the cell membrane. Thus, in principle, all mammalian cell types should be susceptible to protein transduction, and indeed we have used this technology to transduce over 50 proteins ranging in size from 15 to 120 kD into a wide variety of human and murine cell types in vitro. Also, intraperitoneal injection of the 120-kilodalton beta-galactosidase protein, fused to the protein transduction domain from the human immunodeficiency virus TAT protein, results in delivery of the biologically active fusion protein to all tissues in mice, including the brain (11).

6. Summary

Inflammation plays an important role in the progression of atherosclerosis and plaque

destabilization converting a chronic process into an acute disorder with ensuing thromboembolism. Current therapeutic effective in preventing atherosclerosis and stroke such as statins, ASS and RAS inhibitors my exert part of their effects by modulating inflammatory responses in the vessel walls. As alternative approaches, discovery to find having inhibitory action of MMP activity, COX-2, macrophage infiltration, such as APE1/ref-1 and fusion technology for cell permeable protein may provide a new antiatherosclerotic therapy in the future.

Acknowledgements

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