ANTICANCER RESEARCH *41*: 3309-3315 (2021)
doi:10.21873/anticanres.15118

The Association of *MMP9* Promoter Rs3918242 Genotype With Gastric Cancer

CHUN-KAI FU[1,2,3,4*], WEN-SHIN CHANG[1,3*], CHIA-WEN TSAI[1,3*], YUN-CHI WANG[1,3], MEI-DUE YANG[3], HUA-SHAI HSU[5], CHE-YI CHAO[6], CHIEN-CHIH YU[3], JAW-CHYUN CHEN[7], JEN-SHENG PEI[8] and DA-TIAN BAU[1,3,9]

[1]*Graduate Institute of Biomedical Sciences, China Medical University, Taichung, Taiwan, R.O.C.;*
[2]*Taichung Armed Forces General Hospital, Taichung, Taiwan, R.O.C.;*
[3]*Terry Fox Cancer Research Laboratory, Department of Medical Research, China Medical University Hospital, Taichung, Taiwan, R.O.C.;*
[4]*National Defense Medical Center, Taipei, Taiwan, R.O.C.;*
[5]*Department of Medical Imaging, China Medical University Hospital, Taichung, Taiwan, R.O.C.;*
[6]*Department of Food Nutrition and Health Biotechnology, Asia University, Taichung, Taiwan, R.O.C.;*
[7]*Department of Medicinal Botanicals and Health Applications, Da-Yeh University, Changhua, Taiwan, R.O.C.;*
[8]*Department of Pediatrics, Taoyuan General Hospital, Ministry of Health and Welfare, Taoyuan, Taiwan, R.O.C.;*
[9]*Department of Bioinformatics and Medical Engineering, Asia University, Taichung, Taiwan, R.O.C.*

Abstract. *Background/Aim: Matrix metalloproteinase 9 (MMP9) is highly expressed in gastric cancer but the role of MMP9 is unclear. This study aimed at revealing the association of MMP9 promoter rs3918242 genotypes with gastric cancer risk. Materials and Methods: MMP9 rs3918242 genotypes of 121 patients with gastric cancer and 363 healthy individuals were examined by polymerase chain reaction-restriction fragment length polymorphism methodology using serum samples. Results: MMP9 rs3918242 TT genotype carriers had an elevated gastric cancer risk compared to wild-type CC carriers (odds ratio=3.92, 95% confidence interval=1.28-11.99; p=0.0103). Patients with CT/TT genotypes were at higher risk of metastasis (p=0.0178) than those with CC. No correlation was found between MMP9 rs3918242 genotype and gastric cancer risk with smoking or alcohol behavior, nor Helicobacter pylori infection. No correlation was observed for MMP9 rs3918242 genotypic distributions with age, gender, or body mass index. Conclusion: Carrying a T allele for MMP9 rs3918242 may be predictive for higher gastric cancer risk, and as a predictor for higher risk of metastasis.*

Gastric cancer has been listed as the fourth most prevalent cancer and the second most common cause of cancer-related death worldwide (1). Epidemiologically speaking, lifestyle, dietary habits and genomic factors combine to cause the etiology of gastric cancer, while personalized predictive strategy based on genomics for gastric cancer remains unestablished. In literature, dietary habits (2, 3), obesity (4), alcoholism (4-6), cigarette consumption (4), *Helicobacter pylori* infection (7), occupational exposure status (8-10), and inherited genetic polymorphisms (11) may all be involved in the development of gastric carcinogenesis but their interactions are largely unknown. In Taiwan, gastric cancer is the 10th most prevalent type of cancer (7th in males and 10th in females) and the 8th cause of cancer-related deaths (12). In recent years, several population-based genomic studies focusing on evaluating the association of genomic markers with the risk of gastric cancer in Taiwan have some novel findings (13-16), although these have not yet found use in clinical practice.

Matrix metalloproteinases (MMPs), a family of proteins with individual capacities to degrade specific substrates (17), are responsible for the complex and subtle regulation of the extracellular matrix, an imbalance of which may result in tumorigenesis (18). Alteration of expression patterns of MMPs and imbalance of the extracellular microenvironment are frequently observed in various types of cancer, including gastric cancer (19). For instance, *MMP2* and *MMP9* have

*These Authors contributed equally to this study.

Correspondence to: Da-Tian Bau, Terry Fox Cancer Research Laboratory, China Medical University Hospital, 2 Yuh-Der Road, Taichung, 404 Taiwan, R.O.C. Tel: +886 422053366 Ext. 5805, e-mail: artbau2@gmail.com

Key Words: Gastric cancer, genotype, *MMP9*, polymorphism, Taiwan.

