Polymorphisms of matrix metalloproteinase-7 and chymase are associated with susceptibility to and progression of gastric cancer in Japan

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Background. Matrix metalloproteinases (MMPs) are a family of enzymes that degrade most macromolecules making up the extracellular matrix. MMPs are involved in not only the gastric mucosal inflammatory response but also the pathogenesis of Helicobacter pylori-associated diseases. In the renin–angiotensin system, chymase (CMA) is related to gastric carcinogenesis and angiogenesis in H. pylori-infected patients. We aimed to clarify the association of MMP-7-181 and CMA/B polymorphisms with susceptibility to gastric cancer and cancer progression in H. pylori-infected patients.

Methods. We assessed the MMP-7-181 and CMA/B polymorphisms in H. pylori-positive patients with gastric cancer (n = 160), gastric ulcer (n = 157), duodenal ulcer (n = 121), and H. pylori-positive gastritis alone as controls (n = 156).

Results. For gastric cancer risk, the age- and sex-adjusted odds ratio (OR) of the MMP-7-181 G allele carrier relative to the A/A genotype was significantly increased [OR, 2.32; 95% confidence interval (CI), 1.24–4.35], especially in patients with noncardia cancer (OR, 2.31; 95% CI, 1.22–4.36) and those with clinical stage III or IV cancer (OR, 3.66; 95% CI, 1.54–8.73). Carriage of the CMA/B A allele was significantly associated with gastric cancer development (OR, 1.73; 95% CI, 1.10–2.71). Simultaneous carriage of both the MMP-7-181 G allele and the CMA/B A allele dramatically increased the gastric cancer risk (OR, 8.18; 95% CI, 2.79–23.93).

Conclusions. In Japan, carriage of the MMP-7-181 G allele and of the CMA/B A allele were each associated with an increased risk for H. pylori-related noncardia gastric cancer development. MMP-7-181 and CMA/B genotyping tests might be useful tools for screening for individuals with higher gastric cancer risk.

Key words: H. pylori, MMP-7, chymase gastric cancer, polymorphism

Introduction

Gastric cancer is one of the most common cancers in Asian countries, including Japan. Although gastric cancer is generally thought to arise through mucosal changes leading to atrophic gastritis, caused by chronic Helicobacter pylori infection, the development and progression of gastric mucosal atrophy differs depending on the genetic factors of the host, the H. pylori strain, and various environmental factors (e.g., salt intake and smoking habit). Of these factors, H. pylori virulence factors (i.e., cagA, vacA, outer inflammatory protein A, and duodenal ulcer promoting A12) and host genetic factors (i.e., polymorphisms of inflammation-related cytokines,3–6 drug metabolism-related enzymes,7,8 the renin–angiotensin system,9,10 and growth factors) regulate the immune and inflammatory responses of the gastric mucosa and are candidate factors that might explain the interindividual variation observed in the clinical outcome of H. pylori infection, such as gastric cancer, gastric ulcer, duodenal ulcer, and gastritis alone.3–9

The development and progression of cancer comprise several processes, including the initiation, promotion, invasion, migration, and implantation of cancer cells at metastatic sites. These processes are mediated by a variety of molecules related to inflammation and angiogenesis. Recent studies have found that matrix metalloproteinases (MMPs), a family of highly conserved zinc-dependent proteolytic enzymes, and the renin–angiotensin (RA) system are activated by infection with H. pylori,11–15 which is well known to be a definite gastric carcinogen.9,10,16
MMPs play an important role in several steps of cancer development by regulating cancer cell growth, differentiation, apoptosis, angiogenesis, invasion, and metastasis by degrading the extracellular matrix and basement membrane barriers. Interestingly, chronic *H. pylori* infection increases the expression of MMPs in the gastric mucosa, resulting in gastric severe mucosal damage and gastric mucosal atrophy. Among MMPs, MMP-7 (matrilysin, PUMA-1) is a protease with broad substrate specificity, being able to degrade elastin, proteoglycans, fibronectin, and type IV collagen. Moreover, MMP-7 can also cleave nonmatrix substrates from the cell surface.

