Journal of Advanced Biomedical and Pharmaceutical Sciences

Journal Homepage: http://jabps.journals.ekb.eg

Interleukin-27 and Risk of Coronary Artery Disease

Mohamed Abu Khesha Kamel[1], Ahmed Ragaa Nour Ibrahim[2*], Gamal El-Din Abu Rahma[3]

[1] Molecular Biology Center, Faculty of Pharmacy, Minia University, 61519 Minia, Egypt
[2] Department of Biochemistry, Faculty of Pharmacy, Minia University, 61519 Minia, Egypt
[3] Department of Medicinal chemistry, Faculty of Pharmacy, Minia University, 61519 Minia, Egypt

Received: September 3, 2018; revised: October 10, 2018; accepted: November 5, 2018

Abstract

Cardiovascular diseases (CVDs) are the most abundant causative factors of mortality and morbidity in the world specifically in the developing countries. Statistics show that approximately 17.3 million people die every year due to CVDs, equivalent to 30% of all deaths all over the world. Developing countries recently share with more than 80% of all CVD deaths. Atherosclerosis is the main cause of CVDs that include myocardial infarction, heart failure, stroke and peripheral artery disease. Many studies on humans and animals prove that inflammation plays a key role in the initiation and propagation of the process of atherosclerosis. Coronary artery disease (CAD) as a type of atherosclerotic diseases, are recognized to be chronic inflammatory diseases. Interleukin-27 (IL-27) is a new IL-12/IL-6 family member which is a heterodimeric multifunctional pro-and anti-inflammatory cytokine. IL-27 constituted by an IL-27p28 subunit linked to Epstein-Barr virus (EBV)-induced gene3 product (EBI3) subunit. IL-27 one of the important cytokines distinguished in atherosclerotic plaques. Unlike other members of cytokine family IL-27 has dual function, one as an initiator and the other as an attenuator of immune/inflammatory responses. Many studies ascertain the concept that elevated IL-27 may be one of the main cytokines that constitute a regulatory network in immunity and inflammation that affect atherosclerosis. In this review we focus on the coronary artery atherosclerosis its etiology, risk factors and consequences such as myocardial infarction and sudden cardiac death. The pathogenesis of atherosclerosis that include: lipoproteins accumulation, recruitment of immune cells and development of foam cells. Define atherosclerosis as an inflammatory process and the contribution of inflammatory mediators such as TNF-α, IL-6 and IFN-γ in its initiation and progression.IL-27 its construction, production,IL-27 receptors and signaling pathway, the pro and anti-inflammatory characters of IL-27 via T helper1 (Th1), T helper2 (Th2) and IL-10.Finally the effect of IL-27 as an inflammatory regulator in the process of atherogenesis.

Key words
Interleukin-27, Coronary artery disease, Cardiovascular diseases, inflammation, Interleukin.

1. Introduction

Cardiovascular diseases (CVDs) are classified as foremost causative factors of mortality in the world specifically in the developing countries. Coronary artery disease (CAD), precisely atherosclerotic CAD is the major CVD primarily triggered by atherosclerosis. CAD is a multifactorial disease that several environmental and genetic risk factors contribute to its pathogenesis [1]. Inflammation contributes significantly in the initiation and progression of atherosclerosis [2]. Numerous studies suggest that atherosclerosis may be considered as an immune-mediated chronic inflammatory disease [3]. Studies on humans and animals prove vital role of macrophage and T cell infiltrates in the atherosclerotic lesions [4]. Interleukin 27 (IL-27) is a new and important IL-12 family member recovered primarily from Epstein-Barr virus (EBV)-induced transformed B cell lines, which is a heterodimeric multifunctional pro- and anti-inflammatory cytokine. IL-27 is constituted by an IL-27p28 linked to EBV-induced gene3 product (EBI3) subunit [5] (**Figure 1**). The human *IL-27* gene is positioned on chromosome 16p11 and consisted of five exons. This gene is vital for the growth and differentiation of T cells [6]. IL-27 is an immune/inflammatory response regulator with anti-inflammatory character that modulates T cell polarization and production of cytokines by indorsing early T helper 1 (Th1) differentiation and repressing both Th2 and Th17 differentiation [7, 8]. Expression of IL-27 has been also noted in atherosclerotic plaques [9]. During the initiation and progression of atherosclerosis, IL-27 imparts its effect on macrophage as well as T cell functions. As such, the expressed IL-27 in the arterial wall appears not only to regulate macrophage activities, such as phagocytosis and other consecutive cytokine production, but also have crucial impact on other immune and non-immune cells types through controlling the production of cytokines [10].

In this review, the pathogenesis of atherosclerosis and the effect of IL-27 as an inflammatory mediator in the process of atherogenesis will be discussed.

