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Do BRD(4)S of a feather flock together? How an inflammation driven epigenetic regulator may link pulmonary hypertension and coronary artery disease

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Pulmonary arterial hypertension (PAH) is a progressive and ultimately fatal disease characterized by elevated blood pressure in the pulmonary artery¹. The pathogenesis of PAH is driven by hyperproliferation of vascular smooth muscle cells in a hyperinflammatory environment, with local accumulation of innate immune cells, such as macrophages^{2, 3}, and increased levels of inflammatory cytokines such as IL-6, IL-1 β and IL-18. While we traditionally consider PAH a disease that selectively affects the pre-capillary arterioles, coronary artery disease (CAD) is reported to be 4-times more prevalent in patients with PAH, suggesting possible common mechanisms of disease or risk factors. Indeed, PAH is associated with a higher prevalence of the metabolic syndrome, an observation that is particularly notable in patients who develop pulmonary vascular remodeling in the setting of heart failure with preserved ejection fraction⁴.

In this issue of *ATVB*, Meloche and colleagues postulate that systemic inflammation could be a central mechanism linking the development of both coronary and pulmonary vascular smooth muscle cell proliferation. The authors provide rich experimental data demonstrating that, in rat models of PAH and in humans with PAH, there is evident vascular remodeling of the coronary arteries, characterized by the thickening of the arterial wall due to smooth muscle cell proliferation, and reduced cardiac perfusion. They also find elevated IL-6 mRNA levels in the human hearts of patients with PAH and stimulation of coronary artery smooth muscle cells in culture with IL-6 induces smooth muscle cell proliferation and DNA damage.

Investigating mechanisms that might tie vascular IL-6 mediated inflammation and secondary smooth muscle proliferation in PAH and CAD, Meloche et al. demonstrate that the bromodomain containing protein 4 (BRD4) is a key molecule regulating this observed PAH-associated distal coronary artery thickening. BRD4 is an epigenetic “reader” that recognizes histone modification patterns and binds to acetylated lysine residues of the histone tails. BRD4 is a cofactor facilitating transcriptional activation of target genes by stabilizing the recruitment of transcription factors and transcription elongation factors^{5, 6}. The authors

previously showed that BRD4 expression in pulmonary artery smooth muscle cells contributed to the development of experimental PAH⁷. Moreover, BRD4 inhibition with JQ1 inhibitor, which also suppresses BRD2 and BRD3, limits coronary artery atherogenic processes⁸. Based on these reports, Meloche et al., hypothesized that increased BRD4 expression in smooth muscle cells in the coronary vasculature of PAH patients triggers the development of CAD. Indeed, BRD4 expression was increased in both pulmonary artery and coronary artery smooth muscle cells in rat models of PAH and in humans with PAH. IL-6 increased BRD4 expression in smooth muscle cell cultures, which augmented cellular proliferation and suppressed apoptosis. Pharmacological inhibition of BRD4 with JQ1 reduced smooth muscle cell hyperproliferation and enhanced cellular apoptosis. In vivo studies using both a silencing RNA and JQ1 to inhibit BRD4 reduced coronary artery thickness in the sugen-hypoxia rat model of PAH. These studies suggest an important role for IL-6 driven BRD4 expression in the pro-proliferative and anti-apoptotic phenotype of coronary and pulmonary vascular smooth muscle cells in PAH models. It remains to be investigated if other inflammatory cytokines, in addition to IL-6, have an effect on the expression of the epigenetic reader.

The implication of epigenetic “readers” like BRD4 in the inflammation-mediated coronary artery remodeling associated with PAH is of particular interest since it draws a causal association between inflammation and epigenetic reprogramming, which is a new avenue of research in major diseases including cancer, metabolic syndrome and cardiovascular disease^{7–10}. The nature and function of BRD4 as an enzymatically inactive epigenetic “reader” suggest that additional epigenetic and transcriptional mechanisms, such as DNA methylation, histone modification, imbalance of epigenetic “writers” (enzymes adding modifications) and/or epigenetic “erasers” (enzymes removing modifications), as well as microRNA, might be initiated or altered by inflammation. Additional studies are therefore required to delineate the ensemble of mechanisms involved in the inflammation-mediated epigenetic reprogramming in CAD. Importantly, BRD4 can be recruited to both promoter and super enhancer regions⁸. Super enhancers are genomic regulatory elements displaying distinct chromatin landscape and enriched in transcription binding sites. A lineage specific subset of distal enhancers is primed for activation in distinct cell types and there is evidence supporting a critical functional role of enhancers in controlling cell differentiation and cell lineage identity^{11, 12}. This raises the question whether BRD4 binds to specific subsets of enhancers and which target genes are upregulated in smooth muscle cells, endothelial cells and immune cells respectively.

