Inflammatory mechanisms in atherosclerosis

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Summary
Throughout the last two decades inflammation has been recognized as the central mechanism underlying atherogenesis. A multitude of basic science work demonstrates the pivotal role of inflammatory processes during every step of atherosclerotic plaque formation: From initiation via propagation to complication.

This review describes some of the key mechanisms involved with a particular focus on the diverse group of inflammatory cells and their subsets that distinctly contribute to atherogenic and anti-atherogenic phenomena. Furthermore, we summarize the controlling action of a tight network of co-stimulatory molecules and cytokines orchestrating the inflammatory and anti-inflammatory effector functions. Finally, the current status of clinical trials evaluating anti-inflammatory/immune-modulatory treatment strategies is summarized and an outlook for future therapeutic implications is provided.

Inflammation
The link between traditional risk factors and atherogenesis
Cardiovascular disease and its complications represent the leading cause of mortality in the western hemisphere (1).

Particularly, the metabolic syndrome comprising a cluster of risk factors including abdominal obesity, dyslipidaemia, arterial hypertension, and insulin resistance highly predispenses for cardiovascular complications (2). Obese adipose tissue mainly composed of adipocytes and M1-macrophages functions not only as an energy carrier, but also as an endocrine organ expressing various pro-atherogenic adipocytokines such as tumour necrosis factor alpha (TNFα), interleukin 6 (IL-6), IL-1β, and monocyte chemotactic protein (MCP-1) (3, 4). Furthermore, upon the onset of obesity LDL-cholesterol accumulates, which itself is pro-inflammatory and atherogenic (5).

Arterial hypertension on the other hand can damage the arterial wall in areas of disturbed flow and can induce activation of endothelial cells fostering the nascence and growth of plaques. It is known since the beginning of the 20th century that hyperglycaemia induces non-enzymatic transformation of proteins and lipoproteins within the arterial wall forming advanced glycolyzation end products (AGEs).

- AGEs accumulate in the vessel wall and induce the expression of TNFα, IL-1β, and adhesion molecules (6).
- Finally, cigarette smoking increases production of a variety of pro-atherogenic cytokines including TNFα, IL-1β, and IL-8 (7, 8).

Control and promotion of every stage of atherogenesis
Endothelial activation precedes plaque formation
Endothelial cells are permanently exposed to the blood. A crucial step in the initiation of atherosclerosis is the accumulation of low-density lipoprotein (LDL) cholesterol in the intima layer (2). The exact mechanisms, how LDL enters the arterial wall e.g.
Once LDL particles are located in the intimal layer they undergo modification by exposure to reactive oxygen species, myeloperoxidase, and lipoperoxidase.

- The resulting oxidized LDL particles (oxLDL) activate the innate immune system via pattern recognition receptors (PPR).

- Along with several other factors such as shear stress and various cytokines, OxLDL also enhances endothelial expression of adhesion molecules such as E-selectin and vascular cell adhesion molecule 1 (VCAM-1).

- Furthermore, the chemokines MCP-1, IL-8, RANTES, CXCL10, and CX3CL1 are produced to attract leukocytes such as monocytes, T- cells, and dendritic cells to the arterial wall.

These events result in the nascent of early atherosclerotic lesions, so-called “fatty streaks.”

**Inflammatory cell recruitment drives plaque formation and growth**

Rudolf Virchow firstly described atherosclerosis as an inflammatory driven disease 150 years ago. However, the mechanisms underlying inflammatory cell infiltration only emerged during the preceding two decades.

- Activated endothelial cells expressing E- and P-selectin bind leukocytes via interaction with P-selectin glycoprotein ligand-1 (PSGL1) resulting in leukocyte rolling.

- Under the influence of chemokines such as RANTES, MCP-1, and CXCL5, the rolling velocity slows down.

- This is supported by integrin-cell adhesion molecule interaction, a process ultimately mediating firm adhesion through binding between very late antigen 4 (VLA4) and VCAM1 as well as between lymphocyte function-associated antigen 1 (LFA1) and ICAM1.

