Cell. Mol. Life Sci. (2011) 68:3853–3868
DOI 10.1007/s00018-011-0763-x

Cellular and Molecular Life Sciences

REVIEW

Matrix metalloproteinases in tumorigenesis: an evolving paradigm

Hui Hua · Minjing Li · Ting Luo · Yancun Yin · Yangfu Jiang

Received: 20 April 2011 / Revised: 31 May 2011 / Accepted: 21 June 2011 / Published online: 10 July 2011

Abstract Proteases are crucial for development, tissue remodeling, and tumorigenesis. Matrix metalloproteinases (MMPs) family, in particular, consists of more than 20 members with unique substrates and diverse function. The expression and activity of MMPs in a variety of human cancers have been intensively studied. MMPs have well-recognized roles in the late stage of tumor progression, invasion, and metastasis. However, increasing evidence demonstrates that MMPs are involved earlier in tumorigenesis, e.g., in malignant transformation, angiogenesis, and tumor growth both at the primary and metastatic sites. Recent studies also suggest that MMPs play complex roles in tumor progression. While most MMPs promote tumor progression, some of them may protect the host against tumorigenesis in a context-dependent manner. MMPs have been chosen as promising targets for cancer therapy on the basis of their aberrant up-regulation in malignant tumors and their ability to promote cancer metastasis. Although preclinical studies testing the efficacy of MMP suppression in tumor models were so encouraging, the results of clinical trials in cancer patients have been rather disappointing. Here, we review the complex roles of MMPs and their endogenous inhibitors such as tissue inhibitors of metalloproteinase in tumorigenesis and strategies in suppressing MMPs.

H. Hua · M. Li · Y. Yin · Y. Jiang (✉)
State Key Laboratory of Biotherapy, Section of Signal Transduction and Molecular Targeted Therapy, West China Hospital, Sichuan University, Chengdu, China
e-mail: jyangfu@scu.edu.cn

T. Luo
Cancer Center, West China Hospital, Sichuan University, Chengdu, China

Keywords Matrix metalloproteinase · Cancer

Introduction

Tumor development and progression are complex processes that involve oncogenes, tumor suppressor genes, and the tumor microenvironment. Stromal–epithelial interaction plays important roles in the pathogenesis of a variety of tumors [1]. The cross-talk between malignant and surrounding stromal cells (fibroblasts, endothelial cells, and inflammatory cells) may establish fertile soil in which tumor cells grow and metastasize. In addition to the stromal cells within tumors, the extracellular matrix (ECM) also has an important impact on tumor progression [2]. The major components of the ECM include structural proteins such as collagen and elastin; specialized proteins such as fibrillin, fibronectin, and laminin; and proteoglycans. There are diverse types of proteases that control ECM remodeling and drive the dissemination of cancer cells into adjacent tissue. Among them, serine proteases such as urokinase plaminogen activator (uPA)-plasminogen-plasmin system and cysteine cathepsins are involved in the breakdown of multiple ECM proteins during various physiopathological situations, including cell adhesion, invasion, angiogenesis, and metastasis [3]. In addition, matrix metalloproteinases (MMPs) are intensively studied and have been demonstrated to play key roles in inflammation and carcinogenesis [4]. Also, MMPs are involved in cell signaling and are capable of activating specific cell receptors and growth factors or liberating them from the ECM, thereby regulating various cell behaviors, such as cell growth, differentiation, apoptosis, and migration [4, 5].

In light of the critical roles for MMPs in inflammation and tumorigenesis, MMPs appear to be ideal drug targets

[5, 6]. The development of MMP inhibitors has been very attractive within the academy and industry. Many pharmaceutical inhibitors have been developed thus far. Several of them have undergone clinical trials in patients with cancer. Although many of them have shown promising effects in preclinical studies, the results from clinical trials with these agents appear to be disappointing in cancer treatment. These outcomes also stimulate intensive studies on the roles of MMPs in different stages of cancer. It has been recognized that MMPs play complex, and often opposing roles in cancer progression, which raises question marks on how to effectively target MMPs for cancer therapy. In this review, we emphasize on the complex roles of MMPs in tumor progression and recent studies on MMP inhibitors in cancer therapy.

MMPs and the endogenous inhibitors: an overview

MMPs are the principal matrix-degrading proteinases that play key roles in embryonic development, organ morphogenesis, and wound healing [5]. Abnormal activation of MMPs has been implicated in numerous diseases including arthritis, atherosclerosis, and tumorigenesis. The MMPs are endopeptidases that can cleave many components of the ECM. So far, more than 20 MMPs have been identified. Since almost every MMP has an alias that may be confusing, the names of individual MMPs and their aliases are listed in Table 1. Typically, the MMPs contribute to ECM turnover in either secreted or membrane-bound forms. Based on the structure and substrate specificity, MMPs can be classified into several groups, namely collagenases, gelatinases, stromelysins, membrane-type MMPs, and non-classified MMPs (Table 1). In general, the structure of the MMPs includes a signal peptide, a propeptide domain, a catalytic domain, and zinc-binding motif. Except MMP-7 and MMP-26, other MMPs also contain hemopexin-like domain. The most extensively studied MMP-2 and MMP-9 share a gelatin-binding domain [7]. MMP-14 (MT1-MMP), -15 (MT2-MMP), -16 (MT3-MMP), and MMP-24 (MT5-MMP) harbor a transmembrane domain [8]. MMP-17 (MT4-MMP), and MMP-25 (MT6-MMP) are a unique set of membrane-anchored MMPs with a glycosyl-phosphatidyl inositol (GPI) anchor [9]. The diversity of structure

Table 1 Members of the MMP family

Name	Alias	Category
MMP-1	Interstitial collagenase; fibroblast collagenase	Collagenase
MMP-2	72-kDa type IV collagenase; gelatinase A; 72-kDa gelatinase	Gelatinase
MMP-3	Stromelysin-1	Stromelysin
MMP-7	Matrilysin-1; PUMP1	Stromelysin
MMP-8	Neutrophil collagenase; PMNL collagenase	Collagenase
MMP-9	92-kDa type IV collagenase; gelatinase B; 92-kDa gelatinase	Gelatinase
MMP-10	Stromelysin-2; transin-2	Stromelysin
MMP-11	Stromelysin-3	Stromelysin
MMP-12	Macrophage proteinase; macrophage elastase; metalloelastase	Stromelysin
MMP-13	Collagenase 3	Collagenase
MMP-14	MT1-MMP; MT-MMP1	Membrane-type MMP
MMP-15	MT2-MMP; MT-MMP2	Membrane-type MMP
MMP-16	MT3-MMP; MT-MMP3	Membrane-type MMP
MMP-17	MT4-MMP; MT-MMP4	Membrane-type MMP
MMP-19	MMP-18; Matrix metalloproteinase RASI; RASI-1; stromelysin-4	Non-classified
MMP-20	Enamel metalloproteinase; enamelysin	Non-classified
MMP-21	X-MMP	Non-classified
MMP-23A	CA-MMP	Non-classified
MMP-23B	MIFR; MIFR-1	Non-classified
MMP-24	MT5-MMP; MT-MMP5	Membrane-type MMP
MMP-25	MT6-MMP; MT-MMP6	Membrane-type MMP
MMP-26	Matrilysin-2; endometase	Stromelysin
MMP-27	MMP-22; C-MMP	Non-classified
MMP-28	Epilysin	Non-classified

endows individual MMP with different distribution and substrate specificity.

MMPs not only serve as effectors in physiological processes such as embryo implantation and development, branching morphogenesis, bone remodeling, and angiogenesis but also have additional roles in the reorganization of tissues during pathological conditions such as inflammation, wound healing, and invasion of cancer cells. MMPs are frequently overexpressed in a variety of human tumors [10–12]. Ets sites have a critical role in MMP transcription and they often cooperate with AP1 in multiple MMP promoters. Another major cytokine-induced signaling pathway that regulates MMP transcription involves translocation of nuclear factor-κB (NF-κB) family members from the cytoplasm to the nucleus [13]. After being synthesized into proteins, most MMPs require proteolytic cleavage for enzymatic activity after reaching the cell surface or secreting into the extracellular space. A few MMPs, however, are activated intracellularly by a furin-like mechanism [14]. The activities of MMPs can be counteracted by endogenous inhibitors. α2-Macroglobulin is an abundant plasma protein that acts as a main MMPs inhibitor in tissue fluids. α2-Macroglobulin does not inhibit the activation of MMPs, nor does it inhibit MMPs directly. Instead, active MMPs are efficiently captured by α2-macroglobulin in tissue fluids, followed by low-density lipoprotein-receptor-related protein (LRP)-mediated endocytosis and clearance. Interestingly, LRP itself is a target of MT-MMP proteolysis [15]. Thrombospondin (TSP)-2 regulates the clearance of extracellular MMP-2 in a similar way to α2-macroglobulin. Lysosomal degradation of MMP2–TSP2 complexes by LRP is an important mechanism for the regulation of extracellular MMP-2 levels [16]. In contrast, TSP1 binds to pro-MMP2 and pro-MMP9 and directly inhibits their activation [17, 18].

Whereas both α2-macroglobulin and TSP can regulate a broad spectrum of proteases, tissue inhibitors of metalloproteinases (TIMP) are more specific endogenous MMP inhibitors. So far, four TIMPs have been identified, designated as TIMP-1, TIMP-2, TIMP-3, and TIMP-4, respectively. The four TIMPs share common evolutionary structure in their N-terminal regions [17]. Twelve cysteine residues and their relative spacing are highly conserved in all the TIMPs, which form six disulfide bonds that are essential for the native conformations and the MMP-inhibitory activities. The C-terminal regions of the TIMP family are divergent, which may dictate the selectivity of TIMPs to MMPs or confer MMP-independent function. TIMPs inhibit MMPs by forming a strong noncovalent complex with a 1:1 stoichiometry. Although various TIMPs can bind to different MMPs, there is some difference in the inhibitory properties among the TIMPs. TIMP-1 prefers to inhibit MMP-9, stromelysin, and collagenase. Pro-MMP2 prefers to bind to TIMP-2. TIMP-4 is similar to TIMP-2 in that it is a more potent inhibitor of MMP2 than of MMP9 and matrilysin [18]. The type I transmembrane MT-MMPs are relatively well inhibited by TIMP-2, TIMP-3, and TIMP-4, but are poorly inhibited by TIMP-1. In contrast, the GPI-anchored MT-MMPs are inhibited by both TIMP-1 and TIMP-2 [19].

