Atherosclerosis

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Is atherosclerosis a cellular or humoral mediated autoimmune disease?

Atherosclerosis is a complex and indolent histopathological process, which is considered to be the most common underlying process in cardiovascular morbidity and mortality. In recent years it has become apparent that in addition to the traditional risk factors for atherosclerosis, this condition is associated with infectious, inflammatory, and autoimmune factors.[1] Atherosclerosis fulfils all the four criteria delineated by Witebsky and Rose to define a condition as autoimmune in nature, and all arms of the immune system (including cellular components, autoantigens, and autoantibodies) play a part in atherosclerosis.[2] Thus one of the questions raised is whether atherosclerosis, as other autoimmune conditions, is mainly a humoral or cellular mediated autoimmune disease.

INVOLVEMENT OF CELLULAR COMPONENTS IN ATHEROSCLEROSIS

There is good evidence to suggest that cellular components of the immune system are involved in atherosclerosis. Studies using transgenic murine models show recruitment of mononuclear leucocytes through vascular leucocyte adhesion molecules and chemokines, differentiation of monocytes to macrophages, and endocytosis through scavenger receptors of oxidised LDL (oxLDL) in atherogenesis. The importance of T cells in atherosclerosis is emphasised in a study in which CD4+ and CD8+ T cell depletion reduced fatty streak formation in C57BL/6 mice, indicating that T cells aggravate fatty streak formation.[3] A recent study emphasises also the importance of specific lymphocytes. Lymphocytes obtained from LDL receptor deficient mice immunised with β_2 glycoprotein I (β_2GPI) were transferred intraperitoneally into syngeneic mice, producing larger fatty streaks in the recipients than in mice receiving lymphocytes from control mice.[4] T cell depletion of lymphocytes failed to induce this effect. Hence, β_2GPI reactive T cells could promote atherogenesis.

As for other autoimmune diseases, it is also of interest to determine whether atherosclerosis is mainly a Th1 or Th2 mediated condition. Although it is not yet sufficiently clear, a recent study favours the former. In that study apo-E deficient mice were treated with an inhibitor of the Th1 differentiation pathway (PTX) or control for 12 weeks.[5] The former mice reduced the size of atherosclerotic lesions by 60%, and developed lesions that were limited to the degree of fatty streaks. The control mice developed mature fibrofatty atherosclerotic lesions. Of note is that the lesion size correlated with the proportion of interferon γ (IFNγ) positive T cells. In vitro addition of PTX to cultured spleen cells did not modify the production of IFNγ, but increased the production of interleukin 10 (IL10) by T cells, indicating that PTX does not suppress IFNγ production but rather blocks Th1 polarisation while promoting Th2 polarisation.[5] This study suggests that the Th1 immune response associated with atherosclerosis is deleterious. None the less, it seems that atherosclerosis, like other autoimmune conditions, involves both Th1 and Th2 related cytokines (an example with IL4 is provided below).

ASSOCIATION BETWEEN AUTOANTIBODIES AND ATHEROSCLEROSIS

Apart from the above data on the involvement of cellular components in atherosclerosis, the evidence also suggests an association between autoantibodies and atherosclerosis (reviewed by Shoenfeld *et al*[6]). When a pathological state is multifactorial, it is not surprising that many autoantibodies play a part in its pathogenesis. A brief summary of the most investigated autoantibodies follows.

Anti-oxLDL antibodies

Oxidation of LDL probably has an important role in the pathogenesis of atherosclerosis. It is not yet firmly established whether the immune response to oxLDL is pro-atherogenic or anti-atherogenic in vivo, or, alternatively, whether it is merely an epiphenomenon for the presence of oxLDL. Anti-oxLDL antibodies are raised in patients with early onset peripheral vascular disease,[7] severe carotid atherosclerosis,[8] and angiographically verified coronary artery disease.[9 10] In addition, raised levels of oxLDL antibodies were predictive of carotid atherosclerosis progression,[11] and myocardial infarction occurrence and mortality.[12] We have recently found raised levels of these antibodies in patients with coronary artery disease compared with healthy controls, regardless of the amount of coronary calcification.[13] None the less, reduction of oxLDL antibody levels was reported in elderly patients with ischaemic stroke,[14] and in animal models immunisation with oxLDL resulted in suppression rather than aggravation of early atherogenesis.[15 16]

