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Original Article

Clinicopathological features and prognosis of gastric mixed adenoneuroendocrine carcinoma

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Abstract: Gastric mixed adenoneuroendocrine carcinomas (MANECs) are rare malignant tumors. This study aimed to investigate the clinicopathological features, diagnosis, prognosis, and treatment outcome in gastric MANECs patients. Clinicopathological data and the archived slides of 40 cases of MANEC patients were retrospectively reviewed. Immunohistochemistry (IHC) staining was performed to detect expression of synaptophysin (Syn), chromogranin A (CgA), CD56, CKpan, CK7, CK8/18, carcinoembryonic antigen (CEA), CK5/6, P40 and Ki-67. Hematoxylin and eosin staining demonstrated exocrine and neuroendocrine components, each accounting for at least 30% of the whole lesion. Exocrine components diffusely expressed epithelial markers CKpan, CK7, CK8/18, and CEA and endocrine components widely expressed at least one of the markers Syn, CgA, and CD56. Ki-67 index and mitosis determined the endocrine component grade as G3. Thirty-three of 40 patients were successfully followed up for 3 to 105 months with median survival of 12 months. Survival analysis showed a significant difference in prognosis with regard to patient's age, disease stage, tumor relapse status, and distant metastasis status. In conclusion, patient's age, disease stage, tumor relapse status, and distant metastasis status are important contributors to poor prognosis. Old patients with advanced stage, recurrence, or metastasis to the liver, pancreas or other distant organs show a poor prognosis.

Keywords: Mixed adenoneuroendocrine carcinomas, neuroendocrine tumors, gastric, grading, diagnosis, prognosis

Introduction

Gastroenteropancreatic neuroendocrine tumors (GEP-NETs) represent a heterogeneous disease entity with different clinical, histopathological, and biological features. However, there are no universally accepted standards for the classification and treatment of GEP-NETs. In 2010, the World Health Organization (WHO) classification of digestive system tumors put forward the revised nomenclature and grade classification based on the morphological criteria and assessment of proliferation fraction [1]. Thus, grading based on mitotic and proliferation fraction (Ki-67 index) is important for the diagnosis and management of GEP-NET patients. The tumors were classified as NET G1, NET G2, NEC, and mixed adenoneuroendocrine carcinomas (MANEC). High-grade GEP-NETs were scored as G3, which include both NEC and well differentiated neuroendocrine tumors (WD-NETs) with a high proliferative index of Ki-67 staining and/or mitoses. MANECs are defined as tumors that comprise both exocrine and neuroendocrine components at different ratios, with each component representing no less than 30% of the entire lesion.

MANEC is a rare tumor with poor prognosis. High-grade MANECs are generally more aggressive and patients often are in the late stages when diagnosed. Most previous studies on MANECs have been case reports or involved very few subjects in series studies [2-6]. Therefore, the characteristics of MANECs remain unclear. Furthermore, the two components of MANEC may consist of various combinations of neuroendocrine tumors and adenocarcinomas from low-grade to high-grade, making MANEC highly heterogeneous in morphology and immunophenotype. Thus, an accurate diagnosis is necessary and important for MANECs.

In this study, we collected clinicopathological data of 40 cases with gastric MANEC in China. We performed histological and immunohistological analysis on the biopsies from these patients, and thirty-three patients were successfully followed up. We found that age, disease stage, recurrence, or metastasis to the liver, pancreas or other distant organs are major contributors to poor prognosis, whereas other parameters, such as tumor size, location, arrangement, and component ratio have no significant impact on survival rate.

Patients and methods

Patients

This retrospective study was approved by the Ethics committee of Jinling Hospital (Nanjing, Jiangsu, China). Informed consent was waived by the committee because of the retrospective nature of the study. The present study covered 40 cases with gastric MANEC between January 2006 and December 2016 from the Department of Pathology, Jinling Hospital. Patients with tumor components accounting for no less than 30% and MANEC confirmed by immunohistochemistry (IHC), and without other primary malignancies were included. Patients with a few neuroendocrine cells scattered in conventional adenocarcinoma (AC) and the inverse condition or with focal AC in almost pure NEC were excluded. The pathological tumor-node-metastasis (pTNM) stage was defined according to the American Joint Committee on Cancer (AJCC) Cancer staging Manual for carcinoma of the stomach, seventh edition [7].

