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Original contribution

Differences in interleukin 8 expression in *Helicobacter pylori*–infected gastric mucosa tissues from patients in Bhutan and the Dominican Republic☆

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Summary The outcomes of *Helicobacter pylori* infection vary geographically. *H pylori* strains, disease presentation, and environments differ markedly in Bhutan and Dominican Republic. The aims were to compare the strains, histology, and expression of interleukin (IL) 8 and IL-10 from gastric mucosa from the 2 countries. *H pylori* status was assessed by the combination of rapid urease test, culture, and histology. Histology was evaluated using the updated Sydney System, and cytokines in gastric biopsies were measured using real-time polymerase chain reaction (PCR). There were 138 subjects from Bhutan and 155 from Dominican Republic. The prevalence of *H pylori* infection was 65% and 59%, respectively. The genotype of *cagA* was predominantly East Asian type in Bhutan versus Western type

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in Dominican Republic. Gastritis severity was significantly higher in *H pylori*–infected subjects from Bhutan than those from Dominican Republic. IL-8 expression by *H pylori* infection was 5.5-fold increased in Bhutan versus 3-fold in Dominican Republic ($P < .001$); IL-10 expression was similar. IL-8 expression levels among *H pylori*–infected cases tended to be positively correlated with polymorphonuclear leucocyte and monocyte infiltration scores in both countries. IL-8 expression among those with grade 2 and 3 polymorphonuclear leucocyte and monocyte infiltration was significantly higher in Bhutan than in Dominican Republic. The difference in IL-8 expression in the 2 countries is reflected in the different disease pattern between them. Whether the dominant factor is differences in *H pylori* virulence, in host–*H pylori*–environmental interactions, genetic factors or all remains unclear. However, severity of inflammation appears to be a critical factor in disease pathogenesis. We compared IL-8 messenger RNA levels between the high gastric cancer risk country, Bhutan (mainly East Asian–type *H pylori*), and the lower gastric cancer risk country, Dominican Republic (mainly Western-type *H pylori*).

1. Introduction

Helicobacter pylori is a spiral, gram-negative human pathogen and a major cause of peptic ulcer disease and gastric cancer [1,2]. *H pylori* gastritis is characterized by infiltration of the gastric mucosa with both neutrophils and mononuclear cells (MNCs) and is associated with the up-regulation of proinflammatory cytokines such as the potent chemotactic peptide interleukin (IL) 8 [2,3]. The clinical manifestations of *H pylori* infections vary both within and among populations; for example, in some areas such as Japan, where *H pylori* infection is common, gastric cancer is an important clinical problem, whereas in other areas such as South India, gastric cancer is rare, and instead, duodenal ulcer is a more common disease manifestation [4]. Whether these geographic differences correlate with the differences in the gastric mucosal cytokine levels remain unknown because studies comparing the mucosal cytokine levels among individuals from different countries are lacking. Here, we have reported a comparison of gastric mucosal IL-8 and IL-10 expression and the histologic findings by using gastric biopsies obtained from adults in Bhutan and the Dominican Republic. These countries were selected for the study because of the differences in their clinical outcomes of *H pylori* infection as well as in their *cagA* genotype of the pathogenic *H pylori* (ie, *cagA*-negative,Western-type *cagA* in the Dominican Republic, and East Asian–type *cagA* in Bhutan). The tissue samples from both the regions were similarly processed and analyzed.

2. Materials and methods

2.1. Study countries

Bhutan and the Dominican Republic are relatively isolated developing countries where *H pylori* infection is common. These countries show several differences in the age-standardized rate of gastric cancer incidence; the rate is higher in Bhutan (17.2/100 000) than in the Dominican Republic (7.3/100 000) (GLOBOCAN 2012: http://globocan.iarc.fr/). Bhutan is a small, landlocked mountainous country located at the eastern end of the Himalayas, and it shares borders on the south, east, and west with the Republic of India and to the north with the People's Republic of China. In contrast, the Dominican Republic is a Caribbean island; it shares borders on the east with Haiti and with a population of 73% multiracial, 16% whites, and 11% Congoid. We hypothesized that the *cagA* genotype between these 2 countries also differs, with Bhutanese strains being East Asian–type *cagA* and the Dominican Republic strains being Western-type *cagA*. In addition, the presence of *cagA*-negative strains makes these regions ideal for comparing the gastric histology and mucosal cytokine levels in relation to the *cagA* genotype and its outcome.

2.2. Subjects

We recruited individuals with mild dyspeptic symptoms living in Bhutan and outpatients with mild dyspeptic symptoms living in the Dominican Republic. The surveys took place at the Jigme Dorji Wangchuk National Referral Hospital, Thimpu, Bhutan, in December 2010 and at the Dr Luís E. Aybar Health and Hygiene City (Digestive Disease Center), Santo Domingo, Dominican Republic, in February 2012. Written informed consent was obtained from all the participants. The protocol was approved by the ethics committee of Oita University Faculty of Medicine (Japan), Universidad Autonoma de Santo Domingo (Dominican Republic), and by both the hospitals where sample collection was performed.

The tissue sampling procedures as well as the method of sample handling were identical at both the locations. During each endoscopy session, 4 gastric biopsy specimens were obtained from the antrum: 1 each for *H pylori* culturing, rapid urease test (*Campylobacter-like* organism test; Kimberly-Clark Ballard Medical Products, Roswell, GA), histologic examination, and cytokine examination. Rapid