Evidence on the pathogenic role of auto-antibodies in acute cardiovascular diseases

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Summary

Atherothrombosis is the major determinant of acute ischaemic cardiovascular events, such as myocardial infarction and stroke. Inflammatory processes have been linked to all phases of atherogenesis. In particular, the identification of autoimmunity mediators in the complex microenvironment of chronic inflammation has become the focus of attention in both early and advanced atherogenic processes. Auto-antibodies against self-molecules or new epitopes generated by oxidative processes infiltrate atherosclerotic plaques and were shown to modulate the activity of immune cells by binding various types of receptors. However, despite mounting evidence for a pathophysiological role of autoantibodies in atherothrombosis, the clinical relevance for circulating autoantibodies in cardiovascular outcomes is still debated. This review aims at illustrating the mechanisms by which different types of autoantibodies might either promote or repress atherothrombosis and to discuss the clinical studies assessing the role of auto-antibodies as prognostic biomarkers of plaque vulnerability.

Keywords

Atherosclerosis, auto-antibodies, inflammation, acute myocardial infarction, stroke

Introduction

The most relevant acute cardiovascular diseases (CVD) - ischaemic stroke and acute coronary syndromes (ACS) - occur as direct manifestations of atherothrombosis, whose understanding is essential to enable the development of targeted and more effective therapies. Although related in part to alterations in lipid metabolism, atherosclerosis is now considered a primarily immunemediated disease (1). Recent studies have highlighted an early role of endothelial dysfunction (2) through an impaired regulation of the vascular tone (alterations in the endothelial metabolism of nitric oxide [NO], prostacyclin and endothelin) and permeability (increased endothelial expression of adhesion molecules, such as ICAM-1, VCAM-1 and selectins) in the pathogenesis of atherosclerosis (2, 3). Endothelial dysfunction allows low-density lipoproteins (LDL) penetration (4) and leukocyte migration within the sub-endothelial space, which results in a local increase of oxidative and inflammatory processes (5). Chemokines and growth factors released in the inflammatory microenvironment have been reported to regulate both cell-mediated and innate immunity in atherogenesis (6, 7). Although a pathogenetic role of B lymphocyte subsets in atherosclerosis remains unclear (8, 9), mounting evidence indicates a potential direct pro-inflammatory role of immunoglobulins (humoral immunity) in plaque maturation and vulnerability. In the last decade, B cell subsets have been shown to play opposing proatherogenic and atheroprotective roles in atherosclerosis also independently of their antibody production (10). B1 B cells have been described as cells resident in the peritoneum or pleura and are important for production of natural IgM antibodies that might protect from atherosclerosis (10). Another protective activity mediated by B1 B cells is represented by their role as regulatory B cells that produce the anti-atherosclerotic cytokine interleukin (IL)-10 (11). On the other hand, naïve conventional B2 cells can differentiate into Be1 or Be2 effector B cells and play critical pro-atherogenic activities (12). In fact, both Be1 B cells, producing Th1 cytokines might increase atherosclerosis, and Be2 B cells, secreting Th2 cytokines (such as IL-4, IL-6) might increase plaque inflammation and instability (13).

Several mechanisms have been suggested to potentially explain the antibody-mediated autoimmune response in atherosclerosis: 1) molecular mimicry between self-molecules and microorganism-derived epitopes (14); 2) exposure of antigenic molecules at the surface of apoptotic cells (15); 3) immune response to oxidative stress-modified molecules (16).

Despite some limitations (mainly due to the low number of patients investigated and the concomitant high expression of several...
other pro-atherosclerotic factors), clinical observational studies also confirmed an association between some auto-antibodies and cardiovascular diseases (17). This narrative review will discuss evidence from human studies on the role of B cell-mediated immunity in atherosclerosis and the potential prognostic value of auto-antibodies in cardiovascular patients. The potential atherogenic role of auto-antibodies against lipids and their components (such as modified LDL, phosphorylcholine [PC] or anti-[apolipoprotein] apoA-1 and phospholipids) or stress-related proteins (heat shock protein [HSP]) will be commented. On the other hand, the auto-antibody-mediated activities on heart failure or dilated cardiomyopathy will not be addressed in the present review.

Anti-modified LDL antibodies

LDLs are a class of lipoprotein particles consisting of a hydrophobic core (containing triglycerides and cholesterol esters) and in a hydrophilic shell of phospholipids, free cholesterol, and Apo lipoproteins (mostly B-100). The access and retention of LDL in the subendothelial layer mainly depends on its sustained plasma levels (4) but also other possible determinants like lipoprotein size, cholesterol enrichment, endothelial permeability, and biosynthetic activity in endothelial cell (i.e. synthesis of the basement membrane and extracellular matrix [ECM]) (18).

Once retained by the ECM components, LDL particles become modified by oxidants (lipoxygenase, myeloperoxidase, free radicals), proteolytic (trypsin, metalloproteinases, thrombin, etc.), and lipolytic (sphingomyelinase, phospholipase A2, phospholipase C, etc.) enzymes. These chemical and/or structural changes generate many different types of modified LDL particles, such as oxidised LDL (oxLDL), which are highly heterogeneous molecules. In smooth muscle cells, oxLDLs have been shown to promote the activation of pro-inflammatory signalling pathways (19-22) and to impair NO biosynthesis (23). In endothelial cells, ox-LDLs have been shown to increase the expression of adhesion molecules, thus leading to endothelial dysfunction and favouring the leukocyte recruitment within the subendothelial space (24). In addition, oxLDLs may affect fibrinolysis (25) and promote a chronic pro-thrombotic state. In phagocytes, oxLDLs initiate activatory signalling pathways by binding scavenger receptors (26), peroxisome proliferator-activated receptors (PPARs) (27, 28), the platelet-activating factor (PAF) receptor (29) and Toll-like receptors (TLRs) (30, 31). In addition, modified LDLs have been shown to be intrinsically immunogenic (32). Circulating antibodies directed to oxidised LDL were then purified and characterised. IgG (mostly IgG1 and IgG3) (33-35) were the predominant isotypes, followed by IgM and IgA.