2. Coronary artery atherosclerosis

Atherosclerosis can be defined as progressive disorder characterized by low-grade inflammation of the of medium-sized arterial intima (inward lining of the artery) that is augmented by risk factors such as diabetes, hypertension (HTN), dyslipidemia, smoking, and genetic factors. Considering coronary atherosclerosis, this slow progression result in gradual

**** Correspondence: Ahmed Ragaa Nour Ibrahim***
Tel.: *+2 01091227274****; Fax:*** *+20 862369075*
Email Address: *a.ibrahim@mu.edu.eg*

increase in the stiffening of the inner layer of the coronary arteries that on the long run causes narrowing of the arterial lumen to different degrees. Atherosclerosis is the leading cause of coronary syndrome of acute myocardial infarction(AMI) and sudden cardiac death(SCD) [11]. AMI is described as acute disturbance between myocardial blood supply and myocardial oxygen demand leading to myocardial necrosis based on clinical evidences that consistent with the diagnosis. These evidences comprise the presenting features of the patient, the finding of the electrocardiograph (ECG), and/or other indication signifying a recent wall motion abnormality in the segments of the myocardium that assessed by the echocardiography [12]. SCD explains the unpredicted natural death that is triggered by a cardiac cause over a period of very short time, mostly not exceeds one hour from starting the onset of complain, in a person that is free from any previous condition that seems to be fatal. SCD is typically subsequent to a catastrophic cardiac arrhythmia like ventricular fibrillation (VF), which is a common consequence of the acute coronary artery thrombosis that reduces cardiac arrhythmia threshold [13].

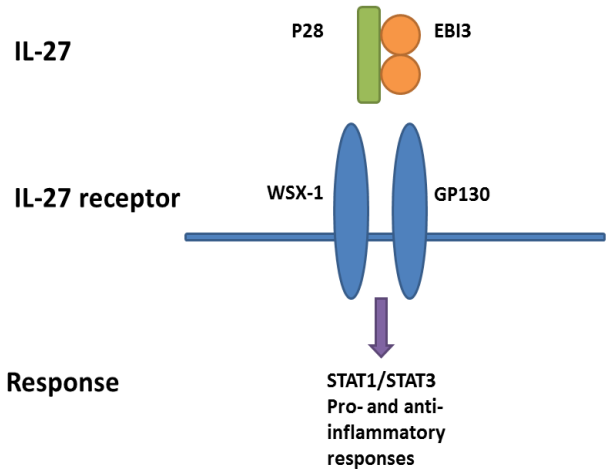

Figure 1: IL-27 cytokine; The IL-27 cytokine has pleiotropic function and plays critical roles in inflammatory response. This cytokine belongs to a family that has very unique characteristics, because they are composed of two different subunits forming a heterodimer. The binding of IL-27 domains to the extracellular domains of IL-27 receptor leads to the activation of STAT1 and STAT3.

3. Pathogenesis of atherosclerosis

3.1. Lipoproteins Accumulation

Buildup of low-density lipoprotein (LDL) in the intima of the arteries is the key step for establishment of atherosclerosis. The retaining of the invading LDL particles on subendothelial proteoglycans is the starting incident in the initiation of atherosclerosis [14]. Proteases and lipases modify the entrapped lipoproteins particles triggering their aggregation thus increasing binding of proteoglycan [15]. Myeloperoxidase, lipoxygenase, and reactive oxygen species induce oxidative modifications which subsequently result in the formation of oxidized form of LDL (ox LDL), which can provoke an innate inflammatory response [16].

3.2. Recruitment of immune cells

Endothelial cells respond to the cumulative effects of the entrapped and modified lipoproteins in the subendothelial space by expressing adhesion protein, like vascular cell adhesion molecule-1 (VCAM-1) [17]. Leukocytes are then recruited to the affected sites. Granulocyte–macrophage colony stimulating factor and macrophage colony-stimulating factor both play crucial role in the differentiation of the invading monocytes into macrophages. Both factors are formed by the endothelium and other numerous cell types. The advanced imaging techniques allowed following the leukocyte migration on the surface of the endothelium in atherosclerotic plaques in live mice [18]. In atherosclerotic plaques, macrophages derived from monocyte remain the foremost cell population due to their continuous local enrollment, differentiation, and proliferation [19].

