Biochimica et Biophysica Acta 1821 (2012) 513–521

Contents lists available at SciVerse ScienceDirect

Biochimica et Biophysica Acta

journal homepage: www.elsevier.com/locate/bbalip

Review

Anti-atherogenic mechanisms of high density lipoprotein: Effects on myeloid cells ☆

Andrew J. Murphy *, Marit Westerterp, Laurent Yvan-Charvet, Alan R. Tall

Division of Molecular Medicine, Department of Medicine, Columbia University, New York, NY, USA

ARTICLE INFO

Article history:
Received 13 June 2011
Received in revised form 27 July 2011
Accepted 9 August 2011
Available online 16 August 2011

Keywords:
High density lipoprotein
Macrophage
Monocyte
Neutrophil
Hematopoietic stem and multipotent progenitor cell
Atherosclerosis

ABSTRACT

In some settings increasing high density lipoprotein (HDL) levels has been associated with a reduction in experimental atherosclerosis. This has been most clearly seen in apolipoprotein A-I (apoA-I) transgenic mice or in animals infused with HDL or its apolipoproteins. A major mechanism by which these treatments are thought to delay progression or cause regression of atherosclerosis is by promoting efflux of cholesterol from macrophage foam cells. In addition, HDL has been described as having anti-inflammatory and other beneficial effects. Some recent research has linked anti-inflammatory effects to cholesterol efflux pathways but likely multiple mechanisms are involved. Macrophage cholesterol efflux may have a role in facilitating emigration of macrophages from lesions during regression. While macrophages can mediate cholesterol efflux by several pathways, studies in knockout mice or cells point to the importance of active efflux mediated by ATP binding cassette transporter (ABC) A1 and G1. In addition to traditional roles in macrophages, these transporters have been implicated in the control of hematopoietic stem cell proliferation, monocytosis and neutrophilia, as well as activation of monocytes and neutrophils. Thus, HDL and cholesterol efflux pathways may have important anti-atherogenic effects at all stages of the myeloid cell/monocyte/dendritic cell/macrophage lifecycle. This article is part of a Special Issue entitled Advances in High Density Lipoprotein Formation and Metabolism: A Tribute to John F. Oram (1945–2010).

1. Introduction

Atherosclerosis is an indolent, macrophage dominated, focal inflammatory disease of the large arteries. This process is initiated by the deposition of ApoB containing lipoproteins on the arterial proteoglycan matrix in regions of disturbed blood flow, followed by their modification and uptake by macrophages [1,2]. Modified lipoproteins also activate combinatorial signaling by toll like receptors (TLR) and scavenger receptors (SR) on macrophages, and the effects of lipid loading and TLR/SR signaling lead to inflammatory and chemokine responses, ER stress, apoptosis and necrosis [3–5]. These latter events are thought to lead to the ultimate complications of plaque rupture and athero-thrombosis. Although traditionally viewed as having a key role in removing the mass of cholesterol from plaques in a process of reverse cholesterol transport, HDL is now seen as having key effects on macrophage inflammation, ER stress and apoptosis (Fig. 1). Some of these effects are dependent on the fundamental ability of HDL and apoA-I to interact with the ATP binding cassette transporters on macrophages, ABCA1 and ABCG1, mediating efflux of cholesterol and oxidized lipids [6–8], but likely multiple mechanisms are involved. Recent studies also point to a role of HDL, ABCA1, ABCG1 in controlling monocyte activation, adhesiveness and inflammation [9,10], and in controlling the proliferation of the stem and progenitor cells [11] that give rise to monocytes and neutrophils that ultimately enter plaques.

2. ABCA1 and ABCG1 are key mediators of cholesterol efflux

Francis and Oram made the seminal discovery that fibroblasts isolated from Tangier Disease (TD) subjects could not promote the efflux of cholesterol or phospholipids to lipid-free apoA-I [12,13]. Several groups discovered through the use of micro-arrays, genetic mapping and biochemical assays that *Abca1* was the defective gene in Tangier Disease [14–18]. Through this discovery and using techniques to specifically knockdown the expression of *Abca1* it was then demonstrated that ABCA1 exports cholesterol from cells to lipid-free apoA-I. Subsequently, it was shown that another transporter, ABCG1, promotes cholesterol efflux to mature HDL particles but not to lipid-poor apoA-I [19,20]. ABCA1 and ABCG1 are target genes of the nuclear receptors, liver X receptor (LXR) and are upregulated in response to