Table I. *Basic characteristics of the control and gastric cancer patient groups.*

Character		Cases (n=121)	Controls (n=363)	*p*-Value[a]
Age, years	Mean±SD	51.3±9.4	53.2±8.1	0.8918
Gender	Female/male	56/65	168/195	0.9999
BMI, kg/m^2	Mean±SD	27.1±5.8	26.7±6.6	0.9344
Alcohol consumption, n (%)	Never drinker	70 (57.9)	268 (73.8)	
	Drinker	39 (32.2)	84 (23.1)	0.0538
	Heavy drinker[b]	12 (9.9)	11 (3.1)	**0.0049**
Cigarette consumption, n (%)	Never smoker	66 (54.6)	286 (78.8)	
	Smoker	42 (34.7)	71 (19.5)	**0.0012**
	Heavy smoker[c]	13 (10.7)	6 (1.7)	**0.0001**
Helicobacter pylori infection, n (%)	Yes	85 (70.2)	188 (51.8)	**0.0005**
Tumor location, n (%)	Upper	17 (14.1)		
	Middle	54 (44.6)		
	Lower	50 (41.3)		
Metastasis, n (%)	No	52 (43.0)		
	Yes	69 (57.0)		

BMI: Body mass index; SD: standard deviation. [a]Based on chi-square test. [b]Unable to walk straight, or respond to common questions properly in speech more than twice weekly after drinking or drinking more than 100 ml per day for at least half year. [c]More than 1 pack per day for at least half a year. Significant *p*-values ($p<0.05$) are shown in bold.

been found to be up-regulated in gastric cancer tissues (20). On the contrary, down-regulation of *MMP3* and *MMP13* have been found to effectively suppress gastric cancer fibroblast proliferation and migration (21, 22). It is evident that MMPs may be involved in the progression of gastric cancer, as part of a complex network.

Twenty-eight MMP family members that are responsible for regulating the components of the extracellular matrix have been identified, and their functions are becoming revealed (23, 24). MMP9 is one of the most important enzymes involved in the breakdown of the extracellular matrix, which plays a crucial role in various types of cancer (25). Among sites of single nucleotide polymorphisms of *MMP9,* C1562T (rs3918242), located in the promoter region, is the most well-known and most frequently examined. *MMP9* rs3918242 polymorphisms have been reported to be associated with risk for several types of cancer, including of the lung (26), prostate (27), breast (28), and colorectum (29, 30). In addition, gain of 20q12-1q13 (where *MMP9* is located) has been identified one of the most frequent regions with genomic gains in gastric cancer (31-34). Moreover, some reports have suggested that the gain at 20q may contribute to gastric cancer lymph node metastasis (35, 36). From the information above, it is apparent that the progression of gastric cancer is influenced by alterations in MMP9 expression, while the effects of genomic variations are largely unknown. Therefore, in the current study, we aimed at evaluating its association with gastric cancer risk in a Taiwanese population and revealing its interactions with clinical and lifestyle factors.

Materials and Methods

Collection of gastric cancer cases and controls. During 2005 to 2007, 121 gastric cancer cases were collected at the China Medical University Hospital by Dr. Yang and his colleagues. Well matched in aspects of age and gender, a three-fold number of healthy people were enrolled from the Health Examination Cohort of the same Hospital. The design and protocol of this study was approved by the Institutional Review Board (IRB number: DMR100-IRB-107) and informed consent was obtained with the great help of the Tissue Bank. Selected demographic characteristics of the two groups are summarized and evaluated in Table I.

Methodology for the determination of MMP9 rs3918242 genotype. DNA was extracted using QIAamp Blood Mini Kit (Blossom, Taipei, Taiwan) as previously described (37-40), and *MMP9* rs3918242 genotyping was conducted as per our previous practice (41, 42). Briefly, the primer sequences for *MMP9* rs3918242 genotyping were 5'-TGGTCAACGTAGTGAAACCCCATCT-3' and 5'-TCCAGCCCCAATTATCACACTTAT-3'. The PCR conditions were set as one cycle at 94˚C for 5 min, then 35 cycles at 94˚C for 30 s, then 59˚C for 30 s and 72˚C for 30 s, and a terminal extension at 72˚C for 10 min. The restriction endonuclease, SphI (New England Biolabs, Taipei, Taiwan, ROC) was applied. The DNA adducts of CC, CT and TT genotypes after enzyme cutting were 386 bp only, 386+320+66 bps, and 320+66 bps, respectively.