- After firm adhesion, leukocytes spread, crawl along the endothelium, and migrate into the subendothelial space.

- Endothelial and smooth muscle cells produce macrophage colony stimulating factor (M-CSF) and promote transformation of monocytes to macrophages.

- Macrophages take up oxLDL via scavenger receptors such as scavenger receptor A (SR-A), SR-B1, CD36, or lectin-type oxLDL-receptor-1 (LOX-1) forming foam cells, which present oxLDL fragments to T-cells via MHCII.

The pro-inflammatory milieu comprising various cytokines and pathogen-associated molecular patterns (PAMPs) further activates macrophages and foam cells fostering the expression of pro-inflammatory cytokines and chemokines.

About 40% of cells within atherosclerotic lesions are macrophages, about 10% CD3+ T-cells. Antigen presenting cells (APCs) such as macrophages and dendritic cells activate T-cells resulting in the expression of mostly Th1-cytokines such as Interferon gamma (IFNγ), IL-12, IL-15, IL-18, and TNFα, a process that perpetuates the inflammatory response.

**Inflammation determines plaque fate and provokes complications**

In the core of the atherosclerotic lesion cholesterol and cell debris from apoptotic cells gather and form the necrotic core. It is highly procoagulant and protected from the blood flow by a fibrous cap consisting of endothelial cells and smooth muscle cells. Under the influence of prolonged inflammation metalloproteinases (MMP) such as MMP1, MMP8, and MMP13 are released within the plaque and start degrading collagen, ultimately compromising the integrity of the fibrous cap.

Upon rupture of the fibrous cap the necrotic core is exposed to the blood stream, tissue factor (TF) activates thrombin, and thus platelet aggregation is initiated.

These events lead to subtotal or total vessel occlusion, the pathophysiologic correlate of the clinical picture of an acute coronary syndrome (ACS).

Thus the degree of plaque inflammation determines its vulnerability: The more inflammatory cells and lipids accumulate the more prone plaque is to rupture.

In contrast, increased content of smooth muscle cells and collagen are associated with stability. The key steps of atherogenesis are simplified (Fig. 1).

**The role of inflammatory cells and their subsets in atherosclerosis**

**Monocytes, macrophages**

Monocytes circulate in the blood stream and can be directed to sites of inflammation. Initiation and progression of atherosclerotic disease depend on the constant influx of monocytes. Monocytes do not necessarily derive from the same splenic reservoir driven by extra-skeletal monocytes such as monocytes that itself give rise to lipid-loaded foam cells.

Alternatively, monocytes can be precursor for dendritic cells in the atherosclerotic plaque. Mouse monocytes can be distinguished by their surface expression of Ly6C/Gr-1: Gr-1<sub>hi</sub>/Ly6C<sub>hi</sub> monocytes are termed inflammatory monocyte and are thought to represent the monocyte subset that is linked to acute inflammation, while Ly6C<sub>low</sub> monocytes may participate in resolution of inflammation and display a wound-healing phenotype and patrolling behaviour in the un-diseased vessels.

Hypercholesterolaemia is associated with an increase in Ly6C<sub>hi</sub> monocytes in mice and Ly6C<sub>hi</sub> monocytes have been shown to preferentially migrate to the atherosclerotic plaque. Notably, Ly6C<sub>hi</sub> monocytes do not necessarily derive from bone marrow, but may also be recruited from a splenic reservoir driven by extra-skeletal haematopoiesis. Recruitment of monocytes to the plaque is linked to a network of chemokines and adhesive receptor-ligand pairs that are summarized in the leukocyte adhesion cascade.

Inhibition of such components limits monocyte recruitment and plaque development.
ment. However, unspecific blockade of adhesion, e.g. inhibition of the leukocyte integrin Mac-1 (CD11b/CD18) or its numerous ligands with blocking antibodies attenuates atherosclerosis (30), but gives rise to unwanted side effect. The latter can be circumvented by selective blocking strategies (31).

