Gut and Liver
https://doi.org/10.5009/gnl220491
pISSN 1976-2283 eISSN 2005-1212

Original Article

Analysis of Characteristics and Risk Factors of Patients with Single Gastric Cancer and Synchronous Multiple Gastric Cancer among 14,603 Patients

Du Hyun Song[1], Nayoung Kim[1,2], Hyeong Ho Jo[1,3], Sangbin Kim[1], Yonghoon Choi[1], Hyeon Jeong Oh[4], Hye Seung Lee[5], Hyuk Yoon[1], Cheol Min Shin[1], Young Soo Park[1], Dong Ho Lee[1,2], So Hyun Kang[6], Young Suk Park[6], Sang-Hoon Ahn[6], Yun-Suhk Suh[6], Do Joong Park[6,7], Hyung Ho Kim[6,7], Ji-Won Kim[1], Jin Won Kim[1], Keun-Wook Lee[1,2], Won Chang[8], Ji Hoon Park[8], Yoon Jin Lee[8], Kyoung Ho Lee[8,9], Young Hoon Kim[8,9], Soyeon Ahn[10], Young-Joon Surh[11,12]

[1]Department of Internal Medicine, Seoul National University Bundang Hospital, Seongnam, Korea; [2]Department of Internal Medicine, Seoul National University College of Medicine, Seoul, Korea; [3]Department of Internal Medicine, Daegu Catholic University School of Medicine, Daegu, Korea; [4]Department of Pathology, Seoul National University Bundang Hospital, Seongnam, Korea; [5]Department of Pathology, Seoul National University College of Medicine, Seoul, Korea; [6]Department of Surgery, Seoul National University Bundang Hospital, Seongnam, Korea; [7]Department of Surgery, Seoul National University College of Medicine, Seoul, Korea; [8]Department of Radiology, Seoul National University Bundang Hospital, Seongnam, Korea; [9]Department of Radiology, Seoul National University College of Medicine, Seoul, Korea; [10]Division of Statistics, Medical Research Collaborating Center, Seoul National University Bundang Hospital, Seongnam, Korea; [11]Department of Molecular Medicine and Biopharmaceutical Sciences, Graduate School of Convergence Science and Technology, Seoul National University, Seoul, Korea; [12]Cancer Research Institute, Seoul National University, Seoul, Korea

Article Info

Received November 17, 2022
Revised February 2, 2023
Accepted February 20, 2023
Published online March 29, 2023

Corresponding Author

Nayoung Kim
ORCID https://orcid.org/0000-0002-9397-0406
E-mail nakim49@snu.ac.kr

Background/Aims: Synchronous multiple gastric cancer (SMGC) accounts for approximately 6% to 14% of gastric cancer (GC) cases. This study aimed to identify risk factors for SMGC.

Methods: A total of 14,603 patients diagnosed with GC were prospectively enrolled. Data including age, sex, body mass index, smoking, alcohol consumption, family history, p53 expression, microsatellite instability, cancer classification, lymph node metastasis, and treatment were collected. Risk factors were analyzed using logistic regression analysis between a single GC and SMGC.

Results: The incidence of SMGC was 4.04%, and that of early GC (EGC) and advanced GC (AGC) was 5.43% and 3.11%, respectively. Patients with SMGC were older (65.33 years vs 61.75 years, p<0.001) and more likely to be male. Lymph node metastasis was found in 27% of patients with SMGC and 32% of patients with single GC. Multivariate analysis showed that SMGC was associated with sex (male odds ratio [OR], 1.669; 95% confidence interval [CI], 1.223 to 2.278; p=0.001), age (≥65 years OR, 1.532; 95% CI, 1.169 to 2.008; p=0.002), and EGC (OR, 1.929; 95% CI, 1.432 to 2.600; p<0.001). Survival rates were affected by Lauren classification, sex, tumor size, cancer type, distant metastasis, and venous invasion but were not related to the number of GCs. However, the survival rate of AGC with SMGC was very high.

Conclusions: SMGC had unique characteristics such as male sex, older age, and EGC, and the survival rate of AGC, in which the intestinal type was much more frequent, was very good (Trial registration number: NCT04973631). **(Gut Liver 2024;18:231-244)**

Key Words: Stomach neoplasms; Multiple primary; Risk factors; Sex

INTRODUCTION

Although the mortality rate is decreasing, the incidence of gastric cancer (GC) is very high in Korea.[1] Therefore, the government recommends performing esophagogastroduodenoscopy every 2 years as a screening test for adults over 40 years of age. Due to the national screening system and improvement in diagnostic ability in esophagogastro-

duodenoscopy, it has become possible to diagnose cancer at an early stage, which has increased rapidly from 54.0% in 2003 to 2007, 63.5% in 2008 to 2012, and 81.0% in 2013 to 2018 in one Korean report.[2] The intestinal type is known to have a better prognosis than the diffuse type GC when classified according to the Lauren classification.[3]

Synchronous multiple GC (SMGC) is a disease in which two or more cancer lesions exist simultaneously and each lesion must exist as a separate lesion regardless of metastasis. Previous studies have shown that the proportion of SMGCs in patients with GC accounts for approximately 6% to 14% of GC cases.[4,5] SMGC is also known to be associated with advanced age, well-differentiated lesion, early stage, microsatellite instability (MSI), and p53 mutations.[6-8] The occurrence of SMGC might be associated with the tumor microenvironment (TME), which consists of cancer cells and various other components, including infiltrating immune cells, blood vessels, signaling molecules, and extracellular matrix proteins.[9] TME is related to chronic inflammation and *Helicobacter pylori* infection of the gastric epithelium is a major risk factor for GC.[10] *H. pylori*-associated gastritis could promote TME including destruction of tight junction protein.[10] This is supported by the beneficial effects of *H. pylori* eradication in reducing the risk of primary GC incidence[11,12] and prevention effect of metachronous GC with a hazard ratio (HR) of 0.32 (95% confidence interval [CI], 0.15 to 0.66; p=0.002).[13] Therefore, we hypothesized that the risk factors and survival rates of SMGC are different from those of single GC. This study aimed to identify the characteristics of single and multiple GC and to determine the risk factors for SMGC and survival rate for GC and SMGC according to the Lauren classification.

