Vascular Smooth Muscle Cells in the Pathogenesis of Vascular Calcification

Keith A. Hruska

Vascular calcification is a major risk factor for cardiovascular morbidity and mortality. In atherosclerotic lesions, calcification is mainly found in the neointima of atheromatous plaques and has been shown to positively correlate with the plaque burden and the risk of myocardial infarction. Calcification of atherosclerotic plaques has been clearly shown to result from osteoblastic differentiation of cells in the neointima forming a mineralized matrix—containing type I collagen similar to bone formation.1–3 The origin of the neointimal osteoblastic cells remains controversial, but evidence that they derive from migratory smooth muscle cells is growing. Atherosclerotic calcification is increased by chronic kidney disease, type 2 diabetes mellitus, and aging.

However, another form of vascular calcification, that of the vascular media, known as Mönckeberg’s medial calcific sclerosis, is also prevalent in patients with chronic kidney disease, in type 2 diabetes mellitus, and in aging patients, especially those with osteoporosis. Medial calcification in these patients can occur independently of intimal atherosclerotic lesions and features linear calcium phosphate deposits along the elastic lamellae, which may form circumferential mineral deposits throughout the media. Medial calcification causes vascular stiffness and increased pulse wave velocity that causes cardiac dysfunction/ischemia. Mineralization of elastin is clearly different from osteoblastic bone formation, and the pathogenesis of medial calcification is less clear than that of atherosclerosis. Osteochondroblastic differentiation has been detected in cells adjacent to medial calcific deposits along with type II collagen in the matrix.4

In 1997, Luo et al reported the important finding of diffuse medial calcification with aortic rupture and death between 6 to 8 weeks of age in mice deficient of the matrix gla protein (MGP).5 The vascular calcification in MGP−/− mice affected the elastic lamellae of elastic and muscular arteries, such as aortas, carotids, and coronary arteries. Calcification in these mice was associated with profound changes in cell differentiation as arterial smooth muscle cells (SMCs) were replaced by chondrocyte-like cells undergoing progressive mineralization. MGP−/− mice are not atherosclerotic. MGP is a calcification inhibitor that accumulates at the border of calcified areas and in normal media of blood vessels and appears to act locally to limit calcium phosphate deposition in the vessel wall. The calcium-binding function of MGP requires vitamin K–dependent γ-carboxylation for activation, and undercarboxylated MGP, mainly resulting from vitamin K insufficiency and/or long-term warfarin treatment, accelerates the development of vascular calcification. In addition, polymorphisms of the MGP gene are associated with an increased risk of myocardial infarction, as well as cardiovascular mortality in chronic kidney disease and hemodialysis patients. Mutation of the MGP gene causes excessive arterial calcification in the human autosomal recessive condition Keutel syndrome.

In this issue of Circulation Research, Speer et al6 attempt to provide definitive evidence that SMCs contribute to the origin of osteochondroblastic cells seen in calcified blood vessels by a fate mapping approach in MGP−/− mice. They bred transgenic mice carrying a SM22-cre transgene and mice carrying the R26R-LacZ transgene into the MGP−/− background. The result of Cre recombinase activity in mice bearing both transgenes is β-galactosidase activity detected by blue staining in cells expressing SM22. Blue-stained cells of MGP mice were limited to smooth muscle–rich tissue, such as the arterial media, and, importantly, the bone marrow was negative. In MGP−/− mice, β-galactosidase–positive cells resembling chondrocytes apparently secreting osteopontin and type II collagen were involved in large calcific medial lesions. The chondrocytic cells were not positive for smooth muscle myosin heavy chain, SM22α, or SMα actin, although they had previously expressed SM22 because they were β-galactosidase–positive. This provides the strong suggestion that the chondrocytic cells had differentiated from SMCs. This is the exciting contribution of this report. Evidence to support circulating or resident multipotent mesenchymal progenitors as the source of the chondrocytic cells was lacking, though not definitive. The time course of expression of Runx2 in the arterial media demonstrated expression in β-galactosidase positive, SM22α-positive medial cells at 2 weeks of age before calcification. After 2 weeks of age, Runx2 and SM22α expression decrease as chondrocytic calcifying cells appear. Because Runx2 is critical for both the chondrocytic and osteoblastic lineage, a weakness of this report is that Sox 9 expression, specific for chondrocytic cells, was not similarly characterized. However, another critical osteoblastic transcription factor, osterix, was not expressed in the MGP−/− arteries.

An important issue not addressed by Speer et al6 is whether or not the vascular calcification of the MGP−/− mice is a model of Mönckeberg’s medial calcific sclerosis. From the
calcific lesions shown in the report, calcification along the elastic lamellae was clearly evident in agreement with Luo et al, albeit at an older age (4 weeks compared to 2 to 3 weeks). However, the presence of chondrocytic cells was limited to larger lesions, and the sections shown in the report bearing chondrocytic cells demonstrated organization into large calcified lesions. Chondrocytic cells were not evident along the elastic lamellae calcifications, raising the issue of whether MGP\(^{-/-}\) deficiency is a model of Mönckeberg’s medial calcific sclerosis and whether the latter is caused by chondrocytic cells mineralizing a type II collagen matrix as in the large lesions of the MGP\(^{-/-}\) mouse. Thus, the nature of Mönckeberg’s medial calcific sclerosis remains an unsolved issue.

Speer et al, when attempting to characterize potential mechanisms of the SMC transdifferentiation that they discovered in the MGP\(^{-/-}\) mice, analyzed a vascular SMC culture system. The weakness of the in vitro system is that it is not a model of the in vivo situation, and the relationship of their in vitro findings to their animal model is not clear. Vascular smooth muscle cells in culture can differentiate along the osteoblastic lineage, as confirmed by Speer et al, but they did not achieve the chondrocytic differentiation and the absence of terminal osteoblastic differentiation shown by lack of osterix expression that they found in their MGP\(^{-/-}\) mice. Speer et al did not analyze the action of MGP\(^{-/-}\) deficiency in stimulating chondrocytic transdifferentiation of SMC, although they allude to the possibility that its role may be to bind bone morphogenetic protein-2 in the extracellular matrix, thereby inhibiting bone morphogenetic protein-2 function as an osteochondrogenic morphogen and osteoinductive factor.

In summary, the report by Speer et al establishes that arterial SMCs developing normally in utero transdifferentiate along the chondrocytic lineage in the first few weeks of postnatal life in MGP\(^{-/-}\) mice. This leads to calcification of large chondrocytic cell bearing medial lesions. In addition, the MGP\(^{-/-}\) mice have calcification of elastic lamellae, in which the presence of the chondrocytic cells is not apparent. Thus, the relationship of the chondrocytic differentiation of SMCs to calcification of elastic lamellae in Mönckeberg’s medial calcific sclerosis is not established by this report, although a role of MGP deficiency is suggested.

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References

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