

# The clinicopathological characteristics and genetic alterations of mucinous carcinoma of the stomach

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## Abstract

**Background:** Mucinous gastric carcinoma (MGC) is rare and often associated with an advanced stage. The clinicopathological features and prognosis of MGC and non-MGC (NMGC) are controversial.

**Methods:** In total, 2637 gastric cancer (GC) patients receiving curative surgery were enrolled. The clinicopathological features and genetic alterations were compared between patients with MGC and NMGC.

**Results:** Among the 2637 GC patients, 92 (3.5%) had MGC. After propensity score matching, compared to patients with NMGC, patients with MGC had more poorly differentiated tumors, medullary stromal reaction-type tumors, tumors with infiltrating Ming's classification, diffuse-type tumors, more abnormal preoperative serum carbohydrate antigen 19-9 levels, and more advanced T categories. After propensity score matching, there were no significant differences between MGC and NMGC regarding the initial recurrence patterns, 5-year overall survival (OS), and disease-free survival (DFS) rates. Multivariate analysis demonstrated that the MGC cell type is not an independent prognostic factor of OS and DFS. No significant differences in microsatellite instability status, Epstein-Barr virus infection, *Helicobacter pylori* infection, or genetic mutations were observed between MGC and NMGC. The expression of programmed death-ligand 1 (PD-L1) was significantly higher in MGC than that in NMGC. MGC was diagnosed at a more advanced stage compared with NMGC.

**Conclusion:** MGC itself was not an independent prognostic factor of worse survival. MGC was correlated with higher PD-L1 expression than NMGC, which may have a clinical impact on the treatment of MGC in the future.

**Keywords:** Genetic alteration; MGC; NMGC; PD-L1

## 1. INTRODUCTION

Gastric cancer (GC) is the sixth most common cancer and the second most common cause of cancer-related deaths worldwide.<sup>1</sup> Among all GC cases, mucinous gastric carcinoma (MGC) is rare and accounts for only 2% to 6%.<sup>2,3</sup> According to the definition by the World Health Organization (WHO), MGC is defined as adenocarcinoma with a substantial amount of extracellular mucin within the gastric tumor ( $\geq 50\%$  of the tumor volume).<sup>4</sup>

The clinicopathological features and prognosis of MGC are controversial. Some authors have reported a worse prognosis in MGC than non-MGC (NMGC)<sup>2,3,5</sup>; however, other studies have shown similar prognoses between MGC and NMGC.<sup>6</sup> Furthermore, differences in the initial recurrence patterns between MGC and NMGC are unclear.

In molecular analysis, MGC was associated with a lower incidence of human epidermal growth factor receptor 2 (HER2) overexpression, HER2 amplification, and epidermal growth factor receptor overexpression than NMGC.<sup>3</sup> To date, whether MGC is associated with genetic mutations is uncertain.

The aim of the study was to compare the clinicopathological features, recurrence patterns, prognoses, genetic alterations, and expression levels of HER2 and programmed death-ligand 1 (PD-L1) between MGC and NMGC.

## 2. METHODS

### 2.1. Ethics statement

All data were prospectively collected from medical records in our hospital. All procedures were performed according to the Declaration of Helsinki of 1964 along with its later versions. The ethics committees of our hospital reviewed and approved this study.

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Conflicts of interest: The authors declare that they have no conflicts of interest related to the subject matter or materials discussed in this article.

Journal of Chinese Medical Association. (2020) 83: 141-147.

Received August 1, 2019; accepted September 4, 2019.

doi: 10.1097/JCMA.000000000000232.

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## 2.2. Patients and sample collection

Between January 1992 and December 2013, 2637 GC patients with adenocarcinoma who underwent curative surgery were enrolled. According to the Japanese classification of gastric carcinoma, curative resection (R0) was defined as a complete resection of the localized tumors along with regional lymph node dissection.<sup>7</sup> As the definition of WHO, MGC was defined as adenocarcinoma with a substantial amount of extracellular mucin ( $\geq 50\%$  of the tumor volume) within the gastric tumor.<sup>4</sup> Among the 2637 patients, 92 (3.5%) were diagnosed with MGC, and 2545 had NMGC.

The tumor tissues and normal gastric mucosa tissues were collected and stored in the liquid nitrogen in biobank at our institution. Pathological staging of the GC was performed according to the eighth American Joint Committee on Cancer/Union for International Cancer Control tumor, node, metastasis (TNM) classification system.<sup>8</sup>

## 2.3. Follow-up

Follow-up examinations were performed every 3 months during the first 5 years after surgery and every 6 months thereafter, including physical examinations, blood tests with measurements of tumor markers (e.g., carcinoembryonic antigen [CEA] and carbohydrate antigen 19-9 [CA19-9]), chest radiography, sonography, or computerized tomography scans of the abdomen.

