Cardiovascular diseases mainly caused by atherosclerosis lead death statistics in Western societies. Atherosclerosis, a chronic inflammation of the vessel wall, is most likely initiated by endothelial dysfunction followed by a constant influx of mononuclear immune cells into the intimal layer. The latter fosters growth of atherosclerotic plaques leading to vessel narrowing and occlusion. Rupture of atherosclerotic lesions can cause myocardial infarction and stroke.1 Myeloid cells, especially macrophages and their secretion products, are known to play an important role in the pathophysiology of atherosclerosis; however, some mediators exert multiple roles and might have to be revisited. In this context, Subramanian et al2 revealed that the hematopoietic growth factor granulocyte-macrophage colony-stimulating factor (GM-CSF; gene name Csf2), to date mostly known for its role in myeloid cell development, growth, and stability, depending on their polarization state and secretion profile.12,13 The secretion profile of M1-polarized macrophages comprises, eg, IL-23 and other proinflammatory mediators12,13, whereas GM-CSF does not only facilitate monocyte to macrophage differentiation but also re-lease by lesional macrophages.2,15 Yet, mouse studies on the role of macrophage apoptosis reveal conflicting results. Some studies suggest a beneficial role of macrophage apoptosis leading to decreased plaque development,4,6 whereas others show that apoptotic macrophages promote necrotic core formation and thereby lesion growth.8,9 In general, it is now agreed that macrophage apoptosis in early lesion development may limit plaque growth by a general dampening of the inflammation. In contrast, advanced lesions depend on adequate apoptotic cell clearance and, therefore, constant macrophage apoptosis and ineffective cell removal promote plaque growth and vulnerability.3,10,11 Undoubtedly, macrophages take over various roles in plaque development, growth, and stability, depending on their polarization state and se cretion profile.12,13 The secretion profile of M1-polarized macrophages comprises, eg, IL-23 and other proinflammatory mediators13,14, whereas GM-CSF does not only facilitate monocyte to macrophage differentiation but also released by lesional macrophages.2,15 Yet, mouse studies on the general role of GM-CSF in atherosclerosis have revealed conflicting results. Although Csf2 knockout mice on the apolipoprotein E (ApoE) background displayed an increase in lesion size (aortic root) after 12 weeks of Western diet,15 injection of GM-CSF into Apoe mice did also enhance plaque formation.16 Furthermore, Shaposhnik et al17 showed a decrease in...
plaque burden (aortic root) in Ldlr<sup>-/-</sup> Csf2<sup>-/-</sup> mice after 3 months of Western diet. In addition, studies modulating the GM-CSF receptor did also reveal inconsistent outcomes. However, the highlighted study and the work by Shaposhnik et al point in the same direction depicting a moderate or no reduction in lesion size and no change in the number of lesional macrophages on the Ldlr<sup>-/-</sup> background, but a more stable plaque phenotype in case of GM-CSF deficiency. Taken together, the above-described mouse models underscore again that changes in systemic lipid profiles may significantly affect the overall immune reaction of a single protein. At that point, one could further speculate that the GM-CSF deficiency in Apoe<sup>-/-</sup> mice leads to an increase in lesion formation because reduced expression of ATP-binding cassette transporter 1 (as GM-CSF has been reported to induce its expression) together with the anyway impaired cholesterol efflux and increased myeloproliferation in case of Apoe<sup>-/-</sup> mice strongly assume that the increased plaque stability in their vaccinated Ldlr<sup>-/-</sup> mice partly depends on IL-23 neutralization. Hence, the presented study revealing a Th17/IL-17–independent role of IL-23 in atherosclerosis supports this finding.

Many mechanisms are discussed to initiate macrophage apoptosis in advanced lesions, eg, cholesterol overload or endoplasmatic reticulum stress inducing the unfolded protein response. In this context, earlier work by the authors of the study discussed here showed that Bcl-2 is an important regulator of lesional macrophage apoptosis and that macrophage-specific deletion of Bcl-2 (LysM-Cre Bcl2-flox on Apoe<sup>-/-</sup> background) resulted in a more vulnerable plaque phenotype. In general, Bcl-2 has been implicated as an important therapeutic target for the treatment of atherosclerosis. Taking this observation further, Subramanian et al now introduce a possible mechanism of Bcl-2 degradation by GM-CSF–induced IL-23 signaling. Simultaneously, IL-23 signaling also increases reactive oxygen species release within these apoptotic lesional macrophages.

**Concluding Remarks**

Understanding the mechanisms favoring macrophage apoptosis in advanced atherosclerotic lesions may reveal promising therapeutic targets for high-risk patients with cardiovascular disease. In support of this idea, vulnerable necrotic human plaques display increased macrophage apoptosis. Along this road, the discussed study highlights IL-23 as such a player. In view of the already ongoing clinical trials for IL-23–blocking antibodies in other chronic inflammatory diseases, this could be a favorable and soon available therapy for patients with elevating plaque burden (aortic root) in Western diet. In addition, studies modulating the GM-CSF receptor did also reveal inconsistent outcomes.16,18,19 However, the highlighted study and the work by Shaposhnik et al point in the same direction depicting a moderate or no reduction in lesion size and no change in the number of lesional macrophages on the Ldlr<sup>-/-</sup> background, but a more stable plaque phenotype in case of GM-CSF deficiency. Taken together, the above-described mouse models underscore again that changes in systemic lipid profiles may significantly affect the overall immune reaction of a single protein. At that point, one could further speculate that the GM-CSF deficiency in Apoe<sup>-/-</sup> mice leads to an increase in lesion formation because reduced expression of ATP-binding cassette transporter 1 (as GM-CSF has been reported to induce its expression) together with the anyway impaired cholesterol efflux and increased myeloproliferation in case of Apoe<sup>-/-</sup> mice strongly assume that the increased plaque stability in their vaccinated Ldlr<sup>-/-</sup> mice partly depends on IL-23 neutralization. Hence, the presented study revealing a Th17/IL-17–independent role of IL-23 in atherosclerosis supports this finding.

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cardiovascular. However, the study by Subramanian et al leaves some open questions. It would have been interesting, for example, to not only inject IL-23 in Ldlr<sup>-/-</sup> Csfs2<sup>-/-</sup> mice but also block IL-23 in the control group. The latter gains exceptional importance thinking of the above-mentioned translation of these findings into humans. Furthermore, one has to dissect carefully which role of different lipid profiles may play in influencing GM-CSF function or vice versa. As apparent from the different GM-CSF knockout mouse models discussed above, and despite the fact that macrophage colony-stimulating factor exacerbate atherosclerosis in apolipoprotein E-deficient mice. Circ Res. 2007;115:2049–2054. doi: 10.1161/CIRCULATIONAHA.106.605570.


Not Growth but Death: GM-CSF/IL-23 Axis Drives Atherosclerotic Plaque Vulnerability by Enhancing Macrophage and DC Apoptosis
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Circ Res. 2015;116:222-224
doi: 10.1161/CIRCRESAHA.114.305674
Circulation Research is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
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Print ISSN: 0009-7330. Online ISSN: 1524-4571

The online version of this article, along with updated information and services, is located on the
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