IHC staining

IHC staining via the EnVision method was performed on 3-μm thick sections of formalin-fixed, paraffin-embedded tumor tissues. Monoclonal antibodies against CKpan (clone AE1/3), CK7 (clone RN7), CK8/18 (clone 5D3), CK5/6 (clone D5/16B4), Chromogranin A (CgA, clone EP38), Ki-67 (clone MIB-1), and P40 (Polyclonal antibody) were obtained from Abcam (Cambridge, United Kingdom). Monoclonal antibodies against carcinoembryonic antigen (CEA) (clone II-7), synaptophysin (Syn, clone DAK-SYNAP), CD56 (clone 123C3) and secondary antibody (HRP Rabbit/Mouse, K5007) were obtained from DakoCytomation (Glostrup, Denmark).

Grading criteria

The criteria according to the 2010 WHO classification for determining the grade of NETs is as follows: G1: mitotic count, <2/10 HPF and/or ≤2% Ki-67 index; G2: mitotic count, 2~20/10HPF and/or 3~20% Ki-67 index; G3: mitotic count, >20/10 HPF and/or >20% Ki-67 index. The grading system requires mitotic count in at least 50 high-power fields (HPFs) and Ki-67 index using the MIB antibody as a percentage of 500-2000 cells counted in areas of strongest nuclear labelling (hot spots). If a grade differs for mitotic count compared with Ki-67 index, it is suggested that the higher grade be assumed. The AC component was divided into three subtypes according to the 2010 WHO classification: well differentiated (WD) AC, moderately differentiated (MD) AC, and poorly differentiated (PD) AC. The degree of malignancy is PD>MD>WD.

Follow-up

All patients were followed up via telephone or consultation through the archived files. The deadline of follow-up was June 15, 2017. A total of 33 patients were successfully followed up.

Statistical analysis

Data were analyzed using SPSS19.0 (SPSS, Inc., Chicago, IL, USA). Continuous data were examined using t-tests or variance analyses and expressed as mean ± SD, and categorical variables were evaluated using χ^2 or Fisher's exact tests. Univariate survival analysis was performed using the Kaplan-Meier method, and differences in survival curves were determined using log-rank tests. Multivariate survival analysis was performed by Cox multivariate regression. The follow-up period extended from the day of diagnosis to the day of death or last visit for assessment. Results were considered significant for *P*-value <0.05.

Results

Clinical characteristics

Among 40 patients, 37 were male and 3 were female. Patient ages ranged from 44 to 83 years (mean ± SD, 63.8±7.2 years). The diameter of the tumor ranged from 1.1 cm to 11 cm (mean ± SD, 4.4±1.9 cm). The clinical symp-

Table 1. Clinicopathological features and follow-up of 40 cases of gastric MANECs

Clinicopathological factors	No.	follow-up available	No. of events	follow-up (month)	Survival (month)
Patients	40	33	17	3-105	12±12.5
Gender					
Male	37 (92.5%)	30 (90.9%)	15	3-105	15±12.8
Female	3 (7.5%)	3 (9.1%)	2	4-12	12±12.5
Age					
<63.8 years	20 (50%)	15 (45.5%)	4	4-90	21±21
≥63.8 years	20 (50%)	18 (54.5%)	13	3-105	12±11.8
Diameter					
<4.4 cm	23 (57.5%)	20 (60.6%)	11	5-90	15±16.5
≥4.4 cm	17 (42.5%)	13 (39.4%)	6	3-105	12±17
Location					
Cardia	23 (57.5%)	15 (45.5%)	9	3-90	12±13
Body and antrum	17 (42.5%)	18 (54.5%)	8	4-105	17±21.8
Two-component ratio					
NEC:AC > 1	31 (77.5%)	25 (75.8)	13	3-90	12±11
NEC:AC < 1	9 (22.5%)	8 (24.2%)	4	5-105	16.5±49.5
Tumor arrangement					
Collision type	14 (35%)	12 (36.4%)	6	3-64	15.5±20.5
Crossing type	26 (65%)	21 (63.6%)	11	4-105	12±12.5
Grading of endocrine component					
G1	0				
G2	0				
G3	40 (100%)	33 (100%)	17	3-105	12±12.5
Grading of exocrine component					
WD	6 (15%)	5 (15.2%)	2	12-31	15±12
MD	24 (60%)	19 (57.7%)	10	3-105	12±16
PD	10 (25%)	9 (27.3%)	5	5-90	12±17
pTNM					
I+II	14 (35%)	10 (30.3%)	2	4-105	19.5±26
III+IV	26 (65%)	23 (69.7%)	15	3-90	12±12
Lymph nodes metastasis					
No	9 (22.5%)	5 (15.2%)	2	4-55	19±38.6
Yes	31 (77.5%)	28 (84.8%)	15	3-105	12±11.3
Recurrence					
No	21 (63.6%)	21 (63.6%)	6	3-105	21±39
Yes	12 (36.4%)	12 (36.4%)	11	9-19	12±1.5
Distant metastasis					
No	14 (42.4%)	14 (42.4%)	1	3-105	21.5±45
Yes	19 (57.6%)	19 (57.6%)	16	5-64	12±10
Chemotherapy					
No	8 (20%)	5 (15.2%)	3	5-105	20±59.5
Yes	32 (80%)	28 (84.8%)	14	3-90	12±12.5