3.3. Development of Foam cells

Macrophages are transformed in to foam cells as a consequence of the unceasing engulfment of lipoprotein particles [20]. Scavenger receptors are expressed by macrophages, among these expressed receptors are CD36 and class A scavenger receptors that have significant role in modified LDL uptake [21]. Unlike LDL receptors, scavenger receptors are not downregulated responding to intracellular cholesterol. The other pathway for lipid uptake by macrophages is through pinocytosis [22]. The entrapped foam cells inside the arterial intima compromise the blood flow and migratory capacity of the artery. Eventually these cells die then generate a core in the plaque area composed of necrotic, apoptotic cells, cholesterol crystals and other extracellular material [23].

4. Inflammation and Atherosclerosis

In the middle of the 19th century, Virchow and von Rokitansky established for the first time the contribution of inflammation to the process of atherosclerosis and reported that atherosclerotic vessels have evidence of cellular inflammation. Subsequently, this concept was extended by Ross and Glomset, who prove that infiltration of the subendothelial layer of the arterial wall by monocyte and macrophage was an initial step of atherosclerosis [24]. Several soluble mediators of the inflammatory response have been suggested to predict cardiovascular risk in patients with atherosclerosis [25]. Following the atherosclerotic progression allows establishment of various evidences that suggest atherosclerosis results from micro inflammation induced by pro-inflammatory cytokines. The evidence that detection of T lymphocytes and monocytes were at all stages of atherosclerotic plaque progression, is consistent with active inflammation in animal models. In addition, marked increase in circulating blood levels of the acute phase reactant like C-reactive protein (CRP) is suggested to be a marker of inflammatory processes and may be of value in the prediction of acute coronary artery events [26].The LDL-induced inflammatory response of endothelial cells and LDL accumulation in the intima are believed to be involved in the initiation of atherosclerosis [27]. Previous studies prove that circulating levels of IL-27 were markedly overstated in patients with CAD, and significantly correlated to ox-LDLox-LDL dose-

dependently up regulated expression of both IL-27 protein and IL-27 (p28 and EBI3) mRNA in vitro, specifying that ox-LDL capable of endorsing production of IL-27 by dendritic cells (DCs). These findings suggested that IL-27 can modulate the network of immunity and inflammation in the pathogenesis of atherosclerosis [28]. Previous studies identified variety of pro-inflammatory cytokines, such as tumor necrosis factor-α (TNF-α), IL-17, IL-6, IL-12, and interferon-γ (IFN-γ), as vital key regulators of atherosclerosis development and progression [29]. The recent literatures buildup a growing body of evidence that support the relationship between various inflammatory biomarkers and potential cardiovascular risk, in healthy individuals as well as in patients with coronary heart disease or heart failure [30]. Activated T cells are differentiated into Th1 effector cells and start to produce the macrophage-activating cytokine INF-γ, which improves the efficiency of antigen presentation and potentiate the synthesis of inflammatory cytokines such as TNF-α and IL-1 [31]. These cytokines work synergistically to promote the production of many inflammatory and cytotoxic molecules in macrophages and vascular cells [27]. The process of inflammation in atherosclerosis may lead to elevated blood levels of inflammatory cytokines and other acute-phase reactants such as level of CRP and IL-6,which are markedly elevated in patients presented with unstable angina and acute myocardial infarction, as well as elevated level indicating bad prognosis [32]. Other inflammatory markers are also elevated in those patients, including fibrinogen, IL-7, IL-8, and the CRP-related protein pentraxin-3Markers of myocardial damage and inflammation in relation to long-term mortality in unstable coronary artery disease [33]. The elevated levels of CRP in patients with unstable angina, a condition that is essentially relies on coronary thrombosis of atherosclerotic plaques, can be used as a method for differentiation from those with vasospastic angina. Consequently, elevated level of CRP in patients with acute coronary syndromes expectedly reflects inflammation in the coronary artery rather than other causes of ischemic myocardium [34]. Endothelial cells express adhesion molecules and secrete several chemokines that lead to the recall of inflammatory cells that participate in the sub-endothelial inflammation. Accumulating monocytes are differentiated into plaque macrophages that take up accumulated LDL-cholesterol forming foam cells [35], these foam cells by the effect of inflammatory cytokines acquire typically activated pro-inflammatory (M1) phenotype that subsequently promote inflammation. Consequently, the formation of primary atherosclerotic plaques is initiated [36]. Once plaque inflammation initiated, there are two consequences, the first is the direction of downregulation in the initial inflammatory reaction with healing and fibrotic processes that resulting in plaque stabilization by the formation of a thick, stable fibrous cap. The second is for vulnerable plaque followed by life threatening consequences, such as stroke and myocardial infarction. In the later, chronic inflammation is accelerated, the plaque becomes unstable, and the fibrous cap becomes thin all of them result in plaque rupture that is complicated by rapid lumen closure with thrombus burden and subsequently an acute clinical event. Such highly unstable plaque is characterized by active chronic inflammatory reactions that include constant infiltration of inflammatory cells and the production of inflammatory cytokines [37].