Paradoxically, TIMP-2 and TIMP-3 are involved in the cell surface activation of proMMP-2. TIMP-2 can bind to proMMP-2 via C-terminal interaction, and to MT1-MMP by its N-terminal domain. This dual binding brings proMMP-2 close to the cell surface, where it can be activated by neighboring TIMP-2-free MT1-MMP molecules [20–23]. It has been reported that the entire propeptide domain of MT1-MMP is required for the TIMP-2 binding and subsequent proMMP-2 activation [24]. Fully functional TIMP-2 is essential for efficient activation of proMMP-2 both in vitro and in vivo [25]. However, the active 65-kDa MMP-2 can be inhibited by plasma membrane-bound TIMP-2 [26]. These results suggest that the pericellular activity of MMP-2 is tightly regulated by membrane-bound TIMP-2 and surrounding ECM components. In addition, TIMP-3 can bind to proMMP-2 with high affinity and promote MT3-MMP mediated activation of proMMP-2 [27], but does not promote MT1-MMP-mediated activation of proMMP-2 [21]. pro-MMP-2 activation by MT3-MMP on the cell surface involves a ternary complex with proMMP-2 assembled with the catalytic domain of MT3-MMP and TIMP-2 or TIMP-3 [27]. In this case, TIMP-2 or TIMP-3 may act as a scaffold to tether proMMP-2 and MT3-MMP. Although TIMP-4 associates with MMP-2 and MT1-MMP in a manner similar to TIMP-3, it does not support MT1-MMP-mediated activation of proMMP-2 [28], nor does it affect MT3-MMP-mediated activation of pro-MMP-2 [27], suggesting that individual TIMP has a different role in the regulation of pro-MMP-2 processing. MT1-MMP also promotes the activation of pro-collagenase 3 (MMP-13) [29], a potent collagenolytic protease. It remains unknown whether TIMPs regulate the activation of proMMP-13.

Complex roles of MMPs in tumorigenesis

Breakdown of extracellular components

The ECM remodeling is an active event during tumor progression. On one hand, ECM serves as a niche for tumor cells to survive and proliferate. On the other hand, it is a barrier that suppresses the spreading of tumor cells. Degradation of ECM is one of the first steps in tumor invasion and metastasis. MMPs promote cell invasion and motility by pericellular ECM degradation. The expression and

activity of MMP-2 and MMP-9, two intensively studied gelatinases, are frequently elevated in human cancer, which correlates with advanced tumor stage, increased metastasis, and poor prognosis. MMP-2 and MMP-9 participate in the degradation of ECM components including the basement membrane, which separates epithelia from stroma [30]. In addition, MMP-1 digests type III collagen more efficiently than collagen types I and II. MMP-3 is known to degrade collagen types III, IV, IX and X, proteoglycans, laminin, elastin, and fibronectin [31]. MT1-MMP, a key MMP that regulates invasion and metastasis, plays a dual role in pathophysiological digestion of the ECM through activation of proMMP-2 and direct cleavage of substrates such as collagen types I, II, and III. Regulated positioning of MT1-MMP to invadopodia, the specialized ECM-degrading membrane protrusions of invasive cells, enables focal degradation of ECM during invasion and metastasis [32].

Release of bioactive molecules

Degradation of structural and specialized components of the ECM by MMPs not only breaks the barrier that restrains tumor cell dissemination but also generates some bioactive fragments. Upon MMP digestion, the ECM also releases biologically active fragments called matrikines, i.e., peptides originating from the fragmentation of matrix proteins and presenting biological activities [33]. For example, cleavage of laminin-5 $\gamma 2$ chains by MMP-2 and MT1-MMP produces a fragment containing epidermal growth factor (EGF)-like motifs that engages EGFR signaling and larger fragments that engage integrin signaling, leading to cell migration [34, 35]. Cleavage of osteopontin by MMP-9 generates a 5-kDa fragment that promotes tumor cell invasion [36]. Moreover, cleavage of insulin-like growth factor-binding protein (IGFBP) by MMP-7 and MMP-9 leads to release of the bioavailable IGF and activation of IGFR signaling [37–39]. Thus, MMPs degrade multiple components of the ECM thereby generating bioactive molecules that promote tumor progression.

Regulation of urokinase plasminogen activator (uPA), growth factor, and cytokine signaling

The cross-talk between MMPs and other proteases also contributes to tumor progression. MMP-9 can regulate the activity of other proteases such as uPA. A recent study reveals that MMP-9 degrades the serpin protease nexin-1, an inhibitor of uPA and the invasion of tumor cells [40]. However, the effects of nexin-1 on tumor metastasis seem to be controversial. Nexin-1 reportedly binds LRP-1 and stimulates extracellular signal-regulated kinase signaling, MMP-9 expression, and metastatic spread of mammary tumors [41]. The inconsistency of these studies is hard to explain. Given that MMP-9 can be upregulated by nexin-1, the degradation of nexin-1 by MMP-9 may represent a negative feedback regulation of nexin-1 activity.

Except for components of the ECM, there are non-ECM substrates for MMPs that include growth factors, kinase, cytokines, chemokines, and receptors. A number of growth factors can stimulate the secretion of MMPs. Conversely, metalloproteinases may release matrix-sequestered growth factors, thereby creating a vicious cycle of autocrine growth. MT1-MMP cleaves HB-EGF and removes the NH(2)-terminal 20 amino acids that are important for binding heparin. The truncated form of HB-EGF is independent of heparin and exhibits enhanced mitogenic activity [42]. Moreover, MT1-MMP degrades the Wnt/planar cell polarity protein-tyrosine kinase-7 (PTK7), an inhibitor of cell invasion. The cleavage of PTK7 by MT1-MMP leads to an increase in cell invasion and migration [43]. Tumor-associated MT1-MMP sheds RANKL and activates src-dependent prostate cancer migration and bone metastasis [44]. Thus, MT1-MMP may play pivotal roles in both the growth and metastasis of tumor cells. Moreover, MMP-13 regulates mammary tumor-induced osteolysis by activating MMP-9 and TGF-β signaling at the tumor–bone interface [45], a common feature of a variety of cancer.

Regulation of angiogenesis

MMPs also have complex roles in angiogenesis. It is known that MMPs can promote endothelial cell migration and trigger angiogenic switch. For example, MMP-9 participates in switching angiogenesis by releasing VEGF from ECM [46]. Also, MMP-2 activity was suggested to be necessary for the switch to angiogenic phenotype in an animal model [47]. Furthermore, MMPs increase the bioavailability of the pro-angiogenic growth factors vascular endothelial growth factor (VEGF), fibroblast growth factor-2 (FGF-2), and TGF-β, which stimulate proliferation and migration of endothelial cells. MT1-MMP regulates VEGF-A expression by promoting VEGFR-2 cell surface localization thereby activating VEGFR-2-Src-Akt-mTOR pathway [48]. MMP-7 degrades soluble VEGF receptor-1 (sVEGFR-1/sFlt-1), an endogenous VEGF inhibitor that sequesters VEGF and blocks its access to VEGF receptors. The degradation of sVEGFR-1 then liberates VEGF from the endogenous trap and allows its access to membrane receptors on endothelial cells [49]. However, MMPs may have adverse effects on angiogenesis. For example, MT1-MMP-mediated endoglin shedding inhibits tumor angiogenesis [50]. The generation of angiostatin and endostatin, two potent angiogenesis inhibitors, also involves MMPs. MMP-7 and MMP-9 hydrolyze plasminogen to generate angiostatin fragments [51]. Endostatin is the C-terminal proteolytic product of the collagen XVIII $\alpha 1$ chain. MMP-7

and MMP-14 can cleave collagen XVIII to generate endostatin-spanning fragment [52, 53]. Hence, we speculate that the effects of MMPs on angiogenesis may be context-dependent. The outcome relies on the availability of specific MMPs and the balance between two opposing effects of individual MMPs on angiogenesis.

Regulation of inflammation

Another notable function for MMPs is their modulatory roles in inflammation. Inflammatory response is a hallmark of cancer. Inflammatory cells, such as macrophages, neutrophils, and mast cells, are components of tumor microenvironment. The tumor-infiltrating inflammatory cells can release cytokines, polypeptide growth factors, and proteases that may regulate stromal remodeling as well as tumor cell invasion. Studies in transgenic mice demonstrate that mast cells, macrophages, and peripheral mononuclear cells amplify neoplastic cell proliferation and angiogenesis largely by release of MMP-9 [54]. The expression of MMPs is regulated by a variety of cytokines and chemokines. Elevated expression and activities of MMPs are observed in acute or chronic inflammatory diseases, such as rheumatoid arthritis and atherosclerosis. Meanwhile, MMPs themselves also manifest as regulators of inflammation. The processing of chemokines may be regulated by MMPs, generating either inactivated fragments or truncated form of chemokines with increased activity (Table 2) [55–66]. Therefore, MMPs may exhibit either pro-inflammatory activities or anti-inflammatory activities in a context-dependent manner. For example, MMP-9 potentiates the activation of pro-inflammatory cytokines such as TNF, interleukin-1β (IL1β), IL6, IL8, and chemokines such as CXCL5, CXCL6, and LIX. However, MMP-2 may dampen inflammation by processing monocyte chemoattractant proteins thereby generating chemokine receptor antagonists with anti-inflammatory properties [62]. Rheumatoid arthritis is characterized by the autoimmune inflammation of the joints and degradation of the joint cartilage. MMPs are not only essential for the migration of inflammatory cells to the damaged joints, but also responsible for the cleavage and degradation of structural components of cartilage [67]. In addition, MMPs play key roles in vascular pathologies. MMP-2 and MMP-9 contribute to the proliferation and migration of vascular smooth muscle cells that is responsible for restenosis [68].

Induction of epithelial–mesenchymal transition

Epithelial–mesenchymal transition (EMT) is a key developmental process characterized by loss of cell adhesion, repression of E-cadherin expression, and increased cell mobility [42]. EMT may be essential for numerous developmental processes including mesoderm formation and neural tube formation. During tumor progression, EMT is often activated to promote cancer cell invasion and metastasis. MMPs are important regulators or mediators of EMT. MMP-2 is necessary for the EMT that generates neural crest cells and plays an essential role in producing epithelial–mesenchymal transformations in the avian embryo [69]. In highly invasive human squamous carcinoma cells, TGF-β-induced EMT leads to the upregulation of MMP-2 through a snail and Ets-1-dependent mechanism [70]. However, overexpression of Ets-1 induced the expression of MMP-2 without EMT, which indicates that MMP-2 alone may be not enough for inducing EMT in squamous carcinoma cells [71]. Nevertheless, EMT can be induced by MMP-3 in mammary epithelial cells [72], and this effect may be antagonized by synthetic MMP inhibitor [62]. MMP-3-induced EMT involves activation of Rac1b

Table 2 The processing of chemokines by MMP

Chemokine	Processing MMP	Outcome	References
CCL2/MCP-1	MMP-1, MMP-3	Inactivation	[63]
CCL7/MCP-3	MMP-1, -2, -3, -13, -14	Inactivation	[62]
CCL8/MCP-2	MMP-3	Inactivation	[63]
CXCL1/GRO	MMP-9	Inactivation	[58]
CXCL4/PF4	MMP-9	Inactivation	[58]
CXCL5/ENA-78	MMP-8, -9	Inactivation	[59]
CXCL6/GCP-2	MMP-8, -9	Inactivation	[59]
CXCL9/MIG	MMP-8, -9		[60, 61]
CXCL10/IP-10	MMP-8, -9		[60]
CXCL12/SDF-1	MMP-1, -2, -3, -9, -13, -14	Inactivation	[56]
CTAPIII	MMP-9	Inactivation	[58]
CXCL8/IL-8	MMP-4, -8, -9, -13	Increased activity	[59]
CXCL5/ENA-78	MMP-8	Increased activity	[59]
LIX	MMP-1, -2, -8, -9, -13	Increased activity	[59]

and induction of Snail, a transcription factor that plays key role in initiating EMT [73]. In this case, MMP-3 induces the expression of Rac1b, which in turn increases the levels of reactive oxygen species, critical stimuli of snail expression and EMT. MMP-3 is also a mediator of wnt1-induced EMT [74]. Moreover, MMP-9 can cleave E-cadherin and promote EMT [65]. The expression of MMP-9 can be stimulated by snail through a MAPK- and PI3K-dependent mechanism [75]. TGF-β is a well-known inducer of EMT. MMP-28 upregulates MT1-MMP and MMP-9, which promote the processing of latent TGF-β complex thereby increasing levels of active TGF-β. Therefore, MMP-28 induces EMT and cell invasion through a TGF-β-dependent mechanism [67]. Recently, it has been demonstrated that MT1-MMP and MT2-MMP are key mediators of snail-driven EMT, tumor angiogenesis, and metastasis [76]. These findings suggest that MMP play a role in cell migration during the EMT and morphogenesis.