Methodology of statistical analysis. Pearson's chi-square test without Yates' correction and Fisher exact test were applied for all comparisons, except Student's *t*-test was applied for the comparison of distribution of the ages between the case and control groups. The associations between the *MMP9* polymorphisms and gastric cancer risk were evaluated with odds ratios (ORs), as well as the corresponding 95% confidence intervals (CIs) after adjustment for

Table II. *Distribution of matrix metalloproteinase-9 rs3918242 genotypic frequencies among the controls and patients with gastric cancer.*

Genotype	Frequency, n (%)		OR (95% CI)[a]	*p*-Value[b]
	Cases (n=121)	Controls (n=363)		
rs3918242				
CC	83 (68.6)	279 (76.9)	1.00 (Reference)	
CT	31 (25.6)	78 (21.5)	1.34 (0.82-2.17)	0.2388
TT	7 (5.8)	6 (1.6)	3.92 (1.28-11.99)	**0.0103**
p_{trend}				**0.0262**
Carrier comparison				
CC+CT	114 (94.2)	357 (98.4)	1.00 (Reference)	
TT	7 (5.8)	6 (1.6)	3.65 (1.20-11.09)	**0.0149**
CC	83 (68.6)	279 (76.9)	1.00 (Reference)	
CT+TT	38 (31.4)	84 (23.1)	1.52 (0.96-2.40)	0.0698

CI: Confidence interval; OR: odds ratio. [a]Adjusted for age, gender, smoking, alcohol and *Helicobacter pylori* infection. [b]Based on chi-square test without Yates' correction. Significant *p*-values ($p<0.05$) are shown in bold.

Table III. *Allelic frequencies for matrix metalloproteinase-9 rs3918242 in the control and gastric cancer patient groups.*

Allelic type	Frequency, n (%)		Adjusted OR (95% CI)[a]	*p*-Value[b]
	Cases (n=242)	Controls (n=726)		
C	197 (81.4)	636 (87.6)	1.00 (Reference)	
T	45 (18.6)	90 (12.3)	1.61 (1.12-2.43)	**0.0159**

CI: Confidence interval; OR: odds ratio. [a]Adjusted for age, gender, smoking, alcohol and *Helicobacter pylori* infection. [b]Based on chi-square test without Yates' correction. Significant *p*-values ($p<0.05$) are shown in bold.

some factors including age, gender and other indices (Table I). Any *p*-value less than 0.05 was considered significant.

Results

Analysis of basic characteristics between the gastric cancer and control groups. The demographic indices of the 121 gastric cancer cases and the 363 controls are shown in Table I. Firstly, age and gender were well matched between the case and control groups ($p=0.8918$ and 0.9999, respectively). Secondly, the average body mass indices were of the same level ($p=0.9344$) for the case and control groups. Thirdly, alcohol consumers comprised a significantly higher percentage of the gastric cancer group than that of the control group (42.1% *versus* 26.2%) ($p<0.05$). Fourthly, the gastric cancer group had a significantly higher percentage of cigarette smokers, especially heavy smokers, than those of the control group (34.7% *vs*. 19.5%, and 10.7% *vs*. 1.7%, $p=0.0012$ and 0.0001, respectively). Fifthly, 70.2% of the patients with gastric cancer were positive for *Helicobacter pylori* infection, significantly higher than that of 51.8% for the control group ($p=0.0005$).

Association of MMP9 rs3918242 genotypes with gastric cancer risk. The genotypic results for *MMP9* rs3918242 among the 121 patients with gastric cancer and the 363 controls are presented in Table II. The genotypic distribution for *MMP9* rs3918242 in the control group fits well under the Hardy–Weinberg equilibrium ($p=0.8386$). The genotypic frequency distributions for *MMP9* rs3918242 were significantly different between the gastric cancer and control groups (p for trend=0.0262) (Table II). In detail, *MMP9* rs3918242 heterozygous CT genotype seemed not to be associated with risk for gastric cancer ($p=0.2388$). On the contrary, the homozygous variant TT genotype exhibited a significant association with gastric cancer risk (adjusted OR=3.92, 95% CI=1.28-11.99; $p=0.0103$). We also compared those carrying the TT genotype with those with CC or CT genotype, and the results showed that the TT genotype at *MMP9* rs3918242 conferred a 3.65-fold odds of gastric cancer (adjusted OR, 95% CI=1.20-11.09, $p=0.0149$) (Table II). On the contrary, people carrying CT or TT genotypes had a non-significant risk of gastric cancer (adjusted OR=1.52, 95% CI=0.96-2.40; $p=0.0698$), compared with the CC genotype (Table Il).

Table IV. *Correlation between matrix metalloproteinase-9 rs3918242 genotype and clinicopathological features of 121 patients with gastric cancer.*

Characteristic	Subgroup	Patients, n	*MMP9* rs3918242 genotype, n (%)			*p*-Value[a]
			CC	CT	TT	
Age	≤50 Years	54	38 (71.7)	12 (22.7)	3 (5.6)	0.7940
	>50 Years	67	45 (66.2)	19 (27.9)	4 (5.9)	
Gender	Male	65	46 (70.8)	16 (24.6)	3 (4.6)	0.7849
	Female	56	37 (66.1)	15 (26.8)	4 (7.1)	
BMI	≤25 kg/m^2	45	28 (62.2)	14 (31.1)	3 (6.7)	0.5057
	>25 kg/m^2	76	55 (72.4)	17 (22.4)	4 (5.2)	
Alcohol drinker	Never	70	51 (72.9)	16 (22.9)	3 (4.2)	0.4538
	Ever	51	32 (62.7)	15 (29.4)	4 (7.9)	
Cigarette smoker	Never	66	47 (70.1)	16 (23.9)	4 (6.0)	0.8874
	Ever	55	36 (66.7)	15 (27.8)	3 (5.5)	
Helicobacter pylori	Negative	36	21 (58.3)	12 (33.3)	3 (8.3)	0.2791
	Positive	85	62 (72.9)	19 (22.4)	4 (4.7)	
Metastasis	Negative	51	42 (82.4)	8 (15.6)	1 (2.0)	**0.0178**
	Positive	70	41 (58.6)	23 (32.8)	6 (8.6)	

[a]Based on Fisher's exact test; Significant *p*-values ($p<0.05$) are shown in bold.