Abbreviations: HDL, High density lipoprotein; apo, Apolipoprotein; ABC, ATP binding cassette transporter; TLR, Toll like receptor; HSPC, Hematopoietic stem and multipotent progenitor cell; IL, Interleukin; CBS, IL-3 receptor common beta-subunit; GM-CSF, Granulocyte macrophage-colony stimulating factor; BM, Bone marrow; Ldlr, Low density lipoprotein receptor; JAK-2, Janus kinase 2; STAT-3, Signal transducer and activator of transcription-3

☆ This article is part of a Special Issue entitled Advances in High Density Lipoprotein Formation and Metabolism: A Tribute to John F. Oram (1945-2010).

* Corresponding author at: Division of Molecular Medicine, 630 W 168th St, Room 8-401, Columbia University, New York, NY, 10032, USA. Tel.: +1 212 305 5789; fax: +1 212 305 5052.
E-mail address: am3440@columbia.edu (A.J. Murphy).

1388-1981/$ – see front matter © 2011 Elsevier B.V. All rights reserved.
doi:10.1016/j.bbalip.2011.08.003

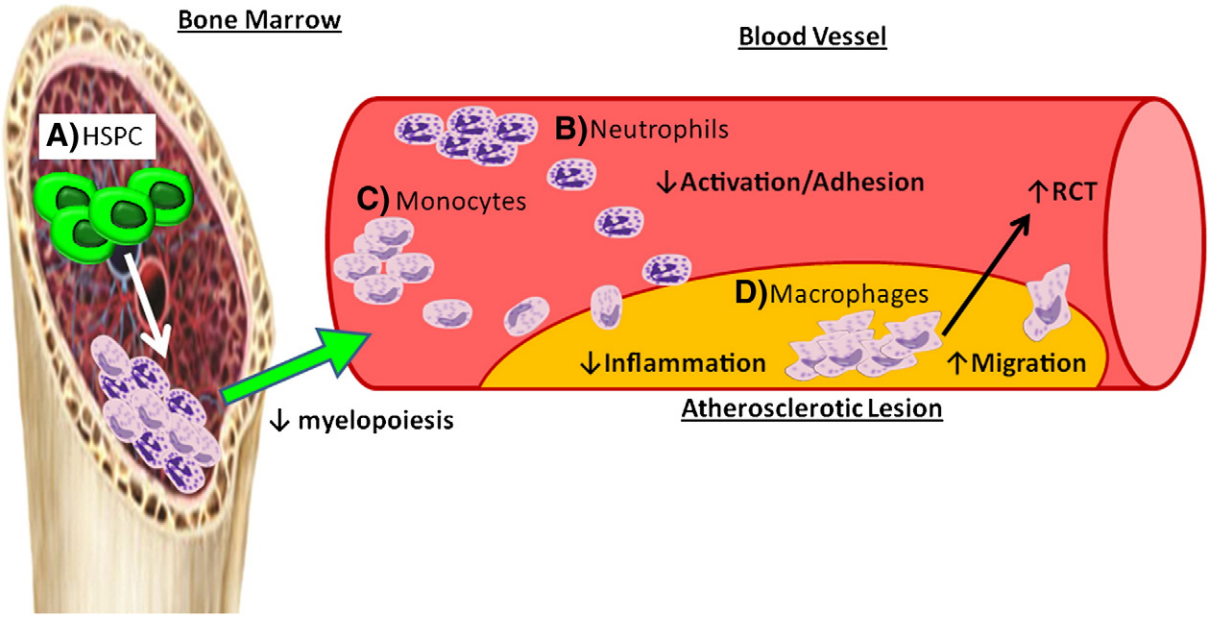

Fig. 1. Anti-atherogenic functions of HDL-sites of action. A) HDL interacts with ABCA1 and ABCG1 on the HSPCs to promote cholesterol efflux and inhibit their proliferation. This regulates the number of mature myeloid cells produced. B, C) HDL and apoA-I can act as an anti-inflammatory reducing monocyte and neutrophil activation. This leads to less recruitment of monocytes/neutrophils to the atherosclerotic lesion. D) HDL interacts with macrophages to regulate a number of cellular functions important to controlling atherosclerosis such as cholesterol efflux, reducing TLR-4 signaling, decreasing apoptosis during efferocytosis and modulating membrane lipid levels to aid in macrophage migration.

sterol loading of macrophages and other cells. Recently it has been shown that ABCA1 (in mouse and human) and ABCG1 (in mouse) are regulated by microRNA-33 (miR-33) [21,22]. The sequence encoding miR-33 is embedded within the sterol response element binding protein-2 (*Srebp2*) gene, so that when cells are deprived of cholesterol, SREBP-2 is upregulated and miR-33 is produced. MiR-33 then binds to a site in the 3'-UTR of ABCA1 and ABCG1 down-regulating their mRNA and protein, thus shutting down cholesterol export [21–25].

