IL-9: a new culprit in atherosclerosis?

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This editorial refers to ‘IL-9 aggravates the development of atherosclerosis in ApoE−/− mice’ by W. Zhang et al., pp. 453–464.

Atherosclerosis is a chronic inflammatory disease of the arterial wall driven by innate and adaptive immunity. Atherosclerotic lesions contain a variety of cells including smooth muscle cells, macrophages, and T lymphocytes, as well as other inflammatory cells, such as mast cells and NKT cells. This collection of cells promotes the development of atherosclerosis through the production of inflammatory factors. Among CD4+ T cells, Th 1 cells have been shown to exert proatherogenic effects, whereas regulatory T cells (Treg) display antiinflammatory properties and the role of Th2 and Th17 cells remains unclear. The differentiation of Th cells into effector subsets that secrete specific pro- or anti-inflammatory cytokines is critical for the initiation and progression of atherosclerosis. The development of Th subpopulations is dependent on the expression of lineage-specific transcription factors that are regulated by specific cell–cell interactions, as well as by the cytokine environment. Among these is the recently described Th9 cell subset that preferentially secretes IL-9. IL-9 is a pleiotropic cytokine that is highly expressed in diseases where Th2 cytokines, including IL-4, IL-5, and IL-13, are up-regulated, which explains why it was initially considered as a Th2 cytokine. In addition to Th9 or Th2 cells, mast cells, eosinophils, innate lymphoid cells (ILCs), and NKT cells have been shown to be sources for this cytokine.

The differentiation of Th9 cells is dependent on transcription factors that include PU.1, downstream of transforming growth factor-β (TGF-β) signals, and IL-4-activated signal transducer and activator of transcription (STAT)-6 that promotes the expression of interferon regulatory transcription factor (IRF)-4. Other factors such as IL-1β or IL-2 have been shown to play important roles for inducing IL-9 production in other cell types aside from Th9 such as, respectively, Th17 and ILC.

Th9 cells also produce IL-21, and the secretion of both IL-9 and IL-21 is enhanced by IL-1β that activates STAT1 and promotes subsequent expression of IRF-1. Moreover, the engagement of OX40, a member of the tumour necrosis factor (TNF) receptor superfamily, by its ligand OX40L, expressed on antigen-presenting cells, is a powerful inducer of Th9 cells. The OX40-induced Th9 cell differentiation is involved in airway inflammation in vivo. Interestingly, previous studies have shown the importance of the OX40/OX40L pathway in atherosclerotic development. Th9 cells, and more generally IL-9, have been described as having proinflammatory properties. The majority of cells, such as Th17, ILC, and mast cells, which produce IL-9, also express IL-9 receptor (Figure 1), suggesting autocrine loop effects. IL-9 was initially shown to have proliferative effects on T cells, and thereafter on other cell types, including mast cells, through activation of the Janus kinase (JAK)-STAT pathway. By using IL-9-deficient mice, the importance of IL-9 was shown in pulmonary mastocytosis and goblet cell hyperplasia. Moreover, IL-9 blockade revealed the pathogenic role of IL-9 as a Th17-derived cytokine that regulates IL-6–producing macrophages in the CNS, as well as mast cell numbers in the regional lymph nodes in a mouse model of multiple sclerosis. Also, overexpression of IL-9 transgene in lungs caused an inflammatory response associated with the expression of Th2 cytokines, responsible for the infiltration of inflammatory cells in a murine model of asthma. Accordingly, through these actions, IL-9 appears to be a pleiotropic cytokine that plays pathogenic roles in a broad range of diseases, including asthma, allergy, and autoimmune diseases, making this cytokine an interesting therapeutic target for new drug development. In the context of cardiovascular disease, it is noteworthy that previous studies reported increased IL-9 plasma levels in patients with acute coronary syndrome, as well as in patients with coronary and carotid atherosclerosis. However, the direct implication of IL-9 and Th9 cells in atherosclerosis had not yet been studied.