toms of gastric MANEC resembled those of general AC, including abdominal discomfort, distension or pain or obstruction while eating. Some patients had hematemesis or black stool. The details of clinicopathological parameters are listed in **Table 1**.

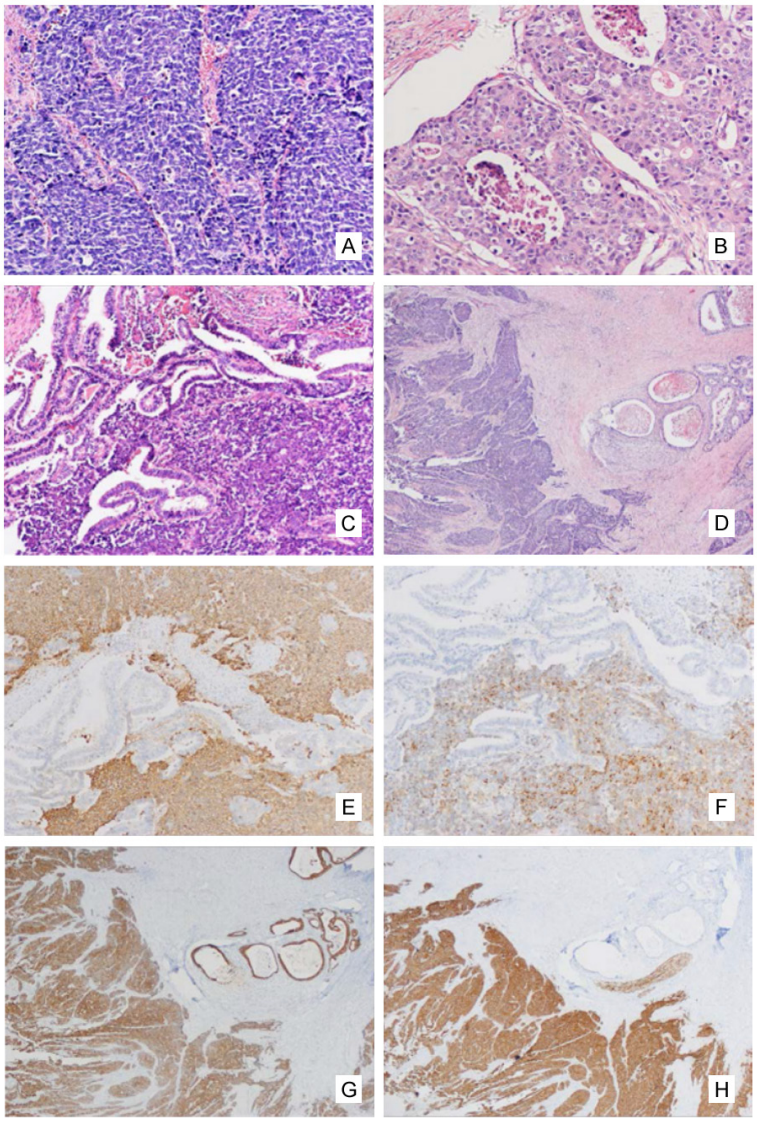

Figure 1. Histological aspects of gastric MANECs. A. SCC demonstrated fusiform nuclei lacking nucleoli with active mitosis. HE×200. B. LCC with geographic necrosis, expansile nests with peripheral palisading, and tubular structures within the large nests. HE×200. C. SCC mixed with AC without boundary. HE×100. D. SCC neighboring AC with a clear boundary. HE×40. E. SCC CgA (2+), AC CgA (-). IHC×100. F. SCC Syn (3+), AC Syn (-). IHC×100. G. Clear boundary between SCC and AC; both are CKpan (3+). IHC×100. H. Clear boundary between SCC and AC. SCC CD56 (3+), AC Syn (-). IHC×100.

Gross inspection

Among 40 gastric cancer radical resection specimens, 36 tumors were ulcerative type, 2 were invasive type, 1 was protruding type, and 1 was superficial erosion type. Sections of these tumors appeared gray in color with invasive boundary, and the tissues appeared brittle.

Histological features

All tumors were composed of PD areas (**Figure 1**) and typical AC. The former could be distinguished as subtypes of small cell and large cell. The cells of small cell type were spindle or fusiform shaped (**Figure 1A**), with minimal cytoplasm, finely granular, and active mitosis arranged in solid nests with necrosis. In three cases, cells were similar to lymphocytes. The cells of large cell type exhibited large cells with abundant cytoplasm, with coarse, granular