Helicobacter pylori infection, garlic intake and precancerous lesions in a Chinese population at low risk of gastric cancer

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Background
Cangshan County of Shandong Province has one of the lowest rates of gastric cancer (GC) in China. While intestinal metaplasia (IM) and dysplasia (DYS) are less common in Cangshan than in areas of Shandong at high risk of GC, these precursor lesions nevertheless affect about 20% of adults age ≥55.

Subjects and Setting
In order to evaluate determinants of IM and DYS in Cangshan County, a low risk area of GC a survey was conducted among 214 adults who participated in a gastroscopic screening survey in Cangshan County in 1994.

Method
A dietary interview and measurement of serum Helicobacter pylori antibodies were performed.

Results
The prevalence of H. pylori was lowest (19%) among those with normal gastric mucosa, rising steadily to 35% for superficial gastritis (SG), 56% for chronic atrophic gastritis (CAG), 80% for IM, and 100% for DYS. The prevalence odds of precancerous lesions were compared with the odds of normal histology or SG. The odds ratio (OR) of CAG associated with H. pylori positivity was 4.2 (95% confidence interval [CI] : 1.7–10.0), while the OR of IM/DYS associated with H. pylori positivity was 31.5 (95% CI : 5.2–187). After adjusting for H. pylori infection, drinking alcohol was a risk factor for CAG (OR = 3.2, 95% CI : 1.1–9.2) and IM/DYS (OR = 7.8, 95% CI : 1.3–47.7). On the other hand, consumption of garlic showed non-significant protective effects and an inverse association with H. pylori infection.

Conclusions
The findings of this study suggest that infection with H. pylori is a risk factor and garlic may be protective, in the development and progression of advanced precancerous gastric lesions in an area of China at relatively low risk of GC.

Keywords
Precancerous gastric lesions, high and low risk, Helicobacter pylori, alcohol, garlic

Accepted 2 June 1998

Gastric cancer (GC) is the most common cancer in China, with mortality rates showing remarkable geographical variation within the country. In Cangshan County of Shandong Province in the northeast of China, GC mortality rates are 5.0/10^5 males and 3.0/10^5 females compared respectively with 70/10^5 and 26/10^5 in Linqu, a county less than 200 miles away. Study of gastric histopathology among adults in the two counties revealed that the prevalence of intestinal metaplasia (IM) and dysplasia (DYS) were also lower in Cangshan than in Linqu. It is noteworthy that Cangshan is one of the leading garlic-producing regions in China, and that garlic consumption has been associated with a reduced risk of GC in both epidemiological and experimental studies. Since Helicobacter pylori infection has been linked to GC and precancerous gastric lesions, this area provides an opportunity to evaluate possible interactions with garlic, which has the potential to inhibit H. pylori growth. There are 28 townships with a population of 1.13 million inhabitants in Cangshan, and 13 of these townships with 700 000 inhabitants are major garlic-producing...
centres. To evaluate the role of \textit{H. pylori}, garlic and other factors in the progression to more advanced gastric lesions in Cangshan, we selected one village from among 37 villages in a garlic-producing township, and we analysed interview and serological data on subjects with precancerous gastric lesions identified in a survey conducted among 214 adults in this relatively low-risk area of China.

**Methods**

Details of the gastroscopic survey have been described elsewhere.\textsuperscript{2,3} In brief, in the spring of 1994, one village was selected at random from a typical garlic farming town in Cangshan for the present study. After the names of all residents aged 35–64 had been transcribed from the village population rosters, health officials visited each person and offered a consent form to invite villagers to participate in a GC screening. A total of 224 adults aged 35–64 participated in this study, representing 30% of the eligible population in the village. The subjects then received gastroscopy, with biopsies taken from four standard sites in the stomach: one from the greater curvature of the body, one from the angulus, and one each from the lesser and greater curvatures of the antrum. Each slide was interpreted by three senior pathologists independently, and doubly blind quality control samples of the slides selected at random were reviewed by experts on gastric pathology in China and the US.\textsuperscript{2,3} All the subjects were classified according to the most severe lesions found in any biopsy specimen (hereafter referred to as the global diagnosis), using the categories of normal, superficial gastritis (SG), chronic atrophic gastritis (CAG), IM or DYS.\textsuperscript{2,3}