Soon after the discovery of anti-oxLDL antibodies, the researchers’ attention focused on their pathogenic role in atherosclerosis. The results of these studies were rather disappointing, as they were frequently contrasting and failed to produce a clear cut indication of the clinical value of oxLDL antibody assays as biomarkers for the development and progression of atherosclerosis. Although the existence of anti-oxLDL antibodies in humans has been known for more than 15 years (36, 37), their potential role in atherogenic processes and their association with CVD is still highly controversial. Historically, the first clinical studies identified several antibodies (both IgG and IgM) against different epitopes on oxidised LDL (such as malondialdehyde [MDA]-LDL, copper-oxidised LDL or [4-hydroxynonenal] 4-HNE LDL) (36, 38). Therefore, as reviewed elsewhere (39), anti-oxLDL antibodies were shown to be neutral, protective, as well as pro-atherogenic in atherosclerosis and CVD risk (37, 40-45). These discrepancies could be explained by the highly heterogeneous and complex nature of oxLDLs, which in turn is expected to stimulate the production of auto-antibodies to different epitopes and with various avidities for the antigens, and belonging to different isotypes including IgA, IgG1, IgG2, IgG3, and IgM. In addition, the pathogenetic potential of anti-oxLDL, as well as that of other antibodies, depends on its isotype and is fully expressed only after the formation of antigen-antibody complexes, mainly IgG1 and IgG3 (34, 35). In the Framingham Offspring Study, the largest prospective study available (with a cohort of over 1,192 men and 1,427 women with a mean follow-up of eight years), no significant association between the serum levels of anti-ox-LDL IgG and cardiovascular events was detected (46). In 2011, a case-control study on participants of the EPIC-Norfolk cohort (748 cases and 1,723 controls; median follow-up six years) showed that both IgM and IgG antibodies against MDA-LDL and their IC are not independent predictors of coronary artery disease (CAD) events (47). The potential association between carotid atherosclerosis (assessed by intima-media thickness [IMT] and plaque number measurements) and plasma levels of antibodies against ox-LDL (IgM, IgG, and IgG2) was evaluated in population-based cohort of 1,022 patients recruited from Finnish national registers (48). The authors showed that high titers of IgM autoantibodies against MDA-LDL has significantly inversely correlated with carotid atherosclerosis independently of age, gender, systolic blood pressure, LDL cholesterol, C-reactive protein (CRP) and smoking (48). This study suggested for the first time a protective role for IgM in atherosclerosis. More recently, Tsimikas et al. investigated the potential associations between IgG and IgM antibodies against ox-LDL (such as MDA-modified LDL, copper-ox-LDL, and oxidised cholesterol linolate) and CAD in 504 patients undergoing coronary angiography for clinical reasons (49). The authors confirmed opposite associations for different antibodies against ox-LDL levels (protective for IgM and deleterious for IgG) also in CAD patients. However, these autoimmune biomarkers failed to predict obstructive CAD independently of age, sex, lipid profile, CRP, hypertension and smoking (49). Clinical studies on the different relationship between atherosclerosis and antibodies against ox-LDL are summarised on Table 1.

As shown above, another interesting potential role underlying the different role of anti-oxLDL antibodies is dependent on immune complex formation since oxLDL-IC interact with Fc receptors (50-52). In fact, as in other autoimmune diseases, immune complexes (IC) IgG-oxLDL have been demonstrated and they were found to be more potent activators of human macrophages than oxLDL (53). A fundamental property of oxLDL-IC is the ability to deliver large concentrations of free and esterified cholesterol.
to macrophages (54). Their uptake is mediated by Fcγ receptors (50, 55) and they block the interaction of oxLDL with CD36 (51). Cross-linking of the Fcγ receptors by OxLDL-IC triggers several pathways (52, 56, 57) that are responsible for the expression of pro-inflammatory genes via nuclear factor-κB activation, and for promoting cell survival through the PI3K/Akt pathway (58). In addition, oxLDL-IC were shown to induce HSP70B expression in macrophages (59). Thus, it is not surprising that the repertoire of oxLDL-IC-induced pro-survival genes is much wider than that induced by oxLDL alone (60). A recent clinical study analysed the level of oxLDL in circulating IC (conjugated with IgG antibody), isolated from sera of 479 patients of the Diabetes Control and Complications Trial/Epidemiology of Diabetes Interventions and Complications (DCCT/EDIC) cohort. After a follow-up of eight and 14 years, IC were significantly associated with increased levels and progression of carotid IMT in type 1 diabetes by 6.11-fold (confidence interval [CI] 2.51–14.8) (61). In the same cohort of patients, it was also shown that increased level of oxLDL-IC were as-

Table 1: Anti-oxLDL antibodies and cardiovascular risk.

<table>
<thead>
<tr>
<th>Author [Reference]</th>
<th>Year</th>
<th>Study design (n)</th>
<th>Antibody</th>
<th>Outcome and relationship</th>
<th>Prediction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bellomo G. et al. [40]</td>
<td>1995</td>
<td>Case-control study (138 patients with non-insulin dependent diabetes mellitus and 80 matched control subjects)</td>
<td>IgG and IgM anti-glycated LDL and anti-glycoxylated LDL</td>
<td>No clinical outcome was investigated.</td>
<td>Diabetic patients have an antibody ratio significantly higher than control subjects for anti-glycated LDL and anti-glycoxylated LDL IgG (1.79 ± 0.38 vs. 1.12 ± 0.23, p &lt;0.01 and 2.55 ± 1.03 vs. 1.39 ± 0.44, p&lt;0.01, respectively), but not IgM.</td>
</tr>
<tr>
<td>Erkkila A.T. et al. [41]</td>
<td>2000</td>
<td>Cohort prospective observational study (415 with CHD)</td>
<td>IgG anti-oxidised LDL</td>
<td>No correlation with lipid profile except a positive correlation with triglycerides in women (r=0.225, p=0.011).</td>
<td>Antibodies against ox-LDL were higher in men with first or recurrent acute myocardial infarction without previous revascularization procedure than in patients with first elective or emergency coronary artery bypass surgery or patients first elective or emergency percutaneous transluminal coronary angioplasty or patients with acute myocardial ischaemia (p=0.022).</td>
</tr>
<tr>
<td>Lehtimaki T. et al. [42]</td>
<td>1999</td>
<td>Case-control study (58 patients with angiographically verified CAD and 34 controls without CAD)</td>
<td>IgG against copper-oxidised LDL</td>
<td>No clinical outcome was investigated.</td>
<td>High antibody levels against oxidized LDL were significantly associated with CAD (p=0.0114), independently of age, gender, body mass index, triglyceride concentration.</td>
</tr>
<tr>
<td>Maggi E. et al. [43]</td>
<td>1994</td>
<td>Case-control study (94 patients with severe carotid atherosclerosis undergoing elective carotid artery endarterectomy and 42 matched controls)</td>
<td>IgG and IgM anti-oxidised LDL, anti-anti-MDA LDL</td>
<td>No clinical outcome was investigated.</td>
<td>Cases had an antibody ratio significantly higher than control subjects in regard to anti-oxLDL IgG (1.78 ± 0.39 vs 1.05±0.3, p&lt;0.01) and IgM (1.98 ± 0.83 vs 1.40 ± 0.09, p&lt;0.05) and anti-MDA-LDL IgG (2.39 ± 0.51 vs 2.04 ± 0.11, p&lt;0.01) and IgM (4.18 ± 1.89 vs 2.9 ± 0.15, p&lt;0.05).</td>
</tr>
<tr>
<td>Virella G. et al. [45]</td>
<td>1993</td>
<td>Case-control study (33 subjects submitted to coronary angiography and split into two subgroups depending on the degree of coronary stenosis and 64 healthy individuals also split into two subgroups according to lipid levels)</td>
<td>IgG anti-oxidised LDL</td>
<td>No clinical outcome was investigated.</td>
<td>Higher levels of antibodies were detected in patients with moderate coronary disease and hyperlipemic healthy individuals. However, the differences did not reach statistical significance (p=N.S.)</td>
</tr>
<tr>
<td>Wilson P.W. et al. [46]</td>
<td>2006</td>
<td>Prospective observational (2,619)</td>
<td>IgG anti-MDA-LDL</td>
<td>No significant independent associations with clinical CHD (angina pectoris, unstable angina pectoris, myocardial infarction, or coronary death) or CVD</td>
<td>In both men and women: for CHD: RR 1.00 (1.00–1.00); for CVD: RR 1.00 (1.00–1.00), p=N.S.</td>
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</tbody>
</table>
associated with the development of coronary calcification as determined by computed tomography (CT) 11-20 years later (62).