5. IL-27

IL-27 is a heterodimeric cytokine and due to its structural characteristics and common receptors IL-27 related to the superfamily IL-6/IL- 12 cytokines. IL-27 constructed of two functional subunits p28 (IL-27a) and EBI3 (Epstein-Barr virus-induced gene 3) (IL-27b) [6]. like most of the IL-6-family members, the alpha-subunit p28 of IL-27 is a four-helical bundle, while EBI3 is the soluble form of alpha receptor that composed of two fibronectin-like domains [38]. Antigen-presenting cells are mainly responsible for production of IL-27p28 and EBI3 consequent to stimulation by inflammatory mediators or microbial products [6]. Myeloid lineage cells primarily monocytes and activated dendritic cell are the main source of p28 and EBI3 [6]. The signal of IL-27 is conducted through receptor complex that is constituted by Glycoprotien130 (gp130) and WSX-1 forming a heterodimer on the surface of the cell that mostly stimulates the Janus kinases (Jak)/signal transducer and activator of transcription (STAT) (**Figure 1**). Unlike the pro-inflammatory properties of IL-6 via gp130 receptor, IL-27 was primarily stated as immunoregulatory mediator. This was mainly depending on the concept that IL-27 stimulates IL-10 secretion as well as confines inflammatory responses in the area of infection [39].

6. IL-27 receptors

IL-27 exerts its action through binding to heterodimeric membrane-bound receptor complex consists mainly of the two distinct subunits the first is gp130, which has predominant expression and considered the leading signal transducing beta-receptor utilized by most IL-6 and IL-12 family members, the second is IL-27R alpha (WSX-1) also called (T cell cytokine receptor, (TCCR)) which imparts ligand specificity and has limited recognized functions other than IL-27 signaling. IL-27ra (WSX-1) is a type I trans-membrane protein receptor which stimulates the Jak/STAT signaling pathway once receptor complex is formed upon ligand binding. Naïve CD4+ T cells show low levels of expression of IL-27ra (WSX-1) while higher levels of expression are noticed on effector and memory T cells that make them the central targets for IL-27 action [40]. IL-27 can bind to IL- 27ra (WSX-1) even if gp130 is absent [41]. Nevertheless, for signal transduction to occur the co-expression of both subunits of the receptor is essential [42]. Not only T cells can expressIL-27 receptor complex but also innate immune cells, like macrophages, endothelial cells and dendritic cells share its expression, which demonstrating that several types of cells can respond to IL-27 signaling [43]. Because of soluble gp130 has not competence to inhibit IL-27 signaling, IL-27 cannot solely bind to gp130 receptor [44]. gp130 expression analysis is not reliably informative as indicator for the signaling pattern of IL-27 because the gp130 chain is widely expressed.

Consequently, IL-27 sensitivity is frequently evaluated by studying expression level of IL-27ra (WSX-1), which is restricted largely to the immune system even if the expression levels in other tissues is low [45]. gp130 mainly stimulates STAT3, while WSX-1 stimulates STAT1, that consequently lead to a diversity of signaling actions, like T-bet–induced induction of IL-12 receptor β chain (*Il12rb2*), in addition to induction of pro-inflammatory IFN-γ and anti-inflammatory IL-10 [43] (**Figure 1**).

