Membrane-type 1 Matrix Metalloproteinase Regulates Macrophage-dependent Elastolytic Activity and Aneurysm Formation in Vivo*

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During arterial aneurysm formation, levels of the membrane-anchored matrix metalloproteinase, MT1-MMP, are elevated dramatically. Although MT1-MMP is expressed predominately by infiltrating macrophages, the roles played by the proteinase in abdominal aortic aneurysm (AAA) formation in vivo remain undefined. Using a newly developed chimeric mouse model of AAA, we now demonstrate that macrophage-derived MT1-MMP plays a dominant role in disease progression. In wild-type mice transplanted with MT1-MMP-null marrow, aneurysm formation induced by the application of CaCl₂ to the aortic surface was almost completely ablated. Macrophage infiltration into the aortic media was unaffected by MT1-MMP deletion, and AAA formation could be reconstituted when MT1-MMP was reintroduced. Macrophage infiltration into the aortic media was unaffected by MT1-MMP deletion, and AAA formation could be reconstituted when MT1-MMP-null marrow recipients. Using a newly developed chimeric mouse model of AAA, we now demonstrate that macrophage-derived MT1-MMP plays a dominant role in disease progression. In wild-type mice transplanted with MT1-MMP-null marrow, aneurysm formation induced by the application of CaCl₂ to the aortic surface was almost completely ablated. Macrophage infiltration into the aortic media was unaffected by MT1-MMP deletion, and AAA formation could be reconstituted when MT1-MMP-null marrow recipients. In vitro studies using macrophages from either WT/MT1-MMP+/- mice, MMP-2-null mice, or MMP-9-null mice demonstrate that MT1-MMP alone plays a dominant role in macrophage-mediated elastolysis. These studies demonstrate that destruction of the elastin fiber network during AAA formation is dependent on macrophage-derived MT1-MMP, which unexpectedly serves as a direct-acting regulator of macrophage proteolytic activity.

Development and progression of abdominal aortic aneurysm (AAA) is a complex process that, untreated, can lead to tissue failure, hemorrhage, and death (1). Destruction of the orderly elastin lamellae of the vessel wall is considered the sine qua non of arterial aneurysm formation (2) as adult tissues cannot regenerate normal elastin fibers (3). Moreover, the elastin degradation products are chemotactic for inflammatory cells and serve to amplify the local injury (4). Although several types of elastolytic proteases are elevated in AAA tissue (5–9), studies using murine models of AAA and targeted protease deletion suggest that matrix metalloproteinases (MMPs), particularly the secreted proteases, MMP-2 and MMP-9, play key roles in the pathologic remodeling of the elastin lamellae that lead to AAA (7, 8).

Membrane-type 1 MMP (MT1-MMP) is the prototypical member of a family of membrane-tethered MMPs (10). Recent studies indicate that MT1-MMP expression is elevated in human AAA tissues and that infiltrating macrophages are the primary source of the proteinase in aortic lesions (11–13). Although indirect evidence suggests that MT1-MMP may participate in the control of monocyte/macrophage motile responses as well as interactions with the vessel wall during transmigration (14, 15), the role(s) played by MT1-MMP in regulating macrophage proteolytic activity or AAA formation remain undefined.

Using a murine model of AAA and mice with a targeted deletion of MT1-MMP in myelogenous cell populations, we now demonstrate that macrophage-derived MT1-MMP is required for elastin degradation and aneurysm formation. Importantly, macrophages are not dependent on MT1-MMP for infiltrating aortic tissues but instead use the protease to directly regulate their elastolytic potential in an MMP-2- and MMP-9-independent fashion. These studies define a new and unexpected role for MT1-MMP in controlling macrophage elastolytic activity in the in vitro and in vivo settings.

EXPERIMENTAL PROCEDURES

Mice—Heterozygous MT1-MMP (MT1-MMP+/-) mice, the generous gift of H. Birkedal-Hansen (NIDCR, National Institutes of Health, Bethesda, MD) were bred and genotyped as described (16). The genetic background of the MT1-MMP-deficient mice is 129/Swiss black (16). Mice with the MMP-2 deletion were obtained from D. Muir (University of Florida, Gainesville, FL) and originally described by Itoh et al. (17). MMP-9-null mice were purchased from The Jackson Laboratory (Bar Harbor, ME). The genetic background of MMP-2-null and MMP-9-null mice is C57/BL6. All experiments were carried out in accordance with the guidelines of the University of Nebraska Medical Center Animal Care Committee.