nuclei arranged in a large expansile growth pattern, with subtle peripheral nuclear palisading and tubule-forming structures within the large nests (**Figure 1B**), and geographic necrosis remained obvious. The exocrine components were composed of three subtypes including: WD papillary AC and tubular AC, with well-formed glands; MD AC, with neoplasms that are intermediate between WD and PD; PD AC, with highly irregular glands that are difficult to be recognized, including solid-type AC, poorly cohesive carcinoma and signet-ring cell carcinoma. Regarding the tumor cell arrangement, 26 cases exhibited crossings without a clear boundary (**Figure 1C**), while the other 14 cases exhibited neighboring arrangements (collision tumor, **Figure 1D**). Thirty one cases had larger proportion of PD areas (endocrine: exocrine >1), and 9 had a preponderance of AC (endocrine: exocrine <1).

IHC showed that PD components diffusely expressed at least one of the following markers: Syn (**Figure 1E**), CgA (**Figure 1F**), and CD56 (**Figure 1H**), which are the most commonly used neuroendocrine markers, with positive rates of

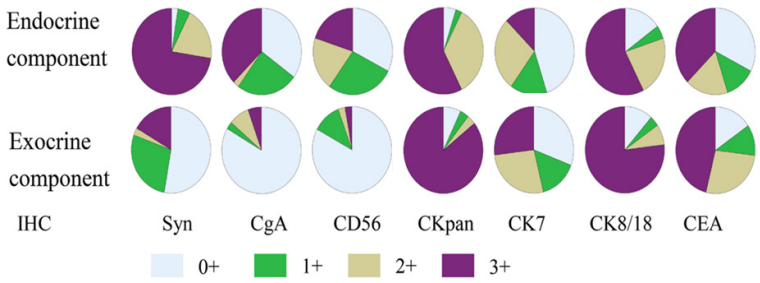

Figure 2. Positive rate of both endocrine and exocrine makers stained in 40 gastric MANECs. Endocrine components: the positive rate of Syn, CgA, and CD56 were 97.5%, 65%, 67.5%, respectively and the positive rate of CKpan, CK8/18, CK7 and CEA were 95%, 55%, 85%, 67.5%, respectively. Exocrine components: the positive rate of CKpan, CK8/18, CK7 and CEA were 92.9%, 69.2%, 88.5%, and 84.6%, respectively, and the positive rate of Syn, CgA and CD56 were 47.2%, 16.7%, and 17.1%, respectively.