The study by Meloche and colleagues focused on the role of BRD4 in proliferation and apoptosis of smooth muscle cells in the coronary artery. However, aside from smooth muscle cells, endothelial cells can contribute to vascular remodeling process in PAH and CAD. Endothelial dysfunction in CAD precedes adherence and extravasation of inflammatory monocytes, which differentiate into pathogenic foam cells in atherosclerotic plaques. Moreover, endothelial cells can indirectly aggravate disease pathogenesis by releasing inflammatory cytokines. Since endothelial dysfunction is a common pathogenic feature in PAH and CAD¹³, BRD4 signaling in endothelial cells may play a role in CAD development in PAH patients. Interestingly, a recent paper¹⁴ revealed the role of BRD4 in producing inflammatory cytokines, such as IL-6 and IL-8, by pulmonary endothelial cells. Clearly,

future studies are warranted to investigate BRD4 signaling in coronary artery endothelial cells in patients with PAH and CAD. Moreover, since inflammatory macrophages and T cells are the major sources of IL-6 in tissues, it is possible that these inflammatory cells are recruited in the perivascular space and then induce smooth muscle cell proliferation by increasing BRD4 expression, suggesting a complex crosstalk between these cell types (Figure 1).

Another hallmark of CAD is presence of atherosclerotic plaques, but it is not currently known if IL-6-mediated DNA damage and epigenetic modifications can mediate plaque development. It would also be interesting to know if the drugs commonly used in CAD, such as statins and aspirin, or anti-inflammatory therapies currently tested in clinical trial like methotrexate¹⁵ (decreasing IL-6 levels) or canakinumab¹⁶ (anti-IL-1 β antibody) can reduce BRD4 expression. As suggested by the authors, smooth muscle cell proliferation directly contributes to distal CAD by inducing vasoconstriction and reducing coronary perfusion. It is also widely assumed that proliferation of smooth muscle cell and their participation in the formation of a protective smooth muscle cell-rich, collagen-rich fibrous cap protects against plaque rupture in coronary arteries^{17, 18}. Whether BRD4 contributes to these disparate properties of the atherosclerotic lesion remains to be studied.

In conclusion, this interesting study provides new evidence suggesting that PAH is a systemic disease affecting other organs such as the systemic vasculature and heart. Future studies will be required to untangle the interactions and clearly define causality and new therapies that capitalize on this new knowledge.

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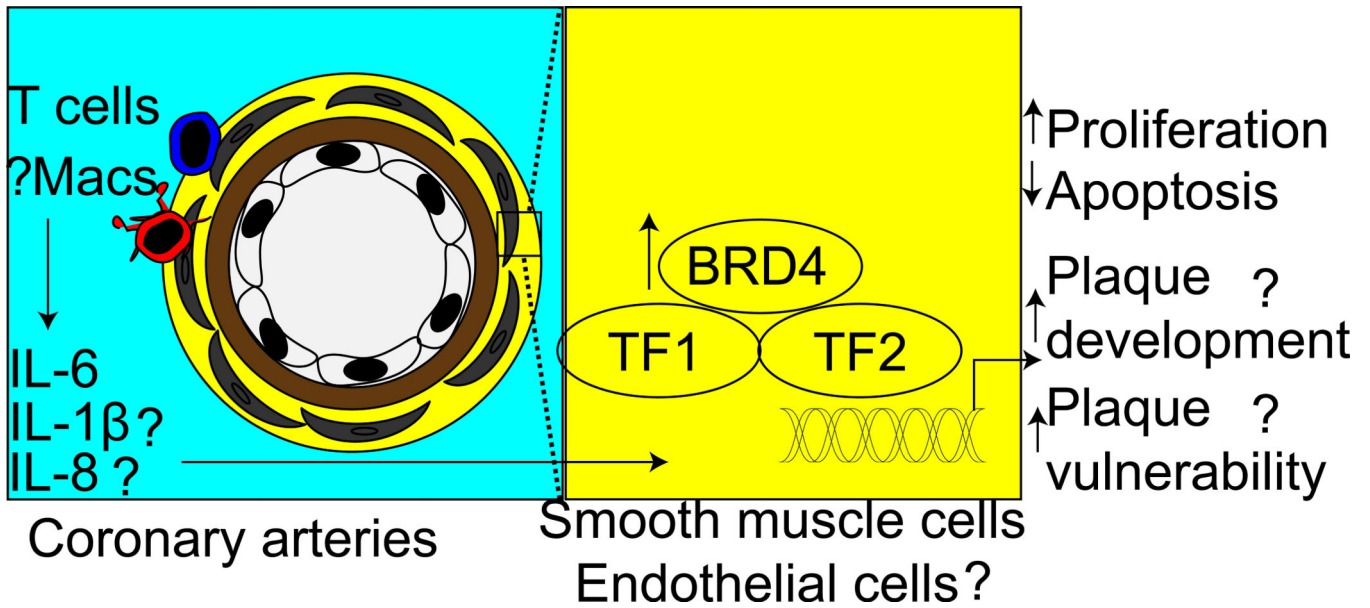


Figure 1.

Hypothesized schematic representation of increased CAD in PAH patients: Inflammatory cytokines, such as IL-6, IL-8 and IL-1 β , produced by recruited macrophages and T cells increase BRD4 expression. Augmented bromodomain containing protein 4 (BRD4) expression in smooth muscle cells or endothelial cells in the coronary arteries facilitates transcription of genes involved in cell proliferation, apoptosis inhibition, plaque development and plaque vulnerability. Question marks indicate unanswered questions. TF, transcription factor.