**Anti-phosphorylcholine (PC) antibodies**

PC is a major antigenic determinant commonly found in oxLDL, apoptotic cells and microorganisms including bacteria (i.e. *Staphylococcus pneumoniae*), and some parasites (63). Bearing in mind that oxidation process of LDL is very complex and might affect the different epitopes of oxLDL, the potential overlap of studies addressing antibodies anti-oxLDL and its epitope PC has to be carefully considered. In the early 1970s, anti-PC antibodies of IgM subclass (anti-PC IgM) were first identified in mice and humans as a major clonal protein synthesised by plasmocytoma cells (64-66), or produced in response to parasitic infections (67). Then, a potential protective role in atherogenesis of these IgM antibodies was shown for the first time in 2000 by Shawn et al. (68), and confirmed in mice 2003 by Binder et al. (69). On the other hand, whether the mechanisms proposed to account for the protective effects of anti-oxLDL IgM, such as reduction of oxLDL uptake by macrophage and endothelial cell inactivation by apoptotic cell (70), applies to anti-PC IgM remains elusive. Nevertheless, anti-PC IgG extracted from pooled human immunoglobulins might inhibit PAF-induced endothelial activation, as determined by the expression of VCAM and ICAM-1 (71).

In accordance with those experimental results, Frostege et al. demonstrated in 2006 that in humans anti-PC IgM levels were inversely associated with carotid atherosclerosis (assessed according to intima-media thickness) in hypertensive patients, while no association with anti-PC IgG was retrieved (72), extending to humans the hypothesis that anti-PC IgM would be protective in atherogenesis. Few years later, the same group demonstrated that low levels of anti-PC IgM were predictors of death in ischemic stroke patients (73-76) (Table 2). In other two case-control studies, no association was found between low IgM and stroke development.

<table>
<thead>
<tr>
<th>Author [Reference]</th>
<th>Year</th>
<th>Study design (n)</th>
<th>Antibody</th>
<th>Outcome and relationship</th>
<th>Prediction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Su J. et al. ELSA Study [72]</td>
<td>2006</td>
<td>Prospective observational (226)</td>
<td>aPC</td>
<td>High levels correlate with slow IMT progression</td>
<td>OR 0.37 (0.15–0.89), p=0.003</td>
</tr>
<tr>
<td>Sjöberg B.G. et al. [73]</td>
<td>2009</td>
<td>Case-control study (349/693)</td>
<td>aPC</td>
<td>Low levels are associated with IS development</td>
<td>RR 1.84 (1.03–3.25), p=0.036</td>
</tr>
<tr>
<td>Carrero J.J. et al. [75]</td>
<td>2009</td>
<td>Prospective observational (203)</td>
<td>aPC</td>
<td>Low levels are predictors of death</td>
<td>HR 0.986 (0.981–0.989), p=0.008</td>
</tr>
<tr>
<td>Fiskesund R. et al. [74]</td>
<td>2010</td>
<td>Nested case-control study (227/455)</td>
<td>aPC</td>
<td>Low levels are associated with IS development</td>
<td>OR 1.62 (1.11–2.35), p=0.05</td>
</tr>
<tr>
<td>Anania C. et al. [76]</td>
<td>2010</td>
<td>Case-control study (114/122)</td>
<td>aPC</td>
<td>Low levels correlate with carotid atherosclerosis occurrence</td>
<td>OR 2.920 (1.08–7.89), p=0.0348</td>
</tr>
</tbody>
</table>

aPC: anti-phosphorylcholine antibodies; IMT: intima media thickness; IS: ischaemic stroke; OR: odds ratio.
between low anti-PC IgM levels and the risk of stroke occurrence, but a significant and modest association with myocardial infarction risk was retrieved, independent of conventional cardiovascular risk factors (77, 78). Recently, a prospective study involving 1,185 patients hospitalised for ACS demonstrated that patients with anti-PC IgM levels below 26 U/ml had a higher risk of major cardiovascular events at six, 12 and 18 months after the primary event, independent of traditional cardiovascular risk factors and other biomarkers, such as cardiac troponins, brain natriuretic peptides and CRP (79). Another smaller prospective study performed on patients presenting to the emergency room for acute chest pain without electrocardiographic changes indicated that low anti-PC IgM levels could be of value for non ST-elevation myocardial infarction (NSTEMI) diagnosis, especially when combined with autoantibodies to apolipoprotein A-1 (apoA-1) (80). Taken together those concordant results support the notion that anti-PC IgM are atheroprotective. Nevertheless, as most studies used a post-hoc defined anti-PC IgM cut-off, the exact anti-PC IgM cut-off to be used for CVD prediction is still unknown and the one used varies significantly (16.9 to 42 U/ml) (72-80). Therefore, further prospective validation studies using a pre-specified cut-off are required before any clinical recommendation can be made.