MATERIALS AND METHODS

1. Study population

A total of 14,598 patients diagnosed with GC between May 2003 and February 2020 at the Seoul National University Bundang Hospital were analyzed. Data were prospectively collected from surgical cohort and medical GC cohort of the Seoul National University Bundang Hospital from 2003. Clinical data warehouses and electronic medical records (EMRs) were also reviewed. Age, sex, primary cancer number, height, weight, body mass index (BMI), smoking, alcohol consumption, family history, p53, MSI, Epstein-Barr virus (EBV), cancer classification, distant metastasis, lymph node metastasis, lymphatic invasion, venous invasion, treatment, histology and tumor diameter were acquired from medical and surgical cohort and EMRs. Age, height, weight and BMI were confirmed at the time of GC diagnosis, and the location, size, number and histology of GC were confirmed by the pathological results after surgery or endoscopy. Alcohol consumption, smoking, and family history of GC were assessed using questionnaire from the medical or surgical GC cohort and by EMR. Among patients with SMGC, those with residual cancer after endoscopic treatment and underwent surgery were excluded. Regular follow-up endoscopy was performed after endoscopic treatment and surgery. Most patients were generally referred to local hospitals after more than 5 years of follow-up. The dates and causes of death of the enrolled patients were cross-reviewed with data from EMR and the National Statistical Office for verification. Random information that guaranteed patient anonymity was compiled and submitted by a third party to the National Statistical Office, and received data related to patient death. This study was reviewed and approved by the Institutional Review Board of the Seoul National University Bundang Hospital (IRB number: B-2006-618-004). This study was performed following the protocols approved by the ethics committee. According to IRB guidelines for unnamed surveys, written informed consent among patients were not required.

2. Data variable

SMGC has been identified by diagnostic endoscopy and pathology of surgery and endoscopic treatment. Regular follow-up endoscopy was performed to check whether GC was developed or recurred at least 1 year after the endoscopic treatment and surgery to distinguish it from missing GC and recurrence. The age was divided into two groups based on the age of 65 as defined by the Welfare of Senior Citizens Act of Korea. The location was classified into three groups: upper, middle, and lower according to surgical pathological report format. BMI was divided into underweight (<18.5 kg/m^2), normal (18.5 to <25 kg/m^2), overweight (25 to <30 kg/m^2) and obese (≥30 kg/m^2) according to World Health Organization criteria. Histological classification was based on Lauren classification and histology was confirmed after surgery and endoscopic treatment. Single GC was classified as intestinal type, diffuse type, and others. The others included both indeterminate and mixed types. SMGC was classified into all intestinal types, all diffuse types, and others according to Lauren classification of each cancer. The others were mixtures of intestinal and diffuse types. SMGC was classified into major and minor lesions according to tumor size. A major lesion was the lesion with the longest diameter or the deepest depth among the lesions that presents simultaneously. Otherwise, it was defined as a minor lesion. Early GC (EGC) and advanced GC (AGC) were classified according to pathologi-

cal findings after endoscopic treatment and surgery well as imaging tests such as computed tomography excluding the metastasis, and EGC was defined as cancer invasion into the submucosal layer. However, if surgery could not be performed or surgery was refused, especially in old age or in the presence of other serious diseases, classification was based on computed tomography findings as well as endoscopy. Actually, these cases were very few not affecting the results. The gross types of EGC and AGC were evaluated based on the pathological and endoscopic findings. EGC was classified into three types (I: polypoid type, II: superficial type, and III: excavated type) according to the Paris classification and AGC was classified according to the Borrmann classification.[14] Tumor size was based on the pathological findings. Lymphovascular invasion was diagnosed after confirming tumor emboli by staining the lymphatic vessels with D2-40 staining. EBV positive was diagnosed when tumor cells were stained blue by EBV RNA in situ hybridization. When staining of tumor cell nuclei regarding p53 immunohistochemistry was more than 10%, it was determined as p53 positivity.

3. Statistical analysis

Statistical analysis was performed using SPSS software version 22.0 (IBM Corp., Armonk, NY, USA). Baseline characteristics and variables were analyzed using univariate analysis by the chi-square test and logistics regression. Risk factors were evaluated using odds ratio (OR) and 95% CI in multivariate analysis using logistics regression. Survival rates according to Lauren classification were compared using the Kaplan-Meier survival analysis and each p-value was confirmed through the log-rank test and Cox proportional hazards regression. Cox proportional hazards regression was used to adjust for various variables related to survival, and multivariate analyses were performed to determine the HRs. A p-value of <0.05 was considered statistically significant.

RESULTS

1. Baseline characteristics of single GC and SMGC

A total of 14,603 patients were included, with 14,013 patients with a single GC (95.96%) and 590 patients (4.04%) with SMGC. In average, patients with single GC were followed for 3.55 years and patients with SMGC were followed for 3.69 years in our study. For those patients with endoscopic treatment, single GC were followed for 4.16 years, and SMGC were followed for 5.02 years. For those patients with the surgery, single GC were followed for 4.17 years, and SMGC for 3.81 years. Among the 14,603 patients, 7,749 had EGC (53%) and 4,968 had AGC (34%), except for 1,872 (13%) without pathological data (Fig. 1).

Table 1 shows the baseline characteristics of the patients. Patients with SMGC were older than those with single GC (65.33 years vs 61.75 years, p<0.001). The number of elderly patients (≥65 years) was 6,287 (44.9%) in the single GC group and 335 (56.8%) in the SMGC group; 9,267 (66.1%) patients in the single GC group and 451 (76.4%) patients in the SMGC group were males. A total of 518 (87.8%) patients with two cancer lesions were the most common in the SMGC group.

According to the Lauren classification, the intestinal type was 7,428 (59.9%) and the diffuse type was 4,605 (37.1%) in a single GC. The other consisted of mixed type and the indeterminate type was 375 (3.0%) in a single GC. In the SMGC group, all lesions were intestinal type in 365 (64.8%) patients and diffuse type in 77 (13.7%) patients.

In the EGC, there were 7,328 (60.4%) patients in single GC group and 421 (73.1%) patients in the SMGC group. Distant metastases were observed in 1,893 (14.6%) patients with a single GC and 15 (3%) patients with SMGC. Surgery was the most common treatment modality for patients with GC (61.6%) and SMGC (82.2%). In EGC group, during the follow-up period, the endoscopic treatment rate increased from 10% in 2003 to 30% recently, and the surgical treatment rate decreased from 80% to 60%. Overexpression of p53 was observed in 2,964 (34.9%) patients with a single GC and 160 (35.1%) patients with SMGC. EBV positivity was observed in 34 (12.3%) and MSI high in 67 (14.9%) of patients with SMGC.

Among patients with SMGC, the total number of le-

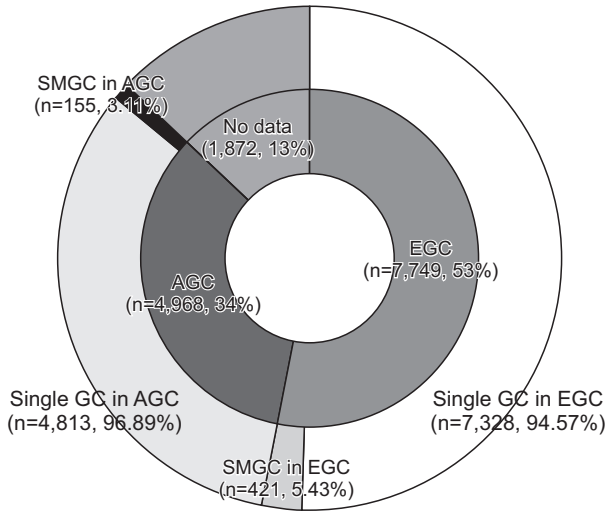

Fig. 1. Cancer classification and distribution of single GC and SMGC. GC, gastric cancer; SMGC, synchronous multiple GC; EGC, early GC; AGC, advanced GC.

