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Original contribution

MMP14 predicts a poor prognosis in patients with colorectal cancer☆

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Summary Matrix metalloproteinases (MMPs) are involved in most biological processes. Recently, MMP14 was reported to be up-regulated in some types of cancer and to promote cancer cell invasion and metastasis. However, there are few reports on the clinical significance of MMP14 in colorectal cancer (CRC). In this study, MMP14 expression was first investigated in The Cancer Genome Atlas (TCGA) and whole-genome expression microarray (GEO; Accession Number GSE39582) and then validated with our database. Univariate and multivariate analyses were performed to assess the association between prognostic factors and survival outcomes. MMP14 was upregulated at both the transcriptional and protein levels in cancer compared with normal tissues ($P < .05$), and high MMP14 expression was associated with advanced tumor stage in the 3 study cohorts. In the univariate Cox proportional hazard ratio analysis, MMP14 correlated significantly with prognosis in both the TCGA and GSE39582 databases ($P < .05$). In the validation cohort, patients with high MMP14 expression had lower 5-year disease-free survival (DFS; hazard ratio [HR] 6.707; 95% confidence interval [CI] 3.184, 14.128; $P < .001$) and overall survival (OS; HR 10.669; 95% CI 3.828, 29.737; $P < .001$) than those with low MMP14 expression. Multivariate survival analysis showed that MMP14 was an independent prognostic marker for both DFS (HR 5.776; 95% CI 2.719, 12.270; $P < .001$) and OS (HR 8.971; 95% CI 3.199, 25.156; $P < .001$). Clearly, MMP14 plays an important role in CRC progression and prognosis and could be a useful biomarker for prediction of survival after colectomy.

1. Introduction

Colorectal cancer (CRC) is the third most commonly diagnosed cancer with a death rate ranking second in men and third in women worldwide [1]. The incidence rates, which are rapidly increasing in many countries including China, are associated with risk factors such as unhealthy diet, smoking, and obesity [2]. Nearly 43% of CRC patients develop liver metastases, and 25% have both liver and lung metastases with a 5-year survival rate in stage IV <10% [3,4]. Understanding the biological mechanisms of metastasis and progression of CRC and developing effective measures to target this process thus are of great importance. Much attention has been paid to molecularly based prognostic markers, which are complementary to the data obtained by pathological diagnosis and may increase the lifespans of patients [5-7].

As is well known, almost all solid tumors are composed not only of tumor cells, but also of a variety of non-tumor cells and

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extracellular matrix (ECM). Prior to metastasis, cancer cells must become motile and detached from the primary tumor and overcome the barrier of extracellular matrix [8]. Matrix metalloproteinases (MMPS), in a complex interplay with their inhibitors, tissue inhibitors of metalloproteinases (TIMPs), serve as key enzymes involved in the degradation of ECM, playing a major role in both normal physiological functions and cancer-related processes such as cell migration, inflammation, invasion, metastasis, angiogenesis, and proliferation [9,10]. As an important member of the MMPs family, MMP14 is the first reported membrane-type matrix metalloproteinase, also referred as membrane type 1-matrix metalloproteinase (MT1-MMP). The enzyme has been suggested to be involved in many biological processes, including proliferation, invasion, angiogenesis, and basement membrane remodeling [11,12]. Generally, MMPs are produced as inactive zymogens that require activation. However, MMP14 does not require activation because of its capacity to be present in its active form on the cell membrane [12,13].

Recent studies demonstrated that the expression of MMP14 is a poor prognostic factor of worse survival outcomes in patients with gastric cancer [12]. But its role in CRC has not been studied. Hence, we performed the present study to investigate the role of MMP14 in CRC. In order to draw a solid conclusion, we first studied MMP14 expression in the publically available The Cancer Genome Atlas (TCGA) and whole-genome expression microarray (GEO, Accession Number GSE39582) databases and then validated it with our own database. All procedures involving human participants were in accordance with the ethical standards of the institutional research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

2. Materials and methods

2.1. Patients in TCGA and GSE39582 database

We initially used The Cancer Genome Atlas (TCGA) database (https://genome-cancer.ucsc.edu/) to forecast MMP14 mRNA expression and the association between its expression and the overall survival (OS) of patients with CRC. The MMP14 expression was simultaneously analyzed using another public database, GSE39582 (https://www.ncbi.nlm.nih.gov/geo/), to investigate the correlation between MMP14 mRNA expression and relapse-free survival (RFS).