Generation of Chimeric Mice Lacking MT1-MMP Expression in Myelogenous Cells—As the short life span of the MT1-MMP+/− mice preclude their use in aneurysm models (16),...
5-week-old wild-type littermates of MT1-MMP-/- mice were irradiated (1200 rads) and transplanted with bone marrow from MT1-MMP+/+ mice (designated as WT/MT1-MMP+/+ mice) or wild-type mice (WT/WT mice). Bone marrow cell suspensions were prepared from the femurs of MT1-MMP+/+ mice or wild-type littermates, and 5 x 10^6 cells were infused via the lateral tail veins.

**Isolation and Infusion of Peritoneal Macrophages and Lymphocytes**—For macrophage isolation, wild-type littermates of MT1-MMP+/+ mice were injected intraperitoneally with 1 ml of 3% Brewer thioglycollate medium. One week later, peritoneal macrophages were collected and suspended in phosphate-buffered saline. For lymphocyte isolation, wild-type spleens were removed, and cells were isolated, resuspended in red blood cell lysis buffer, and washed with RPMI 1640. Macrophages were depleted by allowing the cells to adhere to a plastic substratum, and the non-adherent lymphocytes were resuspended in phosphate-buffered saline. WT/MT1-MMP+/+ mice were injected with 5 x 10^6 peritoneal macrophages (n = 6) or macrophage-depleted splenic lymphocytes (n = 6, as a control group) via the tail vein 1 day before AAA induction. A second cell infusion was repeated 1 week later.

**Aneurysm Induction**—Mice at 9 weeks of age underwent surgery as described previously (7, 18). Briefly, the abdominal aorta between the renal arteries and bifurcation of the iliac arteries was isolated, and the diameter was measured in triplicate in the mid-infrarenal aorta. After baseline measurements, 0.25 mol/liter CaCl_2 was applied to the external surface of the aorta for 15 min, and the incision was closed. NaCl (0.9%) was substituted for CaCl_2 in sham control mice. After 6 weeks, the mice underwent laparotomy, and dissection and aortae were collected for analysis.

**Histology and Immunohistochemistry**—For Masson’s trichrome staining, abdominal aortic tissues were embedded in paraffin and stained as described (7). For macrophage staining, sections were incubated with monoclonal rat antimouse Mac3 antibody (Pharmingen) (7). Macrophage infiltration was graded by a pathologist unaware of genotype.

**RT-PCR**—Total RNA from mouse aortic tissues, macrophages, lymphocytes, and aortic smooth muscle cells was extracted using Trizol reagent (Invitrogen). White blood cells from WT/MT1-MMP+/+ and WT/WT mice were isolated by centrifugation through a Histopaque (Sigma) density gradient, and total RNA was extracted using RNeasy kit (Qiagen, Valencia, CA). RT-PCR was performed using Thermoscript RT-PCR system (Invitrogen) with β-actin or glyceraldehyde-3-phosphate dehydrogenase as an internal reference.

**Gelatin Zymography**—Aortae were extracted for gelatin zymography as previously described (7). Protein concentrations were standardized with Bio-Rad protein assay. The intensity of each band was quantified by densitometry (Amersham Biosciences).

**Elastin Preparation and Elastolytic Assay**—Elastin (type E60; Elastin Product Co., St Louis, MO) was reductively labeled (1300 dpm/μg of elastin) as described (19). Peritoneal macrophages from WT/WT, WT/MT1-MMP+/+, C57/B6L, MMP-2-/-, and MMP-9-/- mice were isolated as above and activated by incubating with 2 units/ml IFN-γ for 4 h and 10 ng/ml LPS for 1 h. After further stimulating cells with 100 ng/ml LPS for 24 h, 1 mg of ([3H]elastin was added to each well in the presence of the cysteine protease inhibitor, E64 (20 μM, Sigma), and the serine protease inhibitor, phenylmethylsulfonyl fluoride (1 mM), in serum-free RPMI 1640 at 37 °C for 5 days. Solubilized [3H]elastin was quantitated by β-scintillation counting. Results are presented as mean ± S.E. of three experiments.

**Statistical Analysis**—Measurements of aortic diameter and gelatinolytic activity are expressed as the mean value ± S.E. A paired Student’s t test was used to compare original and final diameters. Student’s t test was used to compare final aortic diameter between groups. Fisher’s exact test was used to compare the proportion of mice developing aneurysms. Statistical significance was accepted at a p < 0.05. Aortae were scored aneurysm-positive when the diameter increased by 50% or more from the base-line measurement (20).