Regulation of cell survival

MMPs are also emerging as regulators or mediators of cell survival. Different MMPs may have contrasting effects on cell survival. Even for the same MMP, the effects on cell survival may be cell-type-specific or context-dependent. Upon induction of apoptosis, MMP-1 co-localizes with aggregated mitochondria and accumulates around fragmented nuclei, and inhibits apoptosis [77]. MMP-2 silencing causes cancer cell apoptosis by upregulating Fas/Fas-L and FADD [78]. In contrast, MMP-2 promotes endothelial cell migration but reduces cell viability under hypoxia [79]. MMP-3 stimulates ductal proliferation and branching during puberty [72] but induces apoptosis in anchorage-dependent secretory epithelium during pregnancy [80, 81]. Meanwhile, MMP-3 induces spontaneous mammary carcinogenesis [62]. Upon endoplasmic reticulum stress, MMP-3 is increased and promotes neuronal apoptosis downstream of caspase-12 but upstream of caspase-3 [82]. MMP-7 would cleave FASL and this inhibits or induces apoptosis in a context-dependent manner [83–85]. The pro-apoptotic effect of MMP-7 on pancreatic acinar cells is mediated largely by FasL and is responsible for ductal metaplasia [79]. MMP-7 also causes a decrease of FAS at the cell surface, which leads to oxaliplatin-resistance in colon cancer cells [86]. Although MMP-7 cleaves FASL and induces apoptosis, constitutive expression of matrilysin may select for cells with reduced sensitivity to Fas-mediated apoptosis and renders cancer cells more invasive [87]. MMP-9 promotes chronic lymphocytic leukemia B cell survival through its hemopexin domain but not catalytic domain, indicating an enzyme activity-independent action of MMP-9 [82]. In addition to the well-recognized extracellular function, intracellular MMP-9 promotes cell cycle progression and cell growth [88]. However, MMP-9 promotes retinal ganglion cell death after optic nerve ligation [89]. MT1-MMP downregulates the glucose-6-phosphate transporter expression and promotes bone marrow stromal cell death [90]. Active stromelysin-3 (MMP-11), but not the inactive MMP-11, increases MCF-7 survival in three-dimensional Matrigel culture via activation of p42/p44 MAP-kinase and Akt [91]. MMP-15 confers resistance of cancer cells to apoptosis induced by Fas, tumor necrosis factor-related apoptosis-inducing ligand, and serum starvation [92]. Finally, elevation in MMP activity plays a proapoptotic role in β-adrenergic receptor-stimulated apoptosis of adult rat ventricular myocytes [88].

Tumor suppressive roles for MMPs

Although many facets of MMPs action are pro-tumorigenic, some MMPs may also inhibit tumorigenesis. Collagenase-2 (MMP-8) is an MMP mainly produced by neutrophils and associated with many inflammatory conditions. MMP-8 reportedly plays a protective role in cancer through its ability to regulate the inflammatory response induced by carcinogens. Experimental manipulation of the expression levels of this enzyme alters the metastatic behavior of human breast cancer cells [93]. Thus, low levels of MMP-8 expression in tumors may be associated with a poor outcome in patients with cancer. In addition, the tumor suppressive effect of MMP-12 is observed. Overexpression of MMP-12 in colon cancer cells is associated with increased survival [94]. In fact, the complex roles of MMPs in tumor progression remain to be explored. As discussed earlier, several MMPs contribute to the generation of angiogenic inhibitors. MMP-2, MMP-7, MMP-9, and MMP-12 can convert plasminogen into angiostatin [51, 95–98]. The generation of another potent antiangiogenic factor, endostatin, may also require MMPs and elastase [99]. Another notable finding is that silencing of MMP-1 in chondrosarcoma cells inhibits distant metastasis but promotes primary tumor growth and angiogenesis [100]. This evidence suggests that temporal or spatial regulation of MMPs may be essential for angiogenesis.

Biological activities of endogenous TIMPs in cancer

The overexpression and elevated activity of MMPs correlate with tumor progression. The balance between activated MMPs and their inhibitors such as TIMPs may define the net activity of MMPs. Down-regulation of TIMPs may result in an increase in the activity of MMPs and the invasive potential of tumor cells. Conversely, tumor invasion and metastasis can be inhibited by up-regulation of TIMPs in tumor cells. Overexpression of TIMP-1 inhibits

tumor growth and metastasis of melanoma [101], suppresses human gastric cancer metastasis [102], and prevents oral squamous cell carcinoma progression [99]. Adenoviral transfer of TIMP-3 into HeLa, fibrosarcoma, and melanoma cells inhibits the invasiveness and induces apoptosis [103, 104]. Ectopic expression of TIMP-4 in human breast cancer cells inhibits invasion, metastasis, and tumor growth [102]. In addition to the involvement of MMPs in the late stages of tumorigenesis, there is evidence supporting a role for MMPs at the early stage of carcinogenesis and angiogenesis [46, 51, 62, 105, 106]. Therefore, one can speculate that TIMPs may exert tumor suppressive activities at both the early and late stages of tumorigenesis.

Although accumulating data demonstrate that TIMPs have tumor-suppressive activities, studies also show that TIMPs may play contrasting roles in tumor progression. Indeed, the stimulatory effect on cell growth was initially recognized when TIMP-1 and TIMP-2 were found to be of erythroid-potentiating activities [107, 108]. TIMP-1 and TIMP-2 are also mitogenic for non-erythroid cells, including normal keratinocytes [109], fibroblasts, lung adenocarcinoma cells, and melanoma cells [108, 110]. In addition, TIMPs exert diverse effects on apoptosis. Whereas TIMP-3 can induce apoptosis, TIMP-1, -2, -4 can protect cells from apoptosis. TIMP-1 inhibits apoptosis of B cells and human breast epithelial cells in vitro [111, 112], and rescues mammary epithelial cell apoptosis in transgenic mice [79]. TIMP-2 overexpression protects B16F10 melanoma cells from apoptosis [111]. TIMP-4 also protects human breast cancer cells from apoptosis [113]. The effects of TIMPs on cellular proliferation and apoptosis may be mediated by MMP-independent mechanisms. TIMP-1 and TIMP-2 have been shown to stimulate tyrosine kinase, focal adhesion kinase, and mitogen-activated protein kinase activity in a human osteosarcoma cell line MG-63 and in human breast cancer cell lines [113, 114]. TIMP-1, which can be upregulated by Bcl-2, seems to function downstream of Bcl-2 [112]. TIMP-1 also upregulates Bcl-XL expression in B cells [110]. Although overexpression of TIMP-1 in cancer cells inhibited metastasis, recent studies suggested that TIMP-1 may also promote tumorigenesis and metastasis in some animal models [114–116]. It is believed that TIMPs regulate cell growth and apoptosis through paracrine mechanism. Efforts have been placed on the identification of cell surface binding partners for TIMP family members. For the first time, the integrin $\alpha_3\beta_1$ was identified as a cell surface receptor for TIMP-2 [117]. TIMP-2 has been shown to inhibit endothelial cell growth in response to FGF-2, which can be attributed to TIMP-2 binding to endothelial cell surface $\alpha_3\beta_1$ thereby inhibiting FGF2-induced ERK1/2 signaling [118]. TIMP-2 also inhibits VEGF-induced VEGFR-2 phosphorylation and enhances phosphodiesterase activity in endothelial cells [119]. TIMP-3 may bind to VEGFR2 and competitively inhibit VEGF binding and subsequent kinase signaling [120]. In addition, TIMP-1 and TIMP-4 can bind to the tetraspanin CD63 and inhibit both the intrinsic and extrinsic cell death [121, 122].

Strategies for inhibition of MMPs

Synthetic MMP inhibitors

Soon after tumor biologists discovered that MMPs were involved in the invasion and metastasis of cancer cells, intensive efforts have been taken to develop MMP inhibitors (MMPIs) to halt the spreading of cancer cells [5]. Most small-molecule MMPIs are hydroxamate-based in which the hydroxamate acts as a zinc-binding group. Batimastat (BB-94), BB-1101, and marimastat (BB-2516) are the best-known examples of succinyl hydroxamate MMPIs that display efficacy in preclinical studies of cancer therapy [123–127]. Batimastat is a potent MMPI with a broad spectrum of substrates and inhibits tumor growth and metastasis in a variety of tumor models [128, 129]. In addition, batimastat potentiated the growth-suppressive effects of cisplatin and demonstrated synergistic anti-tumor effects with docetaxel and captopril [128, 129]. However, there are some unwanted effects that may hinder the effectiveness of batimastat in cancer therapy. Treatment with batimastat may induce stromal uPA expression that can halt the anti-invasive activity [130]. Moreover, batimastat treatment promotes T-cell lymphoma liver metastasis by inducing liver-specific overexpression of MMP-2, -9, and angiogenesis factors [131]. These contradicting outcomes may be attributed to the redundancy of MMP family, temporal, and spatial determinants of MMP action, and compensatory response to MMP inhibition.

In contrast to batimastat, marimastat is orally active, with an absolute bioavailability of 20–50% in preclinical studies [132]. Inhibition of cervical lymph node metastasis by marimastat (BB-2516) was observed in an orthotopic oral squamous cell carcinoma implantation model [133]. Marimastat also inhibited the peritoneal spread of gastric carcinoma in nude mice [134]. These promising preclinical studies led to the initiation of a series of clinical trials for marimastat in treating advanced tumors. However, a phase III trial involving 505 patients with stage III NSCLC randomized to receive either marimastat or placebo failed to show a significant difference in progression-free survival or overall survival between the two groups [135]. Later on, clinical trials involving cancer patients randomized to receive either chemotherapy or combination of marimastat and chemotherapy were initiated. Once again, marimastat failed in clinical trials as part of a combination therapy for pancreatic, gastric, or ovarian carcinoma, and glioma [136].

Bramhall and colleagues conducted a randomized clinical trial involving 414 patients with advanced pancreatic cancer to receive either gemcitabine or marimastat [137]. Marimastat showed inferior efficacy to gecitabine in treating pancreatic cancer. Musculoskeletal toxicity was reported in 39–55% of patients across different dose levels of marimastat. Combination of mariamastat and gemcitabine did not show superior efficacy compared to gemcitabine alone in treating advanced pancreatic cancer [138].

