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Advances in Brief

An Inverse Relation between *cagA*$^+$ Strains of *Helicobacter pylori* Infection and Risk of Esophageal and Gastric Cardia Adenocarcinoma[1]

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Abstract

Gastric colonization with *Helicobacter pylori*, especially *cagA*$^+$ strains, is a risk factor for noncardia gastric adenocarcinoma, but its relationship with gastric cardia adenocarcinoma is unclear. Although incidence rates for noncardia gastric adenocarcinoma have declined steadily, paralleling a decline in *H. pylori* prevalence, rates for adenocarcinomas of esophagus and gastric cardia have sharply increased in industrialized countries in recent decades. To clarify the role of *H. pylori* infection in these tumors with divergent incidence trends, we analyzed serum IgG antibodies to *H. pylori* and to a recombinant fragment of CagA by antigen-specific ELISA among 129 patients newly diagnosed with esophageal/gastric cardia adenocarcinoma, 67 patients with noncardia gastric adenocarcinoma, and 224 population controls. Cancer risks were estimated by odds ratios (OR) and 95% confidence intervals (CI) using logistic regression models. Infection with *cagA*$^+$ strains was not significantly related to risk for noncardia gastric cancers (OR, 1.4; CI, 0.7–2.8) but was significantly associated with a reduced risk for esophageal/cardia cancers (OR, 0.4; CI, 0.2–0.8). However, there was little association with *cagA*$^-$ strains of *H. pylori* for either cancer site (OR, 1.0 and 1.1, respectively). These findings suggest that the effects of *H. pylori* strains on tumor development vary by anatomical site. Further studies are needed to confirm these results and to assess whether the decreasing prevalence of *H. pylori*, especially *cagA*$^+$ strains, may be associated with the rising incidence of esophageal/gastric cardia adenocarcinomas in industrialized countries.

Introduction

Infection with *Helicobacter pylori* is a risk factor for noncardia gastric adenocarcinoma (1–6). The risk of gastric adenocarcinoma and its precursor state, atrophic gastritis, is associated particularly with *cagA*$^+$ compared with *cagA*$^-$ strains of *H. pylori* (7–9). Although some studies have reported no association between *H. pylori* infection and gastric cardia adenocarcinoma, the number of cardia cases was usually too small for adequate assessment of risk (2, 3, 5, 10). As part of a multicenter case-control study of esophageal and gastric cancers in the United States, we examined the relation of *H. pylori* infection and *cagA* status to adenocarcinomas of esophagus and gastric cardia, the incidence of which is rapidly rising in Western countries (11), compared with noncardia gastric adenocarcinoma, the incidence of which has steadily declined worldwide (12).

Materials and Methods

The methods for this study are described in detail elsewhere (13). Included in the present analysis were data from two of the three participating centers in which blood collection was conducted, *i.e.*, in 9 of the 15-county study area of New Jersey and in a 3-county area of western Washington. Blood specimens were not collected in the remainder of the study areas, mainly for logistical reasons. Residents, ages 30–79, who were newly diagnosed with invasive esophageal or gastric adenocarcinoma between 1993 and 1995 were identified through rapid reporting systems. Population-based controls were selected by random digit dialing (14) for those under 65 years of age and from the Health Care Financing Administration files for those 65 years of age or older (15), frequency-matched to cases by age, sex, and race.

In-person structured interviews were conducted with subjects, or next of kin for deceased subjects, to elicit information on demographic background, tobacco and alcohol use, medication and medical histories, diet, occupation, and height and weight history. The overall interview rate was 81% for cases with esophageal/gastric cardia adenocarcinoma, 69% for the noncardia gastric cases, and 74% for population controls. A 30-ml blood sample was obtained from subjects alive at the time of interview, including 129 esophageal/cardia cases (66% of cases interviewed in person), 67 noncardia cases (58% of cases interviewed in person), and 224 controls (67% of controls interviewed in person).