Lately, it was questioned whether development of lesional macrophages ultimately depends on the influx and differentiation of monocytes. Macrophages are not only present in mouse aortas before atherosclerosis initiates (32), but they also show the ability to proliferate in the aortic plaque as recently demonstrated by Robbins et al. (33). Macrophages have been linked to a broad repertoire of effector functions, including secretion of pro-inflammatory cytokines, lipid uptake and reverse cholesterol transport, and phagocytosis (34). Based on their exclusive gene expression macrophages can be further subdivided into the M1, M2, M4, and Mox phenotype, of which the pro-inflammatory M1 phenotype and the anti-inflammatory M2 phenotype are best characterized (35).

It is likely that both, pro- and anti-inflammatory macrophages reside in the plaque (36). Of note, depletion of cells expressing the leukocyte integrin Mac-1 (including macrophages, but also monocytes and neutrophils) in a model of diphtheria toxin receptor transgenic mice reduced plaque progression (37), supporting the hypothesis that the net effect of macrophages may be pro-atherogenic.

During uptake of lipids, macrophages turn into foam cells and eventually go into apoptosis. Cholesterol in- and efflux critically regulates foam cell formation. In line

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**Fig. 1** Atherosclerotic plaque formation, key events
(1) LDL cholesterol enters the arterial wall, is oxidized to oxLDL and activates endothelial cells (EC) expressing cell adhesion molecules.
(2) Monocytes roll over the endothelium, adhere supported by platelets to the ECs and enter the intima.
(3) Within the intima they transform to macrophages upon M-CSF stimulation.
(4) By intaking oxLDL macrophages form foam cells which produce pro-inflammatory cytokines.
(5) While the inflammation accelerates T-cells, B-cells and dendritic cells enter the plaque and regulate inflammation.
(6) Cell detritus, lipids and extracellular matrix form a necrotic core.
(7) Once the fibrous cap of collagen and smooth muscle cells is degraded by MMPs and ruptures, the necrotic core comes into contact to the blood steam, platelets are activated and the vessel is occluded.
with this concept it has been shown that genetic deficiency of the two regulators of reverse cholesterol transport, ATP-binding cassette transporter A1 (ABCA1) and G1 (ABCGL), aggravates foam cell formation and atherosclerotic lesion development (38). Macrophage apoptosis as ultimate result of foam cell formation is thought to fuel the inflammatory response and to directly promote formation of the necrotic core at later stages. In the advanced plaque, macrophages accumulate in the rupture-prone region and are likely to contribute to fibrous cap thinning and destabilization of the atherosclerotic plaque – a mechanism possibly mediated by secretion of matrix-mestabilizing matrix metalloproteinases (MMPs) (39).

T-cells

T cells can be detected in all stages of the atherosclerotic plaque. Based on their functional and descriptive properties, T cells can be sub-divided in CD4+ T-helper (Th) cells and CD8+ cytotoxic T cells. In the plaques, CD4+ T cells expressing the αβTCR receptor are most common (40). Naïve T-helper cells can undergo further differentiation into their effector subsets, including Th1, Th2, Threg or Th17 cells after contact with antigen presented by antigen presenting cells (APCs). In atherosclerosis most T cells are Th1 polarized expressing the pro-inflammatory cytokines INFγ, IL-2, IL-3, and TNFα and -β. These fuel plaque inflammation by stimulating macrophages and other cells resident in the plaque (40). INFγ as the Th1 key cytokine has been identified in the human atherosclerotic plaque and genetic inhibition of INFγ or its receptor as well as its master transcription factor T-bet abolished murine atherosclerosis (41–43).