The inclusion criteria for the two databases were the following: (1) pathological diagnosis of adenocarcinoma; (2) no neoadjuvant therapy, including chemotherapy and radiotherapy; (3) complete survival information. Patients who died with tumor or recurrence at last follow-up were defined as the clinical endpoint for tumor-specific survival or RFS. Follow-up was completed on April 27, 2016, on the TCGA database and on February 24, 2017, on the GSE39582 database.

2.2. Patients in validation cohort

A total of 218 Chinese patients with CRC diagnosed between January 2005 and December 2010 were used for this study. The group included 126 men and 92 women with a mean age of 58 years (range, 27-85 years). The inclusion criteria were as follows: (1) having a distinctive pathologic diagnosis of adenocarcinoma; (2) surgical resection, defined as complete resection of the primary tumor and regional lymph node dissection with the margins being free of cancer by histologic examination; and (3) having complete clinicopathologic and follow-up data. The exclusion criteria were (1) having distant metastases and (2) receiving anticancer treatment before surgical resection. The tissues were paired tumor and non-tumor from each patient. All patient-derived specimens were collected and archived under protocols approved by the Institutional Review Boards of The First People's Hospital of Shangqiu. All patients were restaged according to the 8th American Joint Committee on Cancer (AJCC) guidelines [14]. The detailed clinicopathologic characteristics of the patients are listed in Table 1.

2.3. Immunohistochemistry study

Immunohistochemistry (IHC) was performed on formalin-fixed, paraffin-embedded tissue sections using a two-step protocol (GTVision III). The MMP14 was detected using the rabbit anti-MMP14 polyclonal antibody ab51074 (Abcam Inc., Cambridge, MA). Negative control samples were treated identically but with the primary antibodies omitted. Briefly, pressure cooker–mediated antigen retrieval was performed in EDTA buffer pH 9.0 for 10 minutes. Sections were incubated with a 1:100 dilution of anti-MMP14 antibody overnight at 4 °C and then incubated with goat anti-rabbit Envision System Plus-HRP (DakoCytomation, Glostrup, Denmark) for 30 minutes at room temperature. After rinsing three times in phosphate-buffered saline (PBS) for 10 minutes each, the sections were incubated with diaminobenzidine (DAB) for 1 minutes, counterstained with Mayer hematoxylin, dehydrated, and mounted.

Data were assessed by two independent single-blinded pathologists. A semiquantitative scoring system [7] was used to evaluate both staining intensity (0, no staining; 1+, weak staining; 2+, moderate staining; 3+, strong staining) and the percentage of stained cells (0, <5%; 1, 5%-25%; 2, 26%-50%; 3, 51%-75%; and 4, >75%). The scores for staining intensity and the percentage of positive cells were multiplied to generate the immunoreactivity score for each case [15]. All cases were sorted into two groups according to the immunoreactivity score. High expression of MMP14 was defined as detectable immunoreactions in the cytoplasmic or membrane with an immunoreactivity score of ≥ 4.

2.4. Statistical analysis

The Pearson χ^2 test was used to analyze the relation between MMP14 expression and clinicopathologic variables

Table 1 Association between MMP14 expression and clinicopathologic features in the validation cohort