Association of MMP9 rs3918242 allelic frequencies with gastric cancer risk. To validate the genotypic findings deduced from Table II, we further conducted allelic frequency analysis for the association of *MMP9* rs3918242 with gastric cancer risk and the results are presented in Table III. There was a significant difference in the distribution of variant alleles between the gastric cancer and control groups for *MMP9* rs3918242 ($p=0.0159$, Table III), with an adjusted OR for those carrying the variant T allele of 1.61 (95% CI=1.12-2.43) compared to those carrying the wild-type C allele (Table III). Thus, the results of allelic frequency analysis support the idea that the T allele was associated with an increased risk of gastric cancer.

Interaction analysis of MMP9 rs3918242 *genotypes and clinical and lifestyle factors*. We are interested in genetic–lifestyle interactions and therefore the interaction between *MMP9* rs3918242 and clinical and lifestyle indices in their effect on gastric cancer risk was evaluated (Table IV). Firstly, we sub-grouped the patients according to their age, finding that the distributions of genotypes of *MMP9* rs3918242 did not differ among those older and younger than 50 years old ($p=0.7940$). Moreover, when we analyzed the distributions of gender, they were also similar ($p=0.7849$) (Table IV). Secondly there was also no difference in distribution observed for those with body mass index using a cut-off of 25 kg/m^2 ($p=0.5057$). Thirdly, for alcohol drinking, cigarette smoking and *H. pylori* infection status, the results remained non-significant (all $p>0.05$). Last but not least, there was an obvious association of metastasis in patients with gastric cancer carrying a T allele for *MMP9* rs3918242 ($p=0.0178$).

Discussion

The overexpression of MMP9 has been observed in many types of cancer, including gastric cancer (43-45). It is reasonable that single nucleotide polymorphisms on the *MMP9* promoter region may control regulation of the expression of MMP9 (46). Among *MMP9* single nucleotide polymorphisms, rs3918242 is the most investigated. For instance, Xing *et al*. has provided evidence supporting the contribution of *MMP9* rs3918242 to colorectal cancer (29). They found that the *MMP9* rs3918242 T allele was associated with an increased risk of colorectal cancer among the elderly (>60 years old) and contributed to elevating the risk of metastasis (29). However, there is no direct evidence of its role in gastric cancer.

In this study, we concluded that the TT genotype of *MMP9* rs3918242 is associated with elevated risk of gastric cancer (Table II), and this finding is consistent with the previous finding in Turkish population with relatively small sample size (case/control=79/65) (47). As far as we are concerned, the present study revealed the genotypic contribution of *MMP9* rs3918242 genotype to gastric cancer in Taiwan. We do not find any difference in age, gender, and other factors (Table IV). A highlight of our finding is that the genetic variants in *MMP9* may play a critical role in metastasis (Table IV). Yang and colleagues found another two *MMP9* polymorphic sites, R279Q and P574R, to be associated with gastric cancer metastasis (48). In literature, there is mounting evidence showing that the expression of MMP9 is correlated with the metastatic behavior of gastric cancer cells (49-52). Further investigations of the underlying

molecular mechanism of its involvement in metastasis using gastric cancer samples are urgently warranted to understand the role of MMP9 in gastric cancer.

There are several directions for further examining the role of *MMP9* in gastric carcinogenesis. One of the missions should be to enlarge the collection sample size. Only in this way could other East-Asian data with similar genetic backgrounds be incorporated together for meta-analyses. A platform of primary cultured cells from tumoral and non-tumoral sites of patients of gastric cancer (and control counterparts) would also be helpful to further understand the role of *MMP9* in aspects of its transcripts, protein level and activity. Although no interaction of body mass index and *MMP9* genotype was found (Table IV), a further understanding of the *MMP9* genotype in joint effects with the diet of patients would be very helpful in understanding the role of *MMP9* in the development of gastric cancer.

In conclusion, this study provided evidence for the significant association of *MMP9* promoter rs3918242 T allele with gastric cancer risk. In addition, *MMP9* rs3918242 appears to play an important role in metastasis. It might serve as both a predictive marker for personal susceptibility and for metastasis of gastric cancer.

Conflicts of Interest

All the Authors declared no conflicts of interest.