7. IL-27 as pro- and anti-inflammatory cytokine

Primarily IL-27 was discovered and specified as a type of cytokine with pro-inflammatory character that induces T helper 1 (Th1) differentiation primary during immune responses [6]. T helper cells are the uppermost antigen recognizing and classified functionally into two distinct subtypes identified as Th1 and Th2. Th1 responds by the amplification of pro-inflammatory pathways through secretion of cytokines such as IFN-γ, so the Th1 response mostly to exacerbate atherosclerosis. Recent studies recognized other T cell subset Th17 cells, also may have predominant pro-inflammatory activities [46]. While IL-27 stimulates Th1 transcription factors (T-bet) as well as (STAT-1) in addition to the up-regulation of the expression of the IL-12Rβ2 chain, EBI3 (*Ebi3*$^{-/-}$) or IL-27Rβ (*Il27ra*$^{-/-}$) deficient mice did not exhibit marked deficiency in the capability to mount Th1 responses, even Th1 responses are delayed in a limited number of infection [46].Subsequent studies revealed that binding of IL-27 to IL-27 receptor and complex formation led to the phosphorylation of signal transducer and activator of transcription (STAT-1) in naïve CD4+ T cells, triggering them to proliferate and become polarized Th1 cells [43]. Consequently, complete response of Th1 is essential to regulate intracellular infection by protozoa. IL-27 signaling deficient mice experienced with protozoal infection were predicted to have a diminished Th1 response. Unexpectedly, Mice deficient with one subunit of the receptor IL-27Ra (WSX-1 –/–) encountered with parasitic protozoa had intact Th1 responses, developed excessive pathological inflammation yet capitulated to CD4+ T cell-mediated immune-pathology, signifying the essential role of IL-27 to control *in vivo* inflammation [39]. Furthermore, Successive studies on several infectious and autoimmune disease models established the anti-inflammatory role for IL-27 in Th1, Th2, and Th17 responses [43], regarding the anti-inflammatory effects of IL-27, current efforts have revealed that IL-27 can trigger the production of the anti-inflammatory cytokine IL-10 by T cells [47]. IL-27 represses the immunologic pro-inflammatory responses of IL-17 producing Th17 cells in various experimental animal models, such as animal model of autoimmune encephalomyelitis (EAE), neurological disease characterized by demyelination and autoimmune inflammation of the central nervous system and human multiple sclerosis model [48]. Several studies find significant relation between the elevated serum levels of IL-27 and other interleukins such as IL-10, IL-17A, IL-6 and TNF-α in inflammatory disease such as asthma, rheumatoid arthritis and Kawasaki disease (KD) providing further evidence about the inflammatory role of IL-27 in KD, These explanations specify that IL-27 play an essential role in the immunity and inflammation response [49]. Therefore, while IL-27 was initially categorized as a pro-inflammatory cytokine, successive studies have concentrated on its anti-inflammatory effects.

8. IL-27, Atherosclerosis and coronary artery disease

IL-27 is a crucial member of cytokines in the inflammatory diseases, specifically CAD [1]. Numerous studies advocate the influential function of Interleukin-27 in the development and progression of atherosclerosis [50]. Atherosclerosis is considered as chronic multifactorial vascular disease diffused by inflammatory mediators, like cytokines, chemokines and other mediators [51]. Inflammatory cytokines produced by immune and vascular cells with pro- and anti-inflammatory properties contribute significantly in the initiation and progression of atherosclerosis via controlling cellular signaling in arterial wall via paracrine and autocrine manner [52]. Previous studies presented numerous pro-inflammatory cytokines, such as TNF-α, IL-6, IL-12, IL-17 and IFN-γ, as key regulators in the developing and progression of atherosclerosis [53]. Nonetheless, studying the anti-inflammatory activity of these mediators as negative inflammatory regulators is limited. Recently, studies have recognized that differentiation of macrophages produces several subsets of cells including M1 macrophage phenotype, subsets stimulated by IFN-mediated Th1 type response and M2 macrophage phenotype, subsets stimulated by Th2 response [54]. Each subset signifies discriminated impact as inflammatory regulators in processes of inflammation and atherogenesis [55]. M1 macrophages impart pro-inflammatory functions *via* Th1 response augmentation and elevated production of pro-inflammatory IL-12 and reduced anti-inflammatory IL-10 production. In contrary, M2 macrophages exhibit immunoregulatory functions *via* phagocytosis Th2 response promotion, and elevated anti-inflammatory IL-10 production. Augmented production of IFN-via IL-27- and IL-27 receptor-deficient macrophages has been demonstrated, signifying that IL-27 impairs macrophage cell-autonomous activation and M1 polarization via regulating IFN secretion not only in paracrine but also in autocrine manner [47]. IL-27 significantly improved recovery of post-ischemic and attenuate tissue damage of isolated perfused hearts upon administration five minute earlier to reperfusion. This finding points to that IL-27 has protective role in the myocardium against ischemia/reperfusion (IR) injury and enhance cardiomyocytes recovery via the gp130/STAT3pathway [56]. IL-27 modulates pathological microenvironment in walls of the arteries that controls atherogenesis via regulation of cytokine induced macrophage production. The activation of macrophage in the arterial wall is a crucial step in atherosclerotic plaque rupture and this process induces thrombotic occlusion in the coronary arteries and consequent acute coronary syndrome. Immunosuppressive cytokines have vital role in local arterial lesions through immunoregulation which is beneficial to halt atherosclerosis and other interrelated diseases [57]. IL-27 solely

can considerably endorse the release of Interferon gamma-induced protein 10 (CXCL10) cytokine. Furthermore IL-27 can markedly augment the up regulation of TNF-α-mediated intercellular adhesion molecules (ICAM-1), vascular cell adhesion protein-1(VCAM-1), inflammatory cytokine IL-6 from the endothelial cells of human coronary arteries. The release of IL-6 and CXCL10 were markedly repressed by particular signaling molecule inhibitors [58]. Therefore, these findings signify the anti-inflammatory function of IL-27 that made it candidate for utilization in the treatment and prevention of atherosclerosis.

References

[1] Widén E, Ripatti. Assessment of multifactorial coronary artery disease by utilizing genomic data. *Duodecim*. 2017;133(8):776-81.

[2] Ross R. Atherosclerosis--an inflammatory disease. *N Engl J Med.* 1999;340(2):115-26.

[3] Raman K, Chong M, Akhtar-Danesh GG, D'Mello M, Hasso R, Ross S, et al. Genetic markers of inflammation and their role in cardiovascular disease. *The Canadian journal of cardiology.* 2013;29(1):67-74.

[4] Westerterp M, Murphy AJ, Wang M, Pagler TA, Vengrenyuk Y, Kappus MS, et al. Deficiency of ATP-binding cassette transporters A1 and G1 in macrophages increases inflammation and accelerates atherosclerosis in mice. *Circulation research.* 2013;112(11):1456-65.

[5] Hunter CA. New IL-12-family members: IL-23 and IL-27, cytokines with divergent functions. *Nature reviews Immunology*. 2005;5(7):521-31.

[6] Pflanz S, Timans JC, Cheung J, Rosales R, Kanzler H, Gilbert J, et al. IL-27, a heterodimeric cytokine composed of EBI3 and p28 protein, induces proliferation of naive CD4+ T cells. *Immunity*. 2002;16(6):779-90.

[7] Takeda A, Hamano S, Yamanaka A, Hanada T, Ishibashi T, Mak TW, et al. Cutting edge: role of IL-27/WSX-1 signaling for induction of T-bet through activation of STAT1 during initial Th1 commitment. *Journal of immunology*. 2003;170(10):4886-90.

[8] Yoshida H, Nakaya M, Miyazaki Y. Interleukin 27: a double-edged sword for offense and defense. *J Leukoc Biol*. 2009;86(6):1295-303.

[9] Gregersen I, Sandanger Ø, Askevold ET, Sagen EL, Yang K, Holm S, et al. Interleukin 27 is increased in carotid atherosclerosis and promotes NLRP3 inflammasome activation. *PLoS One*. 2017;12(11):e0188387.

[10] Koltsova EK, Kim G, Lloyd KM, Saris CJ, von Vietinghoff S, Kronenberg M, et al. Interleukin-27 receptor limits atherosclerosis in Ldlr-/- mice. *Circulation research.* 2012;111(10):1274-85.

[11] Wang JC, Normand SL, Mauri L, Kuntz RE. Coronary artery spatial distribution of acute myocardial infarction occlusions. *Circulation*. 2004;110(3):278-84.

[12] Bonaca MP, Wiviott SD, Braunwald E, Murphy SA, Ruff CT, Antman EM, et al. American College of Cardiology/American Heart Association/European Society of Cardiology/World Heart Federation universal definition of myocardial infarction classification system and the risk of cardiovascular death: observations from the TRITON-TIMI 38 trial (Trial to Assess Improvement in Therapeutic Outcomes by Optimizing Platelet Inhibition With Prasugrel-Thrombolysis in Myocardial Infarction 38). *Circulation*. 2012;125(4):577-83.

[13] Al-Khatib SM, Stevenson WG, Ackerman MJ, Bryant WJ, Callans DJ, Curtis AB, et al. 2017 AHA/ACC/HRS Guideline for Management of Patients With Ventricular Arrhythmias and the Prevention of Sudden Cardiac Death: A Report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines and the Heart Rhythm Society. *Heart Rhythm.* 2017.

[14] Skalen K, Gustafsson M, Rydberg EK, Hulten LM, Wiklund O, Innerarity TL, et al. Subendothelial retention of atherogenic lipoproteins in early atherosclerosis. *Nature*. 2002;417(6890):750-4.

[15] Pentikainen MO, Oorni K, Ala-Korpela M, Kovanen PT. Modified LDL - trigger of atherosclerosis and inflammation in the arterial intima. *J Intern Med*. 2000;247(3):359-70.

[16] Yoshida H, Kisugi R. Mechanisms of LDL oxidation. *Clin Chim Acta.* 2010;411(23-24):1875-82.

[17] Fotis L, Agrogiannis G, Vlachos IS, Pantopoulou A, Margoni A, Kostaki M, et al. Intercellular adhesion molecule (ICAM)-1 and vascular cell adhesion molecule (VCAM)-1 at the early stages of atherosclerosis in a rat model. *In Vivo*. 2012;26(2):243-50.

[18] Chevre R, Gonzalez-Granado JM, Megens RT, Sreeramkumar V, Silvestre-Roig C, Molina-Sanchez P, et al. High-resolution imaging of intravascular atherogenic inflammation in live mice. *Circulation research.* 2014;114(5):770-9.

[19] McArdle S, Mikulski Z, Ley K. Live cell imaging to understand monocyte, macrophage, and dendritic cell function in atherosclerosis. *J Exp Med*. 2016;213(7):1117-31.

[20] Moore KJ, Sheedy FJ, Fisher EA. Macrophages in atherosclerosis: a dynamic balance. *Nature reviews Immunology*. 2013;13(10):709-21.

[21] Kunjathoor VV, Febbraio M, Podrez EA, Moore KJ, Andersson L, Koehn S, et al. Scavenger receptors class A-I/II and CD36 are the principal receptors responsible for the uptake of modified low density lipoprotein leading to lipid loading in macrophages. *J Biol Chem.* 2002;277(51):49982-8.

[22] Buono C, Anzinger JJ, Amar M, Kruth HS. Fluorescent pegylated nanoparticles demonstrate fluid-phase pinocytosis by macrophages in mouse atherosclerotic lesions. *J Clin Invest.* 2009;119(5):1373-81.

[23] Park YM, Febbraio M, Silverstein RL. CD36 modulates migration of mouse and human macrophages in response to oxidized LDL and may contribute to macrophage trapping in the arterial intima. *J Clin Invest.* 2009;119(1):136-45.

[24] Ross R, Glomset JA. Atherosclerosis and the arterial smooth muscle cell: Proliferation of smooth muscle is a key event in the genesis of the lesions of atherosclerosis. *Science*. 1973;180(4093):1332-9.

[25] Blake GJ, Ridker PM. Novel clinical markers of vascular wall inflammation. *Circ Res.* 2001;89(9):763-71.

[26] Naghavi M, Libby P, Falk E, Casscells SW, Litovsky S, Rumberger J, et al. From vulnerable plaque to vulnerable patient: a call for new definitions and risk assessment strategies: *Part II. Circulation.* 2003;108(15):1772-8.

[27] Hansson GK, Hermansson A. The immune system in atherosclerosis. *Nat Immunol.* 2011;12(3):204-12.

[28] Zhao PW, Jiang WG, Wang L, Jiang ZY, Shan YX, Jiang YF. Plasma levels of IL-37 and correlation with TNF-alpha, IL-17A, and disease activity during DMARD treatment of rheumatoid arthritis. *PLoS One.* 2014;9(5):e95346.

[29] von Vietinghoff S, Koltsova EK, Mestas J, Diehl CJ, Witztum JL, Ley K. Mycophenolate mofetil decreases atherosclerotic lesion size by depression of aortic T-lymphocyte and interleukin-17-mediated macrophage accumulation. J *Am Coll Cardiol.* 2011;57(21):2194-204.

[30] Vasan RS. Biomarkers of cardiovascular disease: molecular basis and practical considerations. *Circulation*. 2006;113(19):2335-62.

[31] Szabo SJ, Sullivan BM, Peng SL, Glimcher LH. Molecular mechanisms regulating Th1 immune responses. *Annu Rev Immunol*. 2003;21:713-58.

[32] Lindahl B, Toss H, Siegbahn A, Venge P, Wallentin L. Markers of myocardial damage and inflammation in relation to long-term mortality in unstable coronary artery disease. FRISC Study Group. Fragmin during Instability in Coronary Artery Disease. *N Engl J Med.* 2000;343(16):1139-47.

[33] Damås JK, Waehre T, Yndestad A, Otterdal K, Hognestad A, Solum NO, et al. Interleukin-7-mediated inflammation in unstable angina: possible role of chemokines and platelets. *Circulation.* 2003;107(21):2670-6.

[34] Liuzzo G, Biasucci LM, Rebuzzi AG, Gallimore JR, Caligiuri G, Lanza GA, et al. Plasma protein acute-phase response in unstable angina is not induced by ischemic injury. *Circulation*. 1996;94(10):2373-80.

[35] Yvan-Charvet L, Ranalletta M, Wang N, Han S, Terasaka N, Li R, et al. Combined deficiency of ABCA1 and ABCG1 promotes foam cell accumulation and accelerates atherosclerosis in mice. *J Clin Invest.* 2007;117(12):3900-8.

[36] Glass CK, Witztum JL. Atherosclerosis. the road ahead. *Cell*. 2001;104(4):503-16.

[37] Tabas I. Macrophage death and defective inflammation resolution in atherosclerosis. *Nat Rev Immunol.* 2010;10(1):36-46.

[38] Bazan JF. Haemopoietic receptors and helical cytokines. *Immunol Today*. 1990;11(10):350-4.

[39] Villarino A, Hibbert L, Lieberman L, Wilson E, Mak T, Yoshida H, et al. The IL-27R (WSX-1) is required to suppress T cell hyperactivity during infection. *Immunity*. 2003;19(5):645-55.

[40] Villarino AV, Larkin J, Saris CJ, Caton AJ, Lucas S, Wong T, et al. Positive and negative regulation of the IL-27 receptor during lymphoid cell activation. *J Immunol.* 2005;174(12):7684-91.

[41] Wirtz S, Tubbe I, Galle PR, Schild HJ, Birkenbach M, Blumberg RS, et al. Protection from lethal septic peritonitis by neutralizing the biological function of interleukin 27. *J Exp Med.* 2006;203(8):1875-81.
[42] Pflanz S, Hibbert L, Mattson J, Rosales R, Vaisberg E, Bazan JF, et al. WSX-1 and glycoprotein 130 constitute a signal-transducing receptor for IL-27. *J Immunol*. 2004;172(4):2225-31.
[43] Stumhofer JS, Hunter CA. Advances in understanding the anti-inflammatory properties of IL-27. *Immunol Lett.* 2008;117(2):123-30.
[44] Scheller J, Schuster B, Hölscher C, Yoshimoto T, Rose-John S. No inhibition of IL-27 signaling by soluble gp130. *Biochem Biophys Res Commun.* 2005;326(4):724-8.
[45] Chen Q, Ghilardi N, Wang H, Baker T, Xie MH, Gurney A, et al. Development of Th1-type immune responses requires the type I cytokine receptor TCCR. *Nature*. 2000;407(6806):916-20.
[46] Davenport P, Tipping PG. The role of interleukin-4 and interleukin-12 in the progression of atherosclerosis in apolipoprotein E-deficient mice. *Am J Pathol.* 2003;163(3):1117-25.
[47] Awasthi A, Carrier Y, Peron JP, Bettelli E, Kamanaka M, Flavell RA, et al. A dominant function for interleukin 27 in generating interleukin 10-producing anti-inflammatory T cells. *Nat Immunol.* 2007;8(12):1380-9.
[48] Amadi-Obi A, Yu CR, Liu X, Mahdi RM, Clarke GL, Nussenblatt RB, et al. TH17 cells contribute to uveitis and scleritis and are expanded by IL-2 and inhibited by IL-27/STAT1. *Nat Med.* 2007;13(6):711-8.
[49] Si F, Wu Y, Gao F, Feng S, Liu R, Yi Q. Relationship between IL-27 and coronary arterial lesions in children with Kawasaki disease. *Clin Exp Med.* 2017;17(4):451-7.
[50] Posadas-Sanchez R, Perez-Hernandez N, Rodriguez-Perez JM, Coral-Vazquez RM, Roque-Ramirez B, Llorente L, et al. Interleukin-27 polymorphisms are associated with premature coronary artery disease and metabolic parameters in the Mexican population: the genetics of atherosclerotic disease (GEA) Mexican study. *Oncotarget*. 2017;8(38):64459-70.
[51] Ait-Oufella H, Taleb S, Mallat Z, Tedgui A. Recent advances on the role of cytokines in atherosclerosis. *Arterioscler Thromb Vasc Biol*. 2011;31(5):969-79.
[52] Tedgui A, Mallat Z. Cytokines in atherosclerosis: pathogenic and regulatory pathways. *Physiol Rev*. 2006;86(2):515-81.
[53] Smith E, Prasad KM, Butcher M, Dobrian A, Kolls JK, Ley K, et al. Blockade of interleukin-17A results in reduced atherosclerosis in apolipoprotein E-deficient mice. *Circulation*. 2010;121(15):1746-55.
[54] Murray PJ, Wynn TA. Protective and pathogenic functions of macrophage subsets. *Nat Rev Immunol.* 2011;11(11):723-37.
[55] Woollard KJ, Geissmann F. Monocytes in atherosclerosis: subsets and functions. *Nat Rev Cardiol.* 2010;7(2):77-86.
[56] Fan Q, Nie S, Li S, Liao Y, Zhang H, Zha L, et al. Analysis of the genetic association between IL27 variants and coronary artery disease in a Chinese Han population. *Sci Rep.* 2016;6:25782.
[57] Fuster V, Moreno PR, Fayad ZA, Corti R, Badimon JJ. Atherothrombosis and high-risk plaque: part I: evolving concepts. *J Am Coll Cardiol.* 2005;46(6):937-54.
[58] Qiu HN, Liu B, Liu W, Liu S. Interleukin-27 enhances TNF-alpha-mediated activation of human coronary artery endothelial cells. *Mol Cell Biochem*. 2016;411(1-2):1-10.