**Anti-apoA-1 antibodies**

The anti-atherogenic properties of high-density lipoprotein (HDL) have been well-established: firstly, in the classic concept of reverse cholesterol transport, HDL remove excess cellular cholesterol from cholesterol-loaded vascular macrophage foam cells and transport it back to the liver (81). Secondly, HDL have been shown to protect LDL from oxidation by transferring oxidised lipids from LDL to HDL (82, 83). A third important mechanism is also represented by the inhibition of pro-inflammatory cytokine production (84), and concomitant selective decrease of adhesion molecule expression on endothelial cells (85). The atheroprotective role of HDL can be better understood by observing that conditions characterised by a more rapid progression of atherosclerosis are associated with HDL dysfunction (86). ApoA-1 is the main component protein of HDL conferring to the latter most of its atheroprotective properties (87).

![Figure 1: Transmembrane receptors and intracellular pathways triggered by auto-antibody in atherogenesis.](image)

*Figure 1: Transmembrane receptors and intracellular pathways triggered by auto-antibody in atherogenesis.* TNF: tumour necrosis factor; IL: interleukin; MIP: macrophage infectivity potentiator; MCP: monocyte chemotactic protein-1; ICAM-1: intercellular adhesion molecule 1; VCAM-1: vascular cell adhesion molecule 1; SOD: superoxide dismutase; MMP: matrix metalloproteinases; iNOS: intracellular nitric oxide synthetase; NADPH: nicotinamide adenine dinucleotide phosphate-oxidase; COX-2: cyclooxygenase; GM-CSF: granulocyte macrophage colony-stimulating factor; VEGF: vascular endothelial growth factor.
Anti-apoA-1 antibodies were first identified in patients with Systemic Lupus Erythematosus (SLE) and Anti Phospholipid Syndrome (APS) (88) assuming that these autoantibodies had a part in thrombogenesis (89). Vuilleumier et al. demonstrated that anti-apoA-1 antibody levels were raised in patients with ACS without autoimmune disease (90). In 2010, high levels of anti-apoA-1 IgG were shown to predict acute cardiovascular events in a cohort of 133 patients with rheumatoid arthritis (RA) during a median follow-up period of nine years (hazard ratio [HR] 4.2, 95% CI 1.5-12.1, p = 0.0008) (91). From the same cohort of patients Finckh et al. showed that, when compared to NT-proBNP and oxLDL, anti-apoA-1 IgG was the only biomarker to significantly improve the prognostic ability of the Framingham Risk Score (FRS), with AUCs increasing from 0.72 to 0.81 (92). Furthermore, the same group demonstrated in a prospective cohort of 221 patients with MI that anti-apoA-1 IgG serum levels are independently associated with major adverse cardiovascular events during a follow-up period of 12 months (odds ratio [OR] 4.3; 95% CI 1.46–12.6; p=0.007), independent of traditional cardiovascular risk factors (93). In an ancillary study, the same investigators performed a “head to head” comparison of the prognostic accuracies of several autoantibodies including those against anti-β2-glycoprotein I (β2GPI) domain I and IV, cardiolipin, apoA-1, heat-shock protein 60 (anti-HSP-60), and phosphorylcholine (anti-PC IgM). Receiver-operating characteristics (ROC) curve analyses, indicated that anti-apoA-1 IgG was the only autoantibody to significantly predict the occurrence of subsequent major adverse cardiac events (MACE) at one year, and a trend was observed for anti-cardiolipin and anti-HSP60 antibodies (p=0.05 and p=0.07, respectively) (94). Finally, in a single-centre prospective study, Keller et al. demonstrated that anti-apoA-1 IgG could be of diagnostic value in patients presenting to the emergency room for acute chest pain (80). In this study, the investigators demonstrated that anti-apoA-1 IgG values assessed on the first sample available displayed a good diagnostic accuracy for NSTEMI with an area under the curve (AUC) of 0.75 (p<0.0001) that could be further increased when combined with anti-PC IgM and the NSTEMI-TIMI score. Whether anti-apoA-1 IgG could provide incremental diagnostic value over high sensitive troponins, is still unclear and will need to be tested in further studies. Several studies have been performed to identify the potential mechanisms underlying anti-apoA-1 IgG-mediated pathophysiology in atherogenesis. A correlation between anti-apoA-1 IgG titres and oxLDL levels led to speculate that increased oxLDL levels in ACS could be partly due to the presence of anti-apoA-1 IgG (95). Indeed, anti-apoA-1 antibodies have been shown in vitro to prevent scavenger receptor B-1 function in human endothelial cells and to interfere with HDL-mediated NO production (96). Another mechanism by which anti-apoA-1 IgG may worsen myocardial performance is the ability to increase basal heart rate. This observation was confirmed in vivo (93). Anti-apoA-1 IgG

<table>
<thead>
<tr>
<th>Author [Reference]</th>
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<th>Study design (n)</th>
<th>Target</th>
<th>Outcome and relationship</th>
<th>Prediction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vuilleumier N. et al. [95]</td>
<td>2008</td>
<td>Prospective observational (127)</td>
<td>apoA-1</td>
<td>Associated with higher oxLDL levels in ACS patients</td>
<td>P&lt;0.0001</td>
</tr>
<tr>
<td>Vuilleumier N. et al. [93]</td>
<td>2010</td>
<td>Prospective study, one-year follow-up (221)</td>
<td>apoA-1</td>
<td>After MI, high anti-apoA-1 IgG levels are independently associated with subsequent MACE</td>
<td>Adjusted OR:4.3 (95%CI:1.46–12.6), p=0.007</td>
</tr>
<tr>
<td>Vuilleumier N. et al. [91]</td>
<td>2010</td>
<td>Prospective (133)</td>
<td>apoA-1</td>
<td>In RA patients, high anti-apoA-1 IgG levels are independently associated with MACE</td>
<td>Adjusted HR: 4.2 (95%CI:1.5–12.1)</td>
</tr>
<tr>
<td>Montecucco F. et al. [98]</td>
<td>2011</td>
<td>Case-control (102)</td>
<td>apoA-1</td>
<td>In patients with severe carotid stenosis, high anti-apoA-1 IgG levels associated with serum and intraplaque features of atherosclerotic plaque vulnerability</td>
<td></td>
</tr>
<tr>
<td>Pagano S. et al. [99]</td>
<td>2012</td>
<td>Prospective observational (221)</td>
<td>apoA-1</td>
<td>In MI, high anti-apoA-1 IgG levels associated with higher TNF-α, IL-6, and MMP-9 levels</td>
<td></td>
</tr>
<tr>
<td>Keller P.F. et al. [80]</td>
<td>2012</td>
<td>Prospective (138)</td>
<td>apoA-1</td>
<td>In acute chest pain patients without ECG changes, high anti-apoA-1 IgG levels are predictors of subsequent NSTEMI diagnosis</td>
<td>AUC:0.75, p&lt;0.001 Adjusted OR: 6.4 (95%CI:1.72–24.2)</td>
</tr>
<tr>
<td>Wick P. et al. [109]</td>
<td>2012</td>
<td>Case-control (130/46)</td>
<td>ApoA-1</td>
<td>In young periodontitis patients, high anti-apoA-1 IgG levels are predictive of an increased atherosclerosis burden (ankle brachial index, &lt; 1.11)</td>
<td>AUC: 0.63; p=0.03 OR:4.20, p=0.04</td>
</tr>
<tr>
<td>Finckh A. et al. [92]</td>
<td>2012</td>
<td>Prospective (118)</td>
<td>ApoA-1</td>
<td>Anti-apoA-1 IgG provides incremental prognostic information over Framingham risk score for MACE occurrence in RA</td>
<td>HR:4.12 (95%CI:1.66–10.21) Intergated discrimination index: +175%, p&lt;0.001</td>
</tr>
</tbody>
</table>

