A transcriptional regulator of osteogenesis expressed in calcifying atherosclerotic plaques

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Received 24 August 2001; accepted 30 August 2001

See article by Engelse et al. [9] (pages 281–289) in this issue.

Calcification is a common feature of atherosclerotic plaques. Even early lesions of young adults have been found to contain small aggregates of crystalline calcium within the lipid core [1]. As plaque size and complexity increase the amount of calcium within the lesion grows [1]. The degree of plaque calcification correlates with increased risk of coronary disease-related events [2], though the precise ways in which calcification influences plaque pathology remain to be resolved. Coronary vessels with extensively calcified plaques are more prone to dissections during angioplasty and carry a higher risk of operative complications [3]. Deposition of calcium within vessels also occurs in the medial layer of arteries where it is independent of atherosclerotic disease and can cause decreased compliance [4]. Aortic calcification may thus lead to coronary insufficiency as a result of impaired elastic recoil.

Despite the widespread occurrence of vascular calcification and its association with cardiovascular disease, the mechanisms responsible for mineralization within vessels are not known. Similarities between calcification in bone and the vasculature suggest both tissues share some of the same mechanisms of mineralization. As in bone, the principal form of calcium deposited in the vessel wall is hydroxyapatite. During bone formation osteoblasts synthesize matrix vesicles and a complex extracellular matrix incorporating, among other proteins, type I collagen, osteocalcin, osteopontin and bone sialoprotein [5]. Mineralization of this matrix is regulated by the osteogenic proteins and follows nucleation of calcium crystallization. Even in bone formation the specific roles of many of the proteins produced by the osteoblast, and regulatory mechanisms for mineral deposition and resorption have yet to be defined. Several of the proteins involved in bone calcification and re-modelling are also expressed during vascular calcification. These include osteopontin, osteocalcin and matrix Gla protein [6–8].

In a study reported in this issue of Cardiovascular Research, Engelse et al. [9] have examined expression of another bone-associated protein in human atherosclerotic lesions, core binding factor α1(Cbfa-1). They find this protein expressed in areas of calcification within plaques in aortae and iliac arteries. Cbfa-1 is a transcription factor of the ‘runt’ family and has recently been shown to be both a key molecule in osteoblast differentiation in vivo and regulator of bone deposition by differentiated osteoblasts [10]. The importance of this transcription factor in calcification is demonstrated by the complete absence of bone seen in transgenic mice deficient in Cbfa-1 [11,12]. Putative Cbfa-1 binding sites have been identified in the promoters of osteocalcin, bone sialoprotein, osteopontin and α-1 type I collagen [10]. Furthermore, overexpression of Cbfa-1 in non-osteoblast cells, such as fibroblasts, induces expression of osteocalcin and bone sialoprotein [13]. Sustained function of fully differentiated osteoblasts also appears to require continued expression of Cbfa-1 in the cells [14]. Engelse et al. find Cbfa-1 expression in neointimal smooth muscle cells and macrophages in early plaques. This is consistent with their observations of expression in a cultured monocyte cell line and, in common with other reports [15,16], vascular smooth muscle cells. In advanced plaques, exhibiting extensive calcium accumulation, cells strongly expressing Cbfa-1 did not react with antibodies to smooth muscle cell, macrophage or T-cell markers.

This paper is the first to report Cbfa-1 expression in human plaques. Clearly, the transcription factor has the potential to regulate expression of several osteogenic proteins by plaque cells, including some of those already identified as being present in calcifying lesions. Cbfa-1,
therefore, may have a central role in regulating plaque calcification similar to that in bone formation. If this were the case, the transcription factor would be an attractive target for interventions aimed at influencing plaque mineralization. In addition Cbfa-1 would be a useful marker for identifying early events involved in establishing conditions favouring plaque calcification. The findings are also significant as they raise questions over the phenotypes of the Cbfa-1 positive intimal smooth muscle cells and macrophages seen in early plaques. Both of these cell types have already been shown to express some bone-associated proteins, such as osteopontin, matrix Gla protein and osteoglycin [6,8,17]. Cbfa-1 may be responsible for directing some of this programme of expression. Given the central role of the transcription factor in osteoblast differentiation it is possible that these Cbfa-1 expressing vascular cells are in the process of trans-differentiation into an osteoblast-like cell type. The unidentified Cbfa-1 positive cells found in the more advanced plaques could be trans-differentiated cells descended from the cells seen in the early plaques. Another potential source of these cells would be mesenchymal cells and pericytes that are known to exist within plaques [18] and that can be induced to differentiate towards an osteoblast phenotype [19]. It will be important to determine whether subsets of intimal vascular cells truly trans-differentiate, fully or partially, into osteogenic cells and the role of Cbfa-1 in this.

Whilst Cbfa-1 expressing cells provide a possible source for some of the proteins involved in calcification, their precise contribution to plaque mineralization remains to be defined. The apparent absence of calcification in early plaques, even where Cbfa-1-positive cells are present, suggests calcium deposition requires additional factors. These factors could include further modulators of osteogenic phenotype, as well as stimuli for initiating calcium crystallization. Recent data suggests apoptotic bodies derived from smooth muscle cells within the plaque may act as nucleators of calcium deposition [20]. Calcification in atherosclerotic lesions may therefore require such a trigger, combined with the presence of osteoblast-like cells.

In contrast to the findings in plaques, Cbfa-1 does not localize strongly with medial calcification in normal arteries [9]. This suggests diffuse calcification of the media and focal calcium deposits found in plaques arise via different mechanisms. Matrix Gla protein (MGP) has been proposed as an inhibitor of calcification in the vasculature [5] and the authors do report decreased expression of this protein in areas of medial calcification [9]. Interestingly, they also show that MGP-expressing areas within plaques do not contain Cbfa-1-expressing cells and lack calcification. It is possible that MGP could regulate Cbfa-1 expression. MGP binds avidly to bone-morphogenetic protein-2 (BMP-2), a ligand also known to be present in calcifying plaques [18] and recognized to induce Cbfa-1 expression [21] and initiate osteoblast differentiation in mesenchymal cells [22]. BMP-2 complexed with MGP is unable to induce osteoblast differentiation [23]. Localised MGP may therefore act to suppress the ability of BMP-2 to induce differentiation of osteogenic cells within the plaque. There are differences in the patterns of MGP expression reported by Engelse et al. and previous data, which demonstrate high levels of MGP in areas of plaque calcification. These differences may be related to the stage in calcification of vessels examined in each of the studies. MGP is a secreted protein likely to have complex multiple roles in early and late stages of plaque calcification.

Identification of a key transcriptional regulator of bone formation in human atherosclerotic plaques suggests a mechanism by which osteogenic protein expression in these lesions could be regulated. It will now be important to determine the significance of Cbfa-1 in control of expression of bone-associated proteins by vascular cells and in the process of vascular calcification.

References

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