In contrast, Th2 cells secrete anti-inflammatory cytokines such as IL-4, IL-5, and IL-10 inhibit the release of pro-inflammatory molecules. It is noteworthy that the role of Th2 cells in atherosclerosis has not been completely deciphered. Several reports indicated opposing pro- and anti-inflammatory roles for Th2 cells and their corresponding cytokines in atherosclerosis. For example, Th2 cells are found frequently in advanced atherosclerotic lesions (44), but inhibition of the Th2 cytokine IL-4 had no clear effect (45, 46). Equally inconsistent are reports of Th17 cells. IL-17 has been detected in atherosclerotic lesions (47) and some studies suggested a pro-atherogenic role (48, 49), while others failed to confirm these findings (50, 51).

Beyond Th effector cells also Th regulatory (Threg) cells act as anti-inflammatory immune cells. Threg are now defined by co-expression of CD4+ CD25+ and the forkhead transcription factor Foxp3 (52). Threg bear an anti-inflammatory, immune-suppressing phenotype by expression of IL-10 and TGFβ. The latter inhibits atherosclerosis in mouse models (53, 54). Transfer of CD4+ CD25+ Th cells share features with natural Threg demonstrated that Threg are atheroprotective. Consistently, another study recently found that specific depletion of Threg in a model of diphtheria toxin receptor under control of the Foxp3 promoter accelerated atherosclerosis (55). Temporal and spatial appearance, as well as distinct effector functions of T-cells has been extensively characterized, the events initiating T cell activation have been poorly understood. It has been proposed that a break of self tolerance and the recognition of specific antigens recognized by T-cells initiate a pro-atherogenic immune response (56) and antigens such as native and modified LDL, apoB-100 as main protein component of LDL, and heat-shock protein 60 (HSP60) were suggested as such immunogenic antigens (57–59). Furthermore, stimulation of T cells with LDL provoked expression of INFγ.

Recently, Koltsova et al. showed that CD4+ cells extensively interact with APCs causing T-cells to express IFNγ and TNFα. Notably, these cytokines promoted the uptake of modified LDL in macrophages (60). Thus, antigen presentation to CD4+ cells further corroborates the concept of specific antigen recognition and the role of adaptive immunity in atherosclerosis.

B-cells

Despite their early discovery in the atherosclerotic plaques, the role of B-cells in atherogenesis has long been under-estimated and only recently appreciated. Caligiuri et al. first reported that splenectomy in an ApoE-/- mouse model intensified atherosclerosis. Notably, adoptive transfer of splenic B cells from ApoE-/- mice reversed these effects and protected from atherogenesis (61), suggesting that B cells are athero-protective. Another study confirmed these findings by demonstrating that bone marrow transplantation of B cell deficient mice aggravated disease (62).

However, while some studies confirmed an athero-protective role of B cells in mice bearing non-functional B cells or B cell transfers in B cell-deficient mice (63, 64), other groups suggested B cells might be pro-atherogenic. Ait-Oufella et al. and Kyaw et al. consistently showed that B cell depletion with an anti-CD20 antibody protected from atherosclerosis (65, 66). Furthermore, global or bone marrow deficiency for the receptor of B cell activation factor (Baff), a factor required for B-2 B cell survival, showed smaller atherosclerotic lesions (67, 68). This remarkable repertoire of divergent B cell functions in atherogenesis gave rise to a model in which specific B cell subsets might account for independent effector functions. In line with this concept it is important to note that anti-CD20 treatment and deletion of Baff Receptor depleted B-2 B cells (among those range follicular and marginal zone B cells), but not B-1 B-cells. B-1 cells can be sub-divided in B-1a and B-1b cells and are – in contrast to follicular B cells – largely T-cell independent (69). They are considered to be part of the innate immune system and can be found in serosal cavities and the spleen. They are limited by natural selection and respond by secretion of IgM antibodies recognizing self-antigens.