Zhang et al. report for the first time that IL-9 plays a significant role in atherosclerosis. They showed that anti-IL-9 antibody treatment in ApoE−/− mice decreased T-cell and macrophage infiltration within atherosclerotic lesions, and limited the development of atherosclerosis. Conversely, administration of recombinant IL-9 increased inflammatory cell infiltration and plaque size, indicating that IL-9 exerts proatherogenic effects. In the future, it would be of great interest to use IL-9 or IL-9 receptor-deficient mice on either an ApoE−/− or Ldlr−/− background as a continuation of these interesting findings on the involvement of IL-9 in atherosclerosis. This would be particularly interesting since Zhang et al. found no significant differences in IL-9 mRNA between small and large atherosclerotic lesions, nor between ApoE−/− mice on Chow or high fat diet, which indicates that IL-9 was not necessarily up-regulated in atherosclerotic mice. To identify the precise mechanisms whereby IL-9 aggravates atherosclerosis, it would also be important to more carefully characterize the cells that produce IL-9 in hypercholesterolaemic mice. In the present study, the majority of IL-9 produced by splenocytes did not originate from...
T or B cells, suggesting the involvement of other cell types, including NKT cells, ILC, mast cells, or eosinophils, which are known to express IL-9.3

As regards the mechanisms involved in the proatherogenic effects of IL-9, Zhang et al. provided evidence that IL-9 up-regulated vascular cell adhesion molecule (VCAM-1) expression in murine aortic endothelial cells, and that anti-VCAM-1 antibody treatment partially abrogated the IL-9-induced increase in atherosclerotic lesions. IL-9-induced up-regulation of VCAM-1 likely increased inflammatory cell recruitment and infiltration into atherosclerotic lesions, resulting in exacerbation of atherosclerosis. They also found that the concentration of plasma levels of soluble VCAM-1 was significantly increased in patients with acute coronary syndromes and was significantly correlated with IL-9. In vitro, IL-9 induced VCAM-1 expression through a STAT3-dependent pathway in endothelial cells. Therefore, it would be interesting to examine STAT3 activation in vivo following neutralization or activation of IL-9 pathway. However, as the effects of IL-9 were only partially abolished in the absence of VCAM-1, other mechanisms remain to be identified.

Previous studies highlighted the importance of the synergy between IL-9 and Th2 cytokines (IL-4, IL-5, and IL-13) in promoting the infiltration of inflammatory cells and disease development.14 This could be also the case in atherosclerosis, although the role of Th2 cytokines in atherosclerosis has not been clearly established. IL-4 displaying neutral or proatherogenic activities and IL-5 and IL-13 being anti-atherogenic.1

In addition, IL-9 was previously shown to induce inflammatory response by increasing IgE serum levels.9 Interestingly, IgE has been recently shown to exert deleterious effects in atherosclerosis.17,18 Moreover, IL-9 can induce the expression of the chemokine CCL20, promoting the attraction of CCR6+ dendritic cells (DCs), which elicited an effective antitumor response.19 Interestingly, studies using Apoe-/- mice have implicated the chemokine receptor CCR6 and its ligand CCL20 as a non-redundant ligand-receptor pair with proatherogenic effects, potentially operating on several leucocyte subtypes, including DC, T cells, NKT cells, and neutrophils.20 It would be of great interest to explore in the future the effects of IL-9 on Th2-associated cytokines (especially IL-4) and chemokines (CCL17, CCL22, and CCL20), as well as on IgE levels in atherosclerosis (Figure 1).

Collectively, the present finding by Zhang et al. opens new perspectives that will likely be extended to provide a solid proof of concept for involvement of IL-9 and Th9 cells in atherosclerosis, and future studies will determine the exact mechanisms involved in IL-9-mediated proatherogenic effects.

References

Figure 1 Potential effects of IL-9 in atherosclerosis. (Right) Cells expressing IL-9, as well as factors (IL-4, TGF-β, and IL-1β) and co-stimulatory molecules (OX40–OX40L) involved in the induction of IL-9 gene expression. (Left) IL-9 can promote atherosclerosis by increasing IgE and Th2 cytokine (IL-4) and chemokines (CCL17, CCL20, and CCL22).

Th9 cells seem also to contribute to allergic diseases by promoting the expression of Th2-associated chemokines, including CCL17 and CCL22.16 These chemokines have been shown to exert deleterious effects in atherosclerosis.17,18 Moreover, IL-9 can induce the expression of the chemokine CCL20, promoting the attraction of CCR6+ dendritic cells (DCs), which elicited an effective antitumor response.19 Interestingly, studies using Apoe-/- mice have implicated the chemokine receptor CCR6 and its ligand CCL20 as a non-redundant ligand-receptor pair with proatherogenic effects, potentially operating on several leucocyte subtypes, including DC, T cells, NKT cells, and neutrophils.20