Human Pathology (2017) 63, 128–138

ELSEVIER

Human PATHOLOGY

www.elsevier.com/locate/humpath

Original contribution

Clinical significance of spasmolytic polypeptide-expressing metaplasia and intestinal metaplasia in Epstein-Barr virus–associated and Epstein-Barr virus–negative gastric cancer☆,☆☆

Yu Zhang MD[a,b], Jian-ning Chen MD[a], Min Dong MD[c], Zhi-gang Zhang MD[a], Yi-wang Zhang MD, PhD[a], Jun-yan Wu MD[d], Hong Du MD[e], Hai-gang Li MD[f], Yan Huang MD[g], Chun-kui Shao MD, PhD[a,*]

[a]*Department of Pathology, The Third Affiliated Hospital, Sun Yat-sen University, Guangzhou 510630, China*
[b]*Department of Pathology, Guangdong Provincial Hospital of TCM, Guangzhou University of Chinese Medicine, Guangzhou 510405, China*
[c]*Department of Medical Oncology, The Third Affiliated Hospital, Sun Yat-sen University, Guangzhou 510630, China*
[d]*Zhongshan School of Medicine, Sun Yat-sen University, Guangzhou 510080, China*
[e]*Department of Pathology, Guangzhou First Municipal People's Hospital, Guangzhou Medical University, Guangzhou 510180, China*
[f]*Department of Pathology, Sun Yat-sen Memorial Hospital, Sun Yat-sen University, Guangzhou 510120, China*
[g]*Department of Pathology, The Sixth Affiliated Hospital, Sun Yat-sen University, Guangzhou 510655, China*

Received 14 November 2016; revised 30 January 2017; accepted 23 February 2017

Keywords:
Spasmolytic polypeptide-expressing metaplasia (SPEM);
Intestinal metaplasia (IM);
Epstein-Barr virus;
EBVaGC;
Gastric carcinoma

Summary Spasmolytic polypeptide-expressing metaplasia (SPEM) and intestinal metaplasia (IM) have been recognized as neoplastic precursors in gastric carcinogenesis. We explored the relationship between SPEM and IM in Epstein-Barr virus–associated (EBVaGC) and Epstein-Barr virus–negative (EBVnGC) gastric cancer. Sixty-four EBVaGC and one hundred and fifty-four EBVnGC patients were included. EBV positivity was identified using Epstein-Barr virus–encoded RNA-1 in situ hybridization. SPEM was subclassified into absent, early, and advanced SPEM. Acute and chronic inflammation was graded as absent, mild, moderate, and marked. Univariate and multivariate logistic regression analyses were conducted to analyze the correlation between SPEM, IM, and inflammation. Our study revealed that SPEM was detected in 87.5% EBVaGC and 85.1% EBVnGC patients. Distribution of patients according to the SPEM classification was significantly different between EBVaGC and EBVnGC groups ($P = .038$). IM was observed less frequently in EBVaGC when compared with EBVnGC patients ($P < .001$). No difference was observed between EBVaGC and EBVnGC in the levels of acute and chronic inflammation. A positive correlation between IM and SPEM status was observed in both EBVaGC and EBVnGC patients. Furthermore, advanced SPEM was an independent influential factor to IM in EBVnGC ($P = .013$). In conclusion, SPEM was associated with both EBVaGC and EBVnGC more frequently than IM. Moreover, advanced SPEM

☆ Competing Interests: The authors declare that they have no conflict of interest.

☆☆ Funding/Support: This work was supported by the National Natural Science Foundation of China (Nos. 81572309, 81301694, 81272553, and 81071893), the Guangdong Natural Science Foundation (Nos. 2014A030313034 and S2012020010898), and the Guangzhou Science and Technology Project (No. 2011J4100106), Guangdong Province, China.

* Corresponding author. Department of Pathology, the Third Affiliated Hospital, Sun Yat-sen University, 600 Tianhe Rd, Guangzhou 510630, China.
E-mail address: chunkuishao2011@163.com (C. Shao).

http://dx.doi.org/10.1016/j.humpath.2017.02.016

had a stronger association with IM than early SPEM in EBVnGC. These results suggest that identification of SPEM should be used as a high-risk indicator for detecting early gastric carcinoma, and should be brought to the attention of pathologists and clinicians.

1. Introduction

Although significant advances in gastric cancer treatment have been achieved through endoscopic techniques and new therapeutic approaches, patient prognosis still remains poor. Gastric cancer still represents the third leading cause of cancer-related death worldwide, with a 5-year survival rate of 29.6% [1]. Thus, early detection and treatment of gastric preneoplastic metaplasia is essential in reducing gastric cancer mortality.