IgMs have been implicated in atherosclerosis. Binder et al. demonstrated that immunization of mice with heat-inactivated S. pneumoniae antigen, which shares properties with oxidized LDL, increased IgM levels and diminished atherosclerosis (70). Also, mice with a deficiency in secreting IgM presented increased levels of atherosclerosis (64). These discoveries have led to a challenging model in which particular B cell subsets regulate the complex immune response in atherosclerosis. However, many questions remain unanswered. For example, it has not been demonstrated how IL-10 secreting B regulatory cells (Breg)
figure in atherosclerosis. IL-10 itself has athero-protective properties (53). Also, the lately discovered innate response activator (IRA) B cell can convert to a cell expressing granulocyte-macrophage colony-stimulating factor (GM-CSF). The latter induces generation of Ly6Chi monocytes in the spleen (71), a process that likely affects atherosclerosis. Also, typical B cell pathway associated kinases such as spleen tyrosin kinase (Syk) modulate atherosclerosis in vivo. We recently demonstrated that inhibition of Syk via the inhibitor fostamatinib attenuates atherosclerosis in LDLR-deficient mice (72).

**Dendritic cells**

As essential part of the adaptive and innate immune system, dendritic cells (DCs) are needed to present antigenic peptide-MHC II complexes to naive T cells to induce an antigen-specific immune reaction (73). In the context of atherosclerosis, networks of dendritic cells have been identified in the intima of healthy young individuals (74) and DCs are preferentially found in the atherosclerosis-prone regions of arteries (75).

Interestingly, the number of dendritic cells increases in advanced plaques and co-localize with T cells, indicating a specific, immune-driven interaction of both immune compartments (76).

Functional observations of explanted aortas further revealed dynamic and sustained interactions of DCs and T cells (60). Several subsets of DCs can be distinguished:

- conventional DCs,
- plasmacytoid DCs, and
- inflammatory DCs.

It has been proposed that inflammatory DCs – that are not found in the early plaque – may derive from Ly6Clow monocytes (73). This notion is further supported by the observation that genetic deficiency of the chemokine receptor CX3CR1 characterizing Ly6Clow limited the number of dendritic cell accumulation in the intima and ameliorated lesion development (77). It is thought that DCs can present (self) antigens to induce an adaptive immune response. Several studies pointed out that modified lipids such as oxLDL can stimulate DCs, up-regulate expression of co-stimulatory molecules and secrete T cell attracting chemokines (78). However, a direct functional role of dendritic cells in atherosclerosis has not been reported so far.

**Other cell types**

**Neutrophils**

Coronary heart disease is associated with increased leukocyte numbers in humans. Neutrophils have been identified as the main leukocyte subset contributing to this effect (79, 80). In line, levels of neutrophil myeloperoxidase are increased in human CHD (81). Neutrophil infiltration in the atherosclerotic plaque occurs in the early stages of disease (82) and during plaque rupture (83). Depletion of neutrophils in mice is anti-atherogenic (84), an effect possibly caused by neutrophil effector molecules, such as CRAMP (85). Wantha et al. recently proposed that deposition of CRAMP by neutrophils at the site of the endothelium promotes recruitment of inflammatory monocytes by activating the endothelium and its adhesive systems (86).

**Mast cells**

Mast cells are key players in inflammation, allergy, and host defense. Upon activation they release a variety of soluble mediators, such as interleukins, proteases, and growth factors. Mast cells have been detected in human atherosclerotic plaques (87) and based on the broad functionality of their mediators, many implications for atherosclerotic disease have been proposed, including plaque de-stabilization, lipid accumulation, and vascular remodeling (88).

Indeed, pharmacological inhibition of mast cell activation by cromoglicic acid protected from atherosclerosis. Relevant pro-atherogenic mast cell mediators include IL-6 and IFNγ as revealed by adoptive transfers of cytokine-deficient mast cells into mast cell-deficient mice (89, 90). The functional involvement of mast cells has raised the intriguing question of the role of allergic pathways in atherosclerosis (91). Notably, Wang at al. demonstrated that IgE antibodies and their receptor FceR1α are pro-atherogenic (92).

**Platelets**

In recent years, platelets have emerged as key players in regulating leukocyte recruitment to the growing and rupture-prone plaque. The current paradigm includes either platelet deposition directly at the site of the endothelium and subsequent firm adhesion of leukocytes on bound platelets or platelet pre-binding to circulating leukocytes and subsequent deposition of platelet-leukocyte aggregates.