**RESULTS**

**Role of MT1-MMP in AAA Formation**—After the direct application of CaCl_2 to the external surface of the aorta, a localized inflammatory response is initiated which recapitulates key features of the human AAA, including macrophage and lymphocyte infiltration, increased MMP expression, elastin degradation, and progressive vessel wall dilatation (7, 20). To examine the potential contribution of MT1-MMP to AAA, expression levels of MT1-MMP in the aneurysm tissue of CaCl_2-treated wild-type mice were compared with aortic tissue from sham, NaCl-treated controls by RT-PCR. Consistent with reports documenting increased MT1-MMP levels in human aneurysm tissue (21, 22), mouse MT1-MMP mRNA levels were likewise increased in aneurysmal aorta tissue relative to nonaneurysmal controls (Fig. 1A).

As infiltrating macrophages contribute the bulk of the MT1-MMP found in human AAA lesions (21, 22), chimeric mice were generated by transplanting wild-type or MT1-MMP-/- marrow into sub-lethally, γ-irradiated recipients (i.e. WT/WT or WT/MT1-MMP+/+ mice, respectively). After recovery from bone marrow transplantation, mice underwent CaCl_2 aneurysm induction. RT-PCR confirmed minimal expression of MT1-MMP in myeloid cells from the WT/MT1-MMP+/+ chimeras (Fig. 1B). Furthermore, whereas CaCl_2-treated tissues isolated from WT/WT mice processed endogenous proMMP-2, similarly treated tissues recovered from WT/MT1-MMP+/+ chimeras display a pronounced defect in zymogen processing (Fig. 1C).

Given results demonstrating that marrow-derived cells likely serve as a major source of active MT1-MMP in the CaCl_2-treated aorta wall, the relative ability of WT- versus MT1-MMP-/- derived myeloid cells to support AAA formation was assessed in the respective chimeric mice. As observed previously (7, 18), WT/WT mice developed aneurysms (4 of 5) at a rate similar to that reported for WT mice using the standard clinical definition of a 50% increase in diameter (Fig. 1D; i.e. aortic diameter in the WT/WT chimeric mice increased by 63 ± 8%) (7, 18). In marked contrast, however, only 1 of 11 WT/MT1-MMP+/+ mice developed an aneurysm (Fig. 1D and Table 1). Furthermore, although the CaCl_2-treated WT/WT mice exhibit significant disruption of the elastic lamellae of the
Role of MT1-MMP in Aneurysm Formation

Role of Macrophage MT1-MMP in AAA Formation—Infusion of bone marrow from MT1-MMP knock-out mice into sub-letally γ-irradiated wild-type mice results in deletion of MT1-MMP expression in all myelogenous cells. As AAA lesions are dominated by macrophages as well as lymphocytes and both cell populations can express MT1-MMP (Fig. 2A), WT/MT1-MMP−/− chimeric mice were tail-vein-injected with either macrophages or lymphocytes harvested from wild-type mice 24 h before CaCl2 aneurysm induction. A second infusion of macrophages or lymphocytes was performed 1 week after CaCl2 induction as per protocols established in previous studies where the ability of infused cells to infiltrate aortic tissue has been documented (7, 18). As shown in Fig. 2A, infusion of MT1-MMP−/− macrophages into WT/MT1-MMP−/− chimeric mice resulted in reconstitution of proMMP-2 processing (Fig. 2A) and the induction of aneurysm formation (Fig. 2B and Table 2) as well as degradation of the elastin matrix in vivo (Fig. 2C). By contrast, neither proMMP-2 processing nor aneurysm formation was reconstituted when wild-type lymphocytes were infused into WT/MT1-MMP−/− mice (Fig. 2, A and B). Interestingly, vascular smooth muscle cells express MT1-MMP in aorta recovered from WT/MT1-MMP−/− mice (Fig. 2B), these cells process only small amounts of procMMP-2 (Fig. 2A) and are unable to mediate significant elastin fiber degradation or aneurysm formation (Fig. 1H). Taken together, these studies confirm a central role of macrophage-derived MT1-MMP in supporting aneurysm formation and elastin degradation.