Variable	No.	MMP14 expression		χ^2	*P*
		Low	High		
Sex				0.031	.859
Male	126	56 (57.1)	70 (58.3)		
Female	92	42 (42.9)	50 (41.7)		
Age				0.004	.947
≥60	134	60 (61.2)	74 (61.7)		
>60	84	38 (38.8)	46 (38.3)		
T category				6.100	.101
1	6	5 (5.1)	1 (0.8)		
2	35	19 (19.4)	16 (13.3)		
3	92	41 (41.8)	51 (42.5)		
4	85	33 (33.7)	52 (43.3)		
N stage				6.304	.043 [a]
0	103	54 (55.1)	49 (40.8)		
1	69	30 (30.6)	39 (32.5)		
2	46	14 (14.3)	32 (26.7)		
Differentiation				3.081	.214
High	61	22 (22.4)	39 (32.5)		
Moderate	110	55 (56.1)	55 (45.8)		
Poor	47	21 (21.4)	26 (21.7)		
Lymphovascular invasion				5.914	.015 [a]
Negative	173	85 (86.7)	88 (73.3)		
Positive	45	13 (13.3)	32 (26.7)		
Perineural invasion				0.345	.557
Negative	188	86 (87.8)	102 (85.0)		
Positive	30	12 (12.2)	18 (15.0)		
CEA concentration				9.140	.010 [a,b]
Normal	153	75 (76.5)	78 (65.0)		
High	65	23 (23.5)	42 (35.0)		
Microsatellite status				0.695	.404
MSU	78	38 (38.3)	40 (33.3)		
MSS	140	60 (61.2)	80 (66.7)		

Abbreviations: MSS, microsatellite stable; MSU, microsatellite unstable.x

[a] Statistically significant.

[b] Exact χ^2 test.

(the Fisher exact test was used when needed). The RFS was calculated from the date of surgery to the date of disease relapse (local or distant). The OS was calculated from the date of diagnosis to the date of death or last follow-up. Patients without events or death were censored at the last follow-up. Follow-up data were recorded by telephone or medical records.

Survival curves were constructed using the Kaplan-Meier method, and the differences between the survival curves were examined by the log-rank test. Univariate Cox proportional hazards regressions were applied to estimate the individual hazard ratio (HR) for the disease-free survival (DFS) and OS. The significant variables in the univariate analyses ($P < .05$) were then put into the multivariate analysis. The HR with 95% confidence interval (CI) was measured to estimate the hazard risk of individual factors. All reported *P* values were two sided and were considered significant at .05. The statistical analysis was carried out using SPSS version 17.0 (SPSS Inc, Chicago, IL).

3. Results

3.1. Expression patterns of MMP14 mRNA in CRC predicted by bioinformatics

Initially, the TCGA and GSE39582 databases were used to predict the MMP14 mRNA expression patterns in CRC. First, in 32 paired cancers and their normal tissues, the expression of MMP14 mRNA was dramatically higher in cancer than in the normal tissues ($P < .001$; Fig. 1A). Second, MMP14 mRNA expression was significantly different from stage I to stage III in the TCGA database. The most significant difference was between stage I and stage II tumor ($P = .045$; Fig. 1B). In addition, similar results were found in the GSE39582 database, as shown in Fig. 1C ($P < .001$). The results implied that MMP14 affects the initiation and progression of CRC.