97.5%, 65%, and 67.5%, respectively. AC components widely expressed epithelial or exocrine markers. The positive rates of CKpan (**Figure 1G**), CK8/18, CK7 and CEA were 92.9%, 69.2%, 88.5%, and 84.6%, respectively. The positive rates of Syn, CgA, and CD56 in AC were 47.2%, 16.7%, and 17.1%, respectively. The positive rate of the makers in both components is presented in **Figure 2**. CK5/6 and P40 were negative in all cases, which help distinguish from squamous carcinoma. Furthermore, the Ki-67 proliferation index in PD components ranged from 30% to 90%, which confirmed the G3 criteria.

Pathology diagnosis

According to H&E and IHC staining and grading criteria, all cases were diagnosed with MANEC and the exocrine components of AC were graded as WD, MD, and PD. The neuroendocrine components were PD NEC (PD-NEC), and the grading of G3 was characterized by Ki-67 index >20% and mitosis >20/10 HPF.

Survival analysis

Thirty-three patients were followed up successfully: 16 were alive and 17 were dead. The follow-up was from 3 to 105 months, with a median survival of 12 months. The details of follow-up data are shown in **Table 1**. The median survival of patients aged <63.8 years was 9 months longer than those aged ≥63.8 years. The median survival was 7.5 months longer in stages I and II than in stages III and IV. Similarly, the median survival of patients without recurrence was 8.5 months longer than the recurrence group, and 9 months longer in the group without distant metastasis than in the distant metastasis group.

Kaplan-Meier analysis showed that prognosis was significantly correlated with patient's age (P=0.014), pTNM stage (P=0.034), recurrence status (P<0.01) and distant metastasis status (P<0.01) (**Figure 3**), but not with patient's gender, tumor diameter, location, two-component ratio, arrangement, lymph nodes metastasis, AC grade, and chemotherapy (P>0.05).

The 1-year survival rate was 56.7% and the 3-year survival rate was 13.8% (**Table 2**). Cox multivariate regression analysis demonstrated metastasis as an independent prognostic factor (p=0.022, OR=11.7, 95% CI: 1.4, 95.9) (**Table 3**).

Treatment

All 40 patients underwent radical surgery. Of the 33 available patients, 28 patients received traditional chemotherapy for 2-6 times and the other 5 did not receive the therapeutic schedule.

Discussion

According to the 2010 WHO classification of digestive system tumors, NETs range as a series of neoplasms from indolent to highly invasive and metastatic tumors. Recent studies show a rising incidence in GEP cases worldwide [8-11]. MANEC is a rare malignant tumor, accounting for a small percentage of NETs and can be found in the gastrointestinal tract [3-6, 12, 13], pancreas [14-16], liver [12], bladder [17, 18] or uterine cervix [19].

The diagnosis of MANEC mainly depends on H&E morphology and IHC staining. Markers such as Syn, CD56, and CgA are commonly used to recognize the neuroendocrine component that is essential for the diagnosis. Tumor cells that diffusely express one or more neuro-

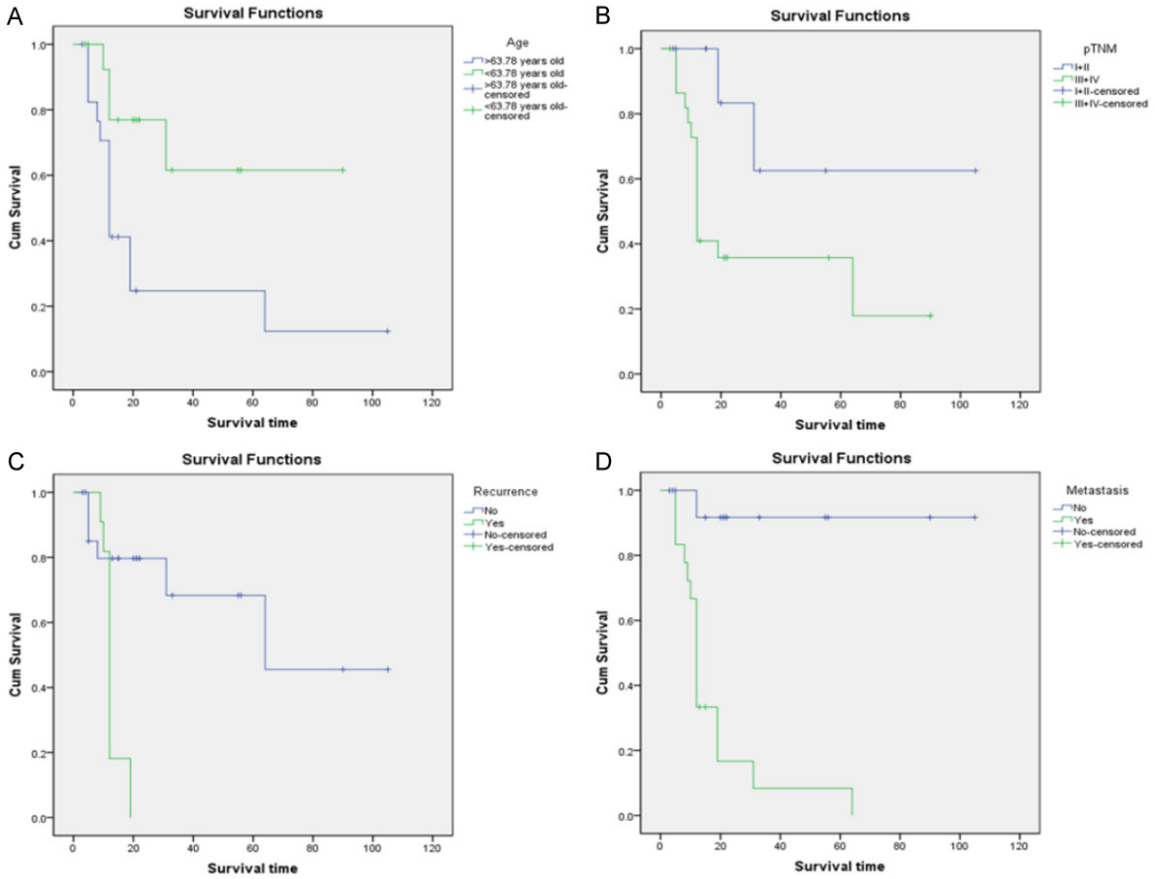

Figure 3. Survival curves of four groups with significant difference in prognosis: (A) age group (χ^2=6.0, P=0.014), (B) pTNM group (χ^2=4.48, P=0.034), (C) recurrence status group (χ^2=15.44, P<0.01) and (D) distant metastasis status group (χ^2=14.93, P<0.01).

endocrine markers can be diagnosed as NET. Conversely, MANEC diagnosis cannot be made if IHC staining showed only a few cells in typical ACs expressing neuroendocrine markers, and hence "adenocarcinoma with neuroendocrine differentiation" should not be recommended. In our study, PD area was composed of small or large cells arranged in solid or nest structure, and three small cell cases even looked like lymphoma. IHC staining showed that the tumor cells widely expressed Syn, CgA, and CD56 markers with a positive rate of 97.5%, 65%, and 67.5%, respectively, confirming the neuroendocrine diagnosis. CK5/6 and P40 were negative markers, excluding the possibility of PD squamous carcinoma. Furthermore, the Ki-67 proliferation index (ranged from 30% to 90%) and a mitotic index >20/10 HPF confirmed G3 criteria of NETs. Therefore, the subtype of neuroendocrine tumors could be divided into small cell carcinoma (SCC) and large cell carcinoma (LCC). The exocrine components that widely expressed epithelial markers determined the diagnosis of AC and the positive rate of CKpan, CK8/18, CK7, and CEA were 92.9%, 69.2%, 88.5%, and 84.6%, respectively. Based on these results, our cases were consistent with the diagnosis criteria of MANEC.

The histogenesis of MANEC has been a controversial issue. There are two main theories: (1) these tumors might arise independently in a synchronous or metachronous manner and later collide with each other; (2) the tumors might be derived from a common multipotent progenitor/stem cell, which then differentiates into different morphologies. However, molecular studies indicated a monoclonal origin of the two components and a possible multistep progression from a common precursor lesion [20-22]. In our study, a large proportion of NEC expressed exocrine markers including CKpan, CK8/18, CK7, and CEA, while some other ACs expressed neuroendocrine markers including

Table 2. Factors affecting survival of gastric MANECs

Clinicopathological factors	Survival rate					
	1-year	χ^2	P	3-year	χ^2	P
Patients	17/30 (56.7%)			4/29 (13.8%)		
Gender						
Male	17/28 (60.7%)	2.802	0.179	4/27 (14.8%)	0.34	0.74
Female	0/3 (0%)			0/2 (0%)		
Age						
<63.8 years	10/13 (76.9%)	3.833	0.055	2/13 (15.4%)	0.05	0.62
≥63.8 years	7/17 (41.2%)			2/16 (12.5%)		
Diameter						
<4.4 cm	6/11 (54.5%)	0.032	0.579	2/19 (10.5%)	0.49	0.43
≥4.4 cm	11/19 (57.9%)			2/10 (20%)		
Location						
Cardia	9/17 (52.9%)	0.22	0.46	1/13 (7.7%)	0.74	0.61
Body and antrum	8/13 (81.5.5%)			3/16 (18.8%)		
Two-component ratio						
NEC:AC>1	12/23 (52.2%)	0.81	0.326	2/23 (8.7%)	2.43	0.18
NEC:AC<1	5/7 (71.4%)			2/6 (33.3%)		
Tumor arrangement						
Collision type	6/10 (60%)	0.07	0.55	1/10 (10%)	0.19	0.57
Crossing type	11/20 (55%)			3/19 (15.8%)		
Grading of endocrine component						
G1						
G2						
G3	17/30 (56.7%)			4/29 (13.8%)		
Grading of exocrine component						
WD	4/5 (80%)	1.66	0.44	0/5 (0%)	1.34	0.51
MD	9/16 (56.3%)			3/15 (20%)		
PD	4/9 (44.4%)			1/9 (11.1%)		
pTNM						
I+II	8/8 (100%)	8.342	0.004	2/8 (25%)	1.17	0.3
III+IV	9/22 (40.9%)			2/21 (9.5%)		
Lymph node metastasis						
No	3/3 (100%)	2.549	0.167	1/3 (33.3%)	1.07	0.37
Yes	14/27 (51.9%)			3/26 (13.8%)		
Recurrence						
No	15/18 (83.3%)	13.0	0.000	4/17 (23.5%)	3.27	0.1
Yes	2/12 (16.7%)			0/12 (0%)		
Distant metastasis						
No	10/11 (90.9%)	8.29	0.005	3/11 (27.3%)	2.7	0.139
Yes	7/19 (36.8%)			1/18 (5.6%)		
Chemotherapy						
No	3/5 (60%)	0.027	0.63	1/5 (20%)	0.196	0.55
Yes	14/25 (56%)			3/24 (12.5%)		

Syn, CgA, and CD56. The expression of markers of each other supports the second theory.

The behavior of gastric MANEC is determined by most of the aggressive components [23-25].

Table 3. Univariate and multivariate analysis of prognostic factors

Clinicopathological factors	Survival (month)	P univariate	P multivariate	Odds ratio (95% CI)
Patients	12±12.5			
Gender				
Male	15±12.8			
Female	8	0.104		
Diameter				
<4.4 cm	15±16.5			
≥4.4 cm	12±17	0.872		
Location				
Cardia	12±13			
Body and antrum	17±21.8	0.819		
Two-component ratio				
NEC:AC>1	12±11			
NEC:AC<1	16.5±49.5	0.769		
Tumor arrangement				
Collision type	15.5±20.5			
Crossing type	12±12.5	0.857		
Grading of endocrine component				
G1				
G2				
G3	12±12.5			
Grading of exocrine component				
WD	15±12			
MD	12±16			
PD	12±17	0.807		
Lymph node metastasis				
No	19±38.6			
Yes	12±11.3	0.864		
Chemotherapy				
No	20±59.5			
Yes	12±12.5	0.912		
Recurrence				
No	21±39			
Yes	12±1.5	P<0.01		
Age				
<63.8 years	21±21			
≥63.8 years	12±11.8	P<0.05	0.458	1.6 (0.5, 5.0)
pTNM				
I+II	19.5±26			
III+IV	12±12	P<0.05	0.243	2.5 (0.5, 11.1)
Distant metastasis				
No	21.5±45			
Yes	12±10	P<0.01	0.022	11.7 (1.4, 95.9)

Patients had a good outcome if the tumors are WD in both the components, even with the occurrence of lymph node metastasis [26]. High grade and a large neuroendocrine component were relevant markers for aggressive behavior and poor clinical outcome. Gastric MANECs with PD AC showed an obviously lower survival rate than MANECs with differentiated

ones [2]. However, in our series, clinical outcome was unlikely to be related to AC grade as the subtype of neuroendocrine component of all cases was PD-NEC (G3), which was more aggressive. The outcome of the group in a large proportion of neuroendocrine components was not worse than AC preponderance group.