## 2.4. DNA extraction

As a previous report,<sup>9</sup> DNA was extracted from tissue specimens using the QIAamp DNA Tissue Kit and MinElute Virus Kit (Qiagen, Valencia, CA).

## 2.5. Identification of *Helicobacter pylori* infection and Epstein-Barr virus DNA detection

Both tumor tissue and nontumor tissues were examined for *Helicobacter pylori* (HP) infection. The polymerase chain

**Table 1**

**Clinical profile in GC patients with MGC and NMGC**

Variables	Original dataset			3:1 matched dataset		
	NMGC n = 2545 n (%)	MGC n = 92 n (%)	<i>p</i>	NMGC n = 276 n (%)	MGC n = 92 n (%)	<i>p</i>
Age			0.511			0.529
<65 y	915 (36.0)	30 (32.6)		100 (36.2)	30 (32.6)	
$\geq 65$ y	1630 (64.0)	62 (67.4)		176 (63.8)	62 (67.4)	
Gender (M/F)	1878/667	74/18	0.153	225/51	74/18	0.817
Tumor size (<5/ $\geq 5$ cm)	1409/1136	14/78	<0.001	42/234	14/78	1.000
Tumor location			0.274			0.641
Upper stomach	428 (16.8)	14 (15.2)		56 (20.3)	14 (15.2)	
Middle stomach	939 (36.9)	30 (32.6)		82 (29.7)	30 (32.6)	
Lower stomach	1116 (43.9)	43 (46.7)		128 (46.4)	43 (46.7)	
Whole stomach	62 (2.4)	5 (5.4)		10 (3.6)	5 (5.4)	
Cell differentiation			<0.001			0.003
Poor	1292 (50.8)	71 (77.2)		160 (58.0)	71 (77.2)	
Moderate	1182 (46.4)	21 (22.8)		112 (40.6)	21 (22.8)	
Well	71 (2.8)	0		4 (1.4)	0	
Gross appearance			<0.001			1.000
Superficial type	997 (39.2)	9 (9.8)		27 (9.8)	9 (9.8)	
Borrmann type 1 and 2	500 (19.6)	26 (28.3)		78 (28.3)	26 (28.3)	
Borrmann type 3 and 4	1048 (41.2)	57 (62.0)		171 (62.0)	57 (62.0)	
Stromal reaction type			<0.001			<0.001
Medullary	665 (26.1)	42 (45.7)		31 (11.2)	42 (45.7)	
Intermediate	1257 (49.4)	47 (51.1)		143 (51.8)	47 (51.1)	
Scirrhus	623 (24.5)	3 (3.3)		102 (37.0)	3 (3.3)	
Ming's classification			<0.001			0.004
Expanding	805 (31.6)	8 (8.7)		62 (22.5)	8 (8.7)	
Infiltrating	1740 (68.4)	84 (91.3)		214 (77.5)	84 (91.3)	
Lauren's classification			<0.001			<0.001
Intestinal type	218 (83.8)	21 (11.6)		151 (54.7)	29 (31.5)	
Diffuse type	42 (16.2)	160 (88.4)		125 (45.3)	63 (68.5)	
Lymphovascular invasion	1402 (55.1)	78 (84.8)	<0.001	235 (85.1)	78 (84.8)	0.933
Lymphoid stroma	237 (9.3)	2 (2.2)	0.019	46 (16.7)	2 (2.2)	<0.001
Preoperative serum CEA (ng/mL)			0.012			0.559
<5/ $\geq 5$	2178/367	70/22		218/58	70/22	
Preoperative serum CA19-9 (U/mL)			<0.001			0.001
<37/ $\geq 37$	2269/276	63/29		233/43	63/29	
Pathological T category			<0.001			0.019
T1/2/3/4	892/379/666/608	4/13/22/53		25/31/104/116	4/13/22/53	
Pathological N category			<0.001			0.809
N0/1/2/3	1245/361/383/556	18/14/20/40		55/49/67/105	18/14/20/40	
Pathological TNM stage			<0.001			0.922
I/II/III	1044/563/938	10/20/62		34/57/185	10/20/62	

CA19-9 = carbohydrate antigen 19-9; CEA = carcinoembryonic antigen; GC = gastric cancer; MGC = mucinous gastric carcinoma; NMGC = non-MGC; TNM = tumor, node, metastasis.

reaction (PCR) method was used as described in a previous study to identify HP infection.<sup>10</sup> The reference sequence of the HP reference genome (GenBank: AE000511.1) was used to design PCR forward (AAGCTTACTTTCTAACACTAACGC) and reverse (AAGCTTTTATAGGGGTGTTAGGGGTTT) primers.

