Review

ROLE OF MATRIX METALLPROTEINASES IN CANCER DEVELOPMENT

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ABSTRACT

In the last twenty years there has been a big interest towards the proteinases and specifically the matrix metalloproteinases (MMPs). MMPs and their impact on the tumor development, including tumor progression, invasion, metastasis, growth and neoangiogenesis has been the focus of many studies for years. MMPs are a large family of zinc-dependent endopeptidases, which are known as the major enzymes responsible for the proteolytic cleavage of proteinaceous components of the extracellular matrix (ECM). Although the main function of MMPs is the degradation of the ECM during tissue resorption and progression of various diseases including cancer, there are also many other functions of the MMP family that control the cancer progression and metastasis. The scope of this review is a discussion regarding the current view and status of MMPs research (specifically MMP-2, MMP-9 and MMP-13) in cancer.

Our review concluded that the MMPs have the ability to process molecules such as growth factors, receptors, adhesion molecules, other proteinases and proteinase inhibitors. MMPs are potent controllers of physiological and pathological events in the cell microenvironment. The overactivation or underactivation of MMPs could be used as diagnostic, prognostic and predictive factors in the cancer treatment.

Key words: matrix metalloproteinases, cancer, cancer development, cancerogenesis

INTRODUCTION

In the past years the matrix metalloproteinases (MMPs) have been extensively investigated in tumor invasion and metastasis. The MMPs play an important role in the development and progression of human malignancies. These enzymes mediate not only the normal physiological processes, such as tissue remodeling and development, but they can also regulate the tumor micro-environment and participate in the different stages of tumor progression, including processes like invasion and neoangiogenesis. Individual MMPs have been identified that show increased expression in the different tumors and tumor stages. It is therefore expected that MMPs could serve as both diagnostic and prognostic biomarkers in cancer patients. Despite a huge number of studies, it is still difficult to establish MMPs as cancer biomarkers. The purpose of this review is to outline the role of MMPs in the progression of cancer.

Characteristics of the MMP family

The matrix metalloproteinases (MMPs) are a relatively large family of structurally and functionally related endopeptidases which are able to degrade all the structural components of the extracellular matrix (ECM). The MMP-

family is characterized by the presence of several conserved protein domains: a prodomain, an active domain and a Zn^{2+} -binding domain. They are first described almost half a century ago and since then more than 24 MMPs have been identified (1). Historically, the MMPs were separated into collagenases - MMP-1, MMP-8, MMP-13, MMP-18; gelatinases - gelatinase A (MMP-2) and gelatinase B (MMP-9); stromelysin 1 - MMP-3 and stromelysin 2 - MMP-10 and -11; matrilysins - matrilysin1 (MMP-7) and matrilysin 2 (MMP-26); membrane-type MMPs (MT-MMPs) - there are six MT-MMP - MMP-14, MMP-15, MMP-16, MMP-17, MMP-24 and MMP-25. Closely related to the MMPs are the so-called ADAM (a disintegrin and metalloproteinase) and ADAMTS (a disintegrin and metalloproteinase with thrombospondin motifs).

Regulation of MMPs

The MMPs enzymes are generally expressed in very low amounts and their transcription is regulated either positively or negatively by cytokines and growth factors such as inflammatory interleukins (IL-6, TNF) or transforming growth factors (TGF). The MMP expression is induced by cytokines, chemical

agents, tumor growth promoters, physical stress, oncogenic transformations etc. (17-21). MMPs are secreted as inactive pro-enzymes and activated by proteinase cleavage. They are kept inactive by an interaction between a cystein-sulphydryl group in the propeptide domain and the zinc ion bound to the catalytic domain. There are several proteinases that activate MMPs zymogens, such as plasmin, furin (2), or active MMPs (3). The activity of MMPs is regulated by five mechanisms: gene transcription, secretion, proteolytic activation of the zymogen form, endogenous inhibition of the active form, and glycosylation (4). The most important natural inhibitors are the tissue inhibitors of metalloproteinases (TIMPs). MMPs are produced by a variety of cell types, including epithelial cells, mesenchymal cells (mainly fibroblasts) and inflammatory cells. The main physiological function firmly established of MMPs was the modulation and regulation of ECM by direct proteolytic degradation of all components of the ECM (5).