Anti-heat shock protein (HSP) 60/65 antibodies

HSPs are a family of proteins that show a highly homologous sequence between different species, from bacteria to man. Sonographic assessment of carotid atherosclerotic lesions showed that subjects with such lesions had significantly raised levels of anti-HSP65 antibodies compared with controls.[17] The possible pathogenicity of anti-HSP antibodies could be learnt from animal models. For example, rabbits that were immunised with material containing HSP65, either in the form of mycobacteria or recombinant HSP65 alone, developed atherosclerotic lesions.[18] In another study C57BL/6 mice were injected with either HSP65, HSP65-rich *Mycobacterium tuberculosis*, or phosphate buffered saline (PBS). Early atherosclerosis was significantly enhanced in mice fed a high cholesterol diet that were immunised with HSP65 or *Mycobacterium tuberculosis*, compared with the mice injected with PBS.[19] Recently, it has been suggested that IL4 has a crucial role in the progression of early atherosclerosis mediated by inflammation, as IL4 knockout mice immunised with HSP65 had significantly less fatty streak formation than lesions in C57BL/6 mice.[20]

Anticardiolipin antibodies

The pathogenic potential of anticardiolipin in atherosclerosis has been demonstrated in a mouse model in which immunisation with this antibody resulted in the development of high titres of mouse anticardiolipin and increased atherosclerosis compared with controls.[21] The presence of high levels of anticardiolipin was found to be an independent risk factor for myocardial infarction or cardiac death in middle aged men.[22] We recently found raised levels of anticardiolipin antibodies in patients with coronary artery disease compared with healthy controls, regardless of the amount of coronary calcification,[13] and these antibodies were raised in patients with typical chest pain compared with patients with atypical or no chest pain.[23]

Anti-β_2GPI antibodies

β_2GPI is currently considered by most researchers as the autoantigen in antiphospholipid syndrome rather than a cofactor. Its association with atherosclerosis has been reviewed by Shoenfeld *et al*.[24] Probably the most important finding comes from animal models, in which immunisation of two different strains of mice with β_2GPI led to the production of anti-β_2GPI antibodies and acceleration of atherosclerosis.[25 26]

INCLUSION OF BOTH CELLULAR AND HUMORAL COMPONENTS IN ATHEROSCLEROSIS

Atherosclerosis may also include both cellular and humoral components in its underlying pathophysiology, as occurs in many autoimmune diseases. A recent study determined the role of cellular and humoral immune responses to HSP65 in murine atherosclerosis. Lymph node cells, splenocytes, and IgG were obtained from the LDL receptor deficient mice immunised with HSP65.[27] Adoptive transfer of HSP65 reactive lymph node cells increased fatty streak formation in comparison with mice treated with bovine serum albumin primed cells. Similarly, repeated intraperitoneal administration of IgG from the serum of HSP65 immunised mice enhanced fatty streak formation in mice in comparison with controls.[27] This study provides direct evidence for the pro-atherogenic properties of cellular and humoral immunity to HSP65, and raises the possibility that both arms of the immune system have a synergistic pro-atherogenic effect.

ANTINUCLEAR ANTIBODIES IN ATHEROSCLEROSIS

In this issue of the *Annals* Grainger and Bethell provide evidence for the presence of raised levels of antinuclear antibodies (ANA) in patients with radiological evidence of advanced atherosclerosis.[28] Their discussion includes the possibility that these antibodies are merely an epiphenomenon or, alternatively, that they have a pathogenic role in atherosclerosis. Even though this study naturally does not provide answers to that crucial question, their finding itself is important and raises several thoughts and assumptions.

"Are ANA markers of advanced atherosclerosis or do they participate in its acceleration?"

The presence of ANA is the hallmark of systemic lupus erythematosus, and they are found in various frequencies in other autoimmune diseases as well. The accelerated atherosclerotic state found in patients with systemic lupus erythematosus and antiphospholipid syndrome might result from the higher frequency of traditional risk factors in these patients as well as from the presence of anticardiolipin and anti-β_2GPI antibodies.[29] However, ANA might also contribute to the accelerated atherosclerosis found in these patients. Further, the association of ANA with atherosclerosis raises the possibility that these antibodies play a part in atherogenesis or arteriosclerosis in other autoimmune and inflammatory states, such as vasculitides. As for other autoantibodies, the frequency of ANA is significantly higher in elderly people (10–37%) than in the young (0–6%).[30] As coronary artery disease resulting from atherosclerosis is also found mostly in the elderly, ANA may be markers of advanced atherosclerosis or, alternatively, participate in its acceleration.

It is obvious now that many autoantibodies, autoantigens, and cellular components of the immune system are involved in atherogenesis and progression of atherosclerosis. The association of ANA with atherosclerosis raises again the "cause and effect" question that is so common in autoimmunity and in the immune aspects of atherosclerosis in particular. The role of ANA in atherosclerosis should be further studied, but the association of ANA with atherosclerosis provides additional evidence for the autoimmune nature of this condition.

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