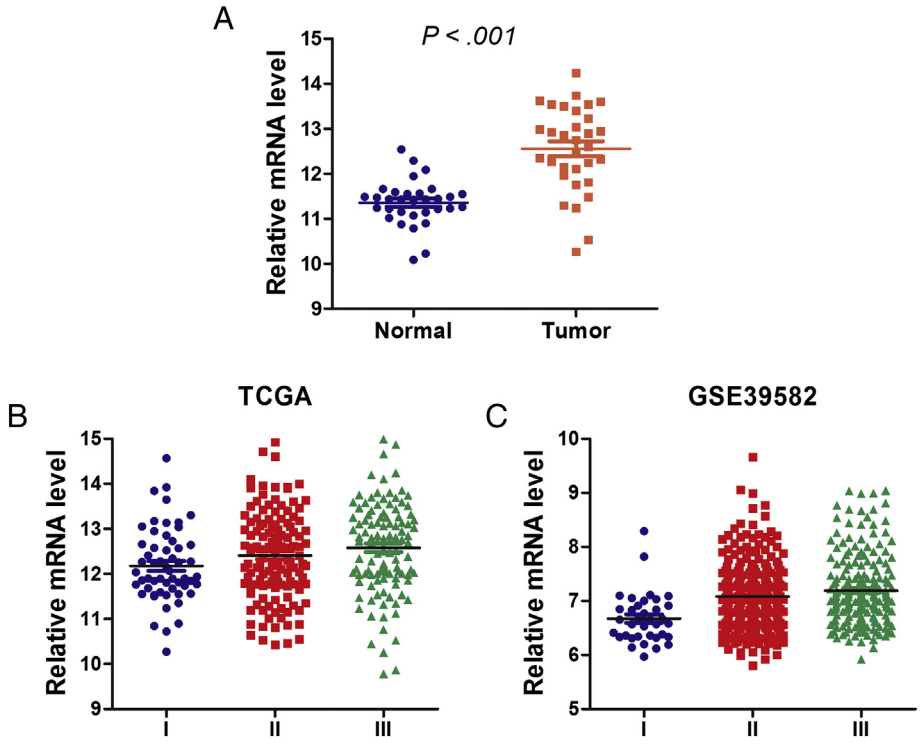

Fig. 1 Expression patterns of MMP14 mRNA in CRC predicted by bioinformatics. A, The mRNA was dramatically more abundant in cancer than in normal colonic tissues in 32 paired colorectal cancers and their normal controls in the TCGA database ($P < .001$). B, Expression was significantly different from stage I to stage III CRC in TCGA database. The most significant difference was between stage I and stage II tissues ($P = .045$). C, Expression increased gradually from stage I to stage III CRC in GSE39582 database ($P < .001$).

3.2. Overexpression of MMP14 was closely associated with poor prognosis of patients in both the TCGA and GSE39582 databases

The expression of MMP14 mRNA was first treated as a continuous variable. Univariate Cox analysis demonstrated that MMP14 was a predictor of OS in the TCGA database (HR 1.238; 95% CI 1.001, 1.533; $P = .049$) and RFS in the GSE39582 database (HR 1.765; 95% CI 1.384, 2.251; $P < .001$). The MMP14 mRNA expression was almost normally distributed in both the TCGA and GSE39582 databases (data not shown). Therefore, we divided the cohorts into low- or high-expression subgroups according to the median expression of MMP14 mRNA. In the TCGA database, the 5-year OS rates were significantly higher among patients with low MMP14 expression than in those with high expression (71.6% versus 51.3%; $P = .003$; Fig. 2A). Similarly, the 5-year RFS rates in the low-expression group were significantly higher than those in the high-expression group (78.4% versus 62.3%; $P < .001$; Fig. 2B).

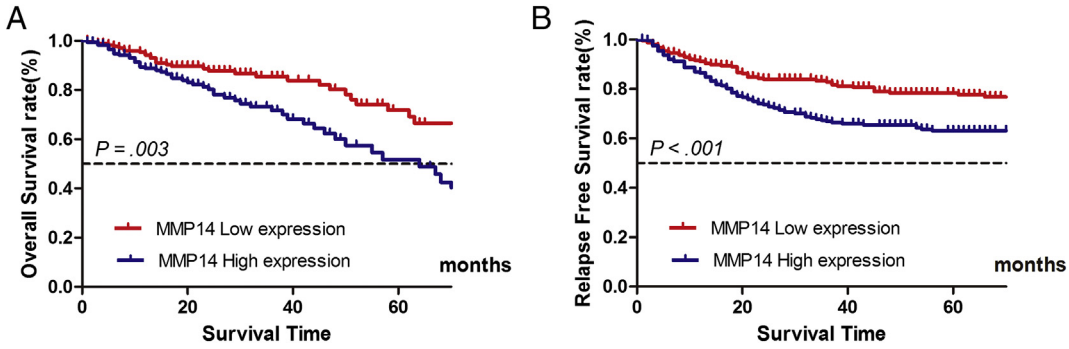

Fig. 2 Overexpression of MMP14 was closely associated with poor prognosis in both the TCGA and GSE39582 databases. A, In TCGA database, 5-year OS rate was significantly higher in patients with low expression than in those with high expression (71.6% vs. 51.3%; $P = .003$). B, The 5-year RFS rate in the low-expression group was significantly higher than that in the high-expression group (78.4% vs. 62.3%; $P < .001$).