MMPs are controlled by endogenous tissue specific inhibitors called tissue inhibitors of matrix metalloproteinases (TIMPS), which are secreted proteins.

TIMPS provide a negative control of the MMP activity. Four various TIMPs have been characterized so far: TIMP 1, TIMP 2, TIMP 3 and TIMP 4. The TIMPs inhibit active MMPs by forming 1:1 molar stoichiometric non-covalent complexes with the endopeptidases. The impact of TIMPs is essential for the homeostasis of the ECM in different physiological processes. The imbalance between TIMPs and MMPs is an essential step in the development of gastro-intestinal malignancies and is of critical importance in early events of tumor progression. TIMPs display a complex influence on tumor progression and metastasis: on one hand, they directly regulate and inhibit MMPs, on the other hand influence angiogenesis, inhibit the apoptosis of tumor cells, malignant transformation and facilitate tumor growth and metastasis (5, 6).

Matrix metalloproteinases, ECM and cancer

The development of malignant neoplasms is a long-term and multi-step process – a complex series of sequential processes that results in rapid growth and invasion of tumor cells into lymphatic and blood vessels. First is the initial transforming event, then the proliferation of transformed cells; the ability of tumor cells to avoid destruction by immune-respose; nutrition supply to the tumor mass; local invasion and

cleavage of the extracellular matrix; migration of tumor cells; penetration of tumor cells through the blood vessel wall; embolization of tumor cells and adhesion to distant organs; arrest of cancer cells in the lumen of small blood vessels and lymphatics; reverse penetration of blood vessels and formation of distant metastasis (7, 8, 9).

The migration of tumor cells is associated with the degradation of the extracellular matrix. The ECM is a dynamic structure composed of various macromolecules such as collagen, fibronectin, laminin and proteoglycans of connective tissue. Their role is the regulation of cells functions and establishment of a specific microenvironment. Transformation of ECM is an important factor for various physiological and pathological processes, tissue development and formation, proliferation and differentiation of cells, as well as invasion and metastasis of neoplastic cells. Three steps describe the sequence of events during tumor cell invasion of ECM: attachment, matrix dissolution and cell migration. The attachment to the basement membrane (BM) is mediated by tumor cell surface receptor together with specific glycoproteins such as fibronectin and laminin. During the second step (local degradation of matrix by tumor cell associated proteinases) tumor cells directly secrete enzymes to degrade ECM. Such proteinases can degrade both structural collagenous proteins of the matrix and the attachment proteins. During the third step (migration), cancer cells are propelled across the BM and stroma through the zone of matrix proteolysis. Chemotactic factors can influence the direction of migration. (10, 11)

The Role of MMPs in Cancer development

The MMPs are major players in the breakdown and reconstruction of ECM in variety of pathological processes: cancer invasion and metastases, rheumatoid arthritis and osteoarthritis, formation of decubitus and various ulcers, neuroinflammatory and cardiovascular diseases, liver cirrhosis, fibrotic lung diseases and others (10-16, 34, 35). The notion that MMPs mediate ECM degradation and this leads to cancer cell invasion and metastases has been a guiding principle in MMP research for many years. The expression and bioactivity of MMPs are increased in almost every type of cancer. Most studies find a negative correlation between MMPs expression and prognosis. Several clinical studies find that high levels of TIMP-1 and -2 also correlate with a poor prognosis. It is now evident that MMPs function is more complex in the cancer development than initially

thought. MMPs mediate a wide range of biological processes, which are likely to have significant consequences on the tumor microenvironment.

Several alterations in cell biology underlie cancer progression: self-support in growth signals; insensitivity to growth-inhibitory signals; escape from apoptosis; infinite replication; sustained angiogenesis and tissue invasion and metastases. MMPs are involved in various steps of cancer development.

MMPs affect Growth signals

Active tumour growth factor- β is derived from an inactive pro-form by proteolytic conversion by furin or other proteinases, such as MMP-9. Similarly, MMP-14 and 2 proteolytically activate TGF- β 1. MMP-2, 9 and 14 also indirectly modulate TGF- β bioactivity by cleaving the ECM component latent TGF- β - binding protein 1 (LTBP-1), thereby solubilizing ECM-bound TGF- β . On other hand, growth factors that are sequestered by ECM proteins become bioavailable once these proteins are degraded by MMPs (36).