Table 1. Baseline Characteristics of Patients with Single GC and with SMGC

Characteristic		Single GC (n=14,013)	SMGC (n=590)	In EGC (n=7,749)		In AGC (n=4,968)	
			Major (n=590)/minor lesion (n=680)*	Single GC (n=7,328)	SMGC (n=421)	Single GC (n=4,813)	SMGC (n=155)
Age, mean±SD, yr		61.75±12.84	65.33±11.21	61.30±11.92	64.85±11.18	62.05±13.51	65.05±11.56
	≤64 yr	7,726 (55.1)	255 (43.2)	4,199 (57.3)	175 (41.6)	2,551 (53.0)	76 (49.0)
	≥65 yr	6,287 (44.9)	335 (56.8)	3,129 (42.7)	246 (58.4)	2,262 (47.0)	79 (51.0)
Sex	Female	4,746 (33.9)	139 (23.6)	2,430 (33.2)	110 (26.1)	1,646 (34.2)	26 (16.8)
	Male	9,267 (66.1)	451 (76.4)	4,898 (66.8)	311 (73.9)	3,167 (65.8)	129 (83.2)
BMI, mean±SD, kg/m^2		23.38±3.43	23.30±3.18	24.08±3.20	24.20±3.12	22.84±3.46	23.30±3.18
Treatment	Operative	8,639 (61.6)	485 (82.2)				
	Endoscopic	2,197 (15.7)	74 (12.5)				
	Chemotherapy	1,435 (10.2)	12 (2.0)				
	Conservative	1,742 (12.4)	19 (3.2)				
No. of lesions	2		518 (87.8)				
	3		63 (10.7)				
	4		5 (0.8)				
	5		1 (0.2)				
	6		2 (0.3)				
	8		1 (0.2)				
Tumor size, mean (range), cm		3.60 (0.1–21.0)	3.09 (0.2–14.5)/1.58 (0–9.5)	2.36 (0.1–14.0)	2.45 (0.2–9.2)	6.05 (0.3–21.0)	4.88 (0.9–14.5)
Gross classification	I			268 (4.0)	16 (3.8)	122 (3.5)	8 (5.5)
	II			6,304 (94.5)	398 (95.2)	489 (14.2)	30 (20.5)
	III			102 (1.5)	4 (1.0)	2,388 (69.2)	103 (70.6)
	IV					450 (13.0)	5 (3.4)
	V					3 (0.1)	
	No data			654	3	1,361	9
Tumor location	Upper	2,525 (19.2)	102 (17.3)/87 (12.8)				
	Middle	3,473 (26.4)	120 (20.3)/157 (23.1)				
	Lower	7,175 (54.5)	367 (62.2)/436 (64.1)				
	Entire		1 (0.2)/0 (0)				
	No data	840					
Lauren type	Intestinal	7,428 (59.9)	365 (64.8)	4,947 (69.0)	292 (70.0)	1,907 (45.1)	73 (50.0)
	Diffuse	4,605 (37.1)	77 (13.7)	2,056 (28.7)	58 (13.9)	2,112 (50.0)	19 (13.0)
	Others	375 (3.0)	121 (21.5)	168 (2.3)	67 (16.1)	207 (4.9)	54 (37.0)
	No data	1,605	27	157	4	587	9
Cancer classification	EGC	7,328 (60.4)	421 (73.1)				
	AGC	4,813 (39.6)	155 (26.9)				
	No data	1,872	14				
Lymph node metastasis	No	7,964 (68.0)	425 (73.0)	6,590 (90.2)	373 (88.8)	1,374 (31.2)	52 (33.5)
	Yes	3,743 (32.0)	157 (27.0)	716 (9.8)	47 (11.2)	3,027 (68.8)	103 (66.5)
	No data	2,306	8	22	1	412	

Table 1. Continued

Characteristic		Single GC (n=14,013)	SMGC (n=590)	In EGC (n=7,749)		In AGC (n=4,968)	
			Major (n=590)/minor lesion (n=680)*	Single GC (n=7,328)	SMGC (n=421)	Single GC (n=4,813)	SMGC (n=155)
Distant metastasis	No	11,052 (85.4)	562 (97.4)	7,291 (99.7)	419 (99.8)	3,761 (78.4)	142 (92.8)
	Yes	1,893 (14.6)	15 (3.0)	21 (0.3)	1 (0.2)	1,037 (21.6)	11 (7.2)
	No data	1,068	13	16	1	15	2
Lymphatic invasion	No	7,068 (68.7)	393 (69.8)	6,006 (88.9)	349 (83.7)	1,048 (30.0)	44 (30.1)
	Yes	3,219 (31.3)	170 (30.2)	752 (11.1)	68 (16.3)	2,447 (70.0)	102 (69.9)
	No data	3,726	27	570	4	1,318	9
Venous invasion	No	9,274 (90.5)	519 (92.2)	6,675 (99.2)	412 (98.8)	2,574 (73.7)	107 (73.3)
	Yes	971 (9.5)	44 (7.8)	51 (0.8)	5 (1.2)	918 (26.3)	39 (26.7)
	No data	3,768	27	602	4	1,321	9
p53	No	5,521 (65.1)	296 (64.9)	3,691 (66.4)	222 (66.1)	1,799 (62.7)	74 (61.7)
	Yes	2,964 (34.9)	160 (35.1)	1,869 (33.6)	114 (33.9)	1,071 (37.3)	46 (38.3)
	No data	5,528	134	1,768	85	1,943	35
MSI	MSS/MSI-L	6,996 (90.5)	382 (85.1)	3,839 (91.5)	260 (83.9)	3,021 (89.1)	122 (87.8)
	MSI-H	731 (9.5)	67 (14.9)	356 (8.5)	50 (16.1)	371 (10.9)	17 (12.2)
	No data	6,286	141	3,133	111	1,421	16
EBV	Negative	3,907 (92.6)	243 (87.7)	2,271 (92.5)	182 (88.8)	1,527 (92.7)	61 (84.7)
	Positive	311 (7.4)	34 (12.3)	183 (7.5)	23 (11.2)	121 (7.3)	11 (15.3)
	No data	9,795	313	4,874	216	3,165	83
Family history	No	3,514 (84.0)	179 (85.2)	2,043 (82.6)	127 (86.4)	1,360 (86.8)	51 (82.3)
	Yes	668 (16.0)	31 (14.8)	429 (17.4)	20 (13.6)	207 (13.2)	11 (17.7)
	No data	9,831	380	4,856	274	3,246	93
Smoker	Never	6,465 (53.8)	251 (44.7)	3,437 (52.6)	185 (46.6)	2,343 (53.6)	62 (40.5)
	Current & past	5,557 (46.2)	310 (55.3)	3,095 (47.4)	212 (53.4)	2,031 (46.4)	91 (59.5)
	No data	1,991	29	796	24	439	2
Drinker	Never	3,876 (54.2)	153 (49.4)	1,971 (50.3)	109 (49.8)	1,483 (57.2)	39 (45.9)
	Current & past	3,277 (45.8)	157 (50.6)	1,948 (49.7)	110 (50.2)	1,111 (42.8)	46 (54.1)
	No data	6,860	280	3,409	202	2,219	70

Data are presented as number (%) unless indicated otherwise.
GC, gastric cancer; SMGC, synchronous multiple GC, EGC, early GC; AGC, advanced GC; BMI, body mass index; MSI, microsatellite instability; MSI-L, MSI low; MSI-H, MSI high; MSS, microsatellite stability; EBV, Epstein-Barr virus.
*Major lesion denotes a lesion with the longest diameter among lesions present at the same time.

sions was 1,270, of which 590 were major, and 680 were minor. The average size of the major lesion was 3.09 cm and that of the minor lesion was 1.58 cm in SMGC. The average size of a single GC was 3.6 cm. The lower third of the tumor location was most common in both major and minor lesions of SMGC. In single GC, intestinal type, lower third, and EGC were the most common.