Authors' Contributions

Research design: Fu CK, Chang WS and Tsai CW; patient and questionnaire summaries: Fu CK, Hsu HS and Yang MD; experimental work: Wang YC, Chang WS and Yu CC; statistical analysis: Chao CY, Chen JS and Pei JS; article writing: Tsai CW and Bau DT; review and revision: Bau DT.

Acknowledgements

The Authors are grateful to Tai-Lin Huang, Yu-Ting Chin and Yu-Chen Hsiau for their technical support. The cooperation of all the participants is highly appreciated. This study was financially supported by Taichung Armed Forces General Hospital to Dr. Fu (grant number: 108A09) and China Medical University Hospital and Asia University (CMU109-ASIA-03).

References

1 Siegel RL, Miller KD, Fuchs HE and Jemal A: Cancer statistics, 2021. CA Cancer J Clin *71(1)*: 7-33, 2021. PMID: 33433946. DOI: 10.3322/caac.21654

2 Buckland G, Travier N, Huerta JM, Bueno-de-Mesquita HB, Siersema PD, Skeie G, Weiderpass E, Engeset D, Ericson U, Ohlsson B, Agudo A, Romieu I, Ferrari P, Freisling H, Colorado-Yohar S, Li K, Kaaks R, Pala V, Cross AJ, Riboli E, Trichopoulou A, Lagiou P, Bamia C, Boutron-Ruault MC, Fagherazzi G, Dartois L, May AM, Peeters PH, Panico S, Johansson M, Wallner B, Palli D, Key TJ, Khaw KT, Ardanaz E, Overvad K, Tjønneland A, Dorronsoro M, Sánchez MJ, Quirós JR, Naccarati A, Tumino R, Boeing H and Gonzalez CA: Healthy lifestyle index and risk of gastric adenocarcinoma in the EPIC cohort study. Int J Cancer *137(3)*: 598-606, 2015. PMID: 25557932. DOI: 10.1002/ijc.29411

3 Lin SH, Li YH, Leung K, Huang CY and Wang XR: Salt processed food and gastric cancer in a Chinese population. Asian Pac J Cancer Prev *15(13)*: 5293-5298, 2014. PMID: 25040991. DOI: 10.7314/apjcp.2014.15.13.5293

4 Vaughan TL, Davis S, Kristal A and Thomas DB: Obesity, alcohol, and tobacco as risk factors for cancers of the esophagus and gastric cardia: adenocarcinoma versus squamous cell carcinoma. Cancer Epidemiol Biomarkers Prev *4(2)*: 85-92, 1995. PMID: 7742727.

5 Jedrychowski W, Wahrendorf J, Popiela T and Rachtan J: A case-control study of dietary factors and stomach cancer risk in Poland. Int J Cancer *37(6)*: 837-842, 1986. PMID: 3710615. DOI: 10.1002/ijc.2910370607

6 Lagergren J, Bergström R, Lindgren A and Nyrén O: The role of tobacco, snuff and alcohol use in the aetiology of cancer of the oesophagus and gastric cardia. Int J Cancer *85(3)*: 340-346, 2000. PMID: 10652424.

7 Warren JR and Marshall B: Unidentified curved bacilli on gastric epithelium in active chronic gastritis. Lancet *1(8336)*: 1273-1275, 1983. PMID: 6134060.

8 Cocco P, Palli D, Buiatti E, Cipriani F, DeCarli A, Manca P, Ward MH, Blot WJ and Fraumeni JF Jr: Occupational exposures as risk factors for gastric cancer in Italy. Cancer Causes Control *5(3)*: 241-248, 1994. PMID: 8061172. DOI: 10.1007/BF01830243

9 Simpson J, Roman E, Law G and Pannett B: Women's occupation and cancer: preliminary analysis of cancer registrations in England and Wales, 1971-1990. Am J Ind Med *36(1)*: 172-185, 1999. PMID: 10361604. DOI: 10.1002/(sici)1097-0274(199907)36:1<172::aid-ajim25>3.0.co;2-x

10 Aragonés N, Pollán M and Gustavsson P: Stomach cancer and occupation in Sweden: 1971-89. Occup Environ Med *59(5)*: 329-337, 2002. PMID: 11983848. DOI: 10.1136/oem.59.5.329

11 La Vecchia C, Negri E, Franceschi S and Gentile A: Family history and the risk of stomach and colorectal cancer. Cancer *70(1)*: 50-55, 1992. PMID: 1606546. DOI: 10.1002/1097-0142(19920701)70:1<50::aid-cncr2820700109>3.0.co;2-i

12 Taiwan Ministry of Health and Welfare Clinical Trial and Research Center of Excellence: Cancer registration annual report. Available at: https://www.hpa.gov.tw/Pages/List.aspx?nodeid=269 [Last accessed on May 25th, 2021]

13 Lin CH, Lin CC, Tsai CW, Chang WS, Yang CW and Bau DT: Association of caveolin-1 genotypes with gastric cancer in Taiwan. Anticancer Res *34(5)*: 2263-2267, 2014. PMID: 24778029.