As reported in a previous study,<sup>10</sup> Epstein-Barr virus (EBV) DNA assays were carried out using the Sequenom MassARRAY system (Sequenom, San Diego, CA).

## 2.6. Microsatellite instability analysis

As described in a previous study,<sup>11</sup> five reference microsatellite markers, including D5S345, D2S123, D17S250, BAT25, and BAT26, were used to determine microsatellite instability (MSI). MSI-high (MSI-H) was defined as  $\geq 2$  loci of instability with five markers, while MSI-low/stable was one locus or without loci of MSI.

## 2.7. MassARRAY-based mutation characterization

Sixty-eight mutation hotspots in eight GC-related genes, including *TP53*, *ARID1A*, *PTEN*, *PIK3CA*, *AKT1*, *AKT2*, *AKT3*, and *BRAF*, were identified using a MassARRAY system (Agena, San Diego, CA).<sup>9</sup> Among them, *PTEN*, *PIK3CA*, *AKT1*, *AKT2*, and *AKT3* were analyzed for *PI3K/AKT* pathway genetic mutations.

## 2.8. Immunohistochemical stains for PD-L1 and HER2

For PD-L1, immunohistochemical (IHC) stains were performed using the PD-L1 IHC 22C3 pharmDx kit on the Dako ASL48 platform.<sup>12</sup> The combined positive score (CPS) was calculated. Positive expression of PD-L1 was defined as a CPS score of  $\geq 1$ . The IHC staining of PD-L1 in GC specimen is shown in Fig. 2.

For HER2 IHC stains, the antihuman c-erbB-2 A0485 polyclonal antibody [dilution 1:500; Agilent (Santa Clara, CA, USA)] was used and IHC stains were performed using a BenchMark Ultra Platform (Ventana Medical Systems, Tucson, AZ) with the Optiview DAB Detection Kit (Ventana Medical Systems).

According to the international guidelines systems, HER2 immunoreactivity was scored as 0, 1+, 2+, or 3+.<sup>13</sup> HER2 positive was defined as an IHC score of 3+ or an IHC score of 2+ with a positive fluorescence in situ hybridization result. The IHC staining of HER2 in GC specimen is shown in Fig. 3.

## 2.9. Propensity score matching strategy

For minimizing the selection bias, propensity score matching through logistic regression modeling based on six covariates (age, gender, tumor size, gross appearance, lymphovascular invasion, and pathological TNM stage) was performed to balance the potential confounders between MGC and NMGC patients. A 1:3 ratio matching for MGC and NMGC patients was paired. A specific caliper width equal to 0.1 SD was used.

## 2.10. Statistical analysis

IBM SPSS Statistics 25.0 (Armonk, NY: IBM Corp) was used for statistical analyses. A  $\chi^2$  test with Yates correction or Fisher's exact test was used for the comparison of the categorical data. The overall survival (OS) was defined from the date of surgery to the date of death or the last follow-up, while the disease-free survival (DFS) was defined as the length of time after surgery during which the patient alive without recurrence of GC. The Kaplan-Meier method was used for the survival analysis and survival curves of OS and DFS. Multivariate analysis with Cox proportional hazards models was performed for analyzing the independent prognostic factors of OS and DFS. A *p* value < 0.05 was defined as statistically significant.

## 3. RESULTS

### 3.1. Clinicopathological features

The clinicopathological characteristics of the MGC and NMGC patients were compared (Table 1). After propensity score