Role of MT1-MMP in Macrophage Transmigration and Elastolytic Activity—MT1-MMP is required for mesenchymal cell migration through collagen-rich tissues, and recent studies have suggested that the proteinase might play similar roles in regulating monocyte/macrophage motility (14, 15, 23–25). As such, the attenuated damage observed in the WT/MT1-MMP−/− chimeric mice might occur as a consequence of a defect in the ability of MT1-MMP-deficient macrophages to invade the aortic wall matrix. To this end sections from WT/WT and WT/MT1-MMP−/− mice were examined using the elastin matrix. As expected, the aortic walls of the NaCl-treated sham control mice from both groups are intact (Fig. 1, E and G).

### TABLE 1

Changes in aortic diameter in WT and MT1-MMP−/− bone marrow transplantation to WT γ-irradiated mice after treatment of NaCl and CaCl2

| Bone marrow transfer | WT | WT | MT1-MMP−/− | MT1-MMP−/− |
|----------------------|----|----|-----------|-----------|
| Treatment Number    | NaCl | CaCl2 | NaCl | CaCl2 |
| AAA development      | 0/5 (0%) | 4/5 (80%) | 0/7 (0%) | 1/11 (9%) |
| Post-treatment (µm)  | 542 ± 16 | 567 ± 14 | 572 ± 117 | 551 ± 62 |
| % Increase           | 13.3 | 63.3 | 7.4 | 32.9 |

*p < 0.01, Paired Student’s t test, compared to pre-treatment value.

*p < 0.05, compared to CaCl2-treated MT1-MMP−/−.

FIGURE 1. A, expression of MT1-MMP. Six weeks after 0.9% NaCl or 0.25 mM CaCl2 treatment, WT mouse aortae were harvested, and total RNA was extracted. MT1-MMP mRNA levels were determined by RT-PCR. Glyceraldehyde-3-phosphate dehydrogenase (GAPDH) was used as the internal standard. Gels are representative of three separate experiments. B, 4 weeks after bone marrow transplantation, mouse blood was obtained. Total RNA from white blood cells was extracted and MT1-MMP mRNA levels were examined by RT-PCR. β-Actin was used as the internal standard. C, gelatin zymographic analysis of latent and processed MMP-2 in the mouse aortae. Six weeks after 0.9% NaCl or 0.25 mM CaCl2 treatment, mouse aortas from WT/WT and WT/MT1-MMP−/− mice were harvested. Aortic proteins were extracted and separated by electrophoresis on a 10% SDS-PAGE containing 0.8% gelatin. A representative gelatin zymography is shown. D, percentage of WT/WT or WT/MT1-MMP−/− chimeric mice developing aneurysms after CaCl2 treatment; E–H, histological changes in the mouse aortae as seen with trichrome stain. Sections were photographed and shown with the lumen at the top (×20). Three different aortas from each of the four groups were stained and representative sections are shown.

Aortic diameters were measured before NaCl or CaCl2 incubation (Pre-treatment) and at sacrifice (Post-treatment). Measurements of aortic diameter were expressed as the mean ± S.E. The percent increase was represented as a percent compared with pre-treatment. The development of an aneurysm was defined as a 50% or greater increase relative to original diameter of aorta.

GAPDH was used as the internal standard. Gels are representative of three separate experiments. As expected, the aortic walls of the NaCl-treated sham control mice from both groups are intact (Fig. 1, E and G).
Role of MT1-MMP in Aneurysm Formation

FIGURE 2. A, expression of MT1-MMP in macrophages (Mφ) and macrophage-depleted splenocytes (Lymphocytes) (left panel). Total RNA from cells was extracted, and MT1-MMP mRNA levels were determined by RT-PCR. β-Actin was used as the internal standard. Gelatin zymographic analysis of latent and processed MMP-2 in the mouse aorta (right panel) is shown. Six weeks after 0.25 mM CaCl2 treatment of wild-type or WT/MT1-MMP mice given infusions of WT lymphocytes or WT peritoneal macrophages, the aortae were harvested. Aortic proteins were extracted and separated by electrophoresis on a 10% SDS-PAGE containing 0.8% gelatin. The gels shown were representative of three trials with similar results. B, percentage of WT/MT1-MMP−/− mice developing aneurysm after CaCl2 treatment. Expression of MT1-MMP in macrophages (Mφ) or lymphocytes developing aneurysm after CaCl2 treatment. Expression of MT1-MMP in aortic smooth muscle cells (SMC) is shown in the inset. C, histological changes in the aorta of WT/MT1-MMP−/− mouse infused with WT Mφ or lymphocytes demonstrating elastin degradation (Fig. 3C). As MMP-2 and MMP-9 have been shown to play important, but mechanistically undefined roles in AAA formation in vivo, the potential impact of these proteases on macrophage-mediated elastolysis was likewise assessed. In a fashion distinct from that observed with MT1-MMP−/− cells, macrophages isolated MMP-2-null retained wild-type elastolytic activity (Fig. 3C). Similarly, consistent with the fact that MMP-9 expression is unaffected in MT1-MMP−/− macrophages (Fig. 3D), MMP-9-null macrophages also degrade elastin at normal rates (Fig. 3C). Taken together, these results demonstrate that MT1-MMP plays a critical role in AAA formation and that the metalloproteinase directly regulates the elastolytic activity of intact macrophages.