14 Kuo WH, Huang CY, Fu CK, Hsieh YH, Liao CH, Hsu CM, Huang YK, Tsai CW, Chang WS and Bau DT: Effects of interleukin-10 polymorphisms and smoking on the risk of gastric cancer in Taiwan. In Vivo *28(5)*: 967-971, 2014. PMID: 25189915.

15 Kuo HW, Huang CY, Fu CK, Liao CH, Hsieh YH, Hsu CM, Tsai CW, Chang WS and Bau DT: The significant association of CCND1 genotypes with gastric cancer in Taiwan. Anticancer Res *34(9)*: 4963-4968, 2014. PMID: 25202078.

16 Ji HX, Chang WS, Tsai CW, Wang JY, Huang NK, Lee AS, Shen MY, Chen WY, Chiang YC, Shih TC, Hsu CM and Bau DT:

Contribution of DNA repair xeroderma pigmentosum group D genotype to gastric cancer risk in Taiwan. Anticancer Res *35(9)*: 4975-4981, 2015. PMID: 26254397.

17 Birkedal-Hansen H, Moore WG, Bodden MK, Windsor LJ, Birkedal-Hansen B, DeCarlo A and Engler JA: Matrix metalloproteinases: a review. Crit Rev Oral Biol Med *4(2)*: 197-250, 1993. PMID: 8435466. DOI: 10.1177/10454411930040020401

18 Lukashev ME and Werb Z: ECM signalling: orchestrating cell behaviour and misbehaviour. Trends Cell Biol *8(11)*: 437-441, 1998. PMID: 9854310. DOI: 10.1016/s0962-8924(98)01362-2

19 Park CC, Bissell MJ and Barcellos-Hoff MH: The influence of the microenvironment on the malignant phenotype. Mol Med Today *6(8)*: 324-329, 2000. PMID: 10904250. DOI: 10.1016/s1357-4310(00)01756-1

20 Yang SF, Lin CY, Yang PY, Chao SC, Ye YZ and Hu DN: Increased expression of gelatinase (MMP-2 and MMP-9) in pterygia and pterygium fibroblasts with disease progression and activation of protein kinase C. Invest Ophthalmol Vis Sci *50(10)*: 4588-4596, 2009. PMID: 19420332. DOI: 10.1167/iovs.08-3147

21 Kim YH, Jung JC, Gum SI, Park SB, Ma JY, Kim YI, Lee KW and Park YJ: Inhibition of pterygium fibroblast migration and outgrowth by bevacizumab and cyclosporine a involves down-regulation of matrix metalloproteinases-3 and -13. PLoS One *12(1)*: e0169675, 2017. PMID: 28068383. DOI: 10.1371/journal.pone.0169675

22 Tsai YY, Chiang CC, Yeh KT, Lee H and Cheng YW: Effect of TIMP-1 and MMP in pterygium invasion. Invest Ophthalmol Vis Sci *51(7)*: 3462-3467, 2010. PMID: 20207965. DOI: 10.1167/iovs.09-4921

23 Cui N, Hu M and Khalil RA: Biochemical and biological attributes of matrix metalloproteinases. Prog Mol Biol Transl Sci *147*: 1-73, 2017. PMID: 28413025. DOI: 10.1016/bs.pmbts.2017.02.005

24 Łukaszewicz-Zając M, Szmitkowski M, Litman-Zawadzka A and Mroczko B: Matrix metalloproteinases and their tissue inhibitors in comparison to other inflammatory proteins in gastric cancer (GC). Cancer Invest *34(7)*: 305-312, 2016. PMID: 27414231. DOI: 10.1080/07357907.2016.1197237

25 Verma S, Kesh K, Gupta A and Swarnakar S: An overview of matrix metalloproteinase 9 polymorphism and gastric cancer risk. Asian Pac J Cancer Prev *16(17)*: 7393-7400, 2015. PMID: 26625734. DOI: 10.7314/apjcp.2015.16.17.7393

26 Bayramoglu A, Gunes HV, Metintas M, Değirmenci I, Mutlu F and Alataş F: The association of MMP-9 enzyme activity, MMP-9 C1562T polymorphism, and MMP-2 and -9 and TIMP-1, -2, -3, and -4 gene expression in lung cancer. Genet Test Mol Biomarkers *13(5)*: 671-678, 2009. PMID: 19814619. DOI: 10.1089/gtmb.2009.0053

27 Schveigert D, Valuckas KP, Kovalcis V, Ulys A, Chvatovic G and Didziapetriene J: Significance of MMP-9 expression and MMP-9 polymorphism in prostate cancer. Tumori *99(4)*: 523-529, 2013. PMID: 24326842. DOI: 10.1700/1361.15105