MMPs regulate neoangiogenesis

MMPs regulate the cancer development and metastasis through the regulation and activation of the neoangiogenesis. The angiogenic response is a complex process of vessels sprouting into tumour tissue from existing neighbouring vessels which is essential for tumour growth. The proteolysis of the ECM plays an important role in the neoangiogenesis. Angiogenic factors are also released in the ECM: vascular endothelial growth factor (VEGF), fibroblasts growth factor (FGF), transforming growth factor (TGF), tumor necrosis factor – α (TNF- α), epidermal growth factors (EGF), interleukin-8 (IL-8) and others (26). These factors stimulate the MMP secretion and activation which then cleave the ECM proteins. Proof of the MMP role is that both the synthetic and the endogenous MMP-inhibitors suppress the angiogenic response (22-25). The MMPs self can generate growth-promoting signals. This is proven in tumours from MMP-9-deficient mice compared with wild-type mice, indication that MMPs generate growth-promoting signals (27).

MMPs regulate Apoptosis

MMP can regulate the apoptosis, which is a key element for the cancer development. The MMPs have both apoptotic and anti-apoptotic actions. MMP-3 induces apoptosis when overexpressed in mammary epithelial cells (28, 29). MMP-7 releases membrane-bound FASL,

a transmembrane stimulator of the death receptor FAS (30). Released FASL induces apoptosis of neighboring cells, or decreases cancer apoptosis, depending on the system. MMP-7 inhibits apoptosis by cleaving pro-heparin-binding epidermal growth factor (pro-HB-EGF) to generate mature HB-EGF, which promotes cell survival. MMP-11 also inhibits cancer-cell apoptosis: overexpression of MMP-11 decreases spontaneous apoptosis in tumour xenografts (31).

MMPs regulate tumor cells invasion and metastases

The proteolytic activity of MMPs is an essential step of tumor invasion. The process includes disruption of the basement membrane with subsequent invasion of malignant cells into the peripheral tissue, neoangiogenesis leading to further tumor growth, intravasation into the blood or lymphatic vessels, transport within the circulation from where they disseminate into secondary organs. MMPs participate not only in the breakdown of ECM, but also in these processes. The first mechanism by which MMPs promote tumor cells proliferate and growth is degradation of proteins in the ECM and basement membrane in such a way that tumor cells may physically penetrate into the surrounding peripheral tissue and blood vessels in order to infiltrate and metastasize. The second mechanism by which MMPs participate in these processes is degradation of matrix components and non-matrix components such as chemokines, growth factors (VEGF, FGF, EGF, TGF), growth factor receptors, adhesion molecules and apoptotic mediators that support tumor progression and invasion.

Matrix metalloproteinases- 2 (MMP-2) and -9 (MMP-9) are overexpressed in a variety of cancer tissues. They participate in the degradation of type IV collagen in ECM. It was revealed that tumor cells are able to produce and release MMP-2 and MMP-9 capable of degrading the basement membrane and IV collagen in extracellular matrix. Increased expression of these enzymes was associated with enhanced invasiveness of the cancer.

The retrospective analysis shows that MMP expression and activity in cancer patients are increased in almost every type of human cancer. Elevated expression of MMP-1, -2, -7, -9, 13 and others are associated with tumor progression – poor prognosis and differentiation, invasive stage of cancer and metastases. Matrix metalloproteinases (MMPs) have been regarded as major critical

molecules assisting tumor cells metastasis (32, 33). Numerous studies linked inhibition of MMPs by synthetic and natural inhibitors with a corresponding inhibition of cell invasion. Groblewska M, et al find that the preoperative serum levels of MMP-9 in esophageal cancer were higher than in healthy subjects, and correlated with clinical stage of disease as well as with tumor size (7). The diagnostic measurement of MMP-9 was higher (70%) than for classical tumor markers (CEA -17% and SCC-Ag-64%).

CONCLUSION

Our review concluded that the MMPs have the ability to process molecules such as growth factors, receptors, adhesion molecules, other proteinases and proteinase inhibitors. MMPs are potent controllers of physiological and pathological events in the cell microenvironment. The overactivation or underactivation of MMPs could be used as diagnostic, prognostic and predictive factors in the cancer treatment.

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