Oxyntic atrophy, the loss of acid-secreting parietal cells, along with prominent inflammation, can progress to metaplasia and gastric adenocarcinoma [2,3]. In humans, 2 types of metaplasia can stem from parietal cells impairment: spasmolytic polypeptide-expressing metaplasia (SPEM) and intestinal metaplasia (IM). SPEM, also known as pseudo-pyloric metaplasia, mucous metaplasia, or antralization of the corpus, displays morphologic features of deep antral glands or Brunner glands with expression of Trefoil factor 2 (TFF2; formerly named *spasmolytic polypeptide*) and Mucin 6 (MUC6) [4,5]. Furthermore, mature, terminally differentiated chief cells, which secrete pepsinogen, can transdifferentiate into SPEM in rodent models [6]. Inflammatory cells accelerate this metaplastic change into a more proliferative lineage and even to dysplasia [7].

IM is commonly considered as the transdifferentiation of a mucosa into an intestinal phenotype, appearing in the context of chronic inflammation [8]. IM is characterized by the presence of mucin-containing goblet cells, Paneth cells, and absorptive cells, with expression of Trefoil factor 3 (TFF3) and Mucin 2 (MUC2) [4,9].

IM can arise from preexisting SPEM under inflammatory influences in animal models, suggesting that SPEM may be a neoplastic precursor in the carcinogenesis cascade [9,10]. In 3 clinical studies with a small number of human gastric cancer patients, SPEM was associated with 90% of resected gastric carcinomas, and both SPEM and IM were linked with progression to intestinal-type gastric carcinoma [5,11,12]. However, the precise correlation between SPEM and IM requires further exploration in large-scale clinical studies.

In the proposed Correa multistep pathway of carcinogenesis, *Helicobacter pylori* (*H pylori*)–mediated chronic gastritis over many years progresses into premalignant atrophic gastritis, SPEM and IM, dysplasia, and, eventually, gastric carcinoma [13]. *H pylori* infection is generally regarded as the fundamental cause of IM and intestinal-type gastric cancer [14]. Epstein-Barr virus (EBV), the first virus to be associated with human malignancy, is another strong risk factor for gastric carcinoma [15]. EBV-associated gastric cancer (EBVaGC), which is characterized by the presence of EBV in gastric cancer cells, represents approximately 9% of all gastric cancer cases worldwide and was recently added as one of the 4 novel molecular subtypes of gastric adenocarcinoma according to The Cancer Genome Atlas project [16,17]. Compared with EBV-negative gastric carcinoma (EBVnGC), EBVaGC represents a distinct subset of gastric cancer in terms of clinicopathological and genetic features [18]. In addition, previous studies proved that *H pylori* infection, a strong risk factor for gastric carcinoma, is not a risk agent for EBVaGC, which indicates that *H pylori* and EBV are involved in different carcinogenic pathways [19].

Therefore, because human EBVaGC has only recently been recognized as a distinct gastric cancer entity, the aim of this study was to explore the clinical and histological features of SPEM and IM in EBVaGC and EBVnGC patients. In addition, a possible relationship between SPEM, IM, and inflammation in gastric cancer etiology, as well as overall impact of SPEM on patient prognosis were also explored in EBVaGC and EBVnGC.

2. Materials and methods

All human subjects and protocols were approved by the ethics committee of the Third Affiliated Hospital, Sun Yat-sen University, Guangzhou, China. Written informed consent was obtained from all patients. Ethical guidelines under the Declaration of Helsinki were followed.

This study included 64 EBVaGC patients identified from 996 gastric adenocarcinoma cases who underwent surgical resection at the Second, Third, and Sixth Affiliated Hospitals of Sun Yat-sen University, and the Guangzhou First Municipal People's Hospital, Guangzhou, China, between January 2000 and December 2012. A total of 154 EBVnGC patients who had similar clinicopathological characteristics were identified and selected from 996 cases to match 64 EBVaGC. The patients enrolled in the study conformed to the following eligibility criteria: (1) histologically confirmed with primary gastric adenocarcinoma, (2) negative resection margin, (3) no history of chemotherapy or radiation therapy, and (4) EBV positivity identified by EBV-encoded RNA in situ hybridization (ISH; this criterion applies only to EBVaGC). Clinicopathological characteristics of patients were acquired by reviewing the medical archives. Tumor stage was classified and staged according to the seventh edition of the Union for International Cancer Control/American Joint Committee on Cancer manual for the stomach [20]. Each tumor tissue specimen