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Apabetalone downregulates factors and pathways associated with vascular calcification

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HIGHLIGHTS

- Bromodomain and extraterminal (BET) proteins are implicated in VSMC transdifferentiation and calcification.
- Apabetalone, a BET inhibitor, prevents calcification of VSMCs by regulating expression of key factors.
- BET protein BRD4 may cooperate with 7 specific transcription factors (TFs) to promote transdifferentiation and calcification.
- Apabetalone is a promising therapeutic for pathological vascular calcification.

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ABSTRACT

Background and aims: Apabetalone is an inhibitor of bromodomain and extraterminal (BET) proteins. In clinical trials, apabetalone reduced the incidence of major adverse cardiac events (MACE) in patients with cardiovascular disease and reduced circulating factors that promote vascular calcification (VC). Because VC contributes to MACE, effects of apabetalone on pro-calcific processes were examined.

Methods and results: Apabetalone inhibited extracellular calcium deposition and opposed induction of transdifferentiation markers in human coronary artery vascular smooth muscle cells (VSMCs) under osteogenic culture conditions. Tissue-nonspecific alkaline phosphatase (TNAP) is a key contributor to VC, and apabetalone suppressed osteogenic induction of the mRNA, protein and enzyme activity. The liver is a major source of circulating TNAP, and apabetalone also downregulated TNAP expression in primary human hepatocytes. BRD4, a transcriptional regulator and target of apabetalone, has been linked to calcification. Osteogenic transdifferentiation of VSMCs resulted in disassembly of 100 BRD4-rich enhancers, with concomitant enlargement of remaining enhancers. Apabetalone reduced the size of BRD4-rich enhancers, consistent with disrupting BRD4 association with chromatin. 38 genes were uniquely associated with BRD4-rich enhancers in osteogenic conditions; 11 were previously associated with calcification. Apabetalone reduced levels of BRD4 on many of these enhancers, which correlated with decreased expression of the associated gene. Bioinformatics revealed BRD4 may cooperate with 7 specific transcription factors to promote transdifferentiation and calcification.

Conclusions: Apabetalone counters transdifferentiation and calcification of VSMCs via an epigenetic mechanism involving specific transcription factors. The mechanistic findings, combined with evidence from clinical trials, support further development of apabetalone as a therapeutic for VC.

1. Introduction

Vascular calcification (VC) is aberrant deposition of calcium phosphate and hydroxyapatite in blood vessels, leading to pathological vascular stiffness [1]. VC is prevalent in chronic kidney disease (CKD), and the extent of VC predicts cardiovascular risk [2,3]. VC arising in CKD patients occurs in the medial layer of the vessel wall, an elastic region comprised of vascular smooth muscle cells (VSMCs), in a process

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