In the classical view platelet deposition occurs only at the site of injured endothelium, e.g. at the stage of plaque rupture, to components of the extracellular matrix, including von Willebrand factor (VWF) and collagen by engagement of platelet adhesion factors glycoprotein Ib/IX/V (GPIb/IX/V) and GPVI, respectively. However, also intact, but inflamed endothelial cells can express P-selectin (CD62P), ICAM-1, or αβ3-integrin, which allow either direct binding of platelet expressed glycoprotein GPIb, PSGL-1, or by platelet αβ, to endothelial-bound fibrinogen and fibronectin. Moreover, various receptor-ligand interactions, such as from leukocyte integrin Mac-1 (CD11b/CD18) and PSGL-1 to platelet expressed GPIb and CD62P can mediate leukocyte/platelet complex formation (93). Accordingly, transplantation of P-selectin-deficient bone marrow reduced atherosclerosis in mice (94). In line, adoptive transfer of activated platelets into ApoE-/- mice exacerbated atherosclerosis, but not the transfer of P-selectin-deficient platelets (95), corroborating a direct role for platelets in atherosclerosis.

Additionally, attraction of leukocytes through platelets is caused by a release of various soluble, pro-inflammatory mediators from their á-granules, including chemokines (see below), CD40L, II-1β (93), or serotonin (96) within seconds of activation. Notably, either a direct pro-atherogenic or pro-inflammatory role was established for most of these soluble mediators. In recent years, much emphasis has been put on the differential role of either platelet-derived or platelet-activating chemokines, their homo- and heterotypic interactions, and...
distinct function in various biological systems (97). In this regard, chemokines, such as CCL17 (TARC), CCL22 (MDC), and CXCL12 (SDF-1α), which can be expressed by many tissues, directly activate platelets, cause translocation of P-selectin to the cell surface and release of α-granules by direct ligation of their according receptors CCR4 and CXCR4 on platelets (97, 98).

Others, including CCL5 (RANTES), CXCL1 (GROα, KC), CXCL7, and CXCL4 (platelet factor 4, PF4) are secreted and deposited at the side of the endothelium to attract monocytes, neutrophils, and T-cells (99). Accordingly, a genetic knock out of CXCL4 (100) and CXCL1 (101) was shown to reduce atherosclerosis. Moreover, the role of platelet expands beyond recruitment of leukocytes, participating in proliferation and differentiation of pro-inflammatory leukocytes and stem cells (97).

Whether therapeutic strategies in humans may arise from inhibition of platelet chemokines is controversial, since some chemokines, such as CCL5, participate critically in immune defense. However, therapeutic disruption of CCL5, and its heterotypic pro-inflammatory interaction to CCL5 limited atherogenesis in an ApoE−/− mouse model without generating deleterious side effects (102, 103).

**Co-stimulatory molecules and cytokines orchestrate inflammation of the vessel wall**

The control of the cellular pathways in atherogenesis reviewed largely relies on a tight network of cytokines and co-stimulatory molecules. Most of the key cytokines have already been discussed in the preceding sections of this review.

However, an adequate T cell response may only be provoked by an antigen in the presence of co-stimulation. Without co-stimulation T cell antigen contact results in anergy, a phenomenon also occurring in the presence of so-called co-inhibitory pathways.

Modulation of the balance between co-stimulatory and co-inhibitory pathways may represent an attractive strategy to modulate atherogenesis and its outcome. We recently discussed this in detail in a full review article on this topic (104). Interestingly, many co-stimulatory or co-inhibitory molecules may also enfold direct pro- or anti-inflammatory effects on various cells beyond classic co-stimulation.