At the very beginning, most hydroxamate-based MMPIs are broad-spectrum inhibitors that lack specificity. Some researchers argue that successful anticancer inhibitor may possess MMP selectivity against the MMP subtype. Later on, new compounds were developed that took advantage of alternative zinc-binding groups, such as carboxylic acids, thiols, and phosphorous-based groups. Prinomastat (AG3340) is an oral active, non-peptide inhibitor of MMPs, including MMP-2, -9, MT1-MMP, and collagenase III [139]. Prinomastat demonstrated anti-tumor activities in a variety of preclinical studies upon oral administration, including cancer of pancreas, breast, lung, melanoma, and glioma models [140–143]. The anti-tumor efficacy of prinomastat in chemoresistant human non-small cell lung cancer was also observed either when prinomastat was used as a single agent or when it was combined with chemotherapy. Unfortunately, this compound failed in phase III clinical trials as a combination with taxol and caboplatin for advanced non-small lung cancer and with mitoxantrone for hormone-resistant prostate cancer, due to the drug's lack of effectiveness in patients with late-stage disease [144, 145]. Patient safety was not a factor in this case. It remains to be known whether such drugs may be more effective in earlier stages of disease, when tumors are smaller and may be more sensitive to these agents. Prinomastat was designed to be highly selective for several specific matrix metalloproteinase enzymes. In addition, the carboxylate BAY12-9566 is a relatively selective inhibitor of gelatinases and stromelysin-1. The clinical trial of Bay 12,9566 was stopped due to more rapid tumor growth in patients treated with the drug. BAY12-9566 proved inferior to gemcitabine in a randomized trial of 277 patients with advanced pancreatic cancer [146]. It is possible that selective inhibition of several MMPs may not be sufficient enough to cause significant inhibition of tumor growth. Furthermore, suppressing several enzymes may also lead to increased production by cancer cells of other enzymes that may compensate the loss of those enzymes, leading to continued tumor growth.

Cis-2-aminocyclohexylcarbamoylphosphonic acid (cis-ACCP) is another selective MMP inhibitor that has been evaluated in vitro and in vivo cancer metastasis models. It significantly inhibits metastasis formation in mice [147]. In addition, S-3304 is a potent, orally active, noncytotoxic inhibitor of MMPs, primarily MMP-2 and MMP-9. S-3304 prolongs survival in mice xenografts and is well tolerated in healthy volunteers [148]. Ro-28-2653, a selective and orally bioavailable MMPI with inhibitory activity against MMPs expressed by tumor and/or stromal cells, is a potent antitumor and antiangiogenic agent [149]. However, it is unknown whether these agents would be effective in a clinical setting.

Pyrimidine dicarboxamides are a class of highly selective MMP-13 inhibitors. These inhibitors do not interact with the catalytic zinc but bind the S1 pocket and extend into an additional S1 side pocket, which is unique to MMP-13 [150]. Compared to the catalytic domain, the binding sites outside the active domain of the MMPs are less conserved and are related to substrate selection [151]. Therefore, targeting the binding sites outside of the catalytic domain represents an alternative approach to achieve MMP- and substrate-specific interference.

The mechanism-based inhibition of selective MMP is an alternative strategy. These inhibitors bind to the active sites of specific MMP and initiate slow binding profile for the onset of inhibition, leading to covalent enzyme modification [152]. In this regard, the specific inhibition of single MMP can be achieved. SB-3CT is a specific gelatinase inhibitor based on this mechanism. The key event in the inhibition of MMP-2 by SB-3CT is enzyme-catalyzed active site ring-opening of the thiirane moiety, giving a stable zinc-thiolate species [153]. It inhibited tumor growth, liver metastasis, and improved survival in an aggressive mouse model of T-cell lymphoma [154]. Also, it reduced prostate cancer growth, osteolysis, and angiogenesis in a bone metastasis model [155].

Antibody-based MMP inhibition

Antibody-based therapy is expanding in controlling diseases including cancer. It remains to be known whether an antibody to a specific MMP will be an effective anti-cancer agent. DX-2400, a highly selective fully human MMP-14 inhibitory antibody discovered using phage display technology, blocked proMMP-2 processing in tumor and stromal cells, inhibited angiogenesis, and slowed tumor progression and metastasis in vivo [156]. The combination of potency, selectivity, and robust in vivo activity shows the potential of a selective MMP-14 inhibitor for the treatment of solid tumors. Certainly, more inhibitory antibodies to individual MMP remain to be developed. The efficacy of antibody-based MMP inhibition, either as a monotherapy or in combination with chemotherapy, in treating human tumors needs to be tested.

Inhibitors of MMPs transcription

Another strategy of inhibiting MMPs is to suppress the expression of MMPs. Chemically modified tetracyclines

(CMTs) without antibiotic activities can bind metal ions such as calcium and zinc, and to affect *MMP* gene transcription [157]. A high-throughput screening system identifies a small molecule 5-methyl-2-(4-methylphenyl)-1H-benzimidazole (MPBD) that can repress MMP-9 expression by antagonizing AP-1 transactivation activity [158]. Consistent with this effect, MPBD inhibits MMP-9-dependent invasion of oral cancer cells, preosteoclast migration, and RANKL-induced osteoclast activity over concentration ranges that repressed MMP-9 expression. In addition, the EGFR inhibitor Gefitinib inhibits MMP-9 and MMP-2 secretion and mRNA expression in HT29 cells [159]. Given that the transcription of MMP is regulated by NF-κB, inhibiting NF-κB activity may reduce the expression of MMP thereby suppressing the intrinsic migration ability and invasive potential of tumor cells [160]. Moreover, another transcription factor, Ets-1, can regulate the transcription of *MMP-2*. The COX-2 product prostaglandin E_2 promotes pancreatic cancer metastasis through an ERK/Ets-1-dependent induction of *MMP-2* expression. Rofecoxib, a COX-2 inhibitor, suppressed Ets-1 binding activity and MMP-2 expression, and cellular migratory and invasive potentials [161].

In addition to synthetic inhibitors of MMP transcription, there are natural compounds capable of inhibiting MMP expression. Cannabinoids are natural compounds that down-regulate MMP-2 expression, inhibit tumor angiogenesis, and halt tumor cells spreading [162]. Curcumin, a polyphenol derived from the plant *Curcuma longa*, inhibited the PMA-induced mRNA expression of *MMP-1*, *-3*, *-9*, and *-14* in human astroglioma cells by repressing the DNA-binding and transcriptional activities of AP-1 and the PMA-induced MAP kinase activities, which were involved in modulating the expression of MMPs [163]. These natural compounds may hold promise in cancer chemoprevention.

Stimulators of endogenous MMP inhibitors

Finally, an alternative strategy for inhibiting MMPs is to up-regulate endogenous MMP inhibitors. miRNA is emerging as another regulator of gene expression. A recent study demonstrated that miR-21 could inhibit the expression of RECK and TIMP-3, two tumor suppressors and MMP inhibitors [164]. miR-21 is significantly elevated in glioblastoma and in many other tumors of various origins [165, 166]. This microRNA has been implicated in various aspects of carcinogenesis, including cellular proliferation, apoptosis, and migration. Therefore, targeting this microRNA may increase the expression of RECK and TIMP3 thereby inhibiting MMPs. In addition, 1-alpha-25-dihydroxyvitamin D3 (1, 25-VD) can inhibit the expression of MMP-9 and cathepsins, while it increases the expression of their counterparts, tissue inhibitors of metalloproteinase-1 (TIMP-1) and cathepsin inhibitors [167].

Concluding remarks

ECM homeostasis is critical for tissue architecture that regulates development and cancer. Among diverse proteases, MMPs play important roles in regulating ECM homeostasis. The best-known function of MMPs is to degrade the components of ECM. During tumor progression, the expression and activation of MMPs is upregulated in the tumor infiltrative frontier. Traditionally, MMPs are known as one of the key promotors of tumor metastasis. However, mounting evidence demonstrates that MMPs also play complex roles in malignant transformation and tumor onset (Fig. 1). In addition to ECM, MMPs can target non-matrix proteins such as growth factors and their receptors, chemokines, adhesive molecules. MMPs not only regulate the invasive potential of cancer cells but also regulate cellular proliferation, apoptosis, EMT, angiogenesis, and inflammatory response. Notably, these processes may be temporally and spatially regulated by MMPs. The individual MMP in such a large family may share some

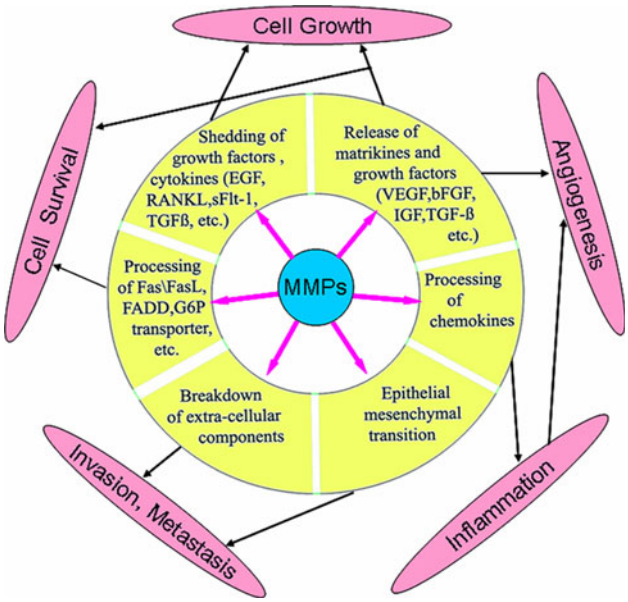

Fig. 1 Complex roles for MMP in cancer. The multifunctional MMPs can not only degrade structural components within the extracellular matrix but also process growth factors and receptors (EGF, TGF-β, sFlt-1, etc.), cytokines and chemokines, and apoptosis-related molecules. Moreover, MMPs are involved in epithelial-mesenchymal transition. Therefore, MMPs have effects on cell growth and survival, angiogenesis, inflammation, invasion, and metastasis. These effects may be context-dependent. While MMPs can stimulate angiogenesis through release of angiogenic factors such as VEGF and bFGF, they are also involved in generating angiogenesis inhibitors such as angiostatin and endostatin

common features but also possesses unique substrates or function.