3. Cholesterol efflux pathways and immune cell production

3.1. HDL, ApoE, ABCA1 and ABCG1 regulate myelopoiesis and monocyte numbers

Hematopoiesis is hierarchical and ordered, and is initiated by long term self-renewing and multi-potent stem cells. Through a process of proliferation, lineage restriction and differentiation, HSPCs give rise to mature lineage committed cells, that ultimately form the mature blood cells. Production of blood cells in the steady state is tightly regulated by a number of well-defined feedback loops. However, production can be increased when required, for instance in response to infection or blood loss. Emerging evidence suggests that cholesterol uptake and efflux can also regulate HSPC proliferation, providing a potential mechanism to explain the association between leukocytosis and atherosclerotic CVD [11].

Recently, Yvan-Charvet et al. [11] described an important role for HDL and cholesterol efflux pathways in the regulation of hematopoietic stem cell proliferation and myelopoiesis. The hematopoietic stem and multipotential progenitor cells (HSPCs) express relatively high levels of *Abca1*, *Abcg1* and *Apoe*[26–29]. Mice deficient in *Abca1* and *Abcg1* develop a myeloproliferative disorder characterized by dramatic monocytosis and neutrophilia, and infiltration of the spleen, heart, liver, small intestine and other organs with macrophage foam cells and neutrophils. This occurs even in chow fed mice and resembles mouse models of chronic myeloid or myelo-monocytic leukemia. The underlying mechanism involves a marked 4- to 5-fold increase in the numbers and proliferation of the HSPC population. There is increased staining of the plasma membrane of HSPCs with cholera toxin B suggesting increased plasma membrane liquid ordered domains. The increased proliferative response of HSPCs reflects an increased responsiveness to the growth factor interleukin (IL) 3 caused by an increased amount of the common beta-subunit (CBS) of the IL-3/GM-CSF receptor in the cell surface of HSPCs (Fig. 2). This mechanism may also explain why myeloid lineage cells such as common myeloid progenitors and granulocyte-macrophage progenitors were expanded and proliferating in these mice, reflecting increased GM-CSF responses. In contrast, lymphoid and megakaryocyte–erythroid progenitor populations were not expanded. Competitive bone marrow transplantation experiments showed that HSPC and myeloid expansion, monocytosis and neutrophilia occurred in a cell autonomous fashion in cells derived from ABCA1/G1 deficient bone marrow, indicating that HSPC expansion and myeloid proliferation did not require increased amounts of exogenous factors such as inflammatory cytokines. Gomes et al. have also reported leukocytosis and thrombocytosis in WT mice after feeding the Paigen diet [30]. This was also due to an expansion of the BM progenitor cells. It was also shown that feeding an atherogenic diet results in BM stem cell mobilization by modulating the SDF-1:CXCR4 axis. While the LDLr, SR-BI and CD36 were found to be expressed in HSPCs in this study, incubation with LDL appeared to have little effect on HSPC proliferation. While our studies have emphasized the role of cholesterol efflux pathways in the regulation of HSPC proliferation, the mechanisms of cholesterol uptake and broader aspects of the regulation of cholesterol homeostasis in HSPCs are worth of further investigation.

In $Ldlr^{+/-}$ mice transplanted with $Abca1^{-/-}Abcg1^{-/-}$ bone marrow (BM) and fed a high fat, high cholesterol diet, atherosclerosis was markedly increased compared to WT or single KO bone marrow recipients, and there was a strong correlation between leukocyte numbers and atherosclerotic lesion size. In contrast an inflammatory marker, apoSAA while increased in DKO BM recipients, did not correlate with lesion area. This suggests a causal relationship between increased leukocytes and accelerated atherosclerosis. Moreover, when $Abca1^{-/-}Abcg1^{-/-}$ BM was transplanted into a mice expressing a human *Apoa-I* transgene the expansion of the HSPCs was almost completely reversed, along with the increased lipid rafts and the expression of the CBS. The blood leukocyte levels were normalized,