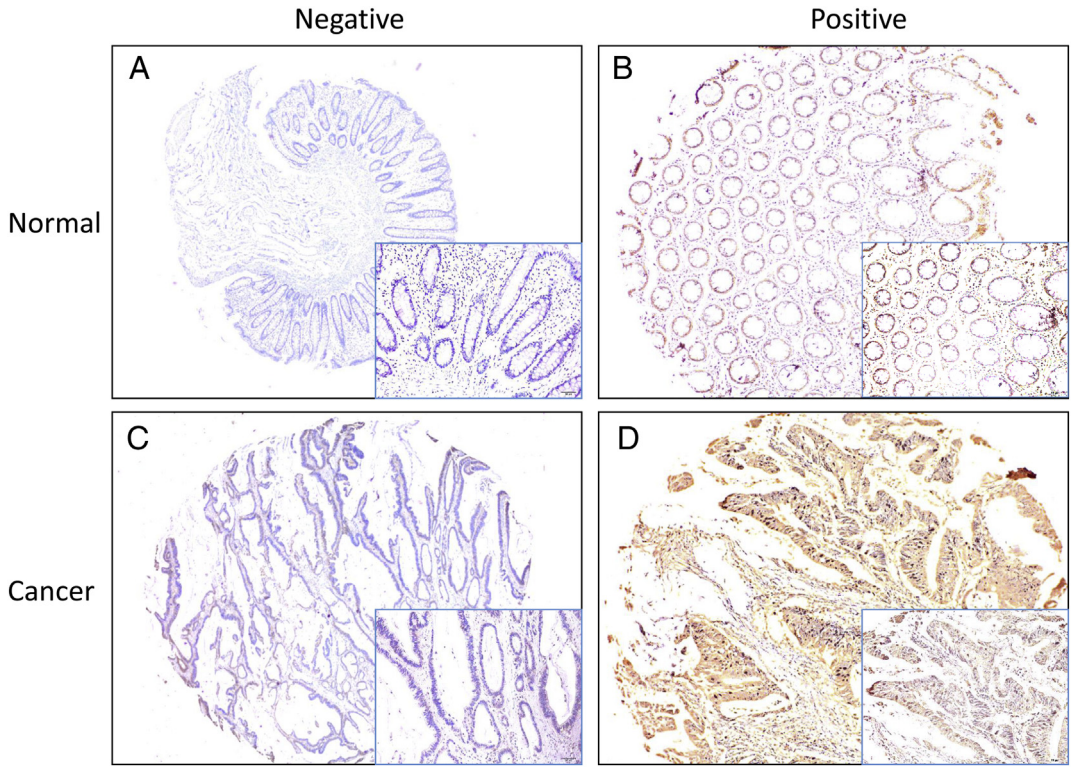

Fig. 3 Representative images of MMP14 expression in CRC tissues and their normal controls. A and B, MMP14 was low (A) or high (B) in normal colonic tissues. C and D, MMP14 was low (C) or high (D) in CRC tissues. Original magnifications ×200 and ×400 (lower panels).

3.3. MMP14 expression correlated with poor clinicopathologic features

To further determine the significance of MMP14 expression, immunohistochemistry staining of a cohort containing 218 cases of primary CRC paired with noncancerous tissue was performed. There were 126 male and 92 female with a mean age of 57 years (range 27–85 years) in the cohort. Of the 218 specimens, 189 cases (86.70%) showed no MMP14 staining in the normal mucosa. In contrast, high MMP14 expression was observed in 55.0% (120/218) of the CRC tissues. Representative images are presented in Fig. 3. Correlation analysis showed that MMP14 protein expression was significantly associated with advanced N stage ($P = .043$), lymphovascular invasion ($P = .015$), and a high concentration of carcinoembryonic antigen (CEA; $P = .010$). No correlation was found between MMP14 expression and other clinicopathologic features (Table 1).