The questionnaire sought information on diet, smoking, alcohol drinking and socioeconomic status. An approximately 5 ml blood specimen was collected from each fasting subject. Serum was separated and aliquoted, stored immediately at \(-20^\circ\text{C}\) and then moved into a \(-70^\circ\text{C}\) freezer. Serum \textit{H. pylori} IgG antibody concentrations were measured using an ELISA procedure.\textsuperscript{14} and an individual was considered to be positive for \textit{H. pylori} infection if the ELISA absorbance reading for IgG was \(>1.0\), a cutoff based on examination of the distribution of such readings in relation to a group of uninfected people and reference sera. Using \(C^\text{15}\) urea breath test as a gold standard, we previously estimated sensitivity and specificity of this assay of 94\% and 83\%, respectively.\textsuperscript{19}

Since the numbers of subjects in each histological state (normal, SG, CAG, IM and DYS) were relatively small, categories were combined into three histological groups: (1) normal and SG, (2) CAG, and (3) IM and DYS. Odds ratios (OR) were calculated comparing the prevalence of CAG to that of normal/SG or the prevalence of IM/DYS to that of normal/SG. A logistic regression analysis\textsuperscript{20} was performed to relate these OR to factors such as \textit{H. pylori} infection, alcohol drinking, smoking and garlic consumption, with adjustments made for gender, age, and other factors.

**Results**

Gastric pathology and interview data were available for 214 subjects, 107 males and 107 females. Gastric pathology and serum IgG antibody for \textit{H. pylori} data were available for a total of 197 subjects (92\% of those screened). Almost 95\% of the participants were garlic farmers or the wives of garlic farmers, and more than 95\% of them were born in Cangshan.

Table 1 shows that the global diagnosis was normal/SG in 18\%, CAG in 69\% and IM/DYS in 14\%. The prevalence rates varied by age, with an upward trend for IM/DYS \((P = 0.12)\) and a downward trend for CAG \((P = 0.16)\). About 20\% of those aged \(>55\) were diagnosed with IM/DYS. Gender was not significantly associated with gastric histopathology \((P = 0.77)\).

Overall, 35\% of the subjects had positive serum antibodies to \textit{H. pylori}, with little variation by sex or age. Furthermore, cigarette smoking, alcohol drinking and dietary factors were not significantly associated with \textit{H. pylori} infection. The prevalence of \textit{H. pylori} was 61\% in those who consumed <5 kg garlic/year, 52\% in those who consumed 5–15 kg/year, and 54\% in those who consumed more than 15 kg/year \((\chi^2 \text{ trend} = 0.91, P = 0.64)\).

As shown in Figure 1, the prevalence of \textit{H. pylori} rose steadily with increasing severity of gastric histopathology. The positivity was 19\% among those with normal gastric mucosa, 35\% with SG, 56\% with CAG, 80\% with IM, and 100\% with DYS. Logistic regression revealed that the OR of CAG associated with \textit{H. pylori} positivity was 4.2 (95\% CI: 1.7–10.0) while the OR of IM/DYS associated with \textit{H. pylori} positivity was 31.5 (95\% CI: 5.2–187) after adjusting for gender, age, cigarette smoking,

Table 1: Age-sex specific prevalence of adults with normal/SG (superficial gastritis), chronic atrophic gastritis (CAG) and intestinal metaplasia (IM)/dysplasia gastric mucosa (DYS) in Cangshan

<table>
<thead>
<tr>
<th>Age</th>
<th>Normal/SG</th>
<th>CAG</th>
<th>IM/DYS</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;40</td>
<td>17</td>
<td>68</td>
<td>74.7</td>
</tr>
<tr>
<td>40-54</td>
<td>10</td>
<td>37</td>
<td>66.1</td>
</tr>
<tr>
<td>&gt;55</td>
<td>11</td>
<td>16.4</td>
<td>62.7</td>
</tr>
</tbody>
</table>

