Fine tuning effector and regulatory T-cell dynamics: a novel tool for plaque regression?

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This editorial refers to 'Regression of atherosclerosis with anti-CD3 antibody via augmenting a regulatory T-cell response in mice' by T. Kita et al., pp. 107–117, this issue.

T cells are present during all stages of atherosclerosis development, and play an essential role in the initiation and progression of plaques. The majority of T lymphocytes in atherosclerotic lesions are CD4+ cells with a phenotype characteristic of the T-helper 1 (Th1) subset, producing high levels of IFN-γ.1,2 The generation of antigen-specific effector T lymphocytes during atherogenesis is likely to occur through self-antigens such as apolipoprotein (apo) B100 and oxidized low-density lipoprotein (LDL).1 Whereas most T effector cell responses have been suggested to aggravate atherosclerosis, regulatory T cells (Tregs) are increasingly recognized as potent atheroprotective players.3 Once activated, Tregs actively suppress responses of effector T cells by contact-dependent mechanisms or through the secretion of cytokines IL-10 and TGF-β. TGF-β plays a dual role in Tregs, as it regulates their differentiation and function but also is secreted as a potent effector cytokine. The functional role of Tregs in atherosclerosis models has been investigated either by direct adoptive transfer or by indirect measures such as vaccination.4,5 Another strategy is the administration of anti-CD3 antibodies (anti-CD3), either systemically or orally, which was shown to reduce atherosclerosis development at early and advanced stage.6,7 The anti-atherogenic effect of anti-CD3 involves TGF-β-dependent mechanisms via expansion and activation of Tregs.

A recent study based on genetic deficiency of Tregs revealed an unexpected novel interaction of Tregs with lipid metabolism. Depletion of Tregs led to doubling of atherosclerotic lesion size and a profound increase in circulating cholesterol concentration, mainly in the very low-density lipoprotein (VLDL) fraction.8 It was further demonstrated that expression of a receptor important in the uptake of cholesterol-rich lipoproteins, sortilin-1, was decreased in the liver and is likely responsible for decreased clearance of pro-atherogenic particles leading to elevated blood cholesterol levels and enhanced atherosclerosis. These findings raise the question which property of Tregs actually controls atherosclerosis at least in this model on LDL receptor-deficiency (LDLR−/−) background, the anti-inflammatory or the lipid-altering function or both. It deserves further investigations to what extent the lipid-altering function might have a therapeutic relevance.

There is evidence from animal models as well as clinical studies that sustained plasma lipid lowering promotes plaque regression and leads to a more stable plaque phenotype.9 Underlying mechanisms promoting plaque regression involve decreased apoB-lipoprotein retention within the arterial wall, cholesterol efflux from the plaques, reduced recruitment of monocytes and emigration of lesional macrophages followed by entry of healthy phagocytes that remove necrotic debris and other plaque components. Different experimental models for plaque regression have been established, including infusion of HDL, overexpression of ApoAI, transplantation of plaque-bearing aortic arches from hypercholesterolemic apoE-deficient mice into recipient mice with a normolipidemic plasma environment, as well as dietary reversal of hypercholesterolaemia.10 Most studies investigated the effects on plaque macrophages and changes in pro- or anti-inflammatory/reparative macrophage phenotype ratios in plaque regression. However, little is known about a potential role of T-cell-mediated responses in this process. Recently, Maganto-Garcia et al. reported dynamic changes of Treg numbers in LDLR−/− mice during prolonged cholesterol-diet feeding, resulting in a decreased ratio between Tregs and effector T cells.11 After an initial increase, Treg numbers decreased in aortas due to apoptotic loss and reduced homing capacity manifested by reduced ability to adhere to endothelium, while effector T-cell numbers continuously increased. The most important observation was that reversal of hypercholesterolaemia prevented the loss of lesional Tregs.

Now, Kita et al. pursued the idea that Tregs are actively involved in the process of plaque regression.12 They performed systemic administration of anti-CD3 in a model of plaque regression mediated by dietary reversal of hypercholesterolaemia. Injection of anti-CD3 for 5 consecutive days at the time point of dietary switch induced rapid plaque regression with concomitant reduction in effector T cell and increase of Treg numbers within lymphoid organs and aortas (Figure 1). The plaques had a more stable phenotype with reduced macrophage content, but higher smooth muscle and increased collagen content. To strengthen the finding that Treg suppressive activity is crucially involved in anti-CD3-stimulated

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Figure 1 Dynamics of Treg and effector T cells in hypercholesterolaemic mouse models of atherosclerosis and plaque regression induced by dietary reversal of hypercholesterolaemia and anti-CD3 administration. Numbers of Tregs within aortas increase in the early phase of atherogenesis, but decrease over time with growing lesion size. The decrease in aortic Tregs is a consequence of enhanced frequency of Treg apoptosis and reduced migratory capacity, thereby leading to increasing numbers of Tregs in the spleen. On the other hand, there is a continuous increase in the number of CD4+ effector T cells (Teff) within the aorta, thereby resulting in an unfavourable ratio between Tregs and effector T cells (Treg:Teff). Reversal of hypercholesterolaemia can prevent the loss of Tregs within the plaque. Therapeutic intervention with anti-CD3 antibody enhances plaque regression, possibly by transient modulation of Teff function and/or regulatory T-cell activation. It is unclear whether there is a direct cross-talk between macrophages and Tregs that contributes to plaque regression and stabilization.
plaque regression and stabilization, the authors depleted Tregs by anti-CD25 injection, which abolished the beneficial effect of anti-CD3. A limitation is, however, that treatment with anti-CD25 does not only deplete Tregs but other CD25+ immune cells as well and therefore does not unambiguously proof the specific role of Tregs in this model. In the same context, a different strategy for Treg expansion based on injection of IL-2 complexes failed to induce plaque regression despite increases in Treg numbers within lymphoid organs. Anti-CD3 induces a sustained immunosuppressive effect as it modulates the CD3/T-cell receptor complex. We previously observed that hypercholesterolaemic LDLR−/− mice treated with the same dose of anti-CD3 dramatically reduced the number of CD3 and TCR expressing cells in blood and lymphoid organs, which only recovered to normal levels after more than 3 weeks. Thus, it is possible that transient immunosuppression rather than long-term immunomodulation by Tregs essentially contributes to the rapid plaque regression found after 2–4 weeks of dietary switch. Whether the reduction in macrophage plaque content is a consequence of the reduced pro-inflammatory environment and thus less recruitment, inhibition of local proliferation, apoptotic clearance, or egression deserves further investigation.

Finally, the authors did not observe differences in lipid profiles between mice treated with anti-CD3 or isotype control, suggesting that direct actions of Tregs on lipid metabolism are not involved in this model. However, it remains unclear how Tregs actually contribute to plaque regression, and whether TGF-β is involved in this process. It further remains to be investigated whether the observed effect is dependent on LDLR deficiency or if it is still operative when the main receptor for uptake of cholesterol, the LDL receptor, is functional. Despite these open questions, this study enlarges our current understanding of the complex role of T-cell-mediated immune mechanisms involved in atherosclerosis and may open new avenues for therapeutic intervention strategies for plaque stabilization and regression.

References