28 Felizi RT, Veiga MG, Carelli Filho I, Souto RPD, Fernandes CE and Oliveira E: Association between Matrix Metallopeptidase 9 Polymorphism and Breast Cancer Risk. Rev Bras Ginecol Obstet *40(10)*: 620-624, 2018. PMID: 30352460. DOI: 10.1055/s-0038-1673366

29 Xing LL, Wang ZN, Jiang L, Zhang Y, Xu YY, Li J, Luo Y and Zhang X: Matrix metalloproteinase-9-1562C>T polymorphism may increase the risk of lymphatic metastasis of colorectal cancer. World J Gastroenterol *13(34)*: 4626-4629, 2007. PMID: 17729419. DOI: 10.3748/wjg.v13.i34.4626

30 Hoelzle CR, Magalhães KC, Carvalho SS, Santos GA, Maia IM, Sousa MC, Andrade-Filho JS, Cruz GM and Simões RT: Matrix metalloproteinase 9 -1562C/T polymorphism increased protein levels in patients with colorectal cancer in a sample from southeastern Brazil. Genet Mol Res *15(1)*, 2016. PMID: 27051027. DOI: 10.4238/gmr.15017478

31 Kimura Y, Noguchi T, Kawahara K, Kashima K, Daa T and Yokoyama S: Genetic alterations in 102 primary gastric cancers by comparative genomic hybridization: gain of 20q and loss of 18q are associated with tumor progression. Mod Pathol *17(11)*: 1328-1337, 2004. PMID: 15154013. DOI: 10.1038/modpathol.3800180

32 Noguchi T, Wirtz HC, Michaelis S, Gabbert HE and Mueller W: Chromosomal imbalances in gastric cancer. Correlation with histologic subtypes and tumor progression. Am J Clin Pathol *115(6)*: 828-834, 2001. PMID: 11392878. DOI: 10.1309/2Q9E-3EP5-KYPK-VFGQ

33 Sakakura C, Hagiwara A, Yasuoka R, Fujita Y, Nakanishi M, Masuda K, Shimomura K, Nakamura Y, Inazawa J, Abe T and Yamagishi H: Tumour-amplified kinase BTAK is amplified and overexpressed in gastric cancers with possible involvement in aneuploid formation. Br J Cancer *84(6)*: 824-831, 2001. PMID: 11259099. DOI: 10.1054/bjoc.2000.1684

34 Nessling M, Solinas-Toldo S, Wilgenbus KK, Borchard F and Lichter P: Mapping of chromosomal imbalances in gastric adenocarcinoma revealed amplified protooncogenes MYCN, MET, WNT2, and ERBB2. Genes Chromosomes Cancer *23(4)*: 307-316, 1998. PMID: 9824203. DOI: 10.1002/(sici)1098-2264(199812)23:4<307::aid-gcc5>3.0.co;2-#

35 Kang JU, Kang JJ, Kwon KC, Park JW, Jeong TE, Noh SM and Koo SH: Genetic alterations in primary gastric carcinomas correlated with clinicopathological variables by array comparative genomic hybridization. J Korean Med Sci *21(4)*: 656-665, 2006. PMID: 16891809. DOI: 10.3346/jkms.2006.21.4.656

36 Hidaka S, Yasutake T, Kondo M, Takeshita H, Yano H, Haseba M, Tsuji T, Sawai T, Nakagoe T and Tagawa Y: Frequent gains of 20q and losses of 18q are associated with lymph node metastasis in intestinal-type gastric cancer. Anticancer Res *23(4)*: 3353-3357, 2003. PMID: 12926075.

37 Chen KY, Chien WC, Liao JM, Tsai CW, Chang WS, Su CH, Hsu SW, Wang HC and Bau DT: Contribution of interleukin-10 genotype to triple negative breast cancer risk. Anticancer Res *41(5)*: 2451-2457, 2021. PMID: 33952470. DOI: 10.21873/anticanres.15020

38 Wu MH, Yueh TC, Chang WS, Tsai CW, Fu CK, Yang MD, Yu CC and Bau DT: Contribution of *Matrix Metalloproteinase-1* genotypes to colorectal cancer in Taiwan. Cancer Genomics Proteomics *18(3)*: 245-251, 2021. PMID: 33893077. DOI: 10.21873/cgp.20255

39 Chen GL, Wang SC, Shen TC, Chang WS, Lin C, Hsia TC, Bau DT and Tsai CW: Significant association of chitinase 3-like 1 genotypes to asthma risk in Taiwan. In Vivo *35(2)*: 799-803, 2021. PMID: 33622872. DOI: 10.21873/invivo.12320

40 Chen LH, Shen TC, Li CH, Chiu KL, Hsiau YC, Wang YC, Gong CL, Wang ZH, Chang WS, Tsai CW, Hsia TC and Bau DT: The significant interaction of excision repair cross-complementing group 1 genotypes and smoking to lung cancer risk. Cancer Genomics Proteomics *17(5)*: 571-577, 2020. PMID: 32859635. DOI: 10.21873/cgp.20213