**Figure 1:** Prevalence of \textit{H. pylori} antibody positivity, Cangshan County (n = 197)
alcohol drinking, and consumption of meat and eggs, garlic and other vegetables (Table 2). Consumption of garlic showed a protective effect with respect to CAG (OR = 0.5–0.7) and IM/DYS (OR = 0.5–0.6), although no significant linear trend was found. Drinking alcohol was a risk factor, with adjusted OR of 3.2 (95% CI: 1.1–9.2) for CAG and 7.8 (95% CI: 1.3–47.7) for IM/DYS, respectively. However, cigarette smoking was not associated with risk of CAG (OR = 0.6, 95% CI: 0.2–1.6) or IM/DYS (OR = 1.0, 95% CI: 0.2–5.1). No effect on risk of CAG or IM/DYS was found for consumption of vegetables other than garlic, salted vegetables, fruits, grains, meat and eggs, or salt. Allium vegetables other than garlic were rarely eaten.

**Discussion**

Although prevalences of IM and DYS were relatively low in Cangshan compared to Linqu, a high-risk area for GC in China, the present study suggests that infection with *H. pylori* is a risk factor for precancerous lesions in Cangshan. Indeed, calculations of attributable risk show that *H. pylori* was potentially responsible for 40% (95% CI: 25–57%) of the CAG cases, and 82% (95% CI: 56–95%) of the IM/DYS cases. In contrast to Linqu, where the *H. pylori* prevalence tended to decrease with more advanced gastric lesions, the prevalence of *H. pylori* tended to rise among subjects with IM and DYS in Cangshan, suggesting that *H. pylori* may play a role in late as well as early stages of carcinogenesis in this low-risk area. It is unclear why the association between *H. pylori* and advanced gastric lesions is stronger in Cangshan than in Linqu, but it seems likely that other aetiological factors in Linqu may contribute to the pattern observed. The overall prevalence of *H. pylori* in Cangshan was about 20% lower than in Linqu. Furthermore, the prevalence of IM and DYS in Cangshan was less than half and the rate of GC was less than one-eighth of that in Linqu. Thus, even though *H. pylori* appears to account for a higher proportion of the precancerous lesions in Cangshan, it seems not to have induced the epidemic levels of gastric disease seen in Linqu.

The present study showed that high consumption of garlic was associated with a nearly 50% reduction in the odds of CAG and IM/DYS. For more than 1700 years, Cangshan has been famous for its garlic products, which have been used widely as a flavour-enhancer and a folk medicine. In 1995, the yearly production of garlic was about 161 525 000 kg, accounting for 25% of the total revenue in this region. Residents of Cangshan consumed an average of 6 kg of fresh garlic per year and 7.5 kg in the study village, compared with 1.5 kg in Linqu and 0.8 kg in the US. We previously reported an inverse dose-response relation between GC risk and consumption of garlic in Linqu and a protective effect has been found also in Italy.

Our study in Cangshan provides further evidence that garlic intake may inhibit gastric carcinogenesis, although the mechanism of action is unclear. S-allyl cysteine sulphoxide (alliiin), a major component in fresh garlic, can be transformed to other garlic compounds which have been shown to inhibit several animal tumours induced by 1,2-dimethylhydrazine, benzo(a)-pyrene, and N-nitromethylbenzylamine. Diallyl sulphide (DSA) and its metabolite, diallyl sulphane, are potent inhibitors of CYP2E1, an enzyme that can activate N-nitroso compounds, while allyl methyl trisulphide, a constituent of garlic oil, can induce increased glutathione S-transferase activity. Extracts of fresh garlic can also inhibit the conversion of nitrate to nitrite, a precursor of N-nitroso compounds. We have no information on the exposures to nitrite and N-nitroso compounds in Cangshan, but data from Linqu indicate that nitrite levels in gastric juice are considerably higher among those with high pH or with IM than among those with less advanced lesions. In addition, we found a weak inverse association between garlic intake and *H. pylori* infection, which supports the finding that *H. pylori* growth can be inhibited by garlic extract in vitro. Thus, the lower prevalence of *H. pylori* infection in Cangshan as compared to Linqu may be partly due to the exceptionally high consumption of fresh garlic in this area. It would be interesting to study possible interactions between *H. pylori* infection and garlic intakes.