Serum levels of IgG antibodies to *H. pylori* were determined by antigen-specific ELISA, which consists of a preparation of sonicated whole bacteria made from five different clinical *H. pylori* isolates (16). An AI[4] was calculated from the mean absorbances at 405 nm of two assays per sample. A previously validated cutpoint of AI $\geq$ 1.0 was used to define *H. pylori* positivity. *cagA* status was determined by an ELISA based on the presence of serum IgG antibodies against orv220, a 65,000 recombinant CagA protein purified from *Escherichia coli*. CagA positivity was defined as an AI $\geq$ 0.3, as validated previously (7). With blinding to the case/control status, serum from each subject was assayed for antibodies to *H. pylori*, and among those found positive, *cagA* status was assessed.

Relative risks associated with *H. pylori* colonization and *cagA* status compared with uninfected subjects were estimated by ORs and 95% CIs using logistic regression models (17). All ORs were adjusted for age, sex, race (white, other), geographic center (New Jersey, western Washington), and education (<12 years, 12 years, vocational school/some college, college graduate and higher). Further adjustment for selected risk factors for esophageal/gastric cardia adenocarcinoma, including cigarette smoking, body mass index, and history of gastroesophageal reflux disease, did not materially alter the findings. Effect modification was assessed by examination of stratum-specific results. Initial analyses conducted separately for adenocarcinomas of the

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[4] The abbreviations used are: AI, absorbance index; OR, odds ratio; CI, confidence interval.

esophagus and gastric cardia found similar results. Hence, the two cancer types were combined in subsequent analyses to provide more stable estimates.

Results

As shown in Table 1, *H. pylori* infection was not significantly related to risk of esophageal/gastric cardia adenocarcinoma (OR, 0.7; 95% CI, 0.4–1.1) or noncardia gastric adenocarcinoma (OR, 1.3; 95% CI, 0.7–2.3). When compared with the *H. pylori* uninfected subjects, no association was observed for carriage of $cagA^-$ strains of *H. pylori*. In contrast, infections with $cagA^+$ strains were associated with a small, nonsignificantly elevated risk for noncardia gastric adenocarcinoma (OR, 1.4; 95% CI, 0.7–2.8) but a significantly reduced risk for esophageal/cardia adenocarcinoma (OR, 0.4; 95% CI, 0.2–0.8). The risk reduction was consistent across strata of several potential confounding factors, including age, sex, educational level, history of gastroesophageal reflux disease, body mass index, cigarette smoking, alcohol drinking, preference for salty foods, and Laurén classification of diffuse *versus* intestinal cell types (data not shown).

Discussion

Our findings suggest that the effect of *H. pylori* colonization on the risk of esophageal and gastric adenocarcinoma varies by anatomical subsite of tumor and bacterial strain. Most notable are the finding for $cagA^+$ strains, which was associated with a significantly reduced risk for esophageal and gastric cardia adenocarcinomas. In interpreting these results, however, some potential limitations of our study should be considered. Among the subjects approached for blood collection, about one-third did not respond. In addition, blood specimens were not collected from subjects who died before interview, accounting for about 30% of eligible cases and 4% of eligible controls. Among both case and control subjects who were alive at time of interview, blood donors were more likely to be men than were nonrespondents, but the two groups were similar in age and race distribution, suggesting that differential selection bias between cases and controls is unlikely. Because nondifferential bias tends on average to attenuate associations, it is possible that the magnitude of the effects observed in our study are underestimated.

Another possible source of underestimation of risk may be the loss of *H. pylori* colonization that can accompany gastric atrophy and later stages of gastric carcinogenesis (18). However, atrophic gastritis has not been described as a common precursor for tumors of the gastric cardia. On the other hand, it has been shown that 5–10% of the subjects that tested negative for *H. pylori* may actually carry $cagA^+$ strains (7). Assuming 7.5% of all *H. pylori*-negative subjects in our study were infected with $cagA^+$ strains, the crude OR for esophageal/gastric cardia adenocarcinoma would increase from 0.4 to 0.5 but would have little effect on risk for noncardia gastric adenocarcinoma or for associations with $cagA^-$ infections.