2. Risk factors of SMGC

Univariate and multivariate analyses were performed using the logistic regression, and the results are summarized in Table 2. In the univariate analysis, age, sex, BMI, tumor size, cancer classification, lymph node metastasis, distant metastasis, MSI, EBV, and smoking were associated with SMGC. Sex, age group, and cancer classification were risk factors according to multivariate analysis. Male patients with SMGC were more common than those with a single GC (OR, 1.669; 95% CI, 1.223 to 2.278; p=0.001). The incidence of SMGC was higher in the elderly (≥65 years) (OR, 1.532; 95% CI, 1.169 to 2.008; p=0.002) and in the EGC (OR, 1.929; 95% CI, 1.432 to 2.600; p<0.001). However, BMI, tumor size, lymph node metastasis, distant metastases, MSI, EBV, and smoking were not significantly associated in the multivariate analysis.

3. Comparison of risk factors in EGC patients with single GC and SMGC

The total number of patients with EGC was 7,749, of which 7,328 had a single GC and 421 had SMGC. In the univariate analysis, sex, age, tumor size, lymphatic invasion, MSI, and smoking were associated with SMGC. According to multivariate analysis, in patients with SMGC compared to patients with single GC, the patients were predominantly male (OR, 1.533; 95% CI, 1.169 to 2.011; p=0.002) and the elderly (≥65 years) was more common (OR, 2.038; 95% CI, 1.599 to 2.599; p<0.001). The incidence of SMGC was lower in tumor size (>2.2 cm) (OR, 0.350; 95% CI, 0.270 to 0.454; p<0.001), and lymphatic invasion was more common (OR, 1.600; 95% CI, 1.169 to 2.190; p=0.003). MSI high patients were more common (OR, 1.520; 95% CI, 1.067 to 2.166; p=0.02).

4. Comparison of risk factors in AGC patients with single GC and SMGC

There were 4,968 patients with AGC (single GC, 4,813; SMGC, 155). Univariate analysis showed that sex, tumor size, distant metastasis, EBV, and smoking were associated with SMGC. According to multivariate analysis, there were significant associations with male sex (OR, 4.711; 95% CI, 1.764 to 12.582; p=0.02), tumor size (>5 cm) (OR, 0.355; 95% CI, 0.179 to 0.705; p=0.003), EBV infection (OR,

Table 2. Univariate and Multivariate Analysis for Patients by Logistic Regression

Factor	Multivariate analysis for single GC and SMGC			Multivariate analysis for single GC and SMGC in EGC			Multivariate analysis for single GC and SMGC in AGC		
	Univariate p-value	Multivariate analysis		Univariate p-value	Multivariate analysis		Univariate p-value	Multivariate analysis	
		OR (95% CI)	p-value		OR (95% CI)	p-value		OR (95% CI)	p-value
Age									
≤64 yr		Reference			Reference				
≥65 yr	<0.001	1.532 (1.169–2.008)	0.002	<0.001	2.038 (1.599–2.599)	<0.001	0.361		
Sex									
Female		Reference			Reference			Reference	
Male	<0.001	1.669 (1.223–2.278)	0.001	0.003	1.533 (1.169–2.011)	0.002	<0.001	4.711 (1.764–12.582)	0.02
Body mass index, kg/m^2									
<18.5 (underweight)		Reference							
18.5 to <25 (normal)	0.002	1.144 (0.521–2.511)	0.737						
25 to <30 (overweight)	<0.001	1.155 (0.518–2.578)	0.725						
≥30 (obesity)	0.004	0.629 (0.193–2.045)	0.441						
Tumor size*									
≤3 cm/≤2.2 cm/≤5 cm					Reference			Reference	
>3 cm/>2.2 cm/>5 cm	0.002	0.780 (0.563–1.080)	0.134	<0.001	0.350 (0.270–0.454)	<0.001	<0.001	0.355 (0.179–0.705)	0.003

Table 2. Continued

Factor	Multivariate analysis for single GC and SMGC			Multivariate analysis for single GC and SMGC in EGC			Multivariate analysis for single GC and SMGC in AGC		
	Univariate p-value	Multivariate analysis		Univariate p-value	Multivariate analysis		Univariate p-value	Multivariate analysis	
		OR (95% CI)	p-value		OR (95% CI)	p-value		OR (95% CI)	p-value
Cancer classification									
AGC		Reference							
EGC	<0.001	1.929 (1.432–2.600)	<0.001						
Gross classification									
I									
II				0.831			0.871		
III				0.462			0.269		
IV							0.002		
Lymph node metastasis									
No									
Yes	0.012	1.099 (0.776–1.556)	0.596	0.353			0.539		
Distant metastasis									
No								Reference	
Yes	<0.001	0.431 (0.058–3.181)	0.409	0.854			<0.001	0	0.998
Lymphatic invasion									
No					Reference				
Yes	0.585			0.001	1.600 (1.169–2.190)	0.003	0.993		
Venous invasion									
No									
Yes	0.189			0.326			0.909		
p53									
No									
Yes	0.946			0.906			0.822		
MSI									
MSS/MSI-L		Reference			Reference				
MSI-H	<0.001	1.369 (0.899–2.084)	0.143	<0.001	1.520 (1.067–2.166)	0.02	0.633		
EBV									
Negative								Reference	
Positive	0.003	1.477 (0.944–2.311)	0.088	0.055			0.016	2.783 (1.136–6.821)	0.025
Family history									
No									
Yes	0.64			0.243			0.306		
Smoker									
Never		Reference			Reference			Reference	
Current & past	<0.001	0.971 (0.694–1.359)	0.864	0.02	1.200 (0.899–1.602)	0.215	0.002	0.468 (0.231–0.949)	0.035
Drinker									
Never								Reference	
Current & past	0.095			0.881			0.04	1.402 (0.702–2.799)	0.338

GC, gastric cancer; SMGC, synchronous multiple GC; EGC, early GC; AGC, advanced GC; OR, odds ratio; CI, confidence interval; MSI, microsatellite instability; MSI-L, MSI low; MSI-H, MSI high; MSS, microsatellite stability; EBV, Epstein-Barr virus.