**Table 2** Age-sex-adjusted odds ratios (OR) and 95% confidence intervals (CI) for precancerous gastric lesions in association with *H. pylori* infection, alcohol drinking and garlic consumption

<table>
<thead>
<tr>
<th></th>
<th>Normal/SG&lt;sup&gt;b&lt;/sup&gt;</th>
<th>CAG&lt;sup&gt;c&lt;/sup&gt;</th>
<th>IM&lt;sup&gt;d&lt;/sup&gt;/DYS&lt;sup&gt;e&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. cases</td>
<td>No. cases</td>
<td>OR</td>
</tr>
<tr>
<td><strong>H. pylori</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Negative</td>
<td>24</td>
<td>61</td>
<td>1.00</td>
</tr>
<tr>
<td>Positive</td>
<td>9</td>
<td>79</td>
<td>4.16</td>
</tr>
<tr>
<td><strong>Garlic (kg)</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0.0–4.9</td>
<td>5</td>
<td>31</td>
<td>1.00</td>
</tr>
<tr>
<td>5.0–15.0</td>
<td>21</td>
<td>79</td>
<td>0.70</td>
</tr>
<tr>
<td>&gt;15.0</td>
<td>8</td>
<td>29</td>
<td>0.46</td>
</tr>
<tr>
<td><strong>Alcohol</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>26</td>
<td>92</td>
<td>1.00</td>
</tr>
<tr>
<td>Yes</td>
<td>7</td>
<td>47</td>
<td>3.17</td>
</tr>
</tbody>
</table>

<sup>a</sup> Odds ratios adjusted for age, gender, salted vegetables, salt fresh vegetables other than allium vegetables, grain, meat, eggs and cigarette smoking.

<sup>b</sup> Superficial gastritis.

<sup>c</sup> Chronic atrophic gastritis.

<sup>d</sup> Intestinal metaplasia.

<sup>e</sup> Dysplasia.
in association with IM or DYS, but our samples were too small to permit this analysis.

There has been no clear evidence of a relationship between alcohol consumption and risk of GC in Linqu and other studies. We previously found in the high-risk area of Linqu that consumption of alcoholic beverages was a risk factor for advanced gastric lesions only in conjunction with cigarette smoking, but the present study in Cangshan, where there may be fewer competing factors, suggests that alcohol may promote the development and progression of gastric lesions independent of cigarette consumption. Alcoholic beverages consumed in Cangshan tend to have especially high ethanol concentrations, generally 80–120%, proof, which might contribute to CAG, and perhaps enhance progression to IM and DYS. This study did not reveal an association between cigarette smoking and progression of advanced gastric lesions, unlike the studies in Linqu. Because of the small numbers of subjects with IM or DYS in Cangshan, we were not able to evaluate the joint effect of cigarette smoking and alcohol drinking.

Some limitations of the present study should be mentioned. Only 30% of those asked to undergo endoscopy agreed to participate in the survey, perhaps because GC is relatively uncommon in Cangshan, indicating potential for selection bias. However, those who participated had the same age and gender distribution as the general population in the village, and we focused on internal comparison among the study participants. Another limitation was the relatively small sample size. Although a reverse association was found between garlic consumption and gastric histopathology, the findings were based on small numbers and were not statistically significant.

In summary, our study in a Chinese population at relatively low risk of GC revealed that _H. pylori_ infection is a risk factor, and there is a suggestion that garlic consumption is protective against progression of precancerous gastric lesions. These findings indicate the need for further studies of the interplay between _H. pylori_ infection and garlic intake in gastric carcinogenesis.

Acknowledgement

Supported in part by National Cancer Institute Contracts NO1-CP-15620, 95660, 05631, 21009 and 33041.

References