Several polymorphisms in the promoters of MMP genes, which are thought to affect the respective MMP production in a genotype-specific manner, have been reported. Recently, the MMP-7-181 polymorphism has been associated with increased risk of development of several cancers, including colorectal cancer, esophageal cancer, gastric cardia cancer, and lung cancer. However, Ghilardi et al. reported that the MMP-7-153 polymorphism is less involved in susceptibility to colorectal cancer. Moreover, the frequency of the MMP-7-153 T allele, a high-producer allele, is rare in Asians. Therefore, although a haplotype analysis of the two polymorphisms may show their additive interaction in gastric carcinogenesis, use of a genotyping test for MMP-7-153 polymorphism in the Asian population may be of limited importance.

Recently, evidence that angiotensin II is involved in the regulation of cell proliferation, angiogenesis, inflammation, and tissue remodeling has been increasing. Moreover, the local overexpression of the RA system in various cancer cells and tissues has suggested that local overexpression of several components of the RA system may be associated with carcinogenesis. In the RA system, chymase (CMA), a chymotrypsin-like serine protease produced in the secretory granules of mast cells, mediates mainly the local, not systemic, generation of angiotensin II. Two polymorphisms in the CMA gene, CMA/A and CMA/B, are localized on chromosome 14. Those two CMA polymorphisms, which have been shown to correlate with the expression of CMA, are candidate markers of neoplastic diseases development. Moreover, because angiotensin II type 1 receptors (AT1Rs) induce vascular endothelial growth factor (VEGF) and angiopoietin II, resulting in angiogenesis in cancer tissues, upregulation of the RA system might be related to metastasis and advanced clinical stage cancer.

Although whether these two molecules interact directly is unclear, MMPs and RA system molecules (i.e., MMP-7 and CMA) might cooperate to regulate inflammation and angiogenesis mediated by inflammatory cytokines and/or growth factors in the gastric mucosa and at metastatic sites, and, therefore, genetic differences in MMPs and the RA system might be associated with interindividual differences in gastric cancer risk and prognosis. However, the exact relationship between gastric cancer risk and the MMP-7-181 and CMA/B polymorphisms remains obscure. Therefore, we examined whether these two polymorphisms are associated with gastric cancer risk and progression in Japan.

**Materials and methods**

**Subjects**

A total of 694 Japanese patients found to be infected with *H. pylori* on the basis of a rapid urease test (RUT) (Helico Check, Otsuka, Tokushima, Japan) were enrolled in this study at the University Hospital of Hamamatsu University School of Medicine from January 2001 to March 2007. Patients with a positive RUT result from biopsied specimens obtained during gastroduodenoscopy were invited to participate in the study. They were enrolled in the study after giving their written informed consent. The study population comprised patients with gastric cancer (*n* = 160), gastric ulcer (*n* = 157), duodenal ulcer (*n* = 121), or gastritis alone (*n* = 156). These patients had endoscopically and histologically proven active chronic gastritis, peptic ulcer, or gastric cancer. Gastric biopsy specimens were stained with hematoxylin and eosin, and the degree of inflammation and atrophy of the gastric antrum and body were assessed according to the updated Sydney system. A single pathologist (H. S.), who was unaware of any clinical information regarding the patients, examined all biopsy specimens. The gastric cancer group was pathologically classified into intestinal-type and diffuse-type gastric cancer subgroups, in accordance with the Lauren classification, and further classified into subgroups based on cancer location (i.e., noncardia or cardia). Moreover, gastric cancer patients were classified according to the clinical stage of the cancer (stages 1–4) according to the TNM classification system. The demographic and clinical characteristics of the patients enrolled in the study are summarized in Table 1. The protocol was approved in advance by the Human Institutional Review Board of Hamamatsu University School of Medicine.