ACS: acute coronary syndrome; CI: confidence interval; MACE: major adverse cardiovascular events; OR: odds ratio; RA: rheumatoid arthritis; MI: myocardial infarction; TNF: tumour necrosis factor; MMP: matrix metalloproteinase.
acts as a positive chronotropic agent in the presence of aldosterone (or corticosterone in oxidised conditions) by enhancing L-type calcium channel activity. This property is dependent on the combination of the non-genomic effects of aldosterone involving phosphatidylinositol 3-kinase (PI3K) activation with a possible constitutive activity of protein kinase A (PKA) (97). Considering atherosclerotic inflammatory processes, we also showed that anti-apoA-1 IgG increase atherosclerotic plaque vulnerability by promoting neutrophil infiltration from the blood stream into the atherosclerotic plaque and by increasing the intraplaque release of chemo-attractants and matrix-metalloproteinase 9 (MMP-9) with consequent collagen degradation (98). A more recent translational study has shown a direct pro-inflammatory effect of anti-apoA-1 IgG, mediated by their interaction with TLR2/CD14-complex leading to a subsequent NF-κB activation and tumour necrosis factor (TNF)-α and interleukin (IL)-6 release (99). These findings are in line with studies showing that some auto-antibodies, such as anti-phospholipid (aPL) and anti- HSP antibodies, can promote inflammation through the engagement of TLR2 and TLR4 (100-103) (Figure 1). Another emerging physiological aspect related to anti-apoA-1 IgG appears to involve HDL function, whose importance in atherogenesis has been described for over 15 years (104). In 2002, Delgado Alves et al. reported for the first time that anti-HDL, and later anti-apoA-1 IgG, could affect HDL function in patients suffering from systemic lupus erythematoses (SLE) and antiphospholipid syndrome (APS) (105-107). In those studies those auto-antibodies were inversely correlated with paraoxonase-1 (PON-1) activity, with the total antioxidant capacity

Table 4: Anti-heat shock protein antibodies and cardiovascular risk.

<table>
<thead>
<tr>
<th>Author [Reference]</th>
<th>Year</th>
<th>Study design (n)</th>
<th>Target</th>
<th>Outcome and relationship</th>
<th>Prediction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Xu Q. et al. Bruneck Study [132]</td>
<td>1993</td>
<td>Prospective observational (867)</td>
<td>Anti HSP65 IgG</td>
<td>High levels correlate with carotid atherosclerosis occurrence</td>
<td>Multiple linear regression: correlation coefficient 0.11 p = 0.003</td>
</tr>
<tr>
<td>Frostegård J. et al. [135]</td>
<td>1997</td>
<td>Case-control (66/67)</td>
<td>Anti HSP65 IgG</td>
<td>Higher levels in pre-hypertensive vs normotensive patients</td>
<td>Student’s t test p=0.34</td>
</tr>
<tr>
<td>Birnie D.H. et al. [136]</td>
<td>1998</td>
<td>Cross-sectional (61/21)</td>
<td>Anti HSP65 IgG</td>
<td>High levels are related with coronary atherosclerosis (extension and severity)</td>
<td>Spearman correlation coefficient r=0.21 p=0.018</td>
</tr>
<tr>
<td>Xu Q. et al. Bruneck Study (FU Sy) [133]</td>
<td>1999</td>
<td>Prospective observational (750)</td>
<td>Anti HSP65 IgG</td>
<td>High levels correlate with IMT</td>
<td>OR 1.42 (1.02–1.98) p=0.039</td>
</tr>
<tr>
<td>Chan Y.C. et al. [137]</td>
<td>1999</td>
<td>Case-control (61/21)</td>
<td>Anti HSP70 IgG</td>
<td>High levels correlate with various vascular disease</td>
<td>Intergroup comparisons with Mann–Whitney U-test p=0.0003</td>
</tr>
<tr>
<td>Burian K. et al. [138]</td>
<td>2001</td>
<td>Case-control (276/129)</td>
<td>Anti-HSP60 IgG</td>
<td>High levels are associated with CAD</td>
<td>OR 5.4 (2.1–14.0) p=0.0004</td>
</tr>
<tr>
<td>Gromadzka G. et al. [139]</td>
<td>2001</td>
<td>Case-control (180/64)</td>
<td>Anti HSP65 IgG</td>
<td>High levels are predictors of IS</td>
<td>OR 54.12 (11.28–259.70) p=0.000001</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Anti HSP70 IgG</td>
<td>High levels are predictors of IS</td>
<td>OR 27.02 (6.49–112.39) p=0.000001</td>
</tr>
<tr>
<td>Huittinen T. et al. Helsinki Heart Study [134]</td>
<td>2002</td>
<td>Prospective, nested, case-control study (239/241)</td>
<td>Anti HSP60 IgA</td>
<td>High levels correlate with coronary events</td>
<td>OR 2 (1.1–3.6) p=0.04</td>
</tr>
<tr>
<td>Veres A. et al. HOPE study [140]</td>
<td>2002</td>
<td>Nested case-control study (386/386)</td>
<td>Anti HSP65 IgG</td>
<td>High levels correlate with cardiovascular events</td>
<td>OR 2.1 (1.2–3.9) p=0.01</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Anti-HSP60 IgA</td>
<td>did not predict any cardiovascular event</td>
<td>OR 1.2 (0.65–2.08) p=0.60</td>
</tr>
<tr>
<td>Kervinen H. et al. Helsinki Heart Study [141]</td>
<td>2003</td>
<td>Nested case-control study (241/241)</td>
<td>Anti HSP60 IgA</td>
<td>High levels correlate with coronary risk</td>
<td>OR 1.41 (0.96–2.05) p-value &lt; 0.05</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>High levels correlate with hypertension</td>
<td>OR 2.32 (1.26–4.27) p-value &lt; 0.05</td>
</tr>
<tr>
<td>Zhu J. et al. [142]</td>
<td>2004</td>
<td>Cross-sectional (198)</td>
<td>Anti HSP65 IgG</td>
<td>High levels correlate with high coronary Ca++</td>
<td>OR 5.53 (1.17–26.09) p=0.012</td>
</tr>
<tr>
<td>Zhang X. et al. [143]</td>
<td>2008</td>
<td>Case-control (1003/1003)</td>
<td>Anti HSP60 IgG</td>
<td>High levels were recorded in CHD patients</td>
<td>OR 3.67 (1.56–8.64) p&lt;0.0001</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>High levels correlate with hypertension</td>
<td>OR 5.17 (3.95–6.75) p&lt;0.0001</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>High levels correlate with DM</td>
<td>OR 6.49 (4.52–9.33) p&lt;0.0001</td>
<td></td>
</tr>
</tbody>
</table>

