Helicobacter pylori infection and subsequent peptic duodenal disease among young adults

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Background Evidence for a causal relationship between presence of Helicobacter pylori (H. pylori) in gastric mucosa and development of peptic disease is based largely on intervention studies in which eradication of H. pylori led to healing of the lesion. The aim of this study was to assess the importance of H. pylori seropositivity for subsequent development of peptic disease in a prospective study design in young Israelis.

Methods A nested case-control serum bank study based on a systematic sample of male and female inductees to the Israel Defense Force. Twenty-nine cases of duodenal ulcer or duodenitis of moderate or higher severity, diagnosed between 1986 and 1995, were individually matched for age, sex, ethnicity, education and year of induction, with five healthy controls each. Presence of anti-H. pylori antibodies in the frozen stored sera was determined by ELISA.

Results The geometric mean titre of anti-H. pylori antibodies at baseline was significantly higher in cases (18.3 U/ml) than controls (6.9 U/ml; P = 0.009). The matched odds ratio for peptic ulcer disease by seropositivity was 3.8 (95% CI: 1.4–10.2). A stronger association was evident for subjects diagnosed ≥2 years after induction than those diagnosed earlier. The population attributable fraction was 56.6% (95% CI: 15.7–81.1).

Conclusions Pre-existing infection with H. pylori, as determined by seropositivity, is an important determinant of development of duodenal ulcer or duodenitis in young Israelis, supporting the generalizability of an apparent causal association to diverse populations.

Keywords Helicobacter pylori, seroprevalence, peptic ulcer disease, duodenal ulcer

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In 1983 the presence of curved bacilli in the mucous layer of the gastric epithelium was described by Warren and Marshall. 1 The bacterium, subsequently classified as Helicobacter pylori (H. pylori), has been associated with chronic active gastritis and peptic disease of the stomach and duodenum. 2–4 Evidence for a causal relationship, now considered overwhelming, between the presence of H. pylori in gastric mucosa and subsequent development of peptic disease is based mainly on two types of research: cross-sectional studies showing higher prevalence of the organism among ulcer patients as compared with healthy controls, and intervention studies in which eradication of H. pylori led to healing of the lesion. 5, 6 However, only one previous study has addressed the issue of time sequence between prior H. pylori infection and the subsequent development of peptic disease. 7 In a prospective study of 50–80-year-old Japanese men on the Hawaiian island of Oahu a significant association was found between presence of anti-H. pylori antibodies in stored serum samples and the later development of duodenal or gastric ulcer disease.

It has not been established whether the association of H. pylori with peptic ulcer disease is present in all populations, in particular since the virulence of the organism may vary from place to place. 8, 9 Furthermore, there are inconsistencies of an ecological nature. In Israel, for example, the prevalence of H. pylori infection was higher among people of Middle Eastern origin, 10, 11 yet their rates of peptic disease were lower than in those of European origin. 12, 13

The Israel Defense Force (IDF) Medical Corps established a serum bank in 1976 for multipurpose cross-sectional and longitudinal studies. Based on this resource we conducted a prospective (nested case-control) study in which we examined...
the association between presence of anti-\textit{H. pylori} antibodies in sera obtained at induction and subsequent development of peptic ulcer during military service.

**Methods**

**Study population**

The Public Health Branch of the IDF Medical Corps continuously draws a systematic, representative sample of male and female recruits on their first day of service, based on digit combinations of the military identification number. These recruits (annual average of 2000 young men and women) are asked to give a blood sample and to undergo a short interview, which includes socio-demographic data and information on smoking behaviour. Additional information is retrieved from the central computerized military personnel file. The entire process has been approved by the IDF Medical Corps Helsinki committee (IRB), and informed consent is obtained from each participant.

Induction into military duty is compulsory in Israel both for Jewish men and women. About 30% of women are exempt from service, mainly due to religious reasons, whereas among Jewish men 85–90% are recruited.

All data about the cohort was collected prior to appearance of symptomatic cases. A case-control analysis was nested in the prospective collection of data. Cases of duodenal peptic disease among IDF personnel in the serum bank cohort, diagnosed between 1986 and 1995 in military gastroenterology clinics were identified by a meticulous record search. Cases among IDF personnel that were diagnosed in civilian facilities were detected by computer linkage with the financial department of the Medical Corps, which reimburses the diagnostic procedure. All diagnoses were made by endoscopy and included duodenitis of moderate or higher severity, haemorrhagic duodenitis and duodenal ulcer. Twenty-nine cases of duodenal peptic disease were identified. Sample size calculations indicated that 25 cases and 125 controls (i.e. a ratio of five controls per case), with \( \alpha = 0.01, 1 - \beta = 0.9, \) are sufficient, assuming a difference of 40% in \textit{H. pylori} seropositivity between cases and controls.