MMPs have been considered as promising targets for cancer therapy on the basis of their aberrant up-regulation in malignant tumors and their ability to degrade components of the ECM. Multiple pharmacological strategies can be taken to target MMPs (Fig. 2). Indeed, preclinical studies testing the efficacy of MMP suppression in tumor models were encouraging. Based on the promising results from preclinical studies on synthetic MMPs inhibitors, the clinical trials on MMPs inhibitors in cancer therapy were conducted. Unfortunately, the results of these trials in cancer patients have been rather disappointing. Recent studies demonstrate that MMPs may have paradoxical roles in tumor progression, with either tumor-promoting effects or tumor suppressive effects in a context-dependent manner. Increase in our knowledge of the molecular mechanisms underlying MMPs action in tumor progression may help understand the failure in previous clinical trials for MMP inhibitors. Meanwhile, these new insights would stimulate the development of novel approaches to target MMPs. Regardless of the clinical outcome, the potential efficacy of MMP inhibitors in cancer therapeutics must be based on the success in inhibiting MMPs. In this respect, imaging activity of specific MMPs in vivo with probes may help evaluate the efficacy of MMP inhibition in certain tumors [168]. Given that members of the MMP family have different roles during cancer progression, the use of broad-spectrum MMP inhibitors may result in unsatisfying clinical outcomes due to the inhibition of tumor suppressive MMPs. Although novel strategies to develop inhibitors of individual MMP are emerging, it is even more difficult to discern what MMPs to target and when to target. Taking into consideration of the redundancy of MMPs, it remains unknown if these specific MMP inhibitors would be successful anticancer agents. In addition, it is possible that

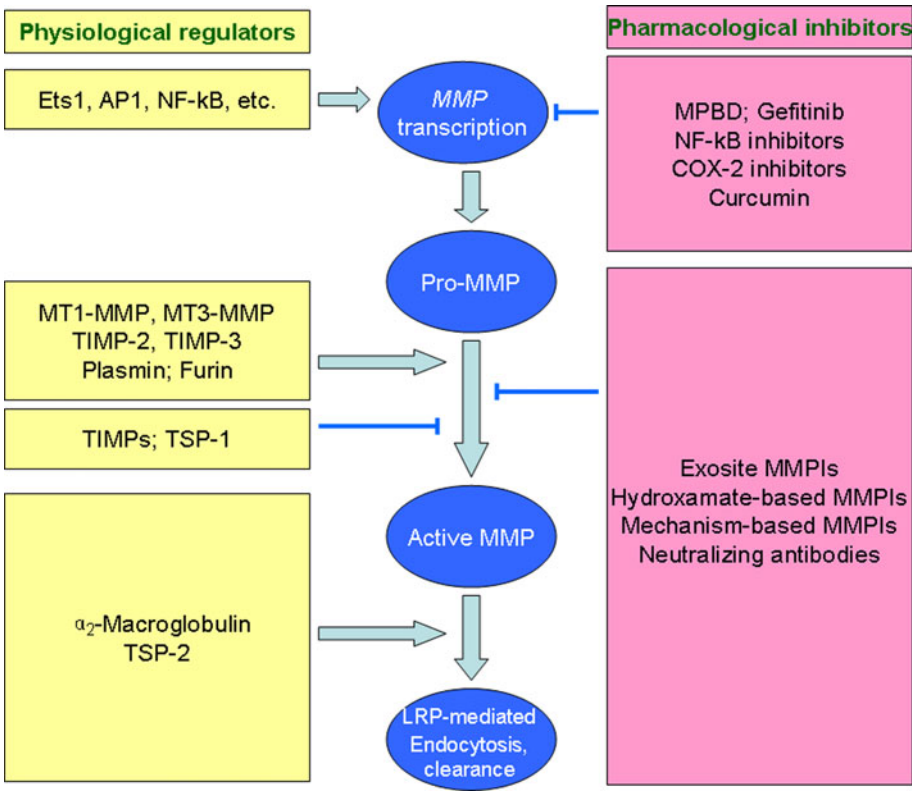

Fig. 2 Physiological and pharmacological regulation of MMP. The inactive pro-MMPs are synthesized from *MMPs* transcripts. The transcription of *MMPs* is regulated by transcription factors including Ets1, AP1, and NF-κB. The mechanisms underlying the activation of pro-MMPs are complex. Both MT1-MMP and MT3-MMP are involved in the activation of pro-MMPs. In addition, MMPs may be activated by plasmin and furin-like mechanism. Tissue inhibitors of metalloproteinase (TIMP) are endogenous MMP inhibitors that prevent the activation of pro-MMPs. TSP1 also inhibits the activation of MMPs. Paradoxically, TIMP-2 and TIMP-3 are involved in the activation of pro-MMP as well. The turnover of active MMPs may involve LRP-mediated endocytosis and clearance. α_2-Macroglobulin and TSP-2 act as endogenous MMP inhibitors through promoting LRP-mediated endocytosis and clearance of active MMPs. The pharmacological inhibitors of *MMPs* transcription include 5-methyl-2-(4-methylphenyl)-1H-benzimidazole (MPBD), Gefitinib, NF-κB inhibitors, COX-2 inhibitors, and curcumin. In addition to hydroxamate-based MMP inhibitors (MMPIs), there are exosite MMPIs and mechanism-based MMPIs. Moreover, neutralizing antibodies to individual MMP are emerging as another choice for selective inhibition of MMP

inhibition of individual MMP will paradoxically up-regulates other MMPs, leading to a compensatory response that hampers the anticancer effects.

Since many clinical trials were conducted in patients with late-stage tumors, it remains to be known whether MMP inhibitors may have benefits in treating early stage tumors. In addition, adverse effects, including musculoskeletal syndrome, limit the maximal-tolerated dose of some early generation of MMP inhibitors such as marimastat and thereby dampen drug efficacy. These side-effects may be the result of broad-spectrum inhibition of the MMP family. Therefore, toxicity effects must be minimalized to ensure that the drug efficacy would not be limited by the maximum-tolerated dose. Moreover, biomarkers may add substantial value to clinical practice by providing an integrated approach to prediction or monitoring drug response using the genetic makeup of the tumor and the genotype of the patient to guide personalized treatment selection. The genetic background of the host might be an issue that determines the efficacy of MMP inhibitors as anticancer agents. A study in a mouse model of breast cancer demonstrated that the anti-metastatic outcome of MMP-9 inhibition was strain-specific [169]. Further work on the identification of specific genetic factors that affect the efficacy of MMP inhibitors are warranted. These studies may be helpful for the use of tumor genotyping to guide the choice of MMP inhibitors. Other predictive biomarkers for MMP inhibitors need to be addressed in future studies. It already takes decades to learn what roles MMPs have in tumor progression. There is still a long way to go to uncover the complex functions of MMPs in tumorigenesis. Certainly, the development of MMP inhibitors and subsequent pharmacological improvements would benefit from knowledge of MMPs.

Acknowledgments The work in the authors' lab was supported by the National Natural Science Foundation of China (Grants 30900554 and 30973435).

References

1. Bhowmick NA, Neilson EG, Moses HL (2004) Stromal fibroblasts in cancer initiation and progression. Nature 432:332–337
2. Stetler-Stevenson WG, Aznavoorian S, Liotta LA (1993) Tumor cell interactions with the extracellular matrix during invasion and metastasis. Annu Rev Cell Biol 9:541–573
3. Friedl P, Wolf K, Travel T (2008) The role of proteases in individual and collective cancer cell invasion. Cancer Res 68:7247–7249
4. Hojilla C, Wood G, Khokha R (2008) Inflammation and breast cancer. Metalloproteinases as common effectors of inflammation and extracellular matrix breakdown in breast cancer. Breast Cancer Res 10:205
5. Overall CM, Lopez-Otin C (2002) Strategies for MMP inhibition in cancer: innovations for the post-trial era. Nat Rev Cancer 2:657–672
6. Hu J, Van den Steen PE, Sang Q-X, Opdenakker G (2007) Matrix metalloproteinase inhibitors as therapy for inflammatory and vascular diseases. Nat Rev Drug Discov 6:480–498
7. Opdenakker G, Van den Steen PE, Van Damme J (2001) Gelatinase B: a tuner and amplifier of immune functions. Trends Immunol 22:571–579
8. Sato H, Seiki M (1996) Membrane-type matrix metalloproteinases (MT-MMPs) in tumor metastasis. J Biochem 119:209–215
9. Sohail A, Sun Q, Zhao H, Bernardo MM, Cho JA, Fridman R (2008) MT4-(MMP17) and MT6-MMP (MMP25), a unique set of membrane-anchored matrix metalloproteinases: properties and expression in cancer. Cancer Metastasis Rev 27:289–302
10. Vincenti MP (2001) The matrix metalloproteinase (MMP) and tissue inhibitor of metalloproteinase (TIMP) genes. Transcriptional and posttranscriptional regulation, signal transduction and cell-type-specific expression. Methods Mol Biol 151:121–148
11. Borden P, Heller RA (1997) Transcriptional control of matrix metalloproteinases and the tissue inhibitors of matrix metalloproteinases. Crit Rev Eukaryot Gene Expr 7:159–178
12. Brinckerhoff CE, Rutter JL, Benbow U (2000) Interstitial collagenases as markers of tumor progression. Clin Cancer Res 6:4823–4830
13. Liacini A, Li WQ, Huang W, Dehnade F, Ahmad M, Zafarullah M (2003) Induction of matrix metalloproteinase-13 gene expression by TNF-α is mediated by MAP kinases, AP-1, and NF-κB transcription factors in articular chondrocytes. Exp Cell Res 288:208–217
14. Remacle AG, Rozanov DV, Fugere M, Day R, Strongin AY (2006) Furin regulates the intracellular activation and the uptake rate of cell surface-associated MT1-MMP. Oncogene 25:5648–5655
15. Rozanov DV, Hahn-Dantona E, Strickland DK, Strongin AY (2004) The low-density lipoprotein receptor-related protein LRP is regulated by membrane type-1 matrix metalloproteinase (MT1-MMP) proteolysis in malignant cells. J Biol Chem 279:4260–4268
16. Yang Z, Strickland DK, Bornstein P (2001) Extracellular matrix metalloproteinase 2 levels are regulated by the low-density lipoprotein-related scavenger receptor and thrombospondin 2. J Biol Chem 276:8403–8408
17. Douglas DA, Shi YE, Sang QA (1997) Computational sequence analysis of the tissue inhibitor of metalloproteinase family. J Protein Chem 16:237–255
18. Bigg HF, Shi YE, Liu YE, Steffensen B, Overall CM (1997) Specific, high affinity binding of tissue inhibitor of metalloproteinases-4 (TIMP-4) to the COOH-terminal hemopexin-like domain of human gelatinase A. J Biol Chem 272:15496–15500
19. Seiki M (1999) Membrane-type matrix metalloproteinases. APMIS 107:137–143
20. Strongin AY, Collier I, Bannikov G, Marmer BL, Grant GA, Goldberg GI (1995) Mechanism of cell surface activation of 72-kDa type IV collagenase. J Biol Chem 270:5331–5338
21. Hernandez-Barrantes S, Toth M, Bernardo MM et al (2000) Binding of active (57 kDa) membrane type 1-matrix metalloproteinase (MT1-MMP) to tissue inhibitor of metalloproteinase (TIMP)-2 regulates MT1-MMP processing and pro-MMP-2 activation. J Biol Chem 275:12080–12089
22. Butler GS, Butler MJ, Butler GS et al (1998) The TIMP2 membrane type 1 metalloproteinase "receptor" regulates the concentration and efficient activation of progelatinase A. J Biol Chem 273:871–880
23. Bigg HF, Morrison CJ, Butler GS et al (2001) Tissue inhibitor of metalloproteinases-4 inhibits but does not support the activation of gelatinase A via efficient inhibition of membrane type 1-matrix metalloproteinase. Cancer Res 61:3610–3618