*Single GC and SMGC/single GC and SMGC in EGC/single GC and SMGC in AGC.

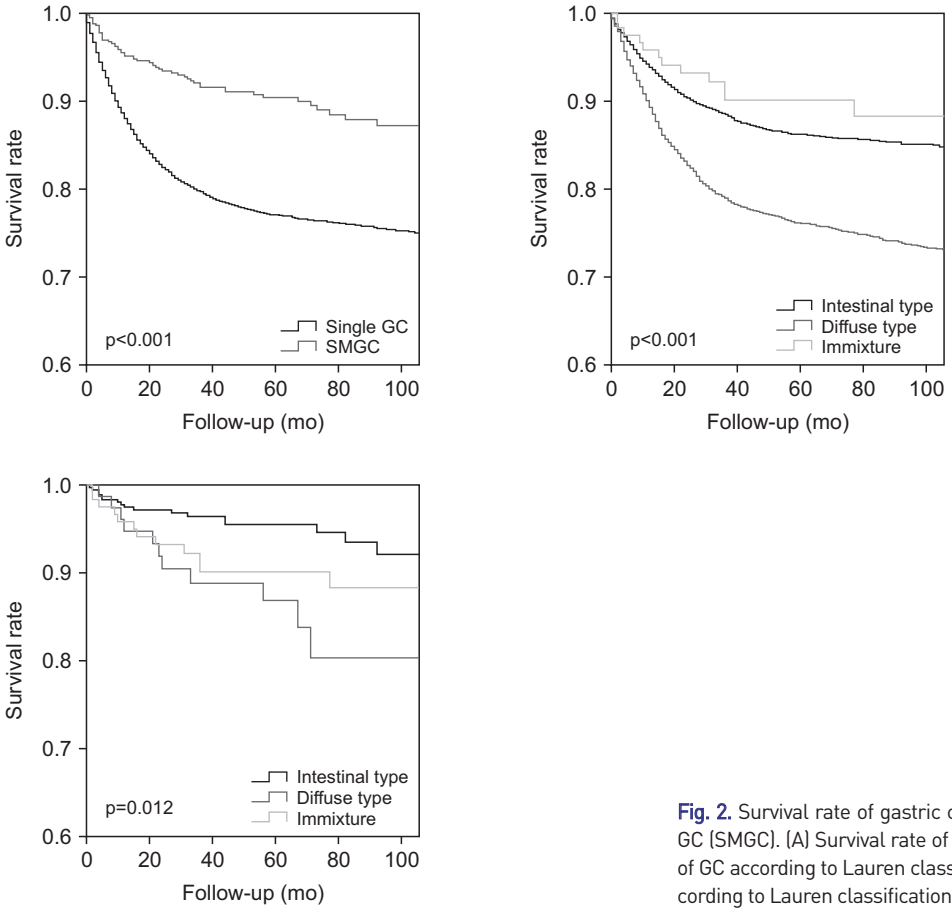

Fig. 2. Survival rate of gastric cancer (GC) and synchronous multiple GC (SMGC). (A) Survival rate of single GC and SMGC. (B) Survival rate of GC according to Lauren classification. (C) Survival rate of SMGC according to Lauren classification.

2.783; 95% CI, 1.136 to 6.821; p=0.025) and smoking (OR, 0.468; 95% CI, 0.231 to 0.949; p=0.035). There was no significant association between age, Borrmann classification, lymphatic invasion, venous invasion, p53, MSI, distant and lymph node metastasis, family history, and alcohol consumption.

5. Survival analysis of patients according to the Lauren classification

According to the Lauren classification, single GCs were divided into intestinal and diffuse types, and SMGCs were classified as all intestinal, all diffuse, and immixture. An immixture is a combination of intestinal and diffuse types. As a result of plotting a Kaplan-Meier graph according to Lauren classification in GC, the survival rate of all intestinal types was higher than that of all diffuse types (Fig. 2).

Cox proportional hazard regression analysis was performed and the results are summarized in Table 3. Age, tumor size, number of primary cancers, cancer type, lymph node metastasis, distant metastasis, lymphatic invasion, venous invasion, p53, MSI, family history, smoking, and alcohol assumption were risk factors in the univariate Cox proportional hazard regression analysis. After adjusting for variables identified by univariate analysis using Cox regression analysis, the risk of all diffuse types was higher than that of all intestinal types (adjusted HR, 1.460; p=0.002). Moreover, when even one intestinal type was included, there were no differences compared to the survival rate of all intestinal types (p=0.412).

6. Risk factor of patients according to the Lauren classification in EGC patients

Fig. 3 shows the result of plotting the Kaplan-Meier graph according to the number of cancers and Lauren classification in EGC. There were no differences in survival rates between the single GC and SMGC groups. There was no difference in survival rate according to the Lauren classification. Multivariate analysis was performed using Cox regression to specifically identify factors that affect the survival rate according to the Lauren classification. The same variables as those of patients with GC were used for the Cox regression analysis. Age, tumor size, lymph node metastasis, lymphatic invasion, and MSI were risk factors in the univariate analysis. After correcting for the variables identified by univariate analysis using Cox regression analysis, as in patients with GC, the risk of all diffuse types was

Table 3. Cox Proportional Hazard Regression for Lauren Classification

Factor	In GC patients			In EGC patients			In AGC patients		
	Univariate p-value	Multivariate		Univariate p-value	Multivariate		Univariate p-value	Multivariate	
		aHR (95% CI)	p-value		aHR (95% CI)	p-value		aHR (95% CI)	p-value
Lauren type									
Intestinal		Reference			Reference			Reference	
Diffuse	<0.001	1.460 (1.143–1.865)	0.002	0.570	1.762 (1.018–3.049)	0.043	<0.001	1.388 (1.063–1.813)	0.016
Immixture	0.286	0.619 (0.196–1.949)	0.412	0.419	1.597 (0.385–6.631)	0.519	0.036	0.442 (0.088–2.234)	0.323
Sex									
Female									
Male	0.32			0.121			0.03	0.786 (0.570–1.084)	0.143
Age	<0.001	1.028 (1.018–1.038)	<0.001	<0.001	1.114 (1.084–1.144)	<0.001	<0.001		
Tumor size	<0.001	1.115 (1.079–1.153)	<0.001	<0.001	0.948 (0.824–1.090)	0.454	<0.001	1.110 (1.073–1.148)	<0.001
No. of primary cancer									
≥2		Reference						Reference	
1	<0.001	0.904 (0.443–1.844)	0.781	0.691			0.001	0.917 (0.403-2.086)	0.837
Cancer type									
EGC		Reference							
AGC	<0.001	4.069 (2.664–6.215)	<0.001						
Lymph node metastasis									
No		Reference			Reference			Reference	
Yes	<0.001	3.407 (2.412–4.813)	<0.001	<0.001	3.364 (2.030–5.574)	<0.001	<0.001	3.455 (2.283–5.230)	<0.001
Distant metastasis									
No		Reference						Reference	
Yes	<0.001	2.584 (1.893–3.529)	<0.001	0.087			<0.001	2.578 (1.885–3.525)	<0.001
Lymphatic invasion									
No		Reference			Reference			Reference	
Yes	<0.001	1.016 (0.753–1.370)	0.919	<0.001	1.579 (0.840 -2.967)	0.156	<0.001	1.018 (0.747–1.387)	0.910
Venous invasion									
No		Reference						Reference	
Yes	<0.001	2.229 (1.743–2.850)	<0.001	0.708			<0.001	2.233 (1.743–2.862)	<0.001
EBV									
Negative									
Positive	0.872			0.479			0.721		
p53									
Negative		Reference							
Over expression	0.003	1.065 (0.849–1.336)	0.585	0.076			0.289		
MSI									
MSI-H		Reference			Reference			Reference	
MSS/MSI-L	0.001	0.734 (0.478–1.128)	0.158	0.020	1.047 (0.533–2.057)	0.895	<0.001	0.720 (0.461–1.123)	0.148