Each case was individually matched with five controls from the study cohort. Matching criteria included sex, age at induction (within 6 months), country of birth (exact match), country of origin (father's birthplace; or if this was Israel, the paternal grandfather's birthplace; exact match or, if unavailable, the closest match), and education (defined as years of schooling). For one case only four controls met these matching criteria.

Data about smoking status at induction were dichotomized into current or past smokers and non-smokers.

**Laboratory analysis**

Blood samples were allowed to clot at room temperature for one hour, then cooled in a refrigerator for 1–2 hours before being centrifuged. Sera were stored at \(-20^\circ\)C. Serum samples of cases and controls experienced identical conditions throughout the storage period.

The anti-\textit{H. pylori} IgG antibody determinations were performed at the IDF Medical Corps laboratories. All samples were masked for case-control status during the laboratory tests. Quantitative analysis of IgG levels was done by enzyme-linked immunosorbent assay (ELISA) using the Cobas-Core anti-\textit{H. pylori} EIA kit (Hoffman-La Roche Ltd).

**Statistical analysis**

A paired t-test was used for quantitative appraisal of the differences between IgG level of cases and controls, after logarithmic transformation to reduce skew in the distribution of antibody levels. The logarithm of antibody concentration in each case was compared with the average log concentration of the five matched controls.

Antibody levels were dichotomized at 6 U/ml, the cutoff value set by the manufacturer. Odds ratios (OR) were calculated using the Mantel-Haenszel method for matched data and exact CI were computed. Odds ratios were also calculated using conditional logistic regression in order to evaluate possible interactions between seropositivity, ethnicity and length of service.

All statistical calculations were performed using PEPI software—computer programs for epidemiological analysis.

**Results**

The 29 patients with duodenal peptic disease and their matched controls were similar with respect to demographic characteristics due to the stringent matching criteria (Table 1). Smoking rates were similar in cases and controls.

The cumulative distribution of cases and controls by \textit{H. pylori} antibody level showed a higher proportion of controls than cases in the area of negative and low antibody levels (Figure 1). The geometric mean titre of antibodies to \textit{H. pylori} among cases was 18.3 U/ml (95% CI : 9.6–34.7) compared with 6.9 U/ml (95% CI : 5.0–9.1) among controls. Twenty-two of 29 cases and 67 of 144 controls were seropositive. In a matched analysis using the paired t-test, the difference was statistically significant \((P = 0.011)\).

The Mantel-Haenszel OR maintaining matched sets was 3.83 (95% CI : 1.4–10.2, \(P = 0.004)\). A similar OR of 3.6 \((P = 0.01)\) was obtained using a conditional logistic regression model that included the matching variables and smoking status.

The association of anti-\textit{H. pylori} with peptic disease was examined by ethnic origin and according to the time elapsed.

<table>
<thead>
<tr>
<th>Characteristics of patients with peptic disease and of controls</th>
<th>Cases (average ± SD)</th>
<th>Controls (average ± SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age at recruitment</strong></td>
<td>18.63 ± 0.68</td>
<td>18.73 ± 0.74</td>
</tr>
<tr>
<td><strong>Age at diagnosis</strong></td>
<td>20.78 ± 1.33</td>
<td>-</td>
</tr>
<tr>
<td><strong>Origin</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- West(^a)</td>
<td>14 (48%)</td>
<td>69 (48%)</td>
</tr>
<tr>
<td>- Asia</td>
<td>5 (17%)</td>
<td>26 (18%)</td>
</tr>
<tr>
<td>- Africa</td>
<td>8 (28%)</td>
<td>42 (29%)</td>
</tr>
<tr>
<td>- Israel</td>
<td>2 (7%)</td>
<td>7 (5%)</td>
</tr>
<tr>
<td><strong>Gender</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- Male</td>
<td>27 (93%)</td>
<td>134 (93%)</td>
</tr>
<tr>
<td>- Female</td>
<td>2 (7%)</td>
<td>10 (7%)</td>
</tr>
<tr>
<td><strong>Smoking</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>- Current or past regular smokers</td>
<td>9 (36%)</td>
<td>44 (33%)</td>
</tr>
<tr>
<td>- Non-smoker</td>
<td>16 (64%)</td>
<td>88 (67%)</td>
</tr>
</tbody>
</table>

\(^a\) Europe, the Americas, Australia, South Africa.