24. Cao J, Drews M, Lee HM, Conner C, Bahou WF, Zucker S (1998) The propeptide domain of membrane type 1 matrix metalloproteinase is required for binding of tissue inhibitor of metalloproteinases and for activation of pro-gelatinase A. J Biol Chem 273:34745–34752
25. Caterina JJ, Yamada S, Caterina NCM et al (2000) Inactivating mutation of the mouse tissue inhibitor of metalloproteinases-2 (Timp-2) gene alters ProMMP-2 activation. J Biol Chem 275:26416–26422
26. Itoh Y, Ito A, Iwata K, Tanzawa K, Mori Y, Nagase H (1998) Plasma membrane-bound tissue inhibitor of metalloproteinases (TIMP)-2 specifically inhibits matrix metalloproteinase 2 (gelatinase A) activated on the cell surface. J Biol Chem 273:24360–24367
27. Zhao H, Bernardo MM, Osenkowski P et al (2004) Differential inhibition of membrane type 3 (MT3)-matrix metalloproteinase (MMP) and MT1-MMP by tissue inhibitor of metalloproteinase (TIMP)-2 and TIMP-3 regulates pro-MMP-2 activation. J Biol Chem 279:8592–8601
28. English JL, Kassiri Z, Koskivirta I et al (2006) Individual *Timp* deficiencies differentially impact pro-MMP-2 activation. J Biol Chem 281:10337–10346
29. Knäuper V, López-Otin C, Smith B et al (1996) Cellular mechanisms for human procollagenase-3 (MMP-13) activation. J Biol Chem 271:17124–17131
30. Lubbe WJ, Zhou Z, Fu W et al (2006) Tumor epithelial cell matrix metalloproteinase 9 is a target for antimetastatic therapy in colorectal cancer. Clin Cancer Res 12:1876–1882
31. Samnegard A, Silveira A, Lundman P et al (2005) Serum matrix metalloproteinase-3 concentration is influenced by MMP-3 1612 5A/6A promoter genotype and associated with myocardial infarction. J Internal Med 258:41–419
32. Poincloux R, Lizárraga F, Chavrier P (2009) Matrix invasion by tumour cells: a focus on MT1-MMP trafficking to invadopodia. J Cell Sci 122:3015–3024
33. Ducaa L, Alixb A, Hayea B, Debellea L (2004) Elastin as a matrikine. Crit Rev Oncol Hematol 49:235–244
34. Koshikawa N, Minegishi T, Sharabi A, Quaranta V, Seiki M (2005) Membrane-type matrix metalloproteinase-1 (MT1-MMP) is a processing enzyme for human laminin gamma 2 chain. J Biol Chem 280:88–93
35. Sadowski T, Dietrich S, Koschinsky F et al (2005) Matrix metalloproteinase 19 processes the laminin 5 gamma 2 chain and induces epithelial cell migration. Cell Mol Life Sci 62:870–880
36. Takafuji V, Forgues M, Unsworth E, Goldsmith P, Wang XW (2007) An osteopontin fragment is essential for tumor cell invasion in hepatocellular carcinoma. Oncogene 26:6361–6371
37. Manes S, Mira E, Barbacid MM et al (1997) Identification of insulin-like growth factor-binding protein-1 as a potential physiological substrate for human stromelysin-3. J Biol Chem 272:25706–25712
38. Manes S, Llorente M, Lacalle RA et al (1999) The matrix metalloproteinase-9 regulates the insulin-like growth factor-triggered autocrine response in DU-145 carcinoma cells. J Biol Chem 274:6935–6945
39. Rorive S, Berton A, D'Haene N et al (2008) Matrix metalloproteinase-9 interplays with the IGFBP2–IGFII complex to promote cell growth and motility in astrocytomas. Glia 56:1679–1690
40. Xu D, McKee CM, Cao Y, Ding Y, Kessler BM, Muschel RJ (2010) Matrix metalloproteinase-9 regulates tumor cell invasion through cleavage of protease nexin-1. Cancer Res 70:6988–6998
41. Fayard B, Bianchi F, Dey J et al (2009) The serine protease inhibitor protease nexin-1 controls mammary cancer metastasis through LRP-mediated MMP-9 expression. Cancer Res 69:5690–5698
42. Koshikawa N, Mizushima H, Minegishi T, Iwamoto R, Mekada E, Seiki M (2010) Membrane type 1-matrix metalloproteinase cleaves off the NH2-terminal portion of heparin-binding epidermal growth factor and converts it into a heparin-independent growth factor. Cancer Res 70:6093–6103
43. Golubkov VS, Chekanov AV, Cieplak P, Aleshin AE, Chernov AV, Zhu W, Radichev IA, Zhang D, Dong PD, Strongin AY (2010) The Wnt/planar cell polarity protein-tyrosine kinase-7 (PTK7) is a highly efficient proteolytic target of membrane type-1 matrix metalloproteinase: implications in cancer and embryogenesis. J Biol Chem 285:35740–35749
44. Sabbota AL, Kim H-RC, Zhe X, Fridman R, Bonfil RD, Cher ML (2010) Shedding of RANKL by tumor-associated MT1-MMP activates Src-dependent prostate cancer cell migration. Cancer Res 70:5558–5566
45. Nannuru KC, Futakuchi M, Varney ML, Vincent TM, Marcusson EG, Singh RK (2010) Matrix metalloproteinase (MMP)-13 regulates mammary tumor-induced osteolysis by activating MMP9 and transforming growth factor-beta signaling at the tumor-bone interface. Cancer Res 70:3494–3504
46. Bergers G, Brekken R, McMahon G et al (2000) Matrix metalloproteinase-9 triggers the angiogenic switch during carcinogenesis. Nat Cell Biol 2:737–744
47. Fang J, Shing Y, Wiederschain D et al (2000) Matrix metalloproteinase-2 is required for the switch to the angiogenic phenotype in a tumor model. Proc Natl Acad Sci USA 97:3884–3889
48. Eisenach PA, Roghi C, Fogarasi M, Murphy G, English WR (2010) MT1-MMP regulates VEGF-A expression through a complex with VEGFR-2 and Src. J Cell Sci 123:4182–4193
49. Ito T-K, Ishii G, Saito S et al (2009) Degradation of soluble VEGF receptor-1 by MMP-7 allows VEGF access to endothelial cells. Blood 113:2363–2369
50. Hawinkels LJ, Kuiper P, Wiercinska E et al (2010) Matrix metalloproteinase-14 (MT1-MMP)–mediated endoglin shedding inhibits tumor angiogenesis. Cancer Res 70:4141–4150
51. Patterson BC, Sang QA (1997) Angiostatin-converting enzyme activities of human matrilysin (MMP-7) and gelatinase B/Type IV collagenase (MMP-9). J Biol Chem 272:28823–28825
52. Lin HC, Chang JH, Jain S et al (2001) Matrilysin cleavage of corneal collagen type XVIII NC1 domain and generation of a 28-kDa fragment. Invest Ophthalmol Vis Sci 42:2517–2524
53. Chang JH, Javier JAD, Chang GY, Oliveira H, Azar D (2005) Functional characterization of neostatins, the MMP-derived, enzymatic cleavage products of type XVIII collagen. FEBS Lett 579:3601–3606
54. Coussens LM, Raymond WW, Bergers G et al (1999) Inflammatory mast cells up-regulate angiogenesis during squamous epithelial carcinogenesis. Genes Devel 13:1382–1397
55. Zhang K, McQuibban GA, Silva C et al (2003) HIV-induced metalloproteinase processing of the chemokine stromal cell derived factor-1 causes neurodegeneration. Nat Neurosci 6:1064–1071
56. McQuibban GA, Butler GS, Gong JH et al (2001) Matrix metalloproteinase activity inactivates the CXC chemokine stromal cell-derived factor-1. J Biol Chem 276:43503–43508
57. Dean RA, Overall CM (2007) Proteomics discovery of metalloproteinase substrates in the cellular context by iTRAQ labeling reveals a diverse MMP-2 substrate degradome. Mol Cell Proteomics 6:611–623
58. Van den Steen PE, Proost P, Wuyts A, Van Damme J, Opdenakker G (2000) Neutrophil gelatinase B potentiates interleukin-8 tenfold by aminoterminal processing, whereas it degrades