3.4. Overexpression of MMP14 predicts poor prognosis in validation cohort

Of the 218 cases, tumor recurred in 60 patients (27.50%), and 47 patients (21.6%) died of the disease during the follow-up. Patients with high expression of MMP14 had a significantly shorter DFS and OS than those with low expression

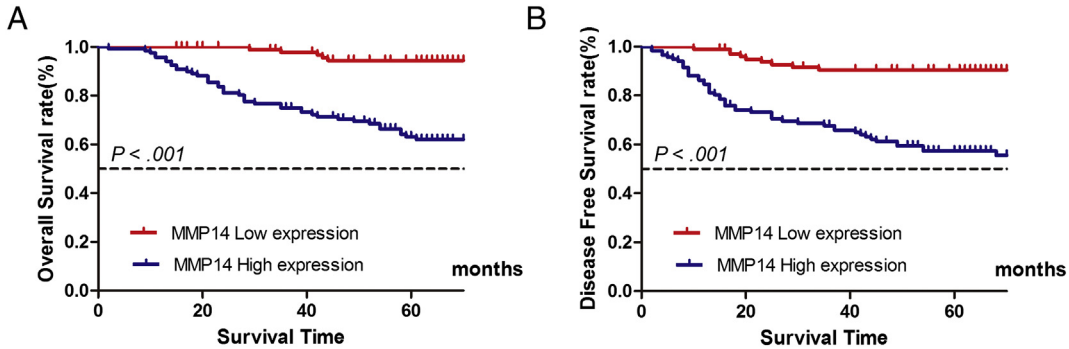

Fig. 4 Overexpression of MMP14 predicted poorer prognosis in the validation cohort. Of the 218 cases, patients with high expressions of MMP14 had significantly shorter OS (A) and DFS (B) compared with those with low expression (both log-rank $P < .001$).

Table 2 Univariate and multivariate Cox proportional hazards analysis of MMP14 expression and OS for patients with CRC in the validation cohort

Factor	Univariate analysis		Multivariate analysis	
	HR (95% CI)	*P*	HR (95% CI)	*P*
Sex	0.938 (0.524, 1.680)	.830		
Age	1.631 (0.920, 2.890)	.094		
Grade	1.279 (0.848, 1.929)	.241		
T stage	2.029 (1.312, 3.138)	.001 [a]	1.470 (0.946, 2.282)	.086
N stage	2.577 (1.784, 3.724)	<.001 [a]	2.048 (1.359, 3.088)	.001 [a]
Lymphovascular invasion	3.154 (1.757, 5.664)	<.001 [a]	1.437 (0.766, 2.696)	.259
Perineural invasion	1.298 (0.606, 2.777)	.502		
CEA concentration	1.036 (0.578, 1.857)	.906		
MMP14	10.669 (3.828, 29.737)	<.001 [a]	8.971 (3.199, 25.156)	<.001 [a]

Abbreviations: CI, confidence interval; HR, hazard ratio.
[a] Statistically significant.

(both log-rank $P < .001$; Fig. 4A and B). In univariate analysis, T stage (HR 2.029; 95% CI 1.312, 3.138; $P = .001$), N stage (HR 2.577; 95% CI 1.784, 3.724; $P < .001$), lymphovascular invasion (HR 3.154; 95% CI 1.757, 5.664; $P < .001$), and MMP14 expression (HR 10.669; 95% CI 3.828, 29.737; $P < .001$) were associated with OS, whereas T stage (HR 1.777; 95% CI 1.233, 2.562; $P = .002$), N stage (HR 2.345; 95% CI 1.703, 3.230; $P < .001$), lymphovascular invasion (HR 2.931; 95% CI 1.737, 4.947; $P < .001$), perineural invasion (HR 2.109; 95% CI 1.159, 3.839; $P = .015$), and MMP14 expression (HR 6.707; 95% CI 3.184, 14.128; $P < .001$) were associated with DFS. In multivariate analysis, only N stage (for OS, HR 2.048; 95% CI 1.359, 3.088; $P = .001$; for DFS, HR 1.913; 95% CI 1.325, 2.76; $P = .001$), and MMP14 expression (for OS, HR 8.971; 95% CI 3.199, 25.156; $P < .001$; for DFS, HR 5.776; 95% CI 2.719, 12.270; $P < .001$) were independent prognostic factors for CRC (Tables 2 and 3).

4. Discussion

Accumulating evidence shows that members of the MMP family of proteins are involved in carcinogenesis and progression of various cancers. Moreover, ectopic MMP expression is correlated with metastasis, invasion, and survival. However, some members of the MMP family remain uncharacterized, which requires further investigation of these genes.