\(^b\) Smoking status of 4 cases and 12 controls was unknown.
between entry into service and endoscopic diagnosis (Table 2). Interaction was tested by introduction of multiplication terms into conditional logistic models. A higher OR was observed among soldiers of European and American origin (OR = 10.5, 95% CI : 1.7–63.8) as compared with their counterparts of Asian and African extraction (OR = 2.3, 95% CI : 0.6–8.6). This difference, however, was not statistically significant (P = 0.34).

Among soldiers diagnosed ≥2 years after induction the association was stronger (OR = 12.8, 95% CI : 2.1–79.6) than among those diagnosed during the first 2 years of service (OR = 1.6, 95% CI : 0.5–5.6). A test for interaction was associated with borderline statistical significance (P = 0.07).

Discussion
In this prospective study there was a significant association between infection with *H. pylori*, as determined by the presence of elevated concentrations of IgG antibodies to the bacterium, and the risk of duodenal ulcer or duodenitis.

A potential limitation of the study relates to possible under-detection of cases of peptic ulcer disease among personnel during service. It seems unlikely however for this to have biased the outcome. The small proportion of women among the cases of duodenal disease limits inference in this study to men. This is partly due to the smaller number of women inducted into service, and their shorter duration of service, but may also relate to a lower risk among female military personnel. Restriction of the analysis to men resulted in an OR of 4.1 (95% CI : 1.5–11.4), similar to the full data set.

Demonstration of a time sequence with the cause preceding the outcome is considered an essential feature in establishing causality. Several studies of the association of *H. pylori* with gastric cancer have successfully utilized the nested case-control design for this purpose. One previous study has used this approach to assess the relationship between *H. pylori* and peptic ulcer. The prospective design of this study, with blood samples being collected before the diagnosis, reduces the possibility that the disease itself affected the chance of infection with *H. pylori*. Restricting the analysis to cases diagnosed >2 years after blood sampling did not attenuate the association; in fact it increased, strengthening the inference that preceding infection with *H. pylori* plays a causal role in the development of peptic duodenal disease.

The estimate of an OR of 3.8 in early adulthood is similar to the OR of 4 for the presence of anti-*H. pylori* antibodies among middle aged and elderly subjects with duodenal ulcer and their controls, found by Nomura et al. The same methodology was utilized, with a close matching of cases with controls according to demographic criteria. Thus, despite the uncertainty of the point estimate of the OR due to small number of cases in each of the studies, the consistency of the OR suggests they may more closely approximate the true estimate of risk for the development of duodenal peptic disease in the presence of infection with the bacterium. Based on the OR point estimate of 3.8, its 95% CI (1.4–10.2), and the prevalence of *H. pylori* infection in the control group, we calculated the proportion of duodenal peptic disease in this young population attributable to the presence of the organism. The substantial attributable fraction, 56.6%, with respective lower and upper bounds of 15.7% and 81.1%, indicates that *H. pylori* infection is an important factor, although not the only one, in the causation of duodenal ulcer or duodenitis.

The seroprevalence of 46.5% in the population-based controls examined between 1986 and 1995 (albeit matched to the characteristics of the cases and therefore possibly not representative of the IDF) is lower than the 1993 estimate of 72% prevalence among residents aged ≥30 years or older of rural communes in Israel, supporting the possibility of a reduced risk of *H. pylori* infection in later birth cohorts. Although the prevalence of infection is high, most of the infected subjects will probably never develop peptic disease. These data indicate that though *H. pylori* may play an important causal role in peptic disease, it is probably neither a necessary nor a sufficient cause. Additional factors are required, such as the presence of gastric metaplasia in the duodenum, dietary factors, and other environmental or genetic determinants. Ethnicity may serve as a proxy for socioeconomic and economic factors.
other environmental factors that affect \textit{H. pylori} infestation and which were not measured directly. The \textit{H. pylori} carriage was much higher among blacks than among whites in several studies performed in the US.\textsuperscript{6,27,28} Ethnic and racial differences were also described in other countries.\textsuperscript{6,21} Similarly, in Israel, ethnicity is an important factor in the determination of the infection rate with seroprevalence higher in people of African and Asian origin,\textsuperscript{10} as confirmed in our study, where the prevalence in controls of Eastern and Western origin was 54\% and 38\%, respectively. Although the prevalence of antibodies to \textit{H. pylori} is higher among Israelis of Eastern origin, the rates of peptic ulcer disease are not similarly elevated. The rates of peptic disease were actually found to be higher in Israelis originