We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

4,800
Open access books available

122,000
International authors and editors

135M
Downloads

Our authors are among the

154
Countries delivered to

TOP 1%
most cited scientists

12.2%
Contributors from top 500 universities

CLARIVATE ANALYTICS BOOK CITATION INDEX INDEXED

WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com

5

CD40 Ligand and Its Receptors in Atherothrombosis

Daniel Yacoub[1], Ghada S. Hassan[1], Nada Alaadine[1], Yahye Merhi[2] and Walid Mourad[1*]

[1]*Laboratoire d'Immunologie Cellulaire et Moléculaire, Centre Hospitalier de l'Université de Montréal, Hôpital Saint-Luc, Montréal,*

[2]*Institut de Cardiologie de Montréal, Université de Montréal, Montréal, Canada*

1. Introduction

Atherothrombosis is the main underlying determinant of cardiovascular diseases, which remain the leading cause of death in developed countries. Multiple lines of evidence now support the concept of atherothrombosis as a chronic inflammatory disease of the arterial wall.[1, 2] This process involves a complex interplay between modified lipids and cells of the immune and vascular system, which usually evolves into the formation of atherosclerotic lesions yielding a stable necrotic plaque. If left untreated, plaque rupture and thrombosis may ensue, leading to important clinical manifestations, such as acute coronary syndromes and sudden death.[3]

As the incorporation of modified low-density lipoproteins in the arterial wall represents a important step in the onset of atherothrombosis, the subsequent recruitment and activation of inflammatory cells, including monocytes, B- and T-lymphocytes, neutrophils and platelets play a critical role in the pathogenesis of this disease.[4] These cells exhibit pro-atherogenic functions through multiple co-stimulatory and immune molecules present on their cell surface. Among these, the CD40L/CD40 receptor-ligand pair has been the focus of much attention, such that this dyad is now regarded as a pivotal contributor to all underlying phases of atherothrombosis.[5-7] Indeed, the CD40L/CD40 interaction exerts a wide array of biological functions at the forefront of the pathophysiology of this disease and disruption of this cascade by both pharmacological and genetic approaches have shown beneficial results in animal and clinical studies.[8-11] While CD40L is known to mainly interact with its classical receptor CD40, additional binding partners have been described, namely the integrins $\alpha_{IIb}\beta_3$, $\alpha_M\beta_2$ and $\alpha_5\beta_1$. This chapter discusses the role of CD40L and its receptors in the pathophysiology of atherothrombosis, while highlighting its therapeutic potentials in the treatment of this chronic inflammatory disease.

* Corresponding Author

2. The CD40L system

2.1 CD40L

CD40L, also known as CD154, is a 39 kDa transmembrane protein belonging to the tumor necrosis factor (TNF) superfamily originally identified on cells of the immune system.[12, 13] The interaction of CD40L with its respective receptor on B cells, CD40, a glycoprotein also from the TNF receptor (TNFR) family, is of critical importance for immunoglobulin isotype switching during the immune response.[14] The importance of this interaction is highlighted by the pathophysiological manifestations seen in patients suffering from the X-linked hyper-immunoglobulin-M syndrome, in which B-cells fail to produce the immunoglobulin's IgG, IgA and IgE as a consequence of a genetic mutation in the CD40L gene.[15] Because of its wide distribution across cells of the vascular system (endothelium, B and T lymphocytes, neutrophils, platelets, monocytes, dentritic cells and smooth muscle cells), the CD40L/CD40 dyad also shares important implications in cell-mediated immunity. CD40L-induced signaling in these cells leads to the up-regulation of adhesion and co-stimulatory molecules, and the production of pro-inflammatory cytokines, chemokines, growth factors, matrix metalloproteinases (MMPs) and procoagulants.[16-19] These cellular events are also the main mechanisms by which CD40L regulates numerous inflammatory disorders, in particular atherothrombosis and its related complications. In fact, circulating levels of soluble CD40L (sCD40L), which originate from the proteolytic cleavage of membrane-bound CD40L at the surface of activated platelets, have now emerged as strong indicators of cardiovascular events such as atherothrombosis and acute coronary syndromes.[20-22]

2.2 CD40L receptors

CD40

CD40 is the classical high affinity receptor for CD40L. It is constitutively or inducibly expressed by most cells of the vascular system (hematopoietic and non-hematopoietic cells) and represents the main signaling molecule in the CD40L/CD40 receptor-ligand pair.[23] The cytoplasmic domain of CD40 bears signaling domains required for the association of binding proteins termed TNF receptor-associated factors (TRAFs).[24] During humoral immunity, a tight interplay between dendritic cells, T-lymphocytes and B-lymphocytes occurs, throughout which the activation of CD40 provides a crucial signal for the activation, differentiation and secretion of immunoglobulins by B cells.[25] Moreover, CD40 activation in these cells induces an important anti-apoptotic signal that facilitates cell survival and differentiation, primarily through activation of the anti-apoptotic proteins Bcl-XL, A20, Bfl-1 and Mcl-1, which protect against Fas ligand and TNF-induced cell death.[26] As discussed above, CD40 signaling plays a significant role in cell-mediated immunity, an important aspect by which the CD40L/CD40 dyad initiates and exacerbates atherosclerotic lesions. For instance, CD40 activation on endothelial cells induces the up-regulation of a plethora of proinflammatory adhesion molecules, cytokines, chemokines, matrix metalloproteinases and procoagulants.[16] In addition, it has been demonstrated that upon CD40L binding CD40 activation on platelets can enhance platelet function and promote the secretion of inflammatory cytokines involved in plaque formation (this aspect will be discussed in greater detail bellow).[27-29]

CD40-TRAF dependent signaling

The engagement of CD40 by CD40L promotes the clustering of CD40 and induces the association of TRAFs to the cytoplasmic domain of CD40.[30] These adapter proteins are essential for the activation of different signaling pathways including the canonical and non-canonical nuclear factor κB (NF-κB)-signaling pathways and the activation of mitogen-activated protein kinases (MAPKs).[30] The TRAF family comprises six known members, among which TRAF-1, -2, -3, -5 and -6 have been shown to drive CD40-dependent cellular responses.

TRAF-1 can only bind weakly to the cytoplasmic tail of CD40 and therefore regulates the signaling of others TRAF members, in particular TRAF-2.[30, 31] Indeed, TRAF-1 deficiency in antigen presenting cells and B-lymphocytes leads to a significant reduction in the recruitment of TRAF-2 to CD40, indicating that TRAF-1 facilitates the association of TRAF-2 to the cytoplasmic domain of CD40.[32, 33] In agreement with these results, it has been shown that the recruitment of both TRAF-1 and TRAF-2 are required for complete activation of NF-κB in B-cells, since the knockout of both genes results in a greater inhibition of the NF-κB signaling pathway, in comparison to single knockouts.[33]

TRAF-2 is an important contributor to CD40 signaling and its major role resides in the activation of the NF-κB signaling pathway, as well as the activation of the p38, Akt and JNK MAPKs. CD40 bears a direct binding site for TRAF-2 and blockage of this interaction leads to immune deficiencies such as B-cell proliferation and isotype switching.[24, 34] Despite its significant implications in CD40 signaling, TRAF-2 deficiency may be overcome by TRAF-6 activation. This was confirmed by data showing that binding of either TRAF-2 or TRAF-6 alone may activate the NF-κB pathway, while inhibition of both these members completely abolishes CD40-dependant B-cell activation, suggesting that both members collaborate for the activation of this critical signaling cascade.[35-37]

TRAF-3 functions as a negative regulator of CD40 signaling through its constitutive association with TRAF-2.[38, 39] In absence of stimulation, TRAF-3 interacts with TRAF-2, which allows the degradation of the NF-κB inducing kinase (NIK) protein, a critical stimulator of NF-κB.[40] TRAF-3 deficiency in B cells exacerbates NF-κB and JNK activation, primarily through cytosolic accumulation of NIK, thus confirming the negative regulatory functions of TRAF-3 in B cells.[41]

Very little information is available regarding the role of TRAF-5 in CD40 signaling. Nevertheless, it appears that TRAF-5 can interact with TRAF-3 to modulate NF-κB activation in B cells. This was shown by experiments in which TRAF-5 deficiency diminishes NF-κB activation, causing a reduction in cell activation, expression of co-stimulatory molecules and antibody production.[42, 43]

TRAF-6 plays a significant role in the activation of key CD40-dependent signaling pathways, such as NF-κB, p38, JNK and Akt.[44] As discussed above, TRAF-6 synergizes with TRAF-2 in order to regulate the activation of NF-κB. Although TRAF-6 contains a direct binding site for CD40, specific inhibition of this domain shows lesser inhibitory effects than ablation of the complete protein, indicating that TRAF-6 may still have a functional role in CD40 signaling without binding directly to CD40.[30, 45] Indeed, one of the main functions of TRAF-6 resides in its ability to interact with TRAF-2, which is already bound to CD40, and facilitate the activation of downstream targets.

Recently, a study aiming at evaluating the implication of TRAF members in neointima formation, a critical step of atherothrombosis, was conducted. Using a CD40 transgenic mouse model, in which mutations at the TRAF2/3/5, TRAF6 or TRAF2/3/5/6 binding sites were carried out, the authors conclude that the CD40-TRAF6 axis is a key regulator of inflammatory cell infiltration and neointima formation at sites of vascular injury.[46]

Although most vascular complications associated to CD40L, including atherothrombosis, have been largely attributed to its interaction with CD40, recently identified additional receptors merit attention. These include the integrins $\alpha_{IIb}\beta_3$, $\alpha_5\beta_1$ and $\alpha_M\beta_2$ and (Figure 1).

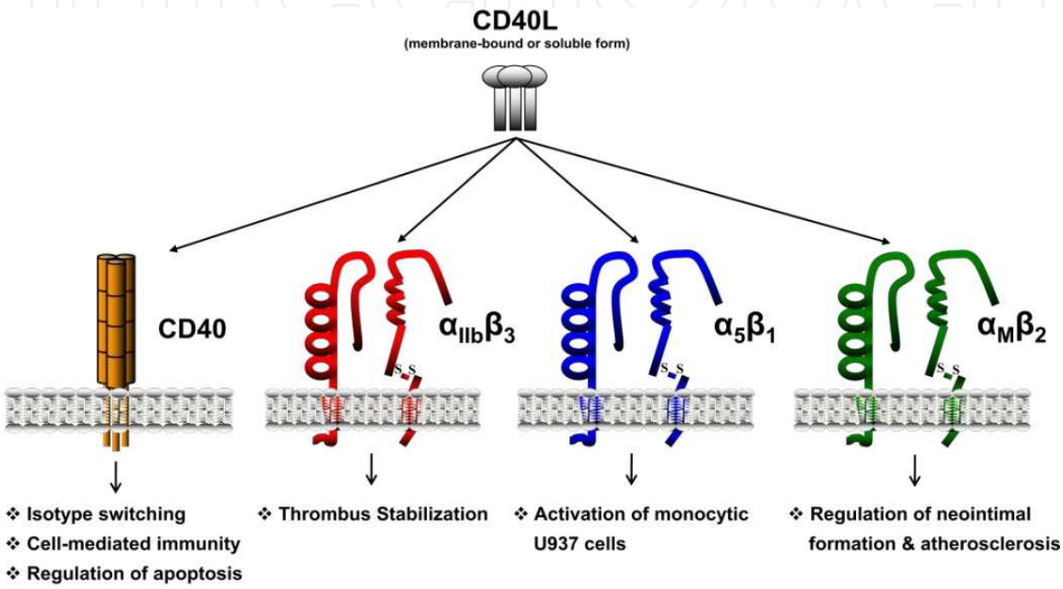


Fig. 1. CD40L and its receptors. The binding of CD40L to its classical CD40 counterreceptor regulates numerous critical biological responses. These mainly include B-cell dependent isotype switching, cell-mediated immunity (production of cytokines, chemokines, adhesion molecules, growth factors, MMPs and procoagulants) and apoptosis. The CD40L/CD40 interaction is at the forefront of the pathogenesis of multiple inflammatory disorders, including atherothrombosis. The interaction of CD40L with the αIIbβ3 platelet integrin is involved in thrombus stabilization and may provide a novel outside-in signaling pathway by which platelets can be activated. CD40L can also bind to the inactive conformation α5β1 and this interaction was shown to induce activation of the human monocytic U937 cell line. Finally, αMβ2 can mediate CD40L-dependent inflammatory responses, in particular leukocyte adhesion and neointimal formation. The pathophysiological relevance of these novel CD40L-mediated interactions in inflammation remains elusive and additional studies will be required to address this issue.

αIIbβ3

The αIIbβ3 integrin is the most abundant receptor of the surface of platelets and mediates platelet adhesion and aggregation. Like all molecules of the integrin family, it will change conformation upon inside-out cellular activation, thereby allowing binding to its natural ligands (fibrinogen, fibronectin, vWF...).[47] These ligands contain KGD sequences and

binding is mediated through the KGD recognition domain present on the αIIbβ3 molecule. Interestingly, CD40L also contains a KGD sequence making its interaction with αIIbβ3 possible. Binding of CD40L to αIIbβ3 was shown to induce phosphorylation of tyrosine residues within the cytoplasmic domain of the β3 subunit and appears essential for thrombus stabilization *in vivo*.[48, 49] Indeed, CD40L$^{-/-}$ mice exhibit unstable thrombi, which can be overcome by infusion of wild type recombinant human CD40L and not CD40L specifically mutated at the site of interaction with αIIbβ3.[48]

α5β1

The *a5β1* integrin was also shown to act as a CD40L receptor.[50] Indeed, sCD40L can bind and activate cells of the undifferentiated human monocytic U937 cell line in a CD40- and αIIbβ3-independent manner. Binding to this cell line was reversed by an anti-*a5β1* antibody, as well as in the presence of soluble *a5β1*, thus confirming *a5β1* specificity. Moreover, this interaction is unaffected by pre-treatment of CD40L with soluble CD40, indicating that CD40L can bind both CD40 and *a5β1* concomitantly.[50] Interestingly, CD40L binds to inactive *a5β1*, contrary to most ligands of the integrin family. However, the physiological relevance of this interaction remains unexplored and additional studies are needed to fully characterize the interplay that might take place between CD40L and *a5β1* in inflammatory disorders.

αMβ2

The αMβ2 (Mac-1) integrin mediates firm adhesion of leukocytes to inflamed vessels by interacting with its endothelial cell counterreceptor intercellular adhesion molecule 1 (ICAM-1).[51] CD40L was also recently shown to bind to active αMβ2 and this interaction may represent an alternative pathway for CD40L-mediated inflammation.[52] Indeed, inhibition of this novel CD40L binding partner significantly attenuates leukocyte accumulation at sites of inflammation and reduces atherogenesis, indicating that CD40L may promote, at least in part, atherosclerotic lesions in a αMβ2-dependent manner. Again, the relative contribution of this CD40L receptor (in comparison to CD40) in the development of inflammatory disorders such as atherothrombosis remains unknown. Perhaps, each of these CD40L receptors may interfere at different stages of the disease, thus contributing to proinflammatory reactions and atherogenesis in their own way.

3. CD40L in atherothrombosis

The involvement of CD40L in the pathogenesis of atherothrombosis is supported by numerous studies. Targeting of CD40L by both pharmacological and genetic approaches has highlighted the importance of this molecule in all stages of the disease. In 1998, Mach et al. showed that treatment of hyperlipidemic LDLR$^{-/-}$ mice with an anti-CD40L antibody significantly ameliorates the size and lipid contents of atherosclerotic lesions.[53] These results were further confirmed by a genetic approach, which showed that CD40L$^{-/-}$/ApoE$^{-/-}$ mice exhibit considerably smaller plaque area than control ApoE$^{-/-}$ mice.[8] Moreover, these animals display enhanced collagen fibrils within the fibrous cap of lesions, a key component of plaque stability. In an additional study, the administration of an anti-CD40L antibody in ApoE$^{-/-}$ mice, at the onset of lesions or once atherosclerotic lesions are fully established, reduces lipid contents and increases plaque stability.[54] Taken together, these studies support the contribution of CD40L in plaque initiation, progression and stability (Figure 2).

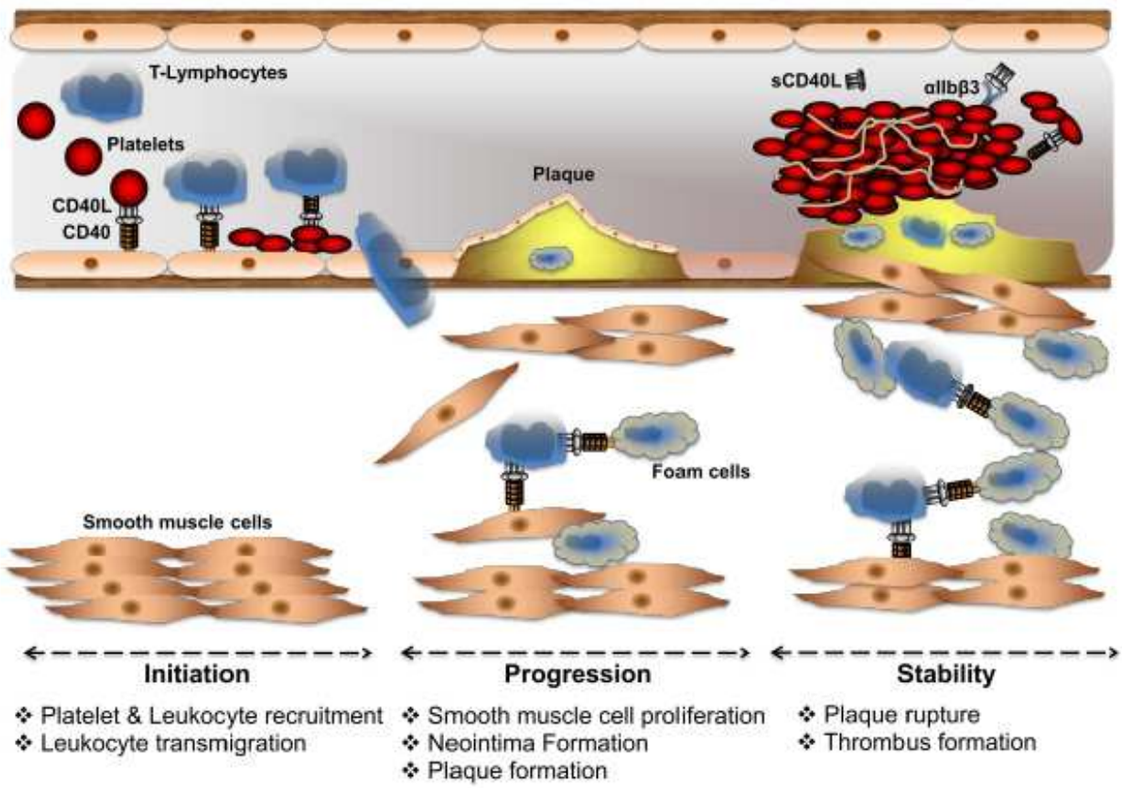


Fig. 2. Role of CD40L in atherothrombosis. The incorporation of oxidized LDLs, among other factors, may upregulate the expression of the CD40L system on the endothelium, thereby promoting the recruitment of platelets and leukocytes at the sites of injury. The CD40L-dependent adhesion of T-lymphocytes and platelets to the endothelium induces an important inflammatory response characterized by the secretion of various cytokines and the upregulation of additional endothelial adhesion molecules. This favors in turn the incorporation and transmigration of additional leukocytes, in particular monocytes. Once in the sub-endothelial space, CD40L/CD40 interactions between foam cells (macrophages which have undergone phagocytosis of oxidized LDL particules), T-lymphocytes and smooth muscle cells take place. These cross talks ultimately lead in part to the proliferation of smooth muscle cells into the intima and promote vascular angiogenesis, primarily through the secretion of key inflammatory and angiogenic cytokines and chemokines. This process eventually yields a stable lipid-enriched atherosclerotic plaque surrounded by a fibrous cap. Plaque stability is threatened by the production of MMPs, which are directly responsible for collagen degradation and rupture of the fibrous cap. The binding of CD40 on endothelial cells, macrophages and smooth muscle cells can provoke the secretion of a long list of MMPs. Following rupture, platelets rapidly adhere to the surface of the highly pro-thrombotic contents of the atherosclerotic plaque, thereby leading to thrombus formation and arterial occlusion. CD40L may also be involved in thrombus formation and procoagulant activity. CD40L binding to the endothelium promotes tissue factor expression, while the binding of CD40L (soluble and membrane-bound forms) to CD40 and αIIbβ3 on platelets enhances platelets aggregation and thrombus stabilization, respectively.

3.1 Initiation of lesions and leukocyte recruitment

Plaque initiation is normally characterized by the accumulation of low-density lipoproteins (LDL) in the arterial wall and the subsequent recruitment and transmigration of leukocytes within the sub-endothelial space.[1] The initial trigger of CD40L (and CD40) expression within cells of the developing atherosclerotic plaque (endothelial cells, lymphocytes, platelets, monocytes/macrophages, and smooth muscle cells) remains elusive. Possible candidates include oxidized LDL, infectious pathogens and alterations in vascular hemodynamic forces.[55-57] For instance it has been demonstrated that lipid lowering reduces CD40L expression in atheroma.[55] In addition, oxidized LDL were reported to induce the expression of CD40 on endothelial cells, which can then bind CD40L from activated T-lymphocytes adherent to the site of injury.[58] CD40 activation on endothelial cells provides a critical proinflammatory signal for the initiation of lesions. Indeed, the CD40L/CD40 interaction favors the up-regulation of adhesion molecules (E-selectin, P-selectin, vascular cell adhesion molecule-1 [VCAM-1] and ICAM-1) and leads to the secretion of proinflammatory cytokines (IL-6, IL-8, IL-15, monocytes chemotactic protein-1 [MCP-1], macrophage inflammatory protein-1 [MIP-1 α/β] and regulated on activation normal T cell expressed and secreted [RANTES]) by the endothelium.[6, 7, 59-62] These reactions induce in turn the incorporation and accumulation of additional leukocytes, in particular monocytes, at the sites of developing lesions.

As discussed above, αMβ2, an integrin expressed on neutrophils and monocytes/ macrophages, has been identified as a receptor for CD40L. This interaction may also mediate adhesion and migration of inflammatory cells at sites of plaque initiation. In agreement with this hypothesis, αMβ2 deficiency attenuates lesion development and reduces lesional macrophage accumulation in LDLR$^{-/-}$ mice, supporting the implication of this integrin in atherothrombosis.[52] However, additional studies will be required to specifically establish the importance of the CD40L/αMβ2 in plaque initiation.

In addition, platelets have been shown to play a crucial role in the initiation of atherothrombosis. Platelets are among the first inflammatory cells at the site of injury and their adhesion to the endothelium provides a fundamental mechanism by which leukocytes are recruited.[63, 64] Because the surface of activated platelets contains a higher density of P-selectin than activated endothelial cells, significantly more leukocytes will incorporate at the sites of injury in their presence.[65] Interestingly, CD40L from activated platelets can also induce a proinflammatory response on endothelial cells, in a similar fashion to that of T-lymphocytes. Henn et al. demonstrated that CD40 ligation on endothelial cells by CD40L from activated platelets induces the expression of numerous adhesion molecules, cytokines, and matrix metalloproteinases involved in the initiation of inflammatory reactions.[16]

3.2 Plaque development and progression

The progression of the atherosclerotic plaque is typically highlighted by the proliferation and migration of smooth muscle cells into the intima, as well as the formation neovessels (angiogenesis), which supports the growth of lesions. This process will eventually yield a stable lipid-enriched necrotic plaque surrounded by a fibrous cap. Once in the sub-endothelial space, macrophages (originally monocytes) undergo phagocytosis of oxidized LDL particles, leading to the formation of foam cells.[66] Thereafter, CD40L from infiltrated T-

lymphocytes will bind to CD40 on the surface of differentiated foam cells, favoring the release of further proinflammatory cytokines (IL-1, IL-6 and IL-12), growth factors (vascular endothelial growth factor [VEGF]) and MMPs (MMP-1 and MMP-3).[7, 67] These responses are intimately involved in the proliferation and migration of smooth muscle cells into the intima layer. In parallel, cross talks between smooth muscle cells and T-lymphocytes may also take place, in which CD40 activation on the former initiates a positive feedback loop enhancing the inflammatory reactions already in place.[6] Indeed, CD40 signaling in smooth muscle cells has been shown to induce the secretion of the cytokines IL-8 and MCP-1.[67, 68] Moreover, the accumulation of migrating fibroblasts within the intima layer exacerbates the atherosclerotic lesions in development and the CD40L/CD40 axis might also takes part in this process.[69] Stimulation of fibroblasts with CD40L was reported to up-regulate the expression of cell surface adhesion molecules, thus facilitating their interaction with immune cells at the site of lesions.[70] This interaction also induces their proliferation and secretion of chemoattractant cytokines such as IL-6 and IL-8.[71-73] Hence, the CD40L/CD40 interaction is at the forefront of a plethora of key inflammatory reactions involved in neointima formation and plaque accumulation. Lesions with eventually develop into the formation of a stable necrotic core consisting of infiltrated leukocytes, foam cells, proliferating smooth muscle cells, extra-cellular matrix proteins and lipids.

The formation of neovessels or angiogenesis plays an integral part in plaque progression and several reports have highlighted the importance of CD40L in this process. For instance, CD40 ligation on endothelial cells and macrophages was shown to upregulate the expression of potent angiogenic factors such as VEGF, fibroblast growth factor and platelet-activating factor, in addition to inducing the synthesis and proteinase activity of various MMPs such as MMP-1, MMP-2, MMP-3 and MMP-9.[18, 74-76] These responses are tightly linked to tubule formation and angiogenesis, essential elements of plaque support and growth. Interestingly, the α5β1 integrin is upregulated on angiogenesis-prone endothelial cells and could also provide a novel mechanism by which CD40L modulates pathological angiogenesis.[77] It would be worthwhile investigating this issue in further details.

3.3 Plaque instability and thrombosis

Plaque stability is regulated by a tight balance between extracellular matrix proteins such as collagen fibers and MMP production. A thin fibrous cap protects the highly thrombotic components of the atherosclerotic plaque. However, upon secretion of MMPs by macrophages and other inflammatory cells present, plaque rupture may ensue following digestion of the collagen fibers within the fibrous cap.[78, 79] This process leads to thrombus formation and may cause complete obstruction of the artery. CD40L mediates several of the processes that set the stage for plaque rupture and its clinical sequelae. CD40L stimulation on endothelial cells, macrophages and smooth muscle cells can provoke the secretion of a long list of MMPs (MMP-1, MMP-2, MMP-3, MMP-8, MMP-9 and MMP-13), the main digestive enzymes of the collagen-rich fibrous cap.[18, 80, 81] Platelets, in addition to their pivotal role in thrombosis, also participate in this process. Indeed, membrane-bound CD154 expressed on the surface of activated platelets can induce MMP upregulation in endothelial cells.[82] MMP secretion and proteolytic activity can be abrogated by physical hindrance of platelet-endothelial contacts, αIIbβ3 interfering agents or anti-CD40L antibodies, thus highlighting in part the importance of platelets and CD40L in this phenomenon.

Following rupture, platelets rapidly adhere to the surface of the highly pro-thrombotic contents of the atherosclerotic plaque, thereby leading to thrombus formation and arterial occlusion.[83, 84] Accumulating evidence also support a role for CD40L in platelet function and thrombus formation, albeit some of the data remain conflicting. For instance, Andre et al. have shown that CD40L plays a role in thrombus stabilization by interacting with αIIbβ3, while we and others have demonstrated that CD40L enhances platelet aggregation and thrombus formation through a CD40-mediated TRAF-2/Rac1/p38 signaling pathway.[27, 28, 48] Indeed, enhanced levels of circulating sCD40L exacerbate thrombus formation *in vivo*, also in a CD40-dependent fashion.[27] Nevertheless, these studies all support the concept of CD40L as a pro-thrombotic agent, predisposing platelets to enhanced cell function. CD40L may also enhance the coagulation system through the induction of tissue factor release from various vascular cells. CD40 engagement on endothelial cells, macrophages and smooth muscle cells by CD40L from activated platelets or T cells induces tissue factor expression and activity.[85-87] Besides its important role in the induction of the extrinsic coagulation cascade, tissue factor also represents a powerful platelet agonist.

4. Soluble CD40L and coronary syndromes

Given the pivotal contribution of the CD40L system in atherothrombosis, multiple clinical studies have evaluated the association between levels of circulating sCD40L and cardiovascular risk, in particular acute coronary syndrome (ACS) such as acute myocardial infarction (AMI) and unstable angina (UA). These studies can be divided into two main categories. The first type of clinical studies has investigated the link between levels of sCD40L and ACS, while the second has determined the link between levels of sCD40L and prognosis and risk prediction.

For the most part, clinical studies demonstrate that circulating sCD40L levels are significantly higher in patients with ACS and stable coronary artery disease (CAD), compared with control subjects.[88-92] Indeed, it appears that a gradual increase in sCD40L levels occurs with ACS progression, with peaks as early as 9 hours following the onset of AMI or UA.[89, 93, 94] For instance, patients suffering from AMI or UA present with levels ranging from 5-25 ng/mL, depending on the study.[88] Moreover, sCD40L levels are independent from other important inflammatory markers, such as IL-6, sICAM-1, sVCAM-1, C-reactive protein and troponin, indicating that sCD40L may represent a more reliable risk factor, as compared to others.[92] Because sCD40L in circulation almost exclusively originates from the shedding of membrane-bound CD40L at the surface of activated platelets, its measuring levels may reflect a state of platelet activation rather than an inflammatory condition.

More importantly, some clinical studies have evaluated the relationship between sCD40L levels and disease prognosis. In the CAPTURE (c7E3 Fab Anti-Platelet Therapy in unstable Refractory Angina) trial, patients with high levels of sCD40L were 3-fold more at risk of developing cardiovascular death or AMI.[21] Moreover, in the MIRACL (Myocardial Ischemia Reduction with Aggressive Cholesterol Lowering) study, sCD40L levels were an independent risk factor for recurrent cardiovascular events, such as death, nonfatal myocardial infarction, cardiac arrest and worsening angina requiring rehospitalization.[95] Interestingly, individuals carrying the -3459A>G polymorphism on the CD40L gene, are more at risk of developing AMI.[96]

Whether enhanced levels of sCD40L seen in patients with ACS are a consequence of increased platelet activation or a predetermining cause of these complications (or perhaps both) is still unknown. Recently, we have shown that enhancing levels of circulating sCD40L in mice to approximately 45 ng/mL exacerbates thrombus formation in a CD40-dependent manner.[27] This observation supports the idea that increased levels of sCD40L in patients may drive, at least in part, the development of certain cardiovascular complications. It would be tempting to speculate the existence of a positive feedback loop taking place in these patients, where disease initiation correlates with platelet activation and release of sCD40L in the circulation. This in turn could further exacerbate pre-existing complications through enhancement of platelet function and thrombus formation.

5. Disruption of the CD40L system as a therapeutic target in atherothrombosis

In light of all the aforementioned data supporting the contribution of CD40L in inflammation, disruption of this system as a therapeutic strategy for the treatment of atherothrombosis and its clinical manifestations has been investigated. Unfortunately, clinical trials using an anti-CD40L antibody were put on hold due to thromboembolic complications.[97, 98] Interactions between CD40 and CD40L-immune complexes at the surface of platelets have been suggested as a possible mechanism by which CD40L therapy induces these complications.[97]

Since circulating levels of sCD40L result from platelet activation, indirect targeting of the CD40L system through anti-platelet therapy may represent an alternative approach to suppress this important component. Clopidogrel, a potent inhibitor of the platelet adenosine diphosphate (ADP) receptor, has been reported to block sCD40L release from ADP-stimulated platelets.[99] Interestingly, clopidogrel regiment significantly reduces platelet CD40L expression and sCD40L levels in patients with stable CAD.[100] Moreover, αIIbβ3 inhibitors, such as abciximab, inhibit platelet aggregation and sCD40L release from activated platelets.[101] In the CAPTURE trial, abciximab significantly reduces sCD40L levels and cardiovascular risk in high-risk ACS patients, confirming a link between αIIbβ3 signaling and platelet sCD40L release.[21]

Statins exert multiple pleiotropic anti-inflammatory effects, in addition to their lipid lowering properties. Several reports have investigated the effects of these drugs on inflammatory markers, including CD40L. Particularly, they have been shown to reduce cytokine-induced CD40L expression on endothelial cells, smooth muscle cells and macrophages.[102] Notably, atorvastatin treatment in the MIRACL trial reduces the risk of recurrent cardiovascular events, which are associated with sCD40L levels.[95]

Most of these agents indirectly target the CD40L system, perhaps through inhibition of platelet activation and the subsequent release of sCD40L. Specific disruption of CD40L or its receptors remains a promising approach for the treatment of atherothrombosis. Although clinical studies using anti-CD40L antibodies have been unsuccessful, alternative targets of this system may render better clinical outcomes. For example, novel anti-CD40L agents that specifically target the interaction of CD40L with its different receptors or inhibition of critical intracellular signaling elements, such as TRAFs, represent valuable approaches.

6. Conclusions

Research over the years overwhelmingly supports the notion of atherothrombosis as a chronic inflammatory disease. Despite the plethora of inflammatory mediators identified thus far as potential contributors to this complication, the CD40L system has attracted a great deal of interest. Besides its pivotal role in humoral immunity, CD40L is now regarded as a key player to all major phases of atherothrombosis, a concept supported in part by the strong relationship between its circulating soluble levels and the occurrence of cardiovascular diseases. In addition to its well-established CD40 counterreceptor, CD40L can also interact with novel binding partners, namely the integrin receptors $\alpha_{IIb}\beta_3$, $\alpha_M\beta_2$ and $\alpha_5\beta_1$. Although most CD40L-mediated functions have been attributed to its interaction with CD40, these novel receptors add complexity to the diverse interplays that might take place during inflammation. The elucidation of the exact physiopathological relevance of these interactions in inflammatory disorders might pave the way for the development of novel anti-CD40L therapeutic targets for the treatment of atherothrombosis and cardiovascular diseases.

7. References

[1] Hansson GK, Libby P. The immune response in atherosclerosis: A double-edged sword. *Nat Rev Immunol*. 2006;6:508-519

[2] Ross R. Atherosclerosis--an inflammatory disease. *N Engl J Med*. 1999;340:115-126

[3] Lusis AJ. Atherosclerosis. *Nature*. 2000;407:233-241

[4] Weber C, Zernecke A, Libby P. The multifaceted contributions of leukocyte subsets to atherosclerosis: Lessons from mouse models. *Nat Rev Immunol*. 2008;8:802-815

[5] Lievens D, Eijgelaar WJ, Biessen EA, Daemen MJ, Lutgens E. The multi-functionality of cd40l and its receptor cd40 in atherosclerosis. *Thromb Haemost*. 2009;102:206-214

[6] Schonbeck U, Libby P. Cd40 signaling and plaque instability. *Circ Res*. 2001;89:1092-1103

[7] Hassan GS, Merhi Y, Mourad WM. Cd154 and its receptors in inflammatory vascular pathologies. *Trends Immunol*. 2009;30:165-172

[8] Lutgens E, Gorelik L, Daemen MJ, de Muinck ED, Grewal IS, Koteliansky VE, Flavell RA. Requirement for cd154 in the progression of atherosclerosis. *Nat Med*. 1999;5:1313-1316

[9] Schonbeck U, Sukhova GK, Shimizu K, Mach F, Libby P. Inhibition of cd40 signaling limits evolution of established atherosclerosis in mice. *Proc Natl Acad Sci U S A*. 2000;97:7458-7463

[10] Davis JC, Jr., Totoritis MC, Rosenberg J, Sklenar TA, Wofsy D. Phase i clinical trial of a monoclonal antibody against cd40-ligand (idec-131) in patients with systemic lupus erythematosus. *J Rheumatol*. 2001;28:95-101

[11] Schuler W, Bigaud M, Brinkmann V, Di Padova F, Geisse S, Gram H, Hungerford V, Kleuser B, Kristofic C, Menninger K, Tees R, Wieczorek G, Wilt C, Wioland C, Zurini M. Efficacy and safety of abi793, a novel human anti-human cd154 monoclonal antibody, in cynomolgus monkey renal allotransplantation. *Transplantation*. 2004;77:717-726

[12] Armitage RJ, Fanslow WC, Strockbine L, Sato TA, Clifford KN, Macduff BM, Anderson DM, Gimpel SD, Davis-Smith T, Maliszewski CR, et al. Molecular and biological characterization of a murine ligand for cd40. *Nature*. 1992;357:80-82

[13] Lederman S, Yellin MJ, Krichevsky A, Belko J, Lee JJ, Chess L. Identification of a novel surface protein on activated cd4+ t cells that induces contact-dependent b cell differentiation (help). *J Exp Med*. 1992;175:1091-1101

[14] Kroczek RA, Graf D, Brugnoni D, Giliani S, Korthuer U, Ugazio A, Senger G, Mages HW, Villa A, Notarangelo LD. Defective expression of cd40 ligand on t cells causes "x-linked immunodeficiency with hyper-igm (higm1)". *Immunol Rev*. 1994;138:39-59

[15] Hill A, Chapel H. X-linked immunodeficiency. The fruits of cooperation. *Nature*. 1993;361:494

[16] Henn V, Slupsky JR, Grafe M, Anagnostopoulos I, Forster R, Muller-Berghaus G, Kroczek RA. Cd40 ligand on activated platelets triggers an inflammatory reaction of endothelial cells. *Nature*. 1998;391:591-594

[17] Karmann K, Hughes CC, Schechner J, Fanslow WC, Pober JS. Cd40 on human endothelial cells: Inducibility by cytokines and functional regulation of adhesion molecule expression. *Proc Natl Acad Sci U S A*. 1995;92:4342-4346

[18] Mach F, Schonbeck U, Fabunmi RP, Murphy C, Atkinson E, Bonnefoy JY, Graber P, Libby P. T lymphocytes induce endothelial cell matrix metalloproteinase expression by a cd40l-dependent mechanism: Implications for tubule formation. *Am J Pathol*. 1999;154:229-238

[19] Schonbeck U, Mach F, Sukhova GK, Herman M, Graber P, Kehry MR, Libby P. Cd40 ligation induces tissue factor expression in human vascular smooth muscle cells. *Am J Pathol*. 2000;156:7-14

[20] Garlichs CD, John S, Schmeisser A, Eskafi S, Stumpf C, Karl M, Goppelt-Struebe M, Schmieder R, Daniel WG. Upregulation of cd40 and cd40 ligand (cd154) in patients with moderate hypercholesterolemia. *Circulation*. 2001;104:2395-2400

[21] Heeschen C, Dimmeler S, Hamm CW, van den Brand MJ, Boersma E, Zeiher AM, Simoons ML. Soluble cd40 ligand in acute coronary syndromes. *N Engl J Med*. 2003;348:1104-1111

[22] Sanguigni V, Pignatelli P, Lenti L, Ferro D, Bellia A, Carnevale R, Tesauro M, Sorge R, Lauro R, Violi F. Short-term treatment with atorvastatin reduces platelet cd40 ligand and thrombin generation in hypercholesterolemic patients. *Circulation*. 2005;111:412-419

[23] Schonbeck U, Libby P. The cd40/cd154 receptor/ligand dyad. *Cell Mol Life Sci*. 2001;58:4-43

[24] Bishop GA. The multifaceted roles of trafs in the regulation of b-cell function. *Nat Rev Immunol*. 2004;4:775-786

[25] Ma DY, Clark EA. The role of cd40 and cd154/cd40l in dendritic cells. *Semin Immunol*. 2009;21:265-272

[26] Dallman C, Johnson PW, Packham G. Differential regulation of cell survival by cd40. *Apoptosis*. 2003;8:45-53

[27] Yacoub D, Hachem A, Theoret JF, Gillis MA, Mourad W, Merhi Y. Enhanced levels of soluble cd40 ligand exacerbate platelet aggregation and thrombus formation through a cd40-dependent tumor necrosis factor receptor-associated factor-2/rac1/p38 mitogen-activated protein kinase signaling pathway. *Arterioscler Thromb Vasc Biol*. 2010;30:2424-2433

[28] Chakrabarti S, Varghese S, Vitseva O, Tanriverdi K, Freedman JE. Cd40 ligand influences platelet release of reactive oxygen intermediates. *Arterioscler Thromb Vasc Biol*. 2005;25:2428-2434

[29] Danese S, de la Motte C, Reyes BM, Sans M, Levine AD, Fiocchi C. Cutting edge: T cells trigger cd40-dependent platelet activation and granular rantes release: A novel pathway for immune response amplification. *J Immunol*. 2004;172:2011-2015

[30] Bishop GA, Moore CR, Xie P, Stunz LL, Kraus ZJ. Traf proteins in cd40 signaling. *Adv Exp Med Biol*. 2007;597:131-151

[31] Pullen SS, Miller HG, Everdeen DS, Dang TT, Crute JJ, Kehry MR. Cd40-tumor necrosis factor receptor-associated factor (traf) interactions: Regulation of cd40 signaling through multiple traf binding sites and traf hetero-oligomerization. *Biochemistry*. 1998;37:11836-11845

[32] Arron JR, Pewzner-Jung Y, Walsh MC, Kobayashi T, Choi Y. Regulation of the subcellular localization of tumor necrosis factor receptor-associated factor (traf)2 by traf1 reveals mechanisms of traf2 signaling. *J Exp Med*. 2002;196:923-934

[33] Xie P, Hostager BS, Munroe ME, Moore CR, Bishop GA. Cooperation between tnf receptor-associated factors 1 and 2 in cd40 signaling. *J Immunol*. 2006;176:5388-5400

[34] McWhirter SM, Pullen SS, Holton JM, Crute JJ, Kehry MR, Alber T. Crystallographic analysis of cd40 recognition and signaling by human traf2. *Proc Natl Acad Sci U S A*. 1999;96:8408-8413

[35] Yeh WC, Shahinian A, Speiser D, Kraunus J, Billia F, Wakeham A, de la Pompa JL, Ferrick D, Hum B, Iscove N, Ohashi P, Rothe M, Goeddel DV, Mak TW. Early lethality, functional nf-kappab activation, and increased sensitivity to tnf-induced cell death in traf2-deficient mice. *Immunity*. 1997;7:715-725

[36] Hsing Y, Hostager BS, Bishop GA. Characterization of cd40 signaling determinants regulating nuclear factor-kappa b activation in b lymphocytes. *J Immunol*. 1997;159:4898-4906

[37] Rothe M, Sarma V, Dixit VM, Goeddel DV. Traf2-mediated activation of nf-kappa b by tnf receptor 2 and cd40. *Science*. 1995;269:1424-1427

[38] Vallabhapurapu S, Matsuzawa A, Zhang W, Tseng PH, Keats JJ, Wang H, Vignali DA, Bergsagel PL, Karin M. Nonredundant and complementary functions of traf2 and traf3 in a ubiquitination cascade that activates nik-dependent alternative nf-kappab signaling. *Nat Immunol*. 2008;9:1364-1370

[39] He JQ, Oganesyan G, Saha SK, Zarnegar B, Cheng G. Traf3 and its biological function. *Adv Exp Med Biol*. 2007;597:48-59

[40] Zarnegar BJ, Wang Y, Mahoney DJ, Dempsey PW, Cheung HH, He J, Shiba T, Yang X, Yeh WC, Mak TW, Korneluk RG, Cheng G. Noncanonical nf-kappab activation requires coordinated assembly of a regulatory complex of the adaptors ciap1, ciap2, traf2 and traf3 and the kinase nik. *Nat Immunol*. 2008;9:1371-1378

[41] Vince JE, Wong WW, Khan N, Feltham R, Chau D, Ahmed AU, Benetatos CA, Chunduru SK, Condon SM, McKinlay M, Brink R, Leverkus M, Tergaonkar V, Schneider P, Callus BA, Koentgen F, Vaux DL, Silke J. Iap antagonists target ciap1 to induce tnfalpha-dependent apoptosis. *Cell*. 2007;131:682-693

[42] Hauer J, Puschner S, Ramakrishnan P, Simon U, Bongers M, Federle C, Engelmann H. Tnf receptor (tnfr)-associated factor (traf) 3 serves as an inhibitor of traf2/5-

mediated activation of the noncanonical nf-kappab pathway by traf-binding tnfrs. *Proc Natl Acad Sci U S A*. 2005;102:2874-2879

[43] Nakano H, Sakon S, Koseki H, Takemori T, Tada K, Matsumoto M, Munechika E, Sakai T, Shirasawa T, Akiba H, Kobata T, Santee SM, Ware CF, Rennert PD, Taniguchi M, Yagita H, Okumura K. Targeted disruption of traf5 gene causes defects in cd40- and cd27-mediated lymphocyte activation. *Proc Natl Acad Sci U S A*. 1999;96:9803-9808

[44] Davies CC, Mak TW, Young LS, Eliopoulos AG. Traf6 is required for traf2-dependent cd40 signal transduction in nonhemopoietic cells. *Mol Cell Biol*. 2005;25:9806-9819

[45] Rowland SL, Tremblay MM, Ellison JM, Stunz LL, Bishop GA, Hostager BS. A novel mechanism for tnfr-associated factor 6-dependent cd40 signaling. *J Immunol*. 2007;179:4645-4653

[46] Donners MM, Beckers L, Lievens D, Munnix I, Heemskerk J, Janssen BJ, Wijnands E, Cleutjens J, Zernecke A, Weber C, Ahonen CL, Benbow U, Newby AC, Noelle RJ, Daemen MJ, Lutgens E. The cd40-traf6 axis is the key regulator of the cd40/cd40l system in neointima formation and arterial remodeling. *Blood*. 2008;111:4596-4604

[47] Parise LV. Integrin alpha(iib)beta(3) signaling in platelet adhesion and aggregation. *Curr Opin Cell Biol*. 1999;11:597-601

[48] Andre P, Prasad KS, Denis CV, He M, Papalia JM, Hynes RO, Phillips DR, Wagner DD. Cd40l stabilizes arterial thrombi by a beta3 integrin--dependent mechanism. *Nat Med*. 2002;8:247-252

[49] Prasad KS, Andre P, He M, Bao M, Manganello J, Phillips DR. Soluble cd40 ligand induces beta3 integrin tyrosine phosphorylation and triggers platelet activation by outside-in signaling. *Proc Natl Acad Sci U S A*. 2003;100:12367-12371

[50] Leveille C, Bouillon M, Guo W, Bolduc J, Sharif-Askari E, El-Fakhry Y, Reyes-Moreno C, Lapointe R, Merhi Y, Wilkins JA, Mourad W. Cd40 ligand binds to alpha5beta1 integrin and triggers cell signaling. *J Biol Chem*. 2007;282:5143-5151

[51] Sigal A, Bleijs DA, Grabovsky V, van Vliet SJ, Dwir O, Figdor CG, van Kooyk Y, Alon R. The lfa-1 integrin supports rolling adhesions on icam-1 under physiological shear flow in a permissive cellular environment. *J Immunol*. 2000;165:442-452

[52] Zirlik A, Maier C, Gerdes N, MacFarlane L, Soosairajah J, Bavendiek U, Ahrens I, Ernst S, Bassler N, Missiou A, Patko Z, Aikawa M, Schonbeck U, Bode C, Libby P, Peter K. Cd40 ligand mediates inflammation independently of cd40 by interaction with mac-1. *Circulation*. 2007;115:1571-1580

[53] Mach F, Schonbeck U, Sukhova GK, Atkinson E, Libby P. Reduction of atherosclerosis in mice by inhibition of cd40 signalling. *Nature*. 1998;394:200-203

[54] Lutgens E, Cleutjens KB, Heeneman S, Koteliansky VE, Burkly LC, Daemen MJ. Both early and delayed anti-cd40l antibody treatment induces a stable plaque phenotype. *Proc Natl Acad Sci U S A*. 2000;97:7464-7469

[55] Aikawa M, Voglic SJ, Sugiyama S, Rabkin E, Taubman MB, Fallon JT, Libby P. Dietary lipid lowering reduces tissue factor expression in rabbit atheroma. *Circulation*. 1999;100:1215-1222

[56] Hakkinen T, Karkola K, Yla-Herttuala S. Macrophages, smooth muscle cells, endothelial cells, and t-cells express cd40 and cd40l in fatty streaks and more advanced human atherosclerotic lesions. Colocalization with epitopes of oxidized

low-density lipoprotein, scavenger receptor, and cd16 (fc gammariii). *Virchows Arch*. 2000;437:396-405

[57] Ruedl C, Bachmann MF, Kopf M. The antigen dose determines t helper subset development by regulation of cd40 ligand. *Eur J Immunol*. 2000;30:2056-2064

[58] Li D, Liu L, Chen H, Sawamura T, Mehta JL. Lox-1, an oxidized ldl endothelial receptor, induces cd40/cd40l signaling in human coronary artery endothelial cells. *Arterioscler Thromb Vasc Biol*. 2003;23:816-821

[59] Lutgens E, Lievens D, Beckers L, Donners M, Daemen M. Cd40 and its ligand in atherosclerosis. *Trends Cardiovasc Med*. 2007;17:118-123

[60] Omari KM, Chui R, Dorovini-Zis K. Induction of beta-chemokine secretion by human brain microvessel endothelial cells via cd40/cd40l interactions. *J Neuroimmunol*. 2004;146:203-208

[61] Pluvinet R, Olivar R, Krupinski J, Herrero-Fresneda I, Luque A, Torras J, Cruzado JM, Grinyo JM, Sumoy L, Aran JM. Cd40: An upstream master switch for endothelial cell activation uncovered by rnai-coupled transcriptional profiling. *Blood*. 2008;112:3624-3637

[62] Thienel U, Loike J, Yellin MJ. Cd154 (cd40l) induces human endothelial cell chemokine production and migration of leukocyte subsets. *Cell Immunol*. 1999;198:87-95

[63] Huo Y, Schober A, Forlow SB, Smith DF, Hyman MC, Jung S, Littman DR, Weber C, Ley K. Circulating activated platelets exacerbate atherosclerosis in mice deficient in apolipoprotein e. *Nat Med*. 2003;9:61-67

[64] Weber C. Platelets and chemokines in atherosclerosis: Partners in crime. *Circ Res*. 2005;96:612-616

[65] Yeo EL, Sheppard JA, Feuerstein IA. Role of p-selectin and leukocyte activation in polymorphonuclear cell adhesion to surface adherent activated platelets under physiologic shear conditions (an injury vessel wall model). *Blood*. 1994;83:2498-2507

[66] Horkko S, Binder CJ, Shaw PX, Chang MK, Silverman G, Palinski W, Witztum JL. Immunological responses to oxidized ldl. *Free Radic Biol Med*. 2000;28:1771-1779

[67] Lutgens E, Daemen MJ. Cd40-cd40l interactions in atherosclerosis. *Trends Cardiovasc Med*. 2002;12:27-32

[68] Mukundan L, Milhorn DM, Matta B, Suttles J. Cd40-mediated activation of vascular smooth muscle cell chemokine production through a src-initiated, mapk-dependent pathway. *Cell Signal*. 2004;16:375-384

[69] Xu F, Ji J, Li L, Chen R, Hu W. Activation of adventitial fibroblasts contributes to the early development of atherosclerosis: A novel hypothesis that complements the "response-to-injury hypothesis" and the "inflammation hypothesis". *Med Hypotheses*. 2007;69:908-912

[70] Springer TA. Adhesion receptors of the immune system. *Nature*. 1990;346:425-434

[71] Yellin MJ, Winikoff S, Fortune SM, Baum D, Crow MK, Lederman S, Chess L. Ligation of cd40 on fibroblasts induces cd54 (icam-1) and cd106 (vcam-1) up-regulation and il-6 production and proliferation. *J Leukoc Biol*. 1995;58:209-216

[72] Danese S, Scaldaferri F, Vetrano S, Stefanelli T, Graziani C, Repici A, Ricci R, Straface G, Sgambato A, Malesci A, Fiocchi C, Rutella S. Critical role of the cd40 cd40-ligand pathway in regulating mucosal inflammation-driven angiogenesis in inflammatory bowel disease. *Gut*. 2007;56:1248-1256

[73] Kawai M, Masuda A, Kuwana M. A cd40-cd154 interaction in tissue fibrosis. *Arthritis Rheum*. 2008;58:3562-3573

[74] Melter M, Reinders ME, Sho M, Pal S, Geehan C, Denton MD, Mukhopadhyay D, Briscoe DM. Ligation of cd40 induces the expression of vascular endothelial growth factor by endothelial cells and monocytes and promotes angiogenesis in vivo. *Blood*. 2000;96:3801-3808

[75] Reinders ME, Sho M, Robertson SW, Geehan CS, Briscoe DM. Proangiogenic function of cd40 ligand-cd40 interactions. *J Immunol*. 2003;171:1534-1541

[76] Russo S, Bussolati B, Deambrosis I, Mariano F, Camussi G. Platelet-activating factor mediates cd40-dependent angiogenesis and endothelial-smooth muscle cell interaction. *J Immunol*. 2003;171:5489-5497

[77] Collo G, Pepper MS. Endothelial cell integrin alpha5beta1 expression is modulated by cytokines and during migration in vitro. *J Cell Sci*. 1999;112 (Pt 4):569-578

[78] Croce K, Libby P. Intertwining of thrombosis and inflammation in atherosclerosis. *Curr Opin Hematol*. 2007;14:55-61

[79] Libby P. Current concepts of the pathogenesis of the acute coronary syndromes. *Circulation*. 2001;104:365-372

[80] Mach F, Schonbeck U, Bonnefoy JY, Pober JS, Libby P. Activation of monocyte/macrophage functions related to acute atheroma complication by ligation of cd40: Induction of collagenase, stromelysin, and tissue factor. *Circulation*. 1997;96:396-399

[81] Schonbeck U, Mach F, Sukhova GK, Murphy C, Bonnefoy JY, Fabunmi RP, Libby P. Regulation of matrix metalloproteinase expression in human vascular smooth muscle cells by t lymphocytes: A role for cd40 signaling in plaque rupture? *Circ Res*. 1997;81:448-454

[82] May AE, Kalsch T, Massberg S, Herouy Y, Schmidt R, Gawaz M. Engagement of glycoprotein iib/iiia (alpha(iib)beta3) on platelets upregulates cd40l and triggers cd40l-dependent matrix degradation by endothelial cells. *Circulation*. 2002;106:2111-2117