MMP14 was one of the first reported membrane-type matrix metalloproteinases [12], and it has been identified as a major physiologic activator of pro-MMP2 [16,17]. Potentially, MMP14 plays a role in various biological processes in both normal and cancerous tissues [18]. Previous studies of MMP14 focused mainly on angiogenesis and invasion of cancers [17,19,20]. Recent studies demonstrated that greater expression of MMP14 correlated with a poor prognosis in Chinese patients with gastric cancer [12]. Down-regulation

Table 3 Univariate and multivariate Cox proportional hazards analysis of MMP14 expression and DFS for patients with CRC in the validation cohort

Factor	Univariate analysis		Multivariate analysis	
	HR (95% CI)	*P*	HR (95% CI)	*P*
Sex	0. 914 (0.545, 1.532)	.733		
Age	1.381 (0.830, 2.297)	.214		
Grade	1.310 (0.909, 1.887)	.147		
T stage	1.777 (1.233, 2.562)	.002 [a]	1.338 (0.923, 1.939)	.125
N stage	2.345 (1.703, 3.230)	<.001 [a]	1.913 (1.325, 2.761)	.001 [a]
Lymphovascular invasion	2.931 (1.737, 4.947)	<.001 [a]	1.401 (0.798, 2.461)	.240
Perineural invasion	2.109 (1.159, 3.839)	.015 [a]	1.244 (0.666, 2.321)	.494
CEA concentration	1.455 (0.879, 2.407)	.145		
MMP14	6.707 (3.184, 14.128)	<.001 [a]	5.776 (2.719, 12.270)	<.001 [a]

Abbreviations: CI, confidence interval; HR, hazard ratio.
[a] Statistically significant.

of MMP14 activity in breast carcinomas reduces primary tumor growth and enhances the response to radiation therapy, especially in tumors with high MMP14 expression [21]. The protein also is the downstream target of some tumor suppressor miRNAs [22-24]. But its role in CRC has not been fully studied. Hence, we conducted the present study to investigate the clinical significance of MMP14 in CRC.

In order to obtain a reliable conclusion, we first explored the prognostic value of MMP14 in the TCGA and GSE39582 databases, and then validated the conclusion in our own database. In the 32 paired CRC and normal tissues from the TCGA database, the expression of MMP14 mRNA was dramatically higher in cancers than in their normal controls. Similarly, the MMP14 protein was significantly more abundant in cancer tissues than in their normal counterparts in the validation cohort. Moreover, MMP14 mRNA expression increased gradually from stage I to stage III in both the TCGA and GSE39582 databases. High MMP14 expression was associated with advanced N stage and lymphovascular invasion in the validation cohort. All these findings suggested that MMP14 plays a critical role in carcinogenesis and progression of CRC.

Overexpression of MMP14 in cancer tissues and its correlation with poor clinicopathologic factors suggested that MMP14 is an oncogene. Hence, we conjectured that MMP14 may be a biomarker in CRC. We first verified our hypotheses in the public databases. MMP14 was a predictor of OS in the TCGA database and of RFS in the GSE39582 database. The TCGA and GSE39582 databases lack some important information, such as the quality of surgery (radical or palliative resection) and adjunctive therapeutic strategies, and OS information was not available in the GSE39592 database, while some RFS information was missing from the TCGA database. We then validated the results in our own database. Higher MMP14 expression indicated shorter OS and DFS in this validation cohort. Importantly, MMP14 was validated as an independent prognostic factor for both OS and RFS in multivariate analysis.

In summary, by employing 3 independent patient cohorts, our study revealed the prognostic value of MMP14 in CRC for the first time. Our findings strongly suggest that MMP14 has potential in predicting the treatment outcome and may be a novel biomarker in CRC. Our results warrant further studies of the mechanisms by which MMP14 promotes tumor progression and metastases in CRC.

References

[1] Siegel RL, Miller KD, Jemal A. Cancer statistics, 2017. CA Cancer J Clin 2017;67:7-30.

[2] Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. CA Cancer J Clin 2011;61:69-90.

[3] Urosevic J, Garcia-Albeniz X, Planet E, et al. Colon cancer cells colonize the lung from established liver metastases through p38 MAPK signalling and PTHLH. Nature Cell Biol 2014;16:685-94.