CTAP-III, PF-4, and GRO-alpha and leaves RANTES and MCP-2 intact. Blood 96:2673–2681
59. Van den Steen PE, Wuyts A, Husson SJ, Proost P, Van Damme J, Opdenakker G (2003) Gelatinase B/MMP-9 and neutrophil collagenase/MMP-8 process the chemokines human GCP-2/CXCL6, ENA-78/CXCL5 and mouse GCP-2/LIX and modulate their physiological activities. Eur J Biochem 270:3739–3749
60. Van den Steen PE, Husson SJ, Proost P, Van Damme J, Opdenakker G (2003) Carboxyterminal cleavage of the chemokines MIG and IP-10 by gelatinase B and neutrophil collagenase. Biochem Biophys Res Commun 310:889–896
61. Liao F, Rabin RL, Yannelli JR, Koniaris LG, Vanguri P, Farber JM (1995) Human Mig chemokine: biochemical and functional characterization. J Exp Med 182:1301–1314
62. McQuibban GA, Gong JH, Tam EM, McCulloch CA, Clark-Lewis I, Overall CM (2000) Inflammation dampened by gelatinase A cleavage of monocyte chemoattractant protein-3. Science 289:1202–1206
63. McQuibban GA, Gong JH, Wong JP, Wallace JL, Clark-Lewis I, Overall CM (2002) Matrix metalloproteinase processing of monocyte chemoattractant proteins generates CC chemokine receptor antagonists with anti-inflammatory properties in vivo. Blood 100:1160–1167
64. Tam EM, Morrison CJ, Wu YI, Stack MS, Overall CM (2004) Membrane protease proteomics: isotope-coded affinity tag MS identification of undescribed MT1-matrix metalloproteinase substrates. Proc Natl Acad Sci USA 101:6917–6922
65. Tester AM, Cox JH, Connor AR et al (2007) LPS responsiveness and neutrophil chemotaxis in vivo require PMN MMP-8 activity. PLoS One 2:e312
66. Balbin M, Fueyo A, Tester AM et al (2003) Loss of collagenase-2 confers increased skin tumor susceptibility to male mice. Nat Genet 35:252–257
67. van den Steen PE, Proost P, Grillet B et al (2002) Cleavage of denatured natural collagen type II by neutrophil gelatinase B reveals enzyme specificity, post-translational modifications in the substrate, and the formation of remnant epitopes in rheumatoid arthritis. FASEB J 16:379–389
68. Cho A, Reidy MA (2002) Matrix metalloproteinase-9 is necessary for the regulation of smooth muscle cell replication and migration after arterial injury. Circ Res 91:845–851
69. Duong TD, Erickson CA (2004) MMP-2 plays an essential role in producing epithelial-mesenchymal transformations in the avian embryo. Dev Dyn 229:42–53
70. Xu J, Lamouille S, Derynck R (2009) TGF-β-induced epithelial to mesenchymal transition. Cell Res 19:156–172
71. Taki M, Verschueren K, Yokoyama K, Nagayama M, Kamata N (2006) Involvement of Ets-1 transcription factor in inducing matrix metalloproteinase-2 expression by epithelial-mesenchymal transition in human squamous carcinoma cells. Int J Oncol 28:487–496
72. Lochter A, Galosy S, Muschler J, Freedman N, Werb Z, Bissell MJ (1997) Matrix metalloproteinase stromelysin-1 triggers a cascade of molecular alterations that leads to stable epithelial-to-mesenchymal conversion and a premalignant phenotype in mammary epithelial cells. J Cell Biol 139:1861–1872
73. Radisky DC, Levy DD, Littlepage LE et al (2005) Rac1b and reactive oxygen species mediate MMP-3-induced EMT and genomic instability. Nature 436:123–127
74. Blavier L, Lazaryev A, Shi XH, Dorey FJ, Shackleford GM, DeClerck YA (2010) Stromelysin-1 (MMP-3) is a target and a regulator of Wnt1-induced epithelial–mesenchymal transition (EMT). Cancer Biol Ther 10:198–208
75. Jordà M, Olmeda D, Vinyals A, Valero E et al (2005) Upregulation of MMP-9 in MDCK epithelial cell line in response to expression of the Snail transcription factor. J Cell Sci 118:3371–3385
76. Ota I, Li X-Y, Hu Y, Weiss SJ (2009) Induction of a MT1-MMP and MT2-MMP-dependent basement membrane transmigration program in cancer cells by Snail1. Proc Natl Acad Sci USA 106:20318–20323
77. Limb GA, Matter K, Murphy G et al (2005) Matrix metalloproteinase-1 associates with intracellular organelles and confers resistance to lamin A/C degradation during apoptosis. Am J Pathol 166:1555–1563
78. Chetty C, Bhoopathi P, Lakka SS, Rao JS (2007) MMP-2 siRNA induced Fas//CD95-mediated extrinsic II apoptotic pathway in the A549 lung adenocarcinoma cell line. Oncogene 26:7675–7683
79. Ben-Yosef Y, Miller A, Shapiro S, Lahat N (2005) Hypoxia of endothelial cells leads to MMP-2-dependent survival and death. Am J Physiol Cell Physiol 289:C1321–C1331
80. Alexander CM, Howard EW, Bissell MJ, Werb Z (1996) Rescue of mammary epithelial cell apoptosis and entactin degradation by a tissue inhibitor of metalloproteinases-1 transgene. J Cell Biol 135:1669–1677
81. Thomasset N, Lochter A, Sympson CJ et al (1998) Expression of autoactivated stromelysin-1 in mammary glands of transgenic mice leads to a reactive stroma during early development. Am J Pathol 153:457–467
82. Kim E-M, Shin E-J, Choi JH et al (2010) Matrix metalloproteinase-3 is increased and participates in neuronal apoptotic signaling downstream of caspase-12 during endoplasmic reticulum stress. J Biol Chem 285:16444–16452
83. Powell WC, Fingleton B, Wilson CL, Boothby M, Matrisian LM (1999) The metalloproteinase matrilysin proteolytically generates active soluble Fas ligand and potentiates epithelial cell apoptosis. Curr Biol 9:1441–1447
84. Mitsiades N, Yu WH, Poulaki V, Tsokos M, Stamenkovic I (2001) Matrix metalloproteinase-7-mediated cleavage of Fas ligand protects tumor cells from chemotherapeutic drug cytotoxicity. Cancer Res 61:577–581
85. Strand S, Vollmer P, van den Abeelen L et al (2004) Cleavage of CD95 by matrix metalloproteinase-7 induces apoptosis resistance in tumour cells. Oncogene 23:3732–3736
86. Almendro V, Ametller E, Garcia-Recio S et al (2009) The role of MMP7 and its cross-talk with the FAS/FASL system during the acquisition of chemoresistance to oxaliplatin. PLoS One 4:e4728
87. Fingleton B, Vargo-Gogola T, Crawford HC, Matrisian LM (2001) Matrilysin [MMP-7] expression selects for cells with reduced sensitivity to apoptosis. Neoplasia 3:459–468
88. Sans-Fons MG, Sole S, Sanfeliu C, Planas AM (2010) Matrix metalloproteinase-9 and cell division in neuroblastoma cells and bone marrow macrophages. Am J Pathol 177:2870–2885
89. Chintala SK, Zhang X, Austin JS, Fini ME (2002) Deficiency in matrix metalloproteinase gelatinase B (MMP-9) protects against retinal ganglion cell death after optic nerve ligation. J Biol Chem 277:47461–47468
90. Currie JC, Fortier S, Sina A, Galipeau J, Cao J, Annabi B (2007) MT1-MMP down-regulates the glucose 6-phosphate transporter expression in marrow stromal cells: a molecular link between pro-MMP-2 activation, chemotaxis, and cell survival. J Biol Chem 282:8142–8149
91. Fromigue O, Louis K, Wu E et al (2003) Active stromelysin-3 (MMP-11) increases MCF-7 survival in three-dimensional matrigel culture via activation of p42/p44 MAP-kinase. Int J Cancer 106:355–363
92. Abraham R, Schafer J, Rothe M, Bange J, Knyazev P, Ullrich A (2005) Identification of MMP-15 as an anti-apoptotic factor in cancer cells. J Biol Chem 280:34123–34132

93. Gutiérrez-Fernández A, Folgueras AR, Garabaya C et al (2008) Matrix metalloproteinase-8 functions as a metastasis suppressor through modulation of tumor cell adhesion and invasion. Cancer Res 68:2755–2763
94. Yang W, Arii S, Gorrin-Rivas MJ, Mori A, Onodera H, Imamura M (2001) Human macrophage metalloelastase gene expression in colorectal carcinoma and its clinicopathologic significance. Cancer 91:1277–1283
95. Cornelius LA, Nehring LC, Harding E et al (1998) Matrix metalloproteinases generate angiostatin: effects on neovascularization. J Immunol 161:6845–6852
96. O'Reilly MS, Holmgren L, Shing Y et al (1994) Angiostatin: a novel angiogenesis inhibitor that mediates the suppression of metastases by a Lewis lung carcinoma. Cell 79:315–328
97. O'Reilly MS, Stetler-Stevenson WG, Folkman J, Moses MA (1999) Regulation of angiostatin production by matrix metalloproteinase-2 in a model of concomitant resistance. J Biol Chem 274:29568–29571
98. Pozzi A, Moberg PE, Miles LA, Wagner S, Soloway P, Gardner HA (2000) Elevated matrix metalloprotease and angiostatin levels in integrin-1 knockout mice cause reduced tumor vascularization. Proc Natl Acad Sci USA 97:2202–2207
99. Wen W, Moses MA, Wiederschain D, Arbiser JL, Folkman J (1999) The generation of endostatin is mediated by elastase. Cancer Res 59:6052–6056
100. Jawad MU, Garamszegi N, Garamszegi SP et al (2010) Matrix metalloproteinase 1: role in sarcoma biology. PLoS One 5:e14250
101. Khokha R (1994) Suppression of the tumorigenic and metastatic abilities of murine B16–F10 melanoma cells in vivo by the overexpression of the tissue inhibitor of the metalloproteinases-1. J Natl Cancer Inst 86:299–304
102. Watanabe M, Takahashi Y, Ohta T, Mai M, Sasaki T, Seiki M (1996) Inhibition of metastasis in human gastric cancer cells transfected with tissue inhibitor of metalloproteinase 1 gene in nude mice. Cancer 77:1676–1680
103. Ahonen M, Baker AH, Kahari V-M (1998) Adenovirus-mediated gene delivery of tissue inhibitor of metalloproteinases-3 inhibits invasion and induces apoptosis in melanoma cells. Cancer Res 58:2310–2315
104. Brand K, Baker AH, Perez-Canto A et al (2000) Treatment of colorectal liver metastases by adenoviral transfer of tissue inhibitor of metalloproteinases-2 into the liver tissue. Cancer Res 60:5723–5730
105. Sternlicht MD, Lochter A, Sympson CJ et al (1999) The stromal proteinase MMP3/stromelysin-1 promotes mammary carcinogenesis. Cell 98:137–146
106. Coussens LM, Tinkle CL, Hanahan D, Werb Z (2000) MMP-9 supplied by bone marrow derived cells contributes to skin carcinogenesis. Cell 103:481–490
107. Docherty AJP, Lyons A, Smith BJ et al (1985) Sequence of human tissue inhibitor of metalloproteinases and its identity to erythroid-potentiating activity. Nature 318:66–69
108. Stetler-Stevenson WG, Bersch N, Golde DW (1992) Tissue inhibitor of metalloproteinase-2 (TIMP-2) has erythroid-potentiating activity. FEBS Lett 296:231–234
109. Bertaux B, Hornebeck W, Eisen AZ, Dubertret L (1991) Growth stimulation of human keratinocytes by tissue inhibitor of metalloproteinases. J Investig Dermatol 97:679–685
110. Hayakawa T, Yamashita K, Tanzawa K, Uchijima E, Iwata K (1992) Growth-promoting activity of tissue inhibitor of metalloproteinases-1 (TIMP-1) for a wide range of cells. A possible new growth factor in serum. FEBS Lett 298:29–32
111. Guedez L, Stetler-Stevenson WG, Wolff L et al (1998) In vitro suppression of programmed cell death of B cells by tissue inhibitor of metalloproteinases-1. J Clin Invest 102:2002–2010
112. Li G, Fridman R, Kim H-RC (1999) Tissue inhibitor of metalloproteinase-1 inhibits apoptosis of human breast epithelial cells. Cancer Res 59:6267–6275
113. Jiang Y, Wang M, Celiker MY et al (2001) Stimulation of mammary tumorigenesis by systemic tissue inhibitor of matrix metalloproteinase 4 gene delivery. Cancer Res 61:2365–2370
114. Bigelow RL, Williams BJ, Carroll JL, Daves LK, Cardelli JA (2009) TIMP-1 overexpression promotes tumorigenesis of MDA-MB-231 breast cancer cells and alters expression of a subset of cancer promoting genes in vivo distinct from those observed in vitro. Breast Cancer Res Treat 117:31–44
115. Kopitz C, Gerg M, Bandapalli OR et al (2007) Tissue inhibitor of metalloproteinases-1 promotes liver metastasis by induction of hepatocyte growth factor signaling. Cancer Res 67:8615–8623
116. Schrotzlmair F, Kopitz C, Halbgewachs B et al (2010) Tissue inhibitor of metalloproteinases-1-induced scattered liver metastasis is mediated by host-derived urokinase-type plasminogen activator. J Cell Mol Med 14:2760–2770
117. Seo DW, Li H, Guedez L et al (2003) TIMP-2 mediated inhibition of angiogenesis: an MMP-independent mechanism. Cell 114:171–180
118. Seo DW, Kim SH, Eom SH et al (2008) TIMP-2 disrupts FGF-2-induced downstream signaling pathways. Microvasc Res 76:145–151
119. Lee SJ, Tsang PS, Diaz TM, Wei BY, Stetler-Stevenson WG (2010) TIMP-2 modulates VEGFR-2 phosphorylation and enhances phosphodiesterase activity in endothelial cells. Lab Invest 90:374–382
120. Qi JH, Ebrahem Q, Moore N et al (2003) A novel function for tissue inhibitor of metalloproteinases-3 (TIMP3): inhibition of angiogenesis by blockage of VEGF binding to VEGF receptor-2. Nat Med 9:407–415
121. Tsagaraki I, Tsilibary EC, Tzinia AK (2010) TIMP-1 interaction with alphavbeta3 integrin confers resistance to human osteosarcoma cell line MG-63 against TNF-alpha-induced apoptosis. Cell Tissue Res 342:87–96
122. Jung KK, Liu XW, Chirco R, Fridman R, Kim HR (2006) Identification of CD63 as a tissue inhibitor of metalloproteinase-1 interacting cell surface protein. EMBO J 25:3934–3942
123. Wang X, Fu X, Brown PD, Crimmin MJ, Hoffman RM (1994) Matrix metalloproteinase inhibitor BB-94 (batimastat) inhibits human colon tumor growth and spread in a patient-like orthotopic model in nude mice. Cancer Res 54:4726–4728
124. Bu W, Tang ZY, Sun FX et al (1998) Effects of matrix metalloproteinase inhibitor BB-94 on liver cancer growth and metastasis in a patient-like orthotopic model LCI-D20. Hepatogastroenterology 45:1056–1061
125. Taraboletti G, Garofalo A, Belotti D et al (1995) Inhibition of angiogenesis and murine hemangioma growth by batimastat, a synthetic inhibitor of matrix metalloproteinases. J Natl Cancer Inst 87:293–298
126. Watson SA, Morris TM, Robinson G, Crimmin MJ, Brown PD, Hardcastle JD (1995) Inhibition of organ invasion by the matrix metalloproteinase inhibitor batimastat (BB-94) in two human colon carcinoma metastasis models. Cancer Res 55:3629–3633
127. Eccles SA, Box GM, Court WJ, Bone EA, Thomas W, Brown PD (1996) Control of lymphatic and hematogenous metastasis of a rat mammary carcinoma by the matrix metalloproteinase inhibitor batimastat (BB-94). Cancer Res 56:2815–2822
128. Giavazzi R, Garofalo A, Ferri C et al (1998) Batimastat, a synthetic inhibitor of matrix metalloproteinases, potentiates the antitumor activity of cisplatin in ovarian carcinoma xenografts. Clin Cancer Res 4:985–992
129. Prontera C, Mariani B, Rossi C, Poggi A, Rotilio D (1999) Inhibition of gelatinase A (MMP-2) by batimastat and captopril