41 Tsai CB, Hsia NY, Wang ZH, Yang JS, Hsu YM, Wang YC, Chang WS, Bau DT, Yin MC and Tsai CW: The contribution of *MMP-9*

genotypes to pterygium in Taiwan. Anticancer Res *40(8)*: 4523-4527, 2020. PMID: 32727782. DOI: 10.21873/anticanres.14457

42 Wu MH, Tzeng HE, Wu CN, Yueh TC, Peng YC, Tsai CH, Wang YC, Ke TW, Pei JS, Chang WS, Tsai CW and Bau DT: Association of matrix metalloproteinase-9 rs3918242 promoter genotypes with colorectal cancer risk. Anticancer Res *39(12)*: 6523-6529, 2019. PMID: 31810917. DOI: 10.21873/anticanres.13867

43 Herszenyi L, Hritz I, Pregun I, Sipos F, Juhasz M, Molnar B and Tulassay Z: Alterations of glutathione S-transferase and matrix metalloproteinase-9 expressions are early events in esophageal carcinogenesis. World J Gastroenterol *13(5)*: 676-682, 2007. PMID: 17278189. DOI: 10.3748/wjg.v13.i5.676

44 Sun WH, Sun YL, Fang RN, Shao Y, Xu HC, Xue QP, Ding GX and Cheng YL: Expression of cyclooxygenase-2 and matrix metalloproteinase-9 in gastric carcinoma and its correlation with angiogenesis. Jpn J Clin Oncol *35(12)*: 707-713, 2005. PMID: 16314343. DOI: 10.1093/jjco/hyi196

45 Pellikainen JM, Ropponen KM, Kataja VV, Kellokoski JK, Eskelinen MJ and Kosma VM: Expression of matrix metalloproteinase (MMP)-2 and MMP-9 in breast cancer with a special reference to activator protein-2, HER2, and prognosis. Clin Cancer Res *10(22)*: 7621-7628, 2004. PMID: 15569994. DOI: 10.1158/1078-0432.CCR-04-1061

46 Wang Y, Fang S, Wei L, Wang R, Jin X, Wen D, Li Y, Guo W, Wang N and Zhang J: No association between the C-1562T polymorphism in the promoter of matrix metalloproteinase-9 gene and non-small cell lung carcinoma. Lung Cancer *49(2)*: 155-161, 2005. PMID: 15949868. DOI: 10.1016/j.lungcan.2005.04.006

47 Matsumura S, Oue N, Nakayama H, Kitadai Y, Yoshida K, Yamaguchi Y, Imai K, Nakachi K, Matsusaki K, Chayama K and Yasui W: A single nucleotide polymorphism in the MMP-9 promoter affects tumor progression and invasive phenotype of gastric cancer. J Cancer Res Clin Oncol *131(1)*: 19-25, 2005. PMID: 15565457. DOI: 10.1007/s00432-004-0621-4

48 Tang Y, Zhu J, Chen L, Chen L, Zhang S and Lin J: Associations of matrix metalloproteinase-9 protein polymorphisms with lymph node metastasis but not invasion of gastric cancer. Clin Cancer Res *14(9)*: 2870-2877, 2008. PMID: 18451255. DOI: 10.1158/1078-0432.CCR-07-4042

49 Song G, Ouyang G, Mao Y, Ming Y, Bao S and Hu T: Osteopontin promotes gastric cancer metastasis by augmenting cell survival and invasion through Akt-mediated HIF-1alpha up-regulation and MMP9 activation. J Cell Mol Med *13(8B)*: 1706-1718, 2009. PMID: 19602039. DOI: 10.1111/j.1582-4934.2008.00540.x

50 Chen Y, Wei X, Guo C, Jin H, Han Z, Han Y, Qiao T, Wu K and Fan D: Runx3 suppresses gastric cancer metastasis through inactivation of MMP9 by upregulation of TIMP-1. Int J Cancer *129(7)*: 1586-1598, 2011. PMID: 21128246. DOI: 10.1002/ijc.25831

51 Pan G, Zhang X, Ren J, Lu J, Li W, Fu H, Zhang S and Li J: Semaphorin 5A, an axon guidance molecule, enhances the invasion and metastasis of human gastric cancer through activation of MMP9. Pathol Oncol Res *19(1)*: 11-18, 2013. PMID: 22821546. DOI: 10.1007/s12253-012-9550-8

52 Zhao C, Yuan G, Jiang Y, Xu J, Ye L, Zhan W and Wang J: Capn4 contributes to tumor invasion and metastasis in gastric cancer via activation of the Wnt/β-catenin/MMP9 signalling pathways. Exp Cell Res *395(2)*: 112220, 2020. PMID: 32777225. DOI: 10.1016/j.yexcr.2020.112220

Received May 26, 2021
Revised June 9, 2021
Accepted June 10, 2021