TABLE 2

Changes in aortic diameter in MT1-MMP−/− mice with macrophage infusion

| Cell transfer | Mφ | Treatment | CaCl2 | Number | AAA development (%) | Pre-treatment (µm) | Post-treatment (µm) | Percent of increase (%) | Range of increase (%) |
|---------------|----|-----------|-------|--------|--------------------|-------------------|---------------------|------------------------|---------------------|
| WT Mφ infusion | WT | | | 6 | 5/6 (83%) | 523 ± 8.2 | 821 ± 8.2 | 57.2 | 42–68 |
| WT lymphocytes infusion | WT/MT1-MMP−/− | | | | | | | | |

**a p < 0.05, paired Student’s t test, compared to CaCl2-treated control.**

Macrophages infiltrating aortic tissue during AAA formation are presumed to play an important role in degrading the elastic lamina that maintains vessel wall integrity in pressure-bearing circuits (6–8). Although macrophages are able to degrade elastin in vitro (26, 27), the role of MT1-MMP in this process has not been characterized previously. As such, wild-type or MT1-MMP-null macrophages were cultured with [H3]elastin in the presence of a mixture of serine and cysteine protease inhibitors to specifically monitor MMP-dependent elastolytic activity. Under these conditions, MT1-MMP-deficient macrophages demonstrated a 64% ± 9 decrease in elastin-degradative activity relative to wild-type macrophage (Fig. 3C). As MMP-2 and MMP-9 have been shown to play important, but mechanistically undefined roles in AAA formation in vivo, the potential impact of these proteases on macrophage-mediated elastolysis was likewise assessed. In a fashion distinct from that observed with MT1-MMP−/− cells, macrophages isolated MMP-2-null retained wild-type elastolytic activity (Fig. 3C). Similarly, consistent with the fact that MMP-9 expression is unaffected in MT1-MMP−/− macrophages (Fig. 3D), MMP-9-null macrophages also degrade elastin at normal rates (Fig. 3C). Taken together, these results demonstrate that MT1-MMP plays a critical role in AAA formation and that the metalloproteinase directly regulates the elastolytic activity of intact macrophages.

DISCUSSION

In human AAA tissues, MT1-MMP expression is increased markedly and localized primarily to infiltrating macrophages (21). Nonetheless, the potential role of macrophage MT1-MMP in AAA formation has remained unexplored largely as a consequence of the fact that MT1-MMP−/− mice are moribund shortly after birth, display multiple musculoskeletal and metabolic defects, and die by 1–3 months of age (16, 28). To circumvent this limitation, we have assessed the role of MT1-MMP in AAA formation in a newly developed chimeric mouse model. Using this protocol, irradiated mice reconstituted with WT marrow (WT/WT chimeras) developed aneurysms at the same rate and mean diameter as CaCl2-treated WT mice. By contrast, only 1/11 mice reconstituted with MT1-MMP-deficient bone marrow developed an aneurysm. The histology co-

mice (2.7 ± 0.2 macrophages/high power field; p = 0.8; n = 9) (Fig. 3, A and B). Furthermore, macrophages were distributed similarly within the media layer of WT/WT and WT/MT1-MMP−/− mouse aortas (Fig. 3A). Taken together, these studies establish a novel role for macrophage-derived MT1-MMP.
Role of MT1-MMP in Aneurysm Formation