**Table 2**

**The initial recurrence pattern in GC patients with MGC or NMGC after curative surgery**

	Original dataset			3:1 matched dataset		
	NMGC n = 2545 n (%)	MGC n = 92 n (%)	<i>P</i>	NMGC n = 276 n (%)	MGC n = 92 n (%)	<i>p</i>
Total patients with recurrence	659 (25.9)	40 (43.5)	<0.001	118 (42.8)	40 (43.5)	0.903
Locoregional recurrence	256 (10.1)	23 (25.0)	<0.001	45 (16.3)	23 (25.0)	0.063
Perigastric area	46 (1.8)	3 (3.3)		10 (3.6)	3 (3.3)	
Hepatoduodenal ligament	124 (4.9)	13 (14.1)		24 (8.7)	13 (14.1)	
Anastomosis	53 (2.1)	6 (6.5)		13 (4.7)	6 (6.5)	
Abdominal wall	89 (3.5)	5 (5.4)		13 (4.7)	5 (5.4)	
Duodenal stump	19 (0.7)	0		2 (0.7)	0	
Distant metastasis	438 (17.2)	24 (26.1)	0.028	86 (31.2)	24 (26.1)	0.357
Peritoneal dissemination	261 (10.3)	16 (17.4)	0.028	50 (18.1)	16 (17.4)	0.875
Hematogenous metastasis	259 (10.2)	14 (15.2)	0.119	46 (16.7)	14 (15.2)	0.745
Liver	169 (6.6)	6 (6.5)		35 (12.7)	6 (6.5)	
Lung	64 (2.5)	5 (5.4)		9 (3.3)	5 (5.4)	
Bone	38 (1.5)	3 (3.3)		7 (2.5)	3 (3.3)	
Brain	6 (0.2)	0		1 (0.4)	0	
Adrenal	8 (0.3)	0		0	0	
Skin	5 (0.2)	0		1 (0.4)	0	
Distant lymphatic recurrence	140 (5.5)	6 (6.5)	0.674	25 (9.1)	6 (6.5)	0.448
Virchow's node	16 (0.6)	0		2 (0.7)	0	
Inguinal lymph node	4 (0.2)	0		0	0	
Lymphangitis carcinomatosa	5 (0.2)	0		1 (0.4)	0	
Para-aortic lymph node	124 (4.9)	6 (6.5)		23 (8.3)	6 (6.5)	

Some patients had more than one recurrence pattern.

GC = gastric cancer; MGC = mucinous gastric carcinoma; NMGC = non-MGC.

matching, compared with the patients with NMGC, the patients with MGC had more poorly differentiated tumors, more medullary tumors, more infiltrating tumors, more diffuse-type tumor, fewer lymphoid stroma, more abnormal preoperative CA19-9 levels, and more advanced T categories.

### 3.2. Initial recurrence patterns

Among the 2637 patients, 699 (26.5%) had recurrence of GC during a median follow-up time of 65.3 months. As shown in Table 2, after propensity score matching, compared with those with NMGC, there was no significant difference in the initial recurrence pattern between patients with MGC and patients with NMGC.