reduces tumor growth and lung metastases in mice bearing Lewis lung carcinoma. Int J Cancer 81:761–766
130. Holst-Hansen C, Low JA, Stephens RW et al (2001) Increased stromal expression of murine urokinase plasminogen activator in a human breast cancer xenograft model following treatment with the matrix metalloprotease inhibitor, batimastat. Breast Cancer Res Treat 68:225–237
131. Kruger A, Soeltl R, Sopov I et al (2001) Hydroxamate-type matrix metalloproteinase inhibitor batimastat promotes liver metastasis. Cancer Res 61:1272–1275
132. Hidalgo M, Eckhardt SG (2001) Development of matrix metalloproteinase inhibitors in cancer therapy. J Natl Cancer Inst 93:178–193
133. Maekarva K, Sato H, Furukawa M, Yoshizaki T (2002) Inhibition of cervical lymph node metastasis by marimastat (BB-2516) in an orthotopic oral squamous cell carcinoma implantation model. Clin Exp Metastasis 19:513–518
134. Kimata M, Otani Y, Kubota T et al (2002) Matrix metalloproteinase inhibitor, marimastat, decreases peritoneal spread of gastric carcinoma in nude mice. Jpn J Cancer Res 93:834–841
135. Shepherd FA, Giaccone G, Seymour L et al (2002) Prospective, randomized, double-blind, placebo-controlled trial of marimastat after response to first-line chemotherapy in patients with small-cell lung cancer: a trial of the National Cancer Institute of Canada-Clinical Trials Group and the European Organization for Research and Treatment of Cancer. J Clin Oncol 20:4434–4439
136. Steward WP, Thomas AL (2000) Marimastat: the clinical development of a matrix metalloproteinase inhibitor. Expert Opin Investig Drugs 9:2913–2922
137. Bramhall SR, Rosemurgy A, Brown PD, Bowry C, Buckels JAC (2001) Marimastat as first-line therapy for patients with unresectable pancreatic cancer: a randomized trial. J Clin Oncol 19:3447–3455
138. Bramhall SR, Schulz J, Nemunaitis J, Brown PD, Baillet M, Buckels JA (2002) A double-blind placebo-controlled, randomised study comparing gemcitabine and marimastat with gemcitabine and placebo as first line therapy in patients with advanced pancreatic cancer. British J Cancer 87:161–167
139. Scatena R (2000) Prinomastat, a hydroxamate-based matrix metalloproteinase inhibitor. A novel pharmacological approach for tissue remodelling-related diseases. Expert Opin Investig Drugs 9:2159–2165
140. Liu J, Tsao MS, Pagura M et al (2003) Early combined treatment with carboplatin and the MMP inhibitor, prinomastat, prolongs survival and reduces systemic metastasis in an aggressive orthotopic lung cancer model. Lung Cancer 42:335–344
141. Alves F, Borchers U, Padge B et al (2001) Inhibitory effect of a matrix metalloproteinase inhibitor on growth and spread of human pancreatic ductal adenocarcinoma evaluated in an orthotopic severe combined immunodeficient (SCID) mouse model. Cancer Lett 165:161–170
142. Shalinsky DR, Brekken J, Zou H et al (1999) Marked antiangiogenic and antitumor efficacy of AG3340 in chemoresistant human non-small cell lung cancer tumors: single agent and combination chemotherapy studies. Clin Cancer Res 5:1905–1917
143. Price A, Shi Q, Morris D et al (1999) Marked inhibition of tumor growth in a malignant glioma tumor model by a novel synthetic matrix metalloproteinase inhibitor AG3340. Clin Cancer Res 5:845–854
144. Frederick R, Ahmann FS, Mercier R et al (2001) Interim results of a phase III study of the matrix metalloprotease inhibitor prinomastat in patients having metastatic, hormone refractory prostate cancer (HRPC). Proc Am Soc Clin Oncol 20:abstract 692
145. Bissett D, O'Byrne KJ, von Pawel J et al (2005) Phase III study of matrix metalloproteinase inhibitor prinomastat in non-small-cell lung cancer. J Clin Oncol 23:842–849
146. Moore MJ, Hamm J, Dancey J et al (2003) Comparison of gemcitabine versus the matrix metalloproteinase inhibitor BAY 12–9566 in patients with advanced or metastatic adenocarcinoma of the pancreas: a phase III trial of the National Cancer Institute of Canada Clinical Trials Group. J Clin Oncol 21:3296–3302
147. Hoffman A, Qadri B, Frant J et al (2008) Carbamoylphosphonate matrix metalloproteinase inhibitors 6: cis-2-aminocyclohexylcarbamoylphosphonic acid, a novel orally active antimetastatic matrix metalloproteinase-2 selective inhibitor-synthesis and pharmacodynamic and pharmacokinetic analysis. J Med Chem 51:1406–1414
148. Chiappori AA, Eckhardt SG, Bukowski R et al (2007) A phase I pharmacokinetic and pharmacodynamic study of S-3304, a novel matrix metalloproteinase inhibitor, in patients with advanced and refractory solid tumors. Clin Cancer Res 13:2091–2099
149. Maquoi E, Sounni NE, Devy L et al (2004) Anti-invasive, antitumoral, and antiangiogenic efficacy of a pyrimidine-2, 4, 6-trione derivative, an orally active and selective matrix metalloproteinases inhibitor. Clin Cancer Res 10:4038–4047
150. Engel CK, Pirard P, Schimanski S et al (2005) Tumor epithelial cell matrix metalloproteinase 9 is a target for antimetastatic therapy in colorectal cancer. Chem Biol 12:181–189
151. Lauer-Fields JL, Whitehead JK, Li S et al (2008) Selective modulation of matrix metalloproteinase 9 (MMP-9) functions via exosite inhibition. J Biol Chem 283:20087–20095
152. Ikejirim M, Bernardo MM, Bonfil RD et al (2005) Potent mechanism-based inhibitors for matrix metalloproteinases. J Biol Chem 280:33992–34002
153. Tao P, Fisher JF, Shi Q et al (2009) Matrix metalloproteinase 2 (MMP2) inhibition: QM/MM studies of the inhibition mechanism of SB-3CT and its analog. Biochemistry 48:9839–9847
154. Krüger A, Arlt MJE, Gerg M et al (2005) Antimetastatic activity of a novel mechanism-based gelatinase inhibitor. Cancer Res 65:3523–3526
155. Bonfil RD, Sabbota A, Nabha S et al (2006) Inhibition of human prostate cancer growth, osteolysis and angiogenesis in a bone metastasis model by a novel mechanism-based selective gelatinase inhibitor. Int J Cancer 118:2721–2726
156. Devy L, Huang L, Naa L et al (2009) Selective inhibition of matrix metalloproteinase-14 blocks tumor growth, invasion, and angiogenesis. Cancer Res 69:1517–1526
157. Lukkonen A, Sorsa T, Salo T et al (2000) Down-regulation of trypsinogen-2 expression by chemically modified tetracyclines: association with reduced cancer cell migration. Int J Cancer 86:577–581
158. Nair RR, Avila H, Ma X et al (2008) A novel high-throughput screening system identifies a small molecule repressive for matrix metalloproteinase-9 expression. Mol Pharmacol 73:919–929
159. Toda D, Ota T, Tsukuda K et al (2006) Gefitinib decreases the synthesis of matrix metalloproteinase and the adhesion to extracellular matrix proteins of colon cancer cells. Anticancer Res 26:129–134
160. Wei LH, Lai KP, Chen CA et al (2005) Arsenic trioxide prevents radiation-enhanced tumor invasiveness and inhibits matrix metalloproteinase-9 through downregulation of nuclear factor kappaB. Oncogene 24:390–398
161. Ito H, Duxbury M, Benoit E et al (2004) Prostaglandin E2 enhances pancreatic cancer invasiveness through an Ets-1-dependent induction of matrix metalloproteinase-2. Cancer Res 64:7439–7446

162. Blazquez C, Salazar M, Carracedo A et al (2008) Cannabinoids inhibit glioma cell invasion by down-regulating matrix metalloproteinase-2 expression. Cancer Res 68:1945–1952
163. Kim SY, Jung SH, Kim HS (2005) Curcumin is a potent broad spectrum inhibitor of matrix metalloproteinase gene expression in human astroglioma cells. Biochem Biophys Res Commun 337:510–516
164. Gabriely G, Wurdinger T, Kesari S et al (2008) MicroRNA 21 promotes glioma invasion by targeting matrix metalloproteinase regulators. Mol Cell Biol 28:5369–5380
165. Chan JA, Krichevsky AM, Kosik KS (2005) MicroRNA-21 is an antiapoptotic factor in human glioblastoma cells. Cancer Res 65:6029–6033
166. Volinia S, Calin GA, Liu CG et al (2006) A microRNA expression signature of human solid tumors defines cancer gene targets. Proc Natl Acad Sci USA 103:2257–2261
167. Bao BY, Yeh SD, Lee YF (2006) 1alpha, 25-dihydroxyvitamin D3 inhibits prostate cancer cell invasion via modulation of selective proteases. Carcinogenesis 27:32–42
168. Sloane BF, Sameni M, Podgorski I et al (2006) Functional imaging of tumor proteolysis. Ann Rev Pharmacol Toxicol 46:301–315
169. Martin MD, Carter KJ, Jean-Philippe SR et al (2008) Effect of ablation or inhibition of stromal matrix metalloproteinase-9 on lung metastasis in a breast cancer model is dependent on genetic background. Cancer Res 68:6251–6259