Table 3. Continued

Factor	In GC patients			In EGC patients			In AGC patients		
	Univariate p-value	Multivariate aHR (95% CI)	Multivariate p-value	Univariate p-value	Multivariate aHR (95% CI)	Multivariate p-value	Univariate p-value	Multivariate aHR (95% CI)	Multivariate p-value
Family history									
No		Reference						Reference	
Yes	0.001	0.774 (0.549–1.092)	0.145	0.979			0.018	0.773 (0.537–1.113)	0.166
Smoking									
No		Reference						Reference	
Yes	<0.001	0.951 (0.738–1.225)	0.968	0.088			<0.001	0.974 (0.723–1.311)	0.860
Alcohol									
No		Reference						Reference	
Yes	<0.001	0.951 (0.738–1.225)	0.696	0.857			<0.001	0.921 (0.707–1.200)	0.543

GC, gastric cancer; EGC, early GC; AGC, advanced GC; aHR, adjusted hazard ratio; CI, confidence interval; EBV, Epstein-Barr virus; MSI, microsatellite instability; MSI-L, MSI low; MSI-H, MSI high; MSS, microsatellite stability.

higher than that of all intestinal types (adjusted HR, 1.762; p=0.043). Moreover, when even one intestinal type was included, there were no differences compared to the survival rate of all intestinal types (p=0.519) (Table 3). In the Cox regression, the univariate variable did not show a significant p-value of 0.05 or more as shown in the Kaplan-Meier graph, but a difference occurred when other variables were adjusted.

7. Risk factor of patients according to the Lauren classification in AGC patients

Fig. 3 is the result of plotting the Kaplan-Meier graph according to number of cancer and Lauren classification in AGC. There were differences in survival rates between single GC and SMGC, and survival rates according to the Lauren classification. As a result of correcting for variables identified through univariate analysis using Cox regression analysis, as in patients with GC, the risk of all diffuse types was higher than that of all intestinal types (adjusted HR, 1.388; p=0.016). And when even one intestinal type was included, there was no difference compared with survival rate of all intestinal types (p=0.323) (Table 3).

DISCUSSION

Our study showed that 4.04% of the patients had SMGC (EGC, 5.43% and AGC, 3.11%), and the patients with SMGC were older and more likely to be male. Multivariate analysis showed that SMGC was associated with sex (male OR, 1.669), age (≥65 years OR, 1.532), and EGC (OR, 1.929). Survival rates were affected by Lauren classification, sex, tumor size, cancer type, distant metastasis, and venous invasion but were not related to the number of GCs. However, the survival rate of AGC with SMGC was high.

Previous studies have shown that the proportion of SMGC in patients with GC accounts for approximately 6% to 14% of GC cases,[4,5,7,15] and in the present study, the incidence of SMGC was about 4.04%. The reason for the difference in the SMGC ratio was that most of the existing studies focused on EGC in which the rate of SMGC was higher than that of AGC, and were included only those that were curative status, or only cases that treated by endoscopic treatment or surgery. However, in our study, all data were included regardless of the treatment method and GC progression.[16-18] In the present study, when the rate of SMGC of EGC was calculated, the incidence rate of SMGC was 5.43%, similar to that of the previous study.[6,7,17]

Risk factors for SMGC are known to be associated with the elderly, males, early T stage, differentiated type tumor, p53 mutations, and MSI.[6-8,17,19,20] Our multivariate analysis

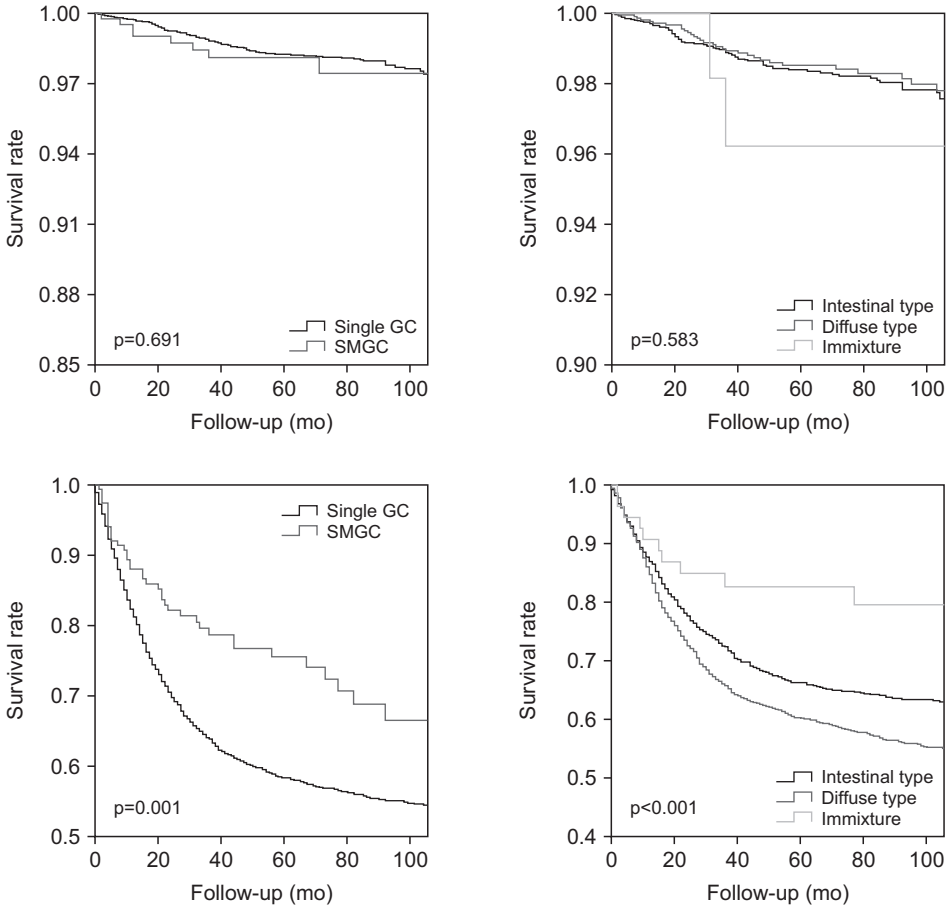

Fig. 3. Survival rate of early gastric cancer (GC) and advanced GC. (A) Survival rate of single GC and synchronous multiple GC (SMGC) in early GC. (B) Survival rate of GC according to Lauren classification in early GC. (C) Survival rate of single GC and SMGC in advanced GC. (D) Survival rate of GC according to Lauren classification in advanced GC.

