G
eometric remodeling is an important component of vascular pathologies, including restenosis and atherosclerosis. Although our understanding of the precise events involved in vascular remodeling is far from complete, it is generally accepted that local breakdown of extracellular matrix (ECM), smooth muscle cell migration, and matrix reorganization are important components. In this respect, particular attention has been directed toward the role of matrix metalloproteinases (MMPs), enzymes capable of remodeling the ECM. However, being able to understand the specific contribution of individual members of a proteinase family that share overlapping substrates is a significant challenge.

MMPs are Zn-containing neutral endopeptidases. At least 23 different MMPs have been identified that, as a family, have the capacity to degrade all components of the ECM, in addition to some nonmatrix substrates. Within the family, several subgroups exist, based on substrate specificities or domain structures. The activity of MMPs is controlled at several distinct levels, including transcription, activation of zymogens, and interaction with specific inhibitors, the TIMPs (tissue inhibitors of MMPs). The two gelatinases MMP-2 and MMP-9 have received particular attention in analysis of vascular remodeling due to their expression by smooth muscle cells and leukocytes and ability to breakdown components of the basement membrane and collagens. At least in vitro, both enzymes have a very similar substrate profile. However, their expression in the vascular wall is differently controlled, in that a basal expression of MMP-2 can be detected within the media, whereas MMP-9 expression is only apparent after injury or inflammatory stimulation. Likewise, activation of these two MMPs can be mediated differently, and they may interact selectively with different TIMPs (MMP-2 with TIMP-2 and MMP-9 with TIMP-1).

A variety of approaches has been used to study the roles of MMPs in models of vascular remodeling, including the use of chemical inhibitors, adenoviral delivery of TIMPs, and more recently, through analysis of the response in knockout (KO) mice lacking specific MMPs. The first two approaches are attractive because they can be carefully controlled since genetically identical mice can be used. However, most inhibitors are active against several MMPs such that the role of a particular MMP is difficult to ascertain. KO models are therefore attractive as they offer the potential to understand the specific role of individual MMPs and may reveal which would be an appropriate therapeutic target. In this issue of Arteriosclerosis, Thrombosis, and Vascular Biology, Johnson and Galis have taken this approach in an attempt to address the relative role of MMP-2 and MMP-9 in a murine model of intimal hyperplasia. While both MMPs appeared to contribute to smooth muscle cell migration and neointima growth, the authors present preliminary evidence that MMP-9 may in addition have a role in collagen assembly and compaction.

Following their previous study using 129/SvEv mice, the authors studied the extent of intimal hyperplasia in MMP-2 and MMP-9 KO mice on a C57/BL6 background after carotid artery ligation. An absence of either MMP significantly reduced the formation of intimal hyperplasia 28 days after ligation, corresponding to fewer numbers of intimal smooth muscle cells and reduced intima thickness compared with wild-type, suggesting a significant decrease in smooth muscle cell migration. In vitro analysis of the migratory capacity of smooth muscle cells indicated that both MMP-2 and MMP-9 are important for migration through a gelatin matrix, and that neither can completely compensate for the other. However, a significant difference was observed when smooth muscle cells were assayed for their effects on collagen assembly and compaction. Thus, whereas absence of MMP-2 had no effect, MMP-9 deficiency impaired collagen assembly and compaction. Although the authors did not prove that wild-type cells release active MMP-9 in their assays (an important test based on the tight control of MMP-9 expression in smooth muscle cells), they were able to rescue the phenotype of MMP-9–deficient cells by adding MMP-9 to the assay. The authors also found that MMP-9 may serve as a bridge between cells and the ECM, in that MMP-9–deficient smooth muscle cells had a reduced capacity to bind to gelatin, and they conclude that this may contribute to traction during cell migration. However, this is not supported by the results with MMP-2–deficient cells, whose migration is apparently impaired to a greater extent than MMP-9–deficient cells. Of particular interest, the authors found that an interaction between MMP-9 and the hyaluronan receptor, CD44, was necessary for the role of MMP-9 in collagen assembly and compaction. A role for CD44 in the contraction of collagen gels containing hyaluronan has previously been reported, but the specific role of MMP-9 was not explored.
The significance of these in vitro observations remain to be thoroughly addressed in vivo. Staining with specific antibodies and picrosirius red suggested that MMP-9 deficiency results in a decreased accumulation and organization of collagen during hyperplasia, but qualitative information on the collagen network is difficult to gauge in the absence of ultrastructure analyses (for example by electron microscopy).

It is interesting that, despite having a larger neointima, the lumen of the MMP-2–deficient mice appeared larger than in the MMP-9–deficient mice 28 days after ligation. This was not apparently due to outward remodeling of the artery, because EEL measurements were not significantly different.

Previously, the authors observed an accumulation of collagen in the adventia of MMP-9–deficient mice after carotid artery ligation. This may contribute to the extra constriction seen in the MMP-9 KO compared with the MMP-2 KO, while the decrease in collagen compaction observed for MMP-9 KO smooth muscle cells in vitro may contribute to the reduced constriction of the lumen compared with wild-type. However, it will also be important to investigate other components of the ECM. In this respect, analysis of the organization of hyaluronan and proteoglycans would be interesting because these high molecular weight hydrophilic complexes can trap water and cause tissue swelling, a factor that could contribute to intima hyperplasia in experimental animal models.

The observations of Johnson and Galis9 begin to address the individual contribution of different MMPs to vascular remodeling and imply that there can be subtle differences in the roles of enzymes with overlapping substrate preferences. However, the chosen model, although technically demanding, produces a relatively straightforward and predictable outcome. A more significant challenge is to understand the contributions of different MMPs to a more complex vascular pathology, such as atherosclerosis where remodeling is complicated by the presence of monocyte/macrophages with the capacity to produce a range of different MMPs, lipid accumulation, and a local inflammatory response. The same authors have previously reported that monocyte/macrophage infiltration after carotid artery ligation in atherosclerotic-prone apoE KO mice can dramatically influence the remodeling process.11 Inhibition of MMP activity has been suggested as an approach to both stabilize vulnerable plaques by reducing matrix breakdown and to limit stenosis by reducing plaque growth. Recent data has addressed the influence of MMPs on plaque stability and, intriguingly, shows opposite effects of MMP-9 and MMP-12.12 Furthermore, investigations using KO or transgenic expression of MMPs or TIMPs have revealed that the contribution of matrix remodeling to lesion growth is complex.13–15 The availability of highly selective inhibitors would be a valuable tool for investigating the relative contribution of individual MMPs in lesion growth, along with the kind of work using KOs and transgenics described by Johnson and Galis.9 However, we can still expect that the task of unraveling the role of individual MMPs will be complex.

References


Matrix Management: Assigning Different Roles for MMP-2 and MMP-9 in Vascular Remodeling
Carl Whatling, William McPheat and Eva Hurt-Camejo

Arterioscler Thromb Vasc Biol. 2004;24:10-11
doi: 10.1161/01.ATV.0000100562.63144.C1
Arteriosclerosis, Thrombosis, and Vascular Biology is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2004 American Heart Association, Inc. All rights reserved.
Print ISSN: 1079-5642. Online ISSN: 1524-4636

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://atvb.ahajournals.org/content/24/1/10

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Arteriosclerosis, Thrombosis, and Vascular Biology can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Arteriosclerosis, Thrombosis, and Vascular Biology is online at:
http://atvb.ahajournals.org//subscriptions/