[83] Stary HC. The sequence of cell and matrix changes in atherosclerotic lesions of coronary arteries in the first forty years of life. *Eur Heart J*. 1990;11 Suppl E:3-19

[84] Sukhova GK, Schonbeck U, Rabkin E, Schoen FJ, Poole AR, Billinghurst RC, Libby P. Evidence for increased collagenolysis by interstitial collagenases-1 and -3 in vulnerable human atheromatous plaques. *Circulation*. 1999;99:2503-2509

[85] Miller DL, Yaron R, Yellin MJ. Cd40l-cd40 interactions regulate endothelial cell surface tissue factor and thrombomodulin expression. *J Leukoc Biol*. 1998;63:373-379

[86] Pradier O, Willems F, Abramowicz D, Schandene L, de Boer M, Thielemans K, Capel P, Goldman M. Cd40 engagement induces monocyte procoagulant activity through an interleukin-10 resistant pathway. *Eur J Immunol*. 1996;26:3048-3054

[87] Zhou L, Stordeur P, de Lavareille A, Thielemans K, Capel P, Goldman M, Pradier O. Cd40 engagement on endothelial cells promotes tissue factor-dependent procoagulant activity. *Thromb Haemost*. 1998;79:1025-1028

[88] Antoniades C, Tousoulis D, Vasiliadou C, Stefanadi E, Marinou K, Stefanadis C. Genetic polymorphisms of platelet glycoprotein ia and the risk for premature myocardial infarction: Effects on the release of scd40l during the acute phase of premature myocardial infarction. *J Am Coll Cardiol*. 2006;47:1959-1966

[89] Aukrust P, Muller F, Ueland T, Berget T, Aaser E, Brunsvig A, Solum NO, Forfang K, Froland SS, Gullestad L. Enhanced levels of soluble and membrane-bound cd40 ligand in patients with unstable angina. Possible reflection of t lymphocyte and platelet involvement in the pathogenesis of acute coronary syndromes. *Circulation*. 1999;100:614-620

[90] Garlichs CD, Eskafi S, Raaz D, Schmidt A, Ludwig J, Herrmann M, Klinghammer L, Daniel WG, Schmeisser A. Patients with acute coronary syndromes express enhanced cd40 ligand/cd154 on platelets. *Heart*. 2001;86:649-655

[91] Tayebjee MH, Lip GY, Tan KT, Patel JV, Hughes EA, MacFadyen RJ. Plasma matrix metalloproteinase-9, tissue inhibitor of metalloproteinase-2, and cd40 ligand levels in patients with stable coronary artery disease. *Am J Cardiol*. 2005;96:339-345

[92] Tousoulis D, Antoniades C, Nikolopoulou A, Koniari K, Vasiliadou C, Marinou K, Koumallos N, Papageorgiou N, Stefanadi E, Siasos G, Stefanadis C. Interaction between cytokines and scd40l in patients with stable and unstable coronary syndromes. *Eur J Clin Invest*. 2007;37:623-628

[93] Wang Y, Li L, Tan HW, Yu GS, Ma ZY, Zhao YX, Zhang Y. Transcoronary concentration gradient of scd40l and hscrp in patients with coronary heart disease. *Clin Cardiol*. 2007;30:86-91

[94] Ohashi Y, Kawashima S, Mori T, Terashima M, Ichikawa S, Ejiri J, Awano K. Soluble cd40 ligand and interleukin-6 in the coronary circulation after acute myocardial infarction. *Int J Cardiol*. 2006;112:52-58

[95] Kinlay S, Schwartz GG, Olsson AG, Rifai N, Sasiela WJ, Szarek M, Ganz P, Libby P. Effect of atorvastatin on risk of recurrent cardiovascular events after an acute coronary syndrome associated with high soluble cd40 ligand in the myocardial ischemia reduction with aggressive cholesterol lowering (miracl) study. *Circulation*. 2004;110:386-391

[96] Malarstig A, Lindahl B, Wallentin L, Siegbahn A. Soluble cd40l levels are regulated by the -3459 a>g polymorphism and predict myocardial infarction and the efficacy of antithrombotic treatment in non-st elevation acute coronary syndrome. *Arterioscler Thromb Vasc Biol*. 2006;26:1667-1673

[97] Langer F, Ingersoll SB, Amirkhosravi A, Meyer T, Siddiqui FA, Ahmad S, Walker JM, Amaya M, Desai H, Francis JL. The role of cd40 in cd40l- and antibody-mediated platelet activation. *Thromb Haemost*. 2005;93:1137-1146

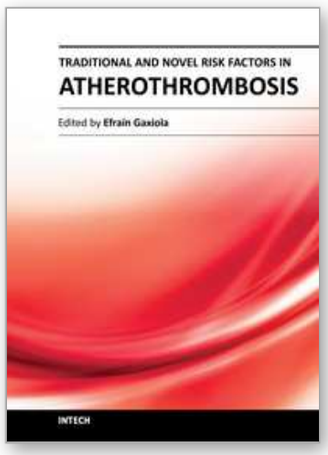
[98] Kawai T, Andrews D, Colvin RB, Sachs DH, Cosimi AB. Thromboembolic complications after treatment with monoclonal antibody against cd40 ligand. *Nat Med*. 2000;6:114

[99] Yip HK, Chang LT, Sun CK, Yang CH, Hung WC, Cheng CI, Chua S, Yeh KH, Wu CJ, Fu M. Impact of clopidogrel on suppression of circulating levels of soluble cd40 ligand in patients with unstable angina undergoing coronary stenting. *Am J Cardiol*. 2006;97:192-194

[100] Azar RR, Kassab R, Zoghbi A, Aboujaoude S, El-Osta H, Ghorra P, Germanos M, Salame E. Effects of clopidogrel on soluble cd40 ligand and on high-sensitivity c-reactive protein in patients with stable coronary artery disease. *Am Heart J*. 2006;151:521 e521-521 e524

[101] Nannizzi-Alaimo L, Alves VL, Phillips DR. Inhibitory effects of glycoprotein iib/iiia antagonists and aspirin on the release of soluble cd40 ligand during platelet stimulation. *Circulation*. 2003;107:1123-1128

[102] Schonbeck U, Gerdes N, Varo N, Reynolds RS, Horton DB, Bavendiek U, Robbie L, Ganz P, Kinlay S, Libby P. Oxidized low-density lipoprotein augments and 3-hydroxy-3-methylglutaryl coenzyme a reductase inhibitors limit cd40 and cd40l expression in human vascular cells. *Circulation*. 2002;106:2888-2893



Traditional and Novel Risk Factors in Atherothrombosis

Edited by Dr. Efrain Gaxiola

ISBN 978-953-51-0561-9
Hard cover, 140 pages
Publisher InTech
Published online 20, April, 2012
Published in print edition April, 2012

Atherothrombosis has reached pandemic proportions worldwide. It is the underlying condition that results in events leading to myocardial infarction, ischemic stroke and vascular death. As such, it is the leading cause of death worldwide manifested mainly as cardiovascular/cerebrovascular death. The complex and intimate relationship between atherothrombosis and traditional and novel risk factors is discussed in the following chapters of Traditional and Novel Risk Factors in Atherothrombosis - from basic science to clinical and therapeutic concerns. Beginning with pathology and pathophysiology of atherothrombosis, plaque rupture/disruption, this book continues with molecular, biochemical, inflammatory, cellular aspects and finally analyzes several aspects of clinical pharmacology.

InTech Europe
University Campus STeP Ri
Slavka Krautzeka 83/A
51000 Rijeka, Croatia
Phone: +385 (51) 770 447
Fax: +385 (51) 686 166
www.intechopen.com

InTech China
Unit 405, Office Block, Hotel Equatorial Shanghai
No.65, Yan An Road (West), Shanghai, 200040, China
中国上海市延安西路65号上海国际贵都大饭店办公楼405单元
Phone: +86-21-62489820
Fax: +86-21-62489821

IntechOpen