HSP: heat shock protein; IMT: intima media thickness; CAD: Coronary Artery Disease; IS: ischaemic stroke; DM: diabetes mellitus; HR: hazard ratio; OR: odds ratio.
of the corresponding sera, and were associated to an increase of pro-inflammatory reactive oxygen species (ROS), which is considered as a feature of HDL dysfunction. The causal nature of this association has lately been suggested in a lupus-prone mice model, where the presence of anti-apoA-1 antibodies was associated with a decrease in the anti-oxidant properties of HDL, and related to a decrease in PON-1 activity (108). Taken together, those observations support the hypothesis that anti-apoA-1 IgG are active mediators of atherogenesis (109) (Table 3). However, further work is required to clarify whether anti-apoA-1 IgG could represent a new potential therapeutic target.

### Anti-HSPs antibodies

HSPs are classified based on their molecular weight: HSPs, HSP10, HSP40, HSP60, HSP70, HSP90 and HSP110. These proteins show high sequence homology between different species (from bacteria to humans), and are involved in maintaining correctly folded forms of cellular proteins (110). Despite their function as chaperones, the different types of HSPs have different bioactivities. For instance, HSP10 is a cofactor for HSP60 and displays an anti-apoptotic role in response to stress (111, 112). HSP27 forms complexes with mitogen-activated protein kinase (MAPK)-activated protein kinase 2 (MK2), and Akt (113). Reduced HSP27 levels during vascular remodeling are thought to reflect proteolytic imbalance (114). HSP60 is normally found inside the cells. However, when found in the extracellular environment, HSP60 is an indicator of cell death and activates macrophages and other immune cells to promote dead cell clearance (115). High levels of HSP60 are induced by many stressful stimuli such as oxidised LDL, biomechanical stress, infections, oxidants, and cytokine (116). HSP70 has many functions such as polypeptide folding modulation, degradation/translocation across membranes, and protein-protein interactions. High levels of HSP70 within atherosclerotic plaques (117-119) have been directly associated with an increased expression of pro-inflammatory cytokines (120). High expression levels of HSPs have been shown to protect the vessel wall against hypercholesterolaemia-induced damage, and to counteract adverse arterial remodelling and stiffness (121).

HSPs have been investigated as a target in the atherosclerotic immune response. In 1986, Srivastava et al. showed that cytoplasmic HSP (such as HSP70 and HSP90) were capable of binding antigenic peptides (122) and promoting their presentation on MCH class I molecules (123, 124). As compared to antigenic peptides alone, HSP-peptide complexes have been shown to strongly activate T cells (125). HSPs also induced the production of pro-inflammatory cytokines (such as TNF-α, IL-1, IL-6, and IL-12), via interactions. High levels of HSP70 within atherosclerotic plaques (117-119) have been directly associated with an increased expression of pro-inflammatory cytokines (120). High expression levels of HSPs have been shown to protect the vessel wall against hypercholesterolaemia-induced damage, and to counteract adverse arterial remodelling and stiffness (121).

### Table 5: Anti-HSP27 antibodies in atherosclerosis.

<table>
<thead>
<tr>
<th>Author [Reference]</th>
<th>Year</th>
<th>Study design (n)</th>
<th>Target</th>
<th>Outcomes and relationship</th>
<th>Prediction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Shams S. et al. [144]</td>
<td>2008</td>
<td>Case-control study (60/63)</td>
<td>Anti HSP27</td>
<td>Higher levels correlate in chest pain population compared to control with -Age -Hyp</td>
<td>Spearman correlation coefficient Age: r=-0.29, p=0.03 Hyp: r=-0.235, p=0.016</td>
</tr>
<tr>
<td>Ghayour-Mobarhan M. et al. [145]</td>
<td>2008</td>
<td>Case-control study (94/81)</td>
<td>Anti HSP27</td>
<td>High levels correlate with MI and UA compared to controls</td>
<td>Data analysis with Kruskal–Wallis test p&lt;0.001</td>
</tr>
<tr>
<td>Burt D. et al. EUROMIDAP Prospective Complications Study [146]</td>
<td>2009</td>
<td>Case-control study (531/168)</td>
<td>Anti HSP27</td>
<td>High levels not correlate with DM complications</td>
<td>OR 1.65 (0.80–3.43) p=0.56</td>
</tr>
<tr>
<td>Burut D.F. et al. [147]</td>
<td>2010</td>
<td>Case-control study (42/26)</td>
<td>Anti HSP27</td>
<td>Higher level in CAD and glucose intolerance population compared to controls</td>
<td>Spearman correlation coefficient r=-0.30 p=0.02</td>
</tr>
<tr>
<td>Azarpazhooh M.R. et al. [148]</td>
<td>2010</td>
<td>Case-control study (168/80)</td>
<td>Anti HSP27</td>
<td>High level in IS population compared to controls</td>
<td>OR 0.18 (0.14–0.28) p&lt;0.001</td>
</tr>
<tr>
<td>Pourghadamfard H. et al. [149]</td>
<td>2011</td>
<td>Case-control study (400)</td>
<td>Anti HSP27</td>
<td>High levels are related to CAD severity</td>
<td>Data analysis with multiple linear regression: correlation coefficient 0.042 p&lt;0.001</td>
</tr>
<tr>
<td>Sahebkar A et al. [150]</td>
<td>2011</td>
<td>Case-control study (161/82)</td>
<td>Anti HSP27</td>
<td>High levels in CAD are more often associated to MS</td>
<td>Data analysis with Mann–Whitney U test p=0.04</td>
</tr>
<tr>
<td>Rahsepar A.A. et al. [151]</td>
<td>2012</td>
<td>Case-control study (30/30)</td>
<td>Anti HSP27</td>
<td>Correlate with VHD</td>
<td>Spearman correlation r=-0.394 p&lt;0.05</td>
</tr>
<tr>
<td>Rahsepar A.A. et al. [152]</td>
<td>2012</td>
<td>Case-control study (53/83)</td>
<td>Anti HSP27</td>
<td>Did not related to different heart function parameters</td>
<td>Spearman correlation coefficient with p&gt;0.05</td>
</tr>
</tbody>
</table>