A number of recent studies have concluded that MT1-MMP can regulate monocyte migration, raising the question of whether MT1-MMP deletion would influence macrophage infiltration into the aortic wall (14, 15). Although these studies were confined to in vitro analyses, we now demonstrate that the macrophage content of CaCl2-treated aortae was unaffected by MT1-MMP deficiency. These results support a model where wild-type as well as MT1-MMP−/− monocytes invade the aortic wall in comparable fashion where they then differentiate into macrophages. Interestingly, similar results were obtained recently where MT1-MMP−/− monocytes were shown to infiltrate atherosclerotic plaques at rates comparable with those exhibited by wild-type cells (30). Although Libby and co-workers (30) have suggested a role for MT1-MMP in effecting the activation of the secreted collagenase, MMP-13, within atherosclerotic lesions, we detect only low levels of active MMP-13 in aneurysm tissue (data not shown). Furthermore, we find that wild-type as well as MT1-MMP−/− macrophages display only low levels of collagenolytic activity.3 We cannot, however, rule out the possibility that monocytes or macrophages use other proteolytic systems to invade the vessel wall matrix, but it should be noted that myeloid cell populations can traverse collagen-rich tissues by protease-independent processes that depend on the ability of the cell to adopt an amoeboid-type of invasive activity (31).

Given the ability of MT1-MMP to degrade collagen types I, II, and III, fibronectin, laminin, and fibrin as well as other extracellular matrix substrates (10), we considered the possibility that MT1-MMP might influence aneurysm formation by regulating macrophage elastolytic activity. Indeed, MT1-MMP−/− macrophages display a profound defect in their ability to degrade insoluble elastin. As MMP-2−/− macrophages did not share a similar loss in elastolytic potential, these results are consistent with a model wherein MT1-MMP directly regulates macrophage elastolytic potential. Interestingly, we have previously reported that macrophage-derived MMP-9 plays a key role in AAA formation in vivo (7). Based, however, on the fact that (i) MMP-9−/− macrophages can now be shown to retain full elastolytic activity in vitro and (ii) MT1-MMP−/− macrophage express MMP-9 at normal, if not heightened levels, these combined findings suggest that MMP-9 must indirectly regulate aneurysm formation. With the increased appreciation that MMP-9 can exert

3 R. Shimizu-Hirota and S. J. Weiss, unpublished observation.
Role of MT1-MMP in Aneurysm Formation

multiple effects on cell behavior ranging from the release of soluble kit-ligand and the regulation of VEGF bioavailability to modulating cell migration and tight junction function (32–34), the role of macrophage-derived MMP-9 is likely complex in the in vivo setting. Nevertheless, our findings indicate that MMP-9 does not play a direct or required role in supporting macrophage-mediated elastolytic activity. From this perspective, caution must also be exercised in terms of speculating the mechanisms by which MT1-MMP controls macrophage-elastin interactions. In the simplest scenario, MT1-MMP might directly display elastolytic activity. In preliminary studies, however, purified transmembrane-deleted, soluble MT1-MMP did not degrade elastin under standard culture conditions.4 Importantly, the soluble enzyme is unlikely to recapitulate the high protease-substrate concentrations found at the macrophage-elastin interface or the specific conditions operative within this specialized microenvironment (e.g. acidic pH) (26, 27). Furthermore, MT1-MMP may additionally control macrophage-mediated elastolytic activity by affecting macrophage signaling pathways (10). Although further studies are needed to characterize the global impact of MT1-MMP activity on macrophage function, our findings demonstrate that macrophages are incapable of exhibiting their full elastolytic potential in the absence of MT1-MMP.

In vivo, we have demonstrated that macrophage-derived MT1-MMP plays a critical role in modulating elastin degradation and aneurysm formation. Although our findings stress a direct role for MT1-MMP in regulating macrophage-mediated proteolysis, cooperative interactions with other cell populations likely occur within the confines of the vessel wall. For example, levels of active MMP-2 are elevated in aneurysm tissues excised during operative repair (9), and as demonstrated in the current study, macrophage-derived MT1-MMP plays a dominant role in activating MMP-2 in our AAA model system. Given the fact that MMP-2+/− mice are resistant to CaCl2-induced aortic damage (7), we posit that macrophage MT1-MMP plays additional roles in AAA by locally activating vessel wall-derived MMP-2. In this scenario, macrophage-derived MT1-MMP would play both MMP-2-dependent and independent roles in driving AAA formation in vivo despite the fact that macrophages can directly degrade elastin in the complete absence of MMP-2 (i.e. Fig. 3C). Nevertheless, given the central role assumed by MT1-MMP in orchestrating elastolytic processes, the proteinase likely represents an important therapeutic target in elastin-destructive disease states ranging from AAA and atherosclerosis to emphysema.

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