The slight inverse association we observed between carriage of *H. pylori* and risk of adenocarcinomas of esophagus and gastric cardia resembles findings reported for tumors of the gastric cardia in prospective studies (2, 3, 10). These observations are consistent also with recent studies indicating similar or lower percentages of *H. pylori* carriage among patients with esophagitis or Barrett's esophagus (25–36%) than normal controls (36–51%) (19, 20), although specific strains were not examined.

Despite the limited sample size, our study is one of the largest to assess *H. pylori* strain-specific risk for esophageal/cardia *versus* noncardia adenocarcinomas. The overall consistency in risk patterns within case groups and across strata of potential confounding variables adds credence to our findings. Although small numbers precluded detailed assessment, the risks associated with $cagA^+$ strains were similarly reduced for adenocarcinomas of esophagus and gastric cardia. It seemed logical to combine these tumor types, particularly in view of shared histological and epidemiological features (11, 21, 22).

Although evidence for the role of *H. pylori* infection in gastric carcinogenesis has accumulated, there remain questions with regard to risks by bacterial strain and tumor subsite (23–25). Our finding of a positive, but nonsignificant, association between $cagA^+$ infection and risk of noncardia gastric cancer is consistent with reports linking $cagA^+$ (in contrast to $cagA^-$) strains of *H. pylori* to more intense gastritis (8, 26, 27) and a greater risk of developing distal stomach cancer (7, 9).

Although a new finding, the reduced risk for adenocarcinomas of the esophagus and gastric cardia associated with $cagA^+$ infection is consistent with recent clinical reports that $cagA^-$ strains predominate among patients with esophageal adenocarcinoma as well as the precursor states of gastroesophageal reflux disease, Barrett's esophagus, and esophageal dysplasia (28, 29). It is also noteworthy that antibiotic therapy in duodenal ulcer patients, over 90% of whom have $cagA^+$ strains of *H. pylori* (30), may promote reflux esophagitis and weight gain (31), both of which are risk factors for esophageal/gastric cardia adenocarcinoma (32, 33).

It is likely that the decline in the incidence of *H. pylori* infection including $cagA^+$ strains (34, 35) has contributed to the downward incidence trends for noncardia tumors of the stomach (12). However, little is known of the reasons for the rising incidence of adenocarcinomas of the esophagus and gastric cardia in industrialized countries (11, 36). Our finding of a reduced risk of these tumors among persons infected with $cagA^+$ strains of *H. pylori* may provide an etiological clue to the upward incidence trends. If confirmed, it will be important to identify the mechanism by which $cagA^+$ strains of *H. pylori* are inversely related to the risk of esophageal and gastric cardia adenocarcinomas, as well as the potential long-term risks (25) and benefits (37) of chemopreventive regimens to eradicate *H. pylori*. Given the increased virulence of the $cagA^+$ strains of *H. pylori* in relation to distal stomach pathology and the lowering of gastric acidity that accompanies progression of atrophic gastritis (8), it is possible that the apparent protection of esophageal and gastric cardia adenocarcinomas was related to a reduced gastric acidity rather than $cagA^+$ strains *per se*. It will be of interest to evaluate whether variations in the acidity and content of refluxate are involved in the mechanism by which *H.*

Table 1 *ORs and 95% CIs for adenocarcinomas of esophagus/gastric cardia and noncardia stomach in relation to H. pylori positivity and cagA status*

		Esophageal/gastric cardia adenocarcinoma			Noncardia gastric adenocarcinoma		
	Control *n*	*n*	OR[a]	95% CI	*n*	OR[a]	95% CI
H. pylori status							
Negative (<1.0)	137	91	1.0	Referent	34	1.0	Referent
Positive (≥1.0)	86	38	0.7	0.4–1.1	33	1.3	0.7–2.3
cagA status (among *H. pylori* positives)							
Negative (<0.30)	40	26	1.0	0.5–1.7	12	1.1	0.5–2.5
Positive (≥0.30)	46	12	0.4	0.2–0.8	21	1.4	0.7–2.8

[a] Adjusted for age, sex, race, education, and geographic center.

pylori strains may affect the risk of adenocarcinoma of the esophagus and gastric cardia.

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