HSP: heat shock protein; Hyp: hypertension; MI: myocardial infarction; UA: unstable angina; DM: diabetes mellitus; CAD: coronary artery disease; VHD: valvular heart disease; OR: odds ratio.

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CD14/TLR (TLR2 and TLR4) pathway, involving the downstream activation of NF-κB and MAPKs (126-130).

HSPs were also described to stimulate auto-antibody formation. Wu et al. showed association between antibodies directed against HSPs and atherosclerosis in the early 1990s (131). Afterward, a large prospective population-based survey, demonstrated that serum antibodies against HSP65 were significantly elevated in subjects with carotid atherosclerosis compared to those without lesions (132). A subsequent follow-up study confirmed that antibody levels remained stable over a five-year observation period, especially in subjects with progressive carotid atherosclerosis (133). In addition, Huittinen et al. demonstrated that patients with high anti-human HSP60 IgA levels in association with elevated serum concentrations of anti-Chlamydia pneumoniae IgA and CRP displayed a higher risk of developing an acute coronary event (134). A direct role of anti-HSP60 antibody levels and in human atherogenesis has been also confirmed by several studies (▶Table 4).

Another promising autoimmune target in atherogenesis might be represented by HSP27. Several clinical studies have recently reported a potential direct association of this anti-HSP27 antibody with CVD (▶Table 5).

However, given the lack of pathophysiological studies and of large randomised clinical trials that confirm this correlation, the association between anti-HSP27 titres and CAD needs further confirmation. Overall, no prospective and/or interventional study confirming the prognostic role of anti-HSP antibodies is currently available. However, the possible relevance of these antibodies is suggested by a study by Mohebbati et al. in which 102 participants were treated with simvastatin (40 mg/day), or placebo in a randomised, double-blind, placebo-controlled, cross-over trial. Treatment with simvastatin was associated with a significant reduction in serum anti-HSP60, 65, and 70 titers in the dyslipidaemic patients (10%, 14%, and 15% decrease, respectively) (p<0.001) (153). We believe that this important study might provide a promising starting point to clarify the role of anti-HSP antibodies as prognostic biomarkers in atherosclerosis.

### Anti-phospholipid antibodies (aPL)

aPL include lupus anticoagulant, anticardiolipin antibodies (aCL) (IgG and IgA, and IgM) and β2GPI (IgG and IgA, and IgM). High titres of these auto-antibodies have been identified in several autoimmune diseases and underlie the so-called “antiphospholipid syndrome” (APS) (154). Despite their name, these antibodies do not only bind phospholipids, but also plasma proteins that have an anionic surface.

Lupus anticoagulant acts as an inhibitor of coagulation, prolonging phospholipid-dependent coagulation time. β2GPI is present at high concentrations in the blood stream and is expressed by many cell populations, including endothelial cells, lymphocytes

### Table 6: Anti-phospholipid antibodies and cardiovascular risk.

<table>
<thead>
<tr>
<th>Author [Reference]</th>
<th>Year</th>
<th>Study design (n) and pathology</th>
<th>Target (aPL)</th>
<th>Outcomes and relationship</th>
<th>Prediction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soltész P. et al. [157]</td>
<td>2003</td>
<td>Retrospective observational (1,519) aPLS and LES</td>
<td>Secondary to LES aPLS vs PaPLS</td>
<td>Most strokes in patients with secondary aPLS compared to primitive aPLS</td>
<td>OR 1.68 (1.10–2.58) p=0.04</td>
</tr>
<tr>
<td>Koskenmies S. et al. [158]</td>
<td>2004</td>
<td>Cross-sectional (292) LES</td>
<td>aCL (IgG, IgA, IgM) anti β2GPI</td>
<td>aCL IgG are more related to higher incidence of TE</td>
<td>OR: 3.9 (1.8–8.0) p&lt;0.005</td>
</tr>
<tr>
<td>Lopez L.R. et al. [159]</td>
<td>2006</td>
<td>Case-control (30/27) LES</td>
<td>β2GPI/oxLDL complexes, IgG or IgM antibodies</td>
<td>High levels are not related to IMT</td>
<td>Pearson’s product moment correlation β2GPI/oxLDL: r=0.002 p=0.456 IgG anti- β2GPI/oxLDL: r=0.205 p=0.271 IgM anti- β2GPI/oxLDL: r=0.079 p=0.128</td>
</tr>
<tr>
<td>Pereira I. et al. [160]</td>
<td>2008</td>
<td>Case-control (71/53) AR</td>
<td>IgG aCL IgMaCL IgG anti β2GPI</td>
<td>Not found a correlation between IMT and antibodies (IgG aCL, IgG aCL, IgG anti β2GPI )</td>
<td>Spearman correlation coefficient IgG aCL r=0.09 p=0.47 IgM aCL r=0.15 p=0.21, IgG anti β2GPI r= 0.07 p=0.57,</td>
</tr>
<tr>
<td>De Laat B. et al. [161]</td>
<td>2009</td>
<td>Cross-sectional (243) APL</td>
<td>Anti β2GPI IgG</td>
<td>High levels are associated to previous obstetric complication history</td>
<td>OR: 3.5 (2.3–5.4) p&lt;0.001</td>
</tr>
<tr>
<td>Gustafsson J. et al. [162]</td>
<td>2009</td>
<td>Prospective observational (182) LES</td>
<td>Any aPL</td>
<td>High levels are associated to endothelial activation marker</td>
<td>HR 4.23 (1.56–14.83) p=0.003</td>
</tr>
<tr>
<td>Gustafsson J. et al. [163]</td>
<td>2012</td>
<td>Prospective observational (208) LES</td>
<td>aCL (IgM + IgG) IgG anti β2GPI</td>
<td>Presence of aPL is a marker of cardiovascular death</td>
<td>aCL RR 2.8(1.0–8.2) p=0.05 Anti β2-GPI IgG RR 3.4(1.2–9.7) p=0.03</td>
</tr>
</tbody>
</table>

aPLS: anti phospholipid syndrome; LES: lupus erythematosus systemicus; PaPLS: primitive anti phospholipid syndrome; aCL: anti cardiolipin antibodies; anti β2GPI: anti β2 glycoprotein I; TE: thrombotic events; oxLDL: oxidised low-density lipoprotein IMT: intima media thickness; aPL: anti phospholipid antibodies.