**The B7 family**

This family of molecules comprises several members regulating T cell biology: B7.1 (CD80), B7.2 (CD86) and their receptor CD28; ICOSL (CD275) and its receptor ICOS (CD278); PD-L1 (CD274) PD-L2 (CD273) and their receptor PD-1 (CD279). B7.1/2+/−/LDLR−/− mice developed smaller atherosclerotic lesions containing less MHCII+ effector cells and lower amounts of IFNγ compared with B7.1/2+/−/LDLR−/− mice (105). In contrast, immunization against ICOS in APOE−/− mice or the transplantation of ICOS-deficient bone marrow into LDLR−/− mice exacerbated atherogenesis compared with respective controls (106, 107). This coincided with increased activation of CD4+ T cells and increased Th1 cytokine production. Similarly, deficiency in PD-L1 and/or PD-L2 aggravated atherosclerotic plaque formation and T cell infiltration (108, 109).

**The TNF superfamily**

Several members of the tumour necrosis factor (TNF) superfamily act as co-stimulators (104). Some of them also enfold direct effects on atherogenesis – independent from co-stimulation.

The classic co-stimulatory pathway within this family of molecules is the CD40L/CD40 pathway. Originally discovered on T cells (CD40L) and B cells (CD40) it has become evident that both molecules are expressed by a variety of cell types. Therefore, they appear to have function beyond co-stimulation and have been associated with various diseases including atherosclerosis (110).

As demonstrated by several groups mice deficient for CD40L or wild-type mice treated with a neutralizing antibody developed smaller atherosclerotic lesions featuring criteria associated with more plaque stability in humans (111–115). Interestingly, we and others demonstrated that CD40L on bone marrow-derived cells does not contribute to atherogenesis warranting a role for CD40L on resident vascular cells (111, 116). This is even more of interest since we recently functionally described a new interaction between CD40L and the leukocyte integrin Mac-1 mediating atherosclerotic plaque formation and inflammatory cell recruitment to the plaque (30, 31) raising the intriguing possibility to selectively inhibit different functions of CD40L through inhibition of the different receptor-associated pathways.

**The action of CD40L also extends to fat inflammation and the associated metabolic syndrome linking metabolic and vascular disease (117–120).**

Downstream of several TNF family members are so-called TNF receptor-associated factors (TRAFs). To date, seven TRAFs are known (121). Our group recently identified a role for TRAF1 and TRAF5 in murine atherosclerosis. TRAF1-deficient animals developed significantly smaller and less inflamed lesions than controls while TRAF5 deficient animals showed increased plaque formation (122, 123).

The role of TRAF6 in atherosclerosis appears to be diverse. While specific interruption of the CD40-TRAF6 axis limited atherogenesis, overall TRAF6 deficiency on bone marrow-derived cells did not affect atherosclerotic plaque formation in mice (124, 125). Several other TRAFs are regulated in atherosclerosis. Their functional contribution, however, remains to be elucidated in vivo (126).

**Conclusion and clinical perspective**

Inflammation initiates, propagates, and complicates the course of atherosclerosis. A multitude of basic science work demonstrates this pathophysiological principle. Of course, a lot of the data has been acquired in mouse models and there are distinct differences between humans and mice (127). However, the basic principles are similar. Beyond that, inflammatory cytokines and
markers are clinically associated with atherosclerosis and its complications.

The most prominent marker in that respect is high sensitive C reactive protein predicting not only risk but even mortality in various collectives.

Although statins enfold some pleiotropic anti-inflammatory actions specific anti-inflammatory or immune-modulatory therapies have not made it into every day clinics so far (128).

Two major clinical trials challenge the inflammatory hypothesis currently:

- The Canakinumab Anti-inflammatory Thrombosis Outcomes Study (CANTOS) that interrogates an antibody neutralizing IL-1β in high risk patients with coronary artery disease on the basis of hard clinical endpoints (129).
- The Cardiovascular Inflammation Reduction Trial (CIRT) evaluating low dose methotrexate in virtually the same collective (130).

These trials could be the begin of a new era of anti-inflammatory/immune-modulatory treatment of atherosclerosis and its sequelae.

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Conflict of interest

The authors report no potential conflict of interest.

References