also showed that older age (≥65 years), males, and EGC were associated with SMGC, similar to previous studies. These associations appear to be related to TME, which causes the intestinal type GC at multifocal sites where atrophy and intestinal metaplasia occur.[21]

In general, tumor multiplicity is related to genetic factors.[19] However, the p53 mutation was not relevant and the MSI lost its significance in multivariate analysis. This difference might be due to the higher incidence of GC in Korea than in the West,[22] meaning that environmental factors were also related. Furthermore, the incidence of MSI in GC varies between East and West.[23] Therefore it might have influenced the association with the incidence of multiple GCs in our study with Koreans.

EBV infection-associated GC (EBVaGC) is more common in young people and males and is located in the upper part of the stomach. According to Lauren classification, diffuse GC was more common in EBVaGC.[24,25] Although EBVaGC rarely has lymphatic metastasis, it is often diagnosed at an advanced stage.[26] In this study, when multivariate analysis was performed by dividing only AGC cases, EBV infection was associated with the risk of SMGC, which is believed to be attributable to the characteristics of EBVaGC.

The gross classification of EGC and AGC was not associated with the risk of multiple GC. In the overall data, tumor size was not associated with the risk of SMGC, but tumor size was associated when EGC and AGC were classified separately. Family history, smoking, and alcohol consumption were factors that increased the risk of GC, but the association with multiple GC was not found. These results were similar to those of previous studies. There were some missing data in the case of not responding to drinking and smoking through the survey in our study, but drinking and smoking did not significantly affect the results. Furthermore, the gross classification of EGC and AGC was not associated with the risk of multiple GC.

In the case of endoscopic treatment, the location of the GC is not important, but in the case of surgery, it becomes an important factor in determining the treatment method (e.g., proximal gastrectomy, distal gastrectomy, total gastrectomy, pylorus-preserving gastrectomy). GC has been known to occur most frequently in the lower third,[27] and when there were multiple lesions, the minor lesions tended to be located adjacent to the major lesions.[6,18] In our study, the major and minor lesions were in the same third in 60%

of the cases and in 90% of the cases, the minor lesions were in the third adjacent to the major lesion. Therefore, when GC is detected, it is important to check the adjacent site during esophagogastroduodenoscopy because it is usually located in the adjacent area of multiple GC, which could prevent further treatment. However, it could occur with a low probability, even in a remote location; therefore, caution is required.

A previous study reported that the survival rates of single GC and SMGC were similar.[21] However, there was a difference in survival rates in this study. To determine the cause of the difference in survival rate, the survival rate was confirmed by dividing EGC and AGC and a survival rate graph was drawn according to the Lauren classification (immixture is a case in which intestinal and diffuse types are mixed in SMGC) This difference was believed to be the result of the integration of the results of the cancer classification (EGC and AGC) and Lauren classification. When survival rates were analyzed in the EGC and AGC groups, there were no differences between single GC and SMGC in EGC (p=0.691); however, a significant difference remained in AGC (p=0.001) (Fig. 3). Considering the factors affecting the survival rate according to the Lauren classification, the number of GC was not related to the survival rate, and the distribution of GC according to the Lauren classification was important. In multiple GCs, all diffuse types were at the highest risk, and there were no differences in survival rate compared with all intestinal types if there was at least one intestinal type. A comprehensive analysis of SMGC, including the tissue type of GC in each AGC/EGC regarding survival rate, has not been reported in a large cohort.

Our study had several limitations. First, although our study had a larger sample size than other studies, it was conducted at a single institution. Second, our study was a prospective observational cohort study, which prospectively collected data from the surgical cohort and the medical cohort, but all data were not filled up consistently. Therefore, there was a limitation that p53, EBV, atrophic gastritis, intestinal metaplasia, *H. pylori* and its eradication treatment which are related with GC,[28] were not performed consistently.[29,30] Although the data are not clean in comparison to complete prospective design, we tried to minimize selection bias during the very long period. In addition, 13% of cases lacked pathological dates mainly because surgery could not be performed or surgery was refused, especially in the old age or in the presence of other serious diseases. Despite these limitations, our study had three strengths. First, the sample size was relatively large. Second, all GCs, including AGC and EGC, were analyzed. Third, in a previous study, only the prognoses of a single GC and multiple GCs were compared. In contrast, our study provided new information by comparing the prognosis according to histology using the Lauren classification.

In conclusion, SMGC had unique characteristics such as male sex, older age, and EGC, and the survival rate was very good in cases of AGC in which the intestinal type was much more frequent.

CONFLICTS OF INTEREST

No potential conflict of interest relevant to this article was reported.

ACKNOWLEDGEMENTS

This work was supported by grant no 13-2019-001 from the Seoul National University Bundang Hospital (SNUBH) Research fund. The authors thank the Division of Statistics in Medical Research Collaborating Center at SNUBH for statistical analysis.

AUTHOR CONTRIBUTIONS

Study concept and design: N.K. Data acquisition: H.H.J., S.K., Y.C., Y.J.S., H.J.O., H.S.L., H.Y., C.M.S., Y.S.P., D.H.L., S.H.K., Y.S.P., S.H.A., Y.S.S., D.J.P., H.H.K., W.C., J.H.P., Y.J.L., K.H.L., Y.H.K., Ji-Won Kim, Jin Won Kim, K.W.L. Data analysis and interpretation: D.H.S. Drafting of the manuscript: D.H.S. Critical revision of the manuscript for important intellectual content: D.H.S., N.K. Statistical analysis: D.H.S., S.A. Approval of final manuscript: all authors.