[4] Chuang HY, Jiang JK, Yang MH, et al. Aminopeptidase a initiates tumorigenesis and enhances tumor cell stemness via TWIST1 upregulation in colorectal cancer. Oncotarget 2017;8:21266-80.

[5] Eschrich S, Yang I, Bloom G, et al. Molecular staging for survival prediction of colorectal cancer patients. J Clin Oncol 2005;23:3526-35.

[6] McLeod HL, Murray GI. Tumour markers of prognosis in colorectal cancer. Br J Cancer 1999;79:191-203.

[7] Li Q, Wu J, Wei P, et al. Overexpression of forkhead box C2 promotes tumor metastasis and indicates poor prognosis in colon cancer via regulating epithelial–mesenchymal transition. Am J Cancer Res 2015;5:2022-34.

[8] Hornebeck W, Emonard H, Monboisse JC, Bellon G. Matrix-directed regulation of pericellular proteolysis and tumor progression. Semin Cancer Biol 2002;12:231-41.

[9] Rempe RG, Hartz AM, Bauer B. Matrix metalloproteinases in the brain and blood–brain barrier: versatile breakers and makers. J Cerebr Blood Flow Metab 2016;36:1481-507.

[10] Baker AH, Edwards DR, Murphy G. Metalloproteinase inhibitors: biological actions and therapeutic opportunities. J Cell Sci 2002;115:3719-27.

[11] Pahwa S, Stawikowski MJ, Fields GB. Monitoring and inhibiting MT1-MMP during cancer initiation and progression. Cancer 2014;6:416-35.

[12] Dong Y, Chen G, Gao M, Tian X. Increased expression of MMP14 correlates with the poor prognosis of Chinese patients with gastric cancer. Gene 2015;563:29-34.

[13] Itoh Y, Seiki M. MT1-MMP: a potent modifier of pericellular microenvironment. J Cell Physiol 2006;206:1-8.

[14] Buyyounouski MK, Choyke PL, Kattan M, Amin MB, editors. AJCC Cancer staging manual. 8th ed. Chicago: Springer; 2017. p. 715-26.

[15] Wang X, Jiang Z, Zhang Y, Wang X, Liu L, Fan Z. RNA sequencing analysis reveals protective role of Kruppel-like factor 3 in colorectal cancer. Oncotarget 2017;8:21984-93.

[16] Sato H, Takino T, Okada Y, et al. A matrix metalloproteinase expressed on the surface of invasive tumour cells. Nature 1994;370:61-5.

[17] Hui P, Xu X, Xu L, Hui G, Wu S, Lan Q. Expression of MMP14 in invasive pituitary adenomas: relationship to invasion and angiogenesis. Int J Clin Exp Pathol 2015;8:3556-7.

[18] Lehti K, Allen E, Birkedal-Hansen H, et al. An MT1-MMP-PDGF receptor-beta axis regulates mural cell investment of the microvasculature. Genes Dev 2005;19:979-91.

[19] Jiang WG, Davies G, Martin TA, et al. Expression of membrane type-1 matrix metalloproteinase, MT1-MMP in human breast cancer and its impact on invasiveness of breast cancer cells. Int J Mol Med 2006;17:583-90.

[20] Sabeh F, Ota I, Holmbeck K, et al. Tumor cell traffic through the extracellular matrix is controlled by the membrane-anchored collagenase MT1-MMP. J Cell Biol 2004;167:769-81.

[21] Ager EI, Kozin SV, Kirkpatrick ND, et al. Blockade of MMP14 activity in murine breast carcinomas: implications for macrophages, vessels, and radiotherapy. J Natl Cancer Inst 2015;107:djv017.

[22] Zuo QF, Cao LY, Yu T, et al. MicroRNA-22 inhibits tumor growth and metastasis in gastric cancer by directly targeting MMP14 and snail. Cell Death Dis 2015;6:e2000.

[23] Jia LF, Wei SB, Mitchelson K, et al. miR-34a inhibits migration and invasion of tongue squamous cell carcinoma via targeting MMP9 and MMP14. PloS One 2014;9:e108435.

[24] Li T, Xie J, Shen C, et al. miR-150-5p inhibits hepatoma cell migration and invasion by targeting MMP14. PloS One 2014;9:e115577.