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Thrombosis and Haemostasis 109.5/2013
and monocytes. It binds negatively charged molecules, including phospholipids, heparin and oxLDL, but also the surface of activated platelets and the membrane of apoptotic cells (155). Antibody binding to β2GPI induces conformational changes and exposure of cryptic epitopes that, in turn, promote its binding to aPL (156).

The role of aPL in atherothrombosis is controversial. In patients with primary or secondary APS, the presence of aPL (β2GPI and aCL) was positively associated with an increased cardiovascular risk, that is statistically significant for thrombotic events such as MI and stroke (Table 6).

However, unexpectedly, two large clinical studies did not confirm these results. The Hopkins Lupus Cohort, a longitudinal study of a population of 1,811 patients with SLE, failed to demonstrate a statistically significant correlation between aPL (aCL and β2GPI) and cardiovascular events and with the atherosclerotic plaque size (164-166). Similar results were obtained in a study by Pahor et al. (167), where high levels of aPL (especially aCL and anti-IgA β2GPI) initially seemed to correlate with accelerated atherosclerosis in a cohort of patients with rheumatoid arthritis (RA) as compared to sex- and age-matched controls (68 pre-menopausal, normotensive and non-diabetic patients with RA vs 40 age- and sex-matched controls). However, subsequent patient follow-up for up to 5.5 years, did not confirm the preliminary data. Although an accelerated atherosclerosis in patients with RA as assessed in terms of IMT and plaque size could be documented, aPL (aCL and especially anti-β2GPI IgA) could not be confirmed as an independent risk factor for atherosclerosis (168).

Artenjak et al. (169) reported on the correlation between aPL and cardiovascular risk in non-autoimmune settings. This study was preceded by a report by Brey et al., who could only detect a statistically significant positive correlation between aCL IgG and stroke (170). Taken together, these results did not demonstrate a clear association between aPL and acute cardiovascular events.

Figure 2: Direct pro-atherosclerotic activities mediated by auto-antibodies on both immune and vascular cells. ROS: reactive oxygen species; MPO: myeloperoxidase; MMP: matrix metalloproteinases; iNOS: intracellular nitric oxide synthetase; TNF: tumour necrosis factor; IL: interleukin; G/M/GM-CSF: granulocyte/macrophage/granulocyte-macrophage colony-stimulating factor; ICAM-1: intercellular adhesion molecule 1; VCAM-1: vascular cell adhesion molecule 1; MCP: monocyte chemotactic protein-1; PDGF: platelet-derived growth factor; VEGF: vascular endothelial growth factor.
Conclusions

Evidence from both basic research and clinical studies indicates that humoral autoimmunity has a dual role in atherogenesis and plaque vulnerability, with some antibodies exerting protective effect and others increasing cardiovascular risk. The reasons underlying this discrepancy are unknown but could be related to the nature of the epitope targeted by the autoantibody, to the antibody subclass, or to the interaction with different Fcγ receptors. While the results available for anti-oxLDL and anti-APS antibodies appear as rather conflicting, the existing data concerning anti-HSP and anti-apoA-1 antibodies are in favour of a pro-inflammatory and pro-atherogenic role of these antibodies mediated through the engagement of innate immune receptors. Thus, these auto-antibodies could represent potential new therapeutic targets. On the other hand, anti-PC antibodies seem to be anti-inflammatory and atheroprotective, raising the possibility that increasing anti-PC IgM (or anti-oxLDL) titres through active or passive immunisation could have therapeutic value in CVD. Overall, a clearer definition of the pathogenetic role of auto-antibodies in CVD and a better understanding of their mode of action are needed. In particular, the cellular mediators activated by auto-antibodies will have to be fully characterised (▶ Figure 2). Importantly, substantial pathophysiological differences between IgM and IgG auto-antibodies have been shown recently. While a potentially direct anti-atherosclerotic role for IgM antibodies has been shown in vivo (171), involving the clearance of apoptotic cells and the release of protective soluble mediators from intraplaque phagocytes (38, 172) (▶ Figure 3), only epidemiological evidence and in vitro studies investigated IgG antibodies in atherosclerosis. In addition, the analytical methods need to be improved, standardised and possibly adapted to provide rapid results in emergency settings. Finally, it will be important to demonstrate in randomised clinical trials whether the presence of some autoantibodies could improve current cardiovascular risk risk stratification approaches and therapeutic algorithm, both in primary and secondary prevention. For those reasons, we believe that the role of autoimmunity in the atherosclerotic domain will represent a hot-topic field for future translational investigations.

Acknowledgements

This research was funded by EU FP7, Grant number 201668, AtheroRemo to Dr. F. Mach. This work was also supported by the Swiss National Science Foundation Grants to Dr. F. Mach (#310030-118245), Dr N. Vuilleumier (#310030-140736), and to Dr. Montecucco (#32003B-134963/1) and by the Italian Ministry of Health (GR-2008-1135635 to A.N.).

What is known about this topic?
• Atherosclerosis is a low-grade inflammatory disease involving both immune and vascular cells and soluble mediators.
• Emerging evidence from both basic research and clinical studies suggests that B cells and antibodies might play a direct pathogenetic role in atherogenesis.

What does this paper add?
• Some auto-antibodies (such as anti-apoA-1 and anti-HSP) have been associated with an increased risk of acute cardiovascular events in human beings.
• Larger clinical trials are needed to validate auto-antibodies as prognostic and therapeutic biomarkers of accelerated atherogenesis and plaque vulnerability.
• Toll-like receptors might represent an important pathophysiological pathway in the autoantibody-mediated immune response in atherogenesis.
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