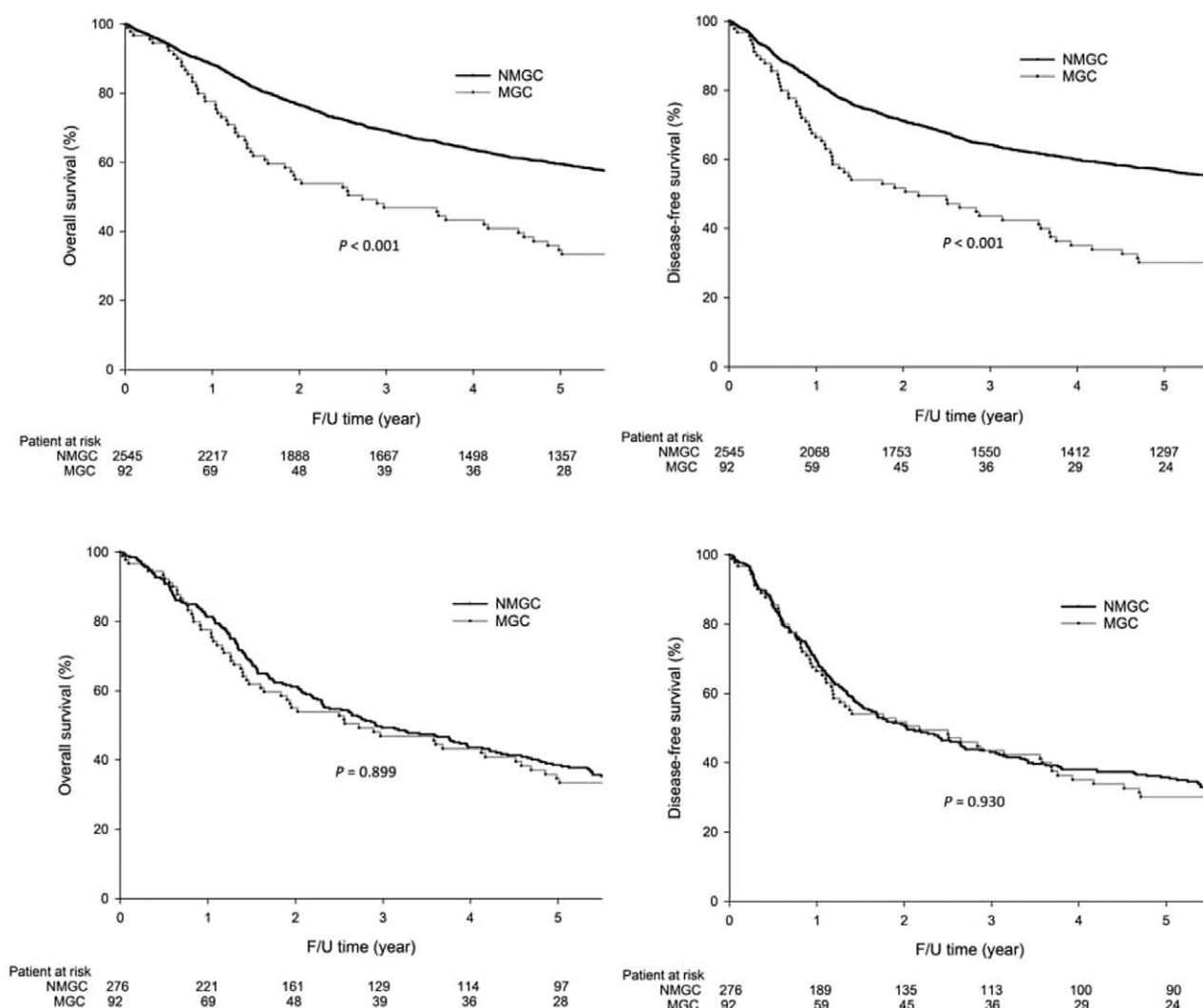
### 3.3. Survival analysis

Among the 2637 patients, the 5-year OS rates (34.6% vs 59.5%,  $p < 0.001$ , Fig. 1A) and DFS rates (30.1% vs 56.8%,  $p < 0.001$ , Fig. 1B) were significantly worse for the MGC patients than those for the NMGC patients. After propensity score matching,

the 5-year OS rates (34.6% vs 38.6%,  $p = 0.899$ , Fig. 1C) and DFS rates (30.1% vs 57.4%,  $p = 0.930$ , Fig. 1D) were not significantly different between MGC and NMGC patients.

As shown in Table 3, multivariate analysis demonstrated that age, gender, tumor size, gross appearance, lymphovascular invasion, pathological TNM stage, and preoperative serum CEA levels were independent prognostic factors of OS. Multivariate analysis of the factors affecting DFS demonstrated that age, gender, tumor size, tumor location, gross appearance, lymphovascular invasion, pathological TNM stage, and preoperative serum CEA levels were independent prognostic factors. The MGC cell type is not an independent prognostic factor for OS or DFS.

After propensity score matching (Table 4), multivariate analysis demonstrated that age, gross appearance, and pathological TNM stage were independent prognostic factors of OS. For DFS, multivariate analysis demonstrated that age, gross appearance, and pathological TNM stage were independent prognostic factors.



**Fig. 1** The 5-year OS rates (34.6% vs 59.5%,  $p < 0.001$ ) and DFS rates (30.1% vs 56.8%,  $p < 0.001$ ) were significantly worse in patients with MGC than in those with NMGC. After propensity score matching, the 5-year OS rates (34.6% vs 38.6%,  $p = 0.899$ ) and DFS rates (30.1% vs 57.4%,  $p = 0.930$ ) were not significantly different between MGC and NMGC patients. The survival curves are shown as follows: (A) OS curves of patients with MGC and NMGC; (B) DFS curves of patients with MGC and NMGC; (C) OS curves of patients with MGC and NMGC after propensity score matching; (D) DFS curves of patients with MGC and NMGC after propensity score matching. DFS = disease-free survival; MGC, mucinous gastric carcinoma; NMGC, non-MGC; OS, overall survival.

**Table 3**  
Multivariate analysis of factors affecting OS and DFS of GC patients after curative surgery

	OS			DFS		
	HR	95% CI	P	HR	95% CI	p
Age (years old)	1.80	1.592–2.027	<0.001	1.69	1.503–1.903	<0.001
Gender	0.81	0.707–0.924	0.002	0.79	0.695–0.905	0.001
Tumor size (cm)	1.20	1.063–1.348	0.003	1.18	1.051–1.327	0.005
Tumor location	1.69	0.920–1.049	0.590	1.14	0.942–1.072	0.003
Gross appearance	1.15	1.068–1.242	<0.001	1.16	1.073–1.246	<0.001
Cell differentiation	1.77	0.883–1.070	0.972	1.02	0.888–1.074	0.621
Lymphovascular invasion	1.31	1.143–1.493	<0.001	1.31	1.151–1.495	<0.001
Adjuvant chemotherapy	0.84	0.698–1.001	0.051	0.93	0.785–1.105	0.413
Pathological TNM stage	1.74	1.592–1.899	<0.001	1.75	1.605–1.908	<0.001
Preoperative serum CEA (ng/mL)	1.34	1.177–1.525	<0.001	1.44	1.271–1.640	<0.001
Preoperative serum CA19-9 (U/mL)	1.11	0.959–1.281	0.196	1.08	0.937–1.250	0.283
MGC cell type	1.01	0.768–1.273	0.928	1.07	0.785–1.105	0.726

CA19-9 = carbohydrate antigen 19-9; CEA = carcinoembryonic antigen; DFS = disease-free survival; GC = gastric cancer; HR = hazard ratio; MGC = mucinous gastric carcinoma; OS, overall survival.

**Table 4**  
Multivariate analysis of factors affecting OS and DFS of GC patients after curative surgery with propensity score matching

	OS			DFS		
	HR	95% CI	p	HR	95% CI	p
Age (years old)	1.73	1.310–2.283	<0.001	1.55	1.184–2.040	0.002
Gender	0.90	0.641–1.268	0.551	0.94	0.670–1.307	0.936
Tumor size (cm)	1.09	0.697–1.707	0.703	1.15	0.737–1.788	0.542
Tumor location	0.89	0.762–1.037	0.135	0.90	0.773–1.043	0.158
Gross appearance	1.34	1.086–1.651	0.006	1.35	1.096–1.661	0.005
Cell differentiation	1.05	0.817–1.358	0.690	1.05	0.821–1.348	0.690
Lymphovascular invasion	1.32	0.861–2.013	0.204	1.16	0.760–1.767	0.494
Adjuvant chemotherapy	0.72	0.474–1.084	0.115	0.83	0.562–1.223	0.344
Pathological TNM stage	2.00	1.574–2.550	<0.001	2.12	1.659–2.700	<0.001
Preoperative serum CEA (ng/mL)	1.08	0.812–1.446	0.587	1.19	0.897–1.579	0.228
Preoperative serum CA19-9 (U/mL)	1.25	0.936–1.681	0.129	1.16	0.865–1.550	0.324
MGC cell type	0.97	0.728–1.304	0.859	1.01	0.761–1.347	0.931

CA19-9 = carbohydrate antigen 19-9; CEA = carcinoembryonic antigen; DFS = disease-free survival; GC = gastric cancer; HR = hazard ratio; MGC = mucinous gastric carcinoma; OS, overall survival.

### 3.4. Analysis of genetic alterations

A total of 370 patients with available tissue samples were enrolled in the analysis of genetic alterations (Table 5). Among them, 40 patients had MGC. As shown in Table 5, there were no significant differences in genetic mutations, *PIK3CA* amplifications, MSI status, and HP or EBV infection between MGC and NMGC.

Regarding the IHC stains for the expression of PD-L1 and HER2, the patients with MGC had significantly higher frequencies of PD-L1 expression than those with NMGC (40.0% vs 19.1%,  $p = 0.002$ ), while HER2 expression was slightly lower in MGC patients than that in the NMGC patients (0% vs 6.1%,  $p = 0.109$ ).

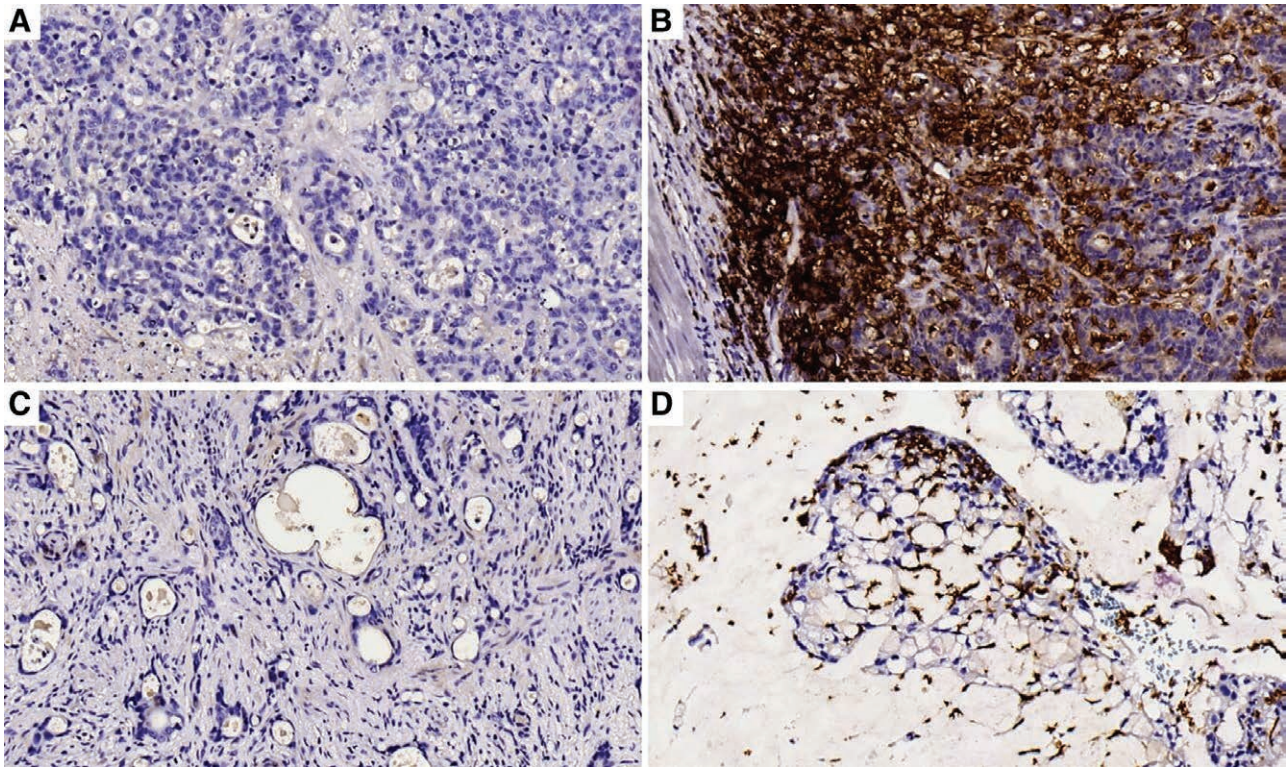
## 4. DISCUSSION

The present study showed that compared with NMGC, MGC is diagnosed at a more advanced stage and is associated with more unfavorable pathological features and worse 5-year OS and DFS rates; however, after propensity score matching, the 5-year OS and DFS rates were not significantly different between MGC and NMGC. Multivariate analysis confirmed that MGC itself is not an independent prognostic factor for OS and DFS, which is consistent with the findings of other studies.<sup>2,3,6</sup> In addition, the expression of PD-L1 was significantly higher in the MGC

**Table 5**  
Comparison of the molecular differences between MGC and NMGC

Variables	NMGC n = 330 n (%)	MGC n = 40 n (%)	p
MSI status			0.210
MSI-H	292 (88.5)	36 (90.0)	
MSI-L/S	38 (11.5)	4 (10.0)	
HP infection	82 (24.8)	15 (37.5)	0.086
EBV infection	38 (11.5)	4 (10.0)	0.775
<i>PIK3CA</i> amplification	121 (36.7)	12 (30.0)	0.407
Genetic mutations			
<i>PI3K/AKT</i> pathway	41 (12.4)	6 (15.0)	0.644
<i>TP53</i>	37 (11.2)	2 (5.0)	0.227
<i>ARID1A</i>	31 (9.4)	4 (10.0)	0.902
<i>BRAF</i>	2 (0.6)	0	0.622
PD-L1 expression	63 (19.1)	16 (40.0)	0.002
HER2 expression	20 (6.1)	0	0.109

EBV = Epstein-Barr virus; HER2 = human epidermal growth factor receptor 2; HP = *Helicobacter pylori*; MGC = mucinous gastric carcinoma; MSI = microsatellite instability; MSI-H = MSI-high; MSI-L/S = MSI-low/stable; NMGC = non-MGC; PD-L1 = programmed death-ligand 1.



**Fig. 2** The IHC staining of PD-L1 in GC specimen is shown as follows: (A) negative expression of PD-L1 in NMGC; (B) positive expression of PD-L1 in NMGC; (C) negative expression of PD-L1 in MGC; (D) positive expression of PD-L1 in MGC. GC = gastric cancer; IHC = immunohistochemical; MGC = mucinous gastric carcinoma; NMGC = non-MGC; PD-L1 = programmed death-ligand 1.

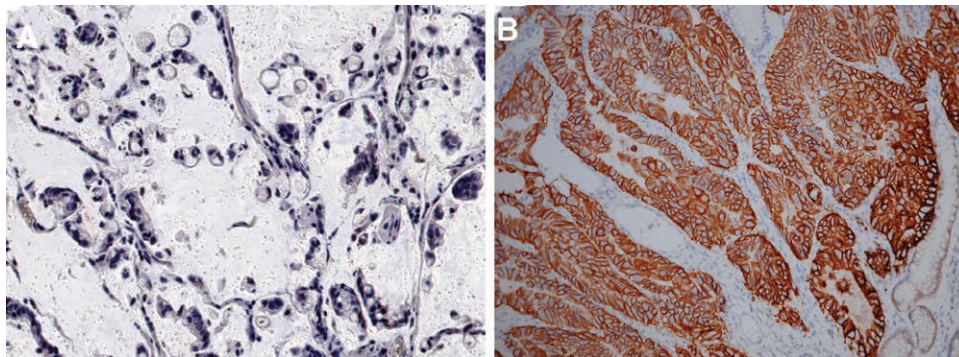
patients than that in the NMGC patients. Therefore, the cause of the worse prognosis in MGC patients vs NMGC patients may be due to the late diagnosis or other confounding factors that were identified as independent prognostic factors.

No reports regarding the genetic alterations in MGC are currently available. We compared the differences in genetic mutations of common GC-related genes between MGC and NMGC. However, we found that there was no significant difference in genetic alterations between MGC and NMGC. Furthermore, MSI status was similar between MGC and NMGC, which is similar with another report.<sup>3</sup> Our results also demonstrated similar frequencies of HP and EBV infection between MGC and NMGC, which has not yet been reported. Therefore, HP or EBV infection and common GC-related genes may not play an important role in the development of GC in MGC compared with

NMGC. Further next-generation sequencing is required in the future to identify novel genes with different expression patterns between MGC and NMGC.

One of our novel findings was a higher frequency of PD-L1 expression in MGC than that in NMGC (40% vs 19.1%,  $p = 0.002$ ). Immunotherapy targeting PD-L1 has been undergone rapid development as oncotherapy for various cancers.<sup>12</sup> For gastrointestinal tract cancers, blocking the ligand PD-L1 is effective in approximately 20% to 40% of patients. GC is one of the malignancies approved by the FDA for the use of PD-L1 blockers. Since MGC is often diagnosed at an advanced stage with a poor outcome, our results may have a clinical impact and provide useful information for the treatment of MGC in the future.

Target therapy with an anti-HER2 monoclonal antibody, such as trastuzumab, has been proven to have a clinical benefit



**Fig. 3** The IHC staining of HER2 in GC specimen is shown as follows: (A) negative expression of HER2 in MGC; (B) positive expression of HER2 in NMGC. GC = gastric cancer; HER2 = human epidermal growth factor receptor 2; IHC = immunohistochemical; MGC = mucinous gastric carcinoma; NMGC = non-MGC.

for HER2-positive GC.<sup>13</sup> Our results demonstrated a slightly lower HER2 expression (0% vs 6.1%,  $p = 0.109$ ) in MGC than that in NMGC, and the frequency was similar with the report of Choi et al (1.5% vs 6.2%,  $p = 0.046$ ).<sup>3</sup> HER2 does not seem to play an important role in the development of MGC.

One of the interesting findings in the present study is that MGC was associated with higher preoperative serum CEA and CA19-9 levels than NMGC. After propensity score matching, MGC was associated with higher preoperative CA19-9 levels than NMGC. A more mucinous component was reported to be associated with greater positivity for CEA and CA19-9 in GC,<sup>14</sup> which may be the reason why the preoperative serum CEA and CA19-9 levels were higher in MGC than those in NMGC in the present study. Another reason may be that MGC was diagnosed at a more advanced stage than NMGC, which would lead to higher preoperative serum CEA and CA19-9 levels in MGC. As shown in Table 3, preoperative serum CEA level rather than preoperative serum CA19-9 level was an independent prognostic factor. However, after propensity score matching, neither preoperative CEA level nor preoperative CA19-9 level was an independent prognostic factor. After propensity score matching, most early-stage NMGC patients were not included in the multivariate analysis of prognostic factors. Elevated preoperative CEA and CA19-9 levels were associated with advanced GC.<sup>15</sup> Consequently, after propensity score matching, most patients had advanced GC and the preoperative serum CEA and CA19-9 levels may become confounding factors in the multivariate analysis of prognostic factors. Our results demonstrated that pathological TNM stage was superior to preoperative CEA and CA19-9 levels as a significant prognostic predictor of OS and DFS.

Some limitations exist in the present study. First, this was a retrospective study and there might be a selection bias. The present study enrolled a large population and investigated the clinicopathological characteristics, recurrence patterns, and genetic alterations associated with MGC and NMGC. More patients of different races and from different countries are needed to validate our results.

In conclusion, our results demonstrated that MGC was often diagnosed at a more advanced stage and a worse prognosis than NMGC. The expression of PD-L1 was significantly higher in MGC than that in NMGC, and immunotherapy may be applicable for the treatment of MGC in the future.

## ACKNOWLEDGMENTS

This study was supported by the grants from the Ministry of Science and Technology, Taiwan (107-2314-B-075-007). The

funding sources had no role in the study design, data analysis, writing, and submission of the manuscript.

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