ORCID

Du Hyun Song https://orcid.org/0000-0002-2903-0786
Nayoung Kim https://orcid.org/0000-0002-9397-0406
Hyeong Ho Jo https://orcid.org/0000-0002-4950-5435
Sangbin Kim https://orcid.org/0000-0002-6884-9516
Yonghoon Choi https://orcid.org/0000-0002-1331-969X
Hyeon Jeong Oh https://orcid.org/0000-0002-9998-3988
Hye Seung Lee https://orcid.org/0000-0002-1667-7986
Hyuk Yoon https://orcid.org/0000-0002-2657-0349
Cheol Min Shin https://orcid.org/0000-0003-2265-9845
Young Soo Park https://orcid.org/0000-0003-1893-7726
Dong Ho Lee https://orcid.org/0000-0002-6376-410X
So Hyun Kang https://orcid.org/0000-0002-8248-9043

Young Suk Park https://orcid.org/0000-0002-6352-9759
Sang-Hoon Ahn https://orcid.org/0000-0001-8827-3625
Yun-Suhk Suh https://orcid.org/0000-0003-3319-8482
Do Joong Park https://orcid.org/0000-0001-9644-6127
Hyung Ho Kim https://orcid.org/0000-0002-8916-0048
Ji-Won Kim https://orcid.org/0000-0001-6426-9074
Jin Won Kim https://orcid.org/0000-0002-1357-7015
Keun-Wook Lee https://orcid.org/0000-0002-8491-703X
Won Chang https://orcid.org/0000-0001-7367-9841
Ji Hoon Park https://orcid.org/0000-0002-6794-4909
Yoon Jin Lee https://orcid.org/0000-0002-3572-029X
Kyoung Ho Lee https://orcid.org/0000-0001-6045-765X
Young Hoon Kim https://orcid.org/0000-0001-5554-3828
Soyeon Ahn https://orcid.org/0000-0003-3440-2027
Young-Joon Surh https://orcid.org/0000-0001-8310-1795

REFERENCES

1. Jung KW, Won YJ, Hong S, Kong HJ, Im JS, Seo HG. Prediction of cancer incidence and mortality in Korea, 2021. Cancer Res Treat 2021;53:316-322.
2. Lee JW, Kim N, Kwon YJ, Lee HS. Changes in the clinical and pathological characteristics of gastric cancer during the last 16 years: a study of a single institution in Korea. Korean J Helicobacter Up Gastrointest Res 2019;19:120-126.
3. Petrelli F, Berenato R, Turati L, et al. Prognostic value of diffuse versus intestinal histotype in patients with gastric cancer: a systematic review and meta-analysis. J Gastrointest Oncol 2017;8:148-163.
4. Lee HJ, Lee YJ, Lee JY, et al. Characteristics of synchronous and metachronous multiple gastric tumors after endoscopic submucosal dissection of early gastric neoplasm. Clin Endosc 2018;51:266-273.
5. Isobe T, Hashimoto K, Kizaki J, et al. Characteristics and prognosis of synchronous multiple early gastric cancer. World J Gastroenterol 2013;19:7154-7159.
6. Kim HJ, Lee JH, Lee JS, et al. Clinicopathlogic characteristics of multiple synchronous early gastric cancers. Korean J Med 2007;72:360-367.
7. Kim DH, Kim SM, Choi MG, Sohn TS, Bae JM, Kim S. Multiple primary malignancies in patients with multiple early gastric cancer. J Gastric Cancer 2017;17:154-161.
8. Noguchi Y, Ohta H, Takagi K, et al. Synchronous multiple early gastric carcinoma: a study of 178 cases. World J Surg 1985;9:786-793.
9. Balkwill FR, Capasso M, Hagemann T. The tumor microenvironment at a glance. J Cell Sci 2012;125(Pt 23):5591-5596.
10. Choi S, Kim N, Park JH, Nam RH, Song CH, Lee HS. Effect of Helicobacter pylori infection and its eradication on the expression of tight junction proteins in the gastric epithelium in relation to gastric carcinogenesis. Helicobacter 2022;27:e12929.
11. Kim N. Chemoprevention of gastric cancer by Helicobacter pylori eradication and its underlying mechanism. J Gastroenterol Hepatol 2019;34:1287-1295.
12. Seta T, Takahashi Y, Noguchi Y, et al. Effectiveness of Helicobacter pylori eradication in the prevention of primary gastric cancer in healthy asymptomatic people: a systematic review and meta-analysis comparing risk ratio with risk difference. PLoS One 2017;12:e0183321.
13. Choi IJ, Kook MC, Kim YI, et al. Helicobacter pylori therapy for the prevention of metachronous gastric cancer. N Engl J Med 2018;378:1085-1095.
14. Kim WH, Park CK, Kim YB, et al. A standardized pathology report for gastric cancer. Korean J Pathol 2005;39:106-113.
15. Otsuji E, Kuriu Y, Ichikawa D, Okamoto K, Hagiwara A, Yamagishi H. Clinicopathologic characteristics and prognosis of synchronous multifocal gastric carcinomas. Am J Surg 2005;189:116-119.
16. Kato M, Nishida T, Yamamoto K, et al. Scheduled endoscopic surveillance controls secondary cancer after curative endoscopic resection for early gastric cancer: a multicentre retrospective cohort study by Osaka University ESD study group. Gut 2013;62:1425-1432.
17. Lee IS, Park YS, Kim KC, et al. Multiple synchronous early gastric cancers: high-risk group and proper management. Surg Oncol 2012;21:269-273.
18. Takeshita K, Tani M, Honda T, et al. Treatment of primary multiple early gastric cancer: from the viewpoint of clinicopathologic features. World J Surg 1997;21:832-836.
19. Kang GH, Kim CJ, Kim WH, Kang YK, Kim HO, Kim YI. Genetic evidence for the multicentric origin of synchronous multiple gastric carcinoma. Lab Invest 1997;76:407-417.
20. Eom BW, Lee JH, Choi IJ, et al. Pretreatment risk factors for multiple gastric cancer and missed lesions. J Surg Oncol 2012;105:813-817.
21. Kitamura K, Yamaguchi T, Okamoto K, et al. Clinicopathologic features of synchronous multifocal early gastric cancers. Anticancer Res 1997;17(1B):643-646.
22. Rawla P, Barsouk A. Epidemiology of gastric cancer: global trends, risk factors and prevention. Prz Gastroenterol 2019;14:26-38.
23. Marrelli D, Polom K, Pascale V, et al. Strong prognostic value of microsatellite instability in intestinal type non-cardia gastric cancer. Ann Surg Oncol 2016;23:943-950.
24. Lee BE. Epstein-Barr virus-associated gastric carcinoma. Korean J Helicobacter Up Gastrointest Res 2021;21:22-28.
25. Tavakoli A, Monavari SH, Solaymani Mohammadi F, et al. Association between Epstein-Barr virus infection and gastric cancer: a systematic review and meta-analysis. BMC Cancer 2020;20:493.

26. Sun K, Jia K, Lv H, et al. EBV-positive gastric cancer: current knowledge and future perspectives. Front Oncol 2020;10:583463.
27. Kim DY, Joo JK, Ryu SY, Park YK, Kim YJ, Kim SK. Clinicopathological characteristics of patients with proximal third gastric carcinoma. Acta Chir Belg 2004;104:677-682.
28. Yoon K, Kim N. Significance of Helicobacter pylori eradication on atrophic gastritis and intestinal metaplasia. Korean J Helicobacter Up Gastrointest Res 2020;20:107-116.
29. Jo HH, Kim N, Jang J, et al. Impact of body mass index on survival depending on sex in 14,688 patients with gastric cancer in a tertiary hospital in South Korea. Gut Liver 2023;17:243-258.
30. Jo HH, Kim N, Oh HJ, et al. The clinicopathological features of mixed carcinoma in 7,215 patients with gastric cancer in a tertiary hospital in South Korea. Gut Liver 2023;17:731-740.