Studies on NETs indicate that stage, grade, and age are prognostic factors for overall survival [27]. Kaplan-Meier analysis showed that patients of older age (aged ≥63.8 years) or at stage III or IV had a worse outcome than their counterparts. Cox multivariate regression analysis suggested that metastasis is the independent factor determining the prognosis. NETs tend to metastasize to lymph nodes, the liver, and the bones [28]. The incidence of regional nodal metastasis was high and associated with a survival disadvantage [29]. Although our data showed no significant difference, patients without lymph node metastasis had a 7 months longer survival than those with lymph nodes metastasis, meanwhile, the 1-year survival rate (100%) and the 3-year survival rate (33.3%) in the former group was better than in the latter (51.9%, 13.8%, respectively). Nineteen patients had metastasis, among them 17 had cancer metastasized to the liver, including 3 cases with additional metastasis to the bone, and the other two cases had cancer metastasized to the pancreas and peritoneum. In most of the metastases cases, both components coexisted, while only one component existed in a few cases. Recurrence and metastasis are the two main factors that lead to death. Most of the patients with tumor recurrence or metastasis died within one year.

Everolimus [30] and 177Lu-Octreotate [31] are effective for patients with WD GEP-NETs (WHO G1, WHO G2). Patients with metastases may have a good survival rate after undergoing curative surgery. Long-time follow-up on rectal WHO G1/G2 patients showed that the 5-year overall survival rate was pretty high (98.61%) [32]. In gastric NETs, the outcome of WD-NETs WHO G3 was worse than WHO G1/G2 [33], but much better than PD-NECs. Tang et al. [34] reported that the median disease-specific survival of WD-NETs (WHO G3) was 55 months, and the 2-year and 5-year disease specific survival (DSS) was 88% and 49%, respectively, significantly better than that of PD-NEC. In contrast, survival was much lower in high-grade GEP-NETs. Korse et al. [35] showed that the 5-year survival for Grade 3 large cell and small cells NETs was 20% and 6%, respectively. Fazio et al. reported that the 5-year survival rate of high-grade NETs was less than 10% [36]. However, Mosquera et al. [29] reported that the 5-year survival rate of high-grade GEP-NETs was 63.3%, which was better than other studies. The reason may be that tumors arise from various sites including small bowel, gastric, appendix, colon/rectum, and pancreas and tumors located in small bowel have a better outcome. In our series, the survival was much shorter than G1/G2 tumors that are previously reported and was similar to the small cell/large cell NETs. The median survival was only 12 months, the 1-year overall survival rate was 56.7% and 3-year overall survival rate was only 13.8%, and the tumor sites showed no influence on the outcome. **Table 2** demonstrates that pTNM stage, tumor relapse status, and distant metastasis status significantly influence 1-year survival rate, while patient's age shows no influence. However, no factors affected the 3-year survival rate. The majority of the patients (86.2%) died within 3 years regardless of clinicopathological parameters, perhaps due to that all the cases were high grade (PD-NEC+AC) and most of the patients were in advanced stages without effective therapeutic strategy. Most of the patients received oxaliplatin-based multidrug therapy. Nevertheless, the side effects were often too strong to tolerate, leading to the discontinuation of treatment. Traditional chemotherapy did not improve the outcome in our series.

In conclusion, 40 cases of Chinese patients with gastric MANEC, along with their long-term follow-up were reported in this study. The symptom of gastric MANEC is nonspecific to general gastric malignant tumors and difficult to be diagnosed at early stages. A combination of multiple biomarkers is necessary to make accurate diagnosis and the mutual expression of markers indicates monoclonal origin. The patient's age, pTNM stage, tumor relapse status, and distant metastasis status are the main predictors of prognosis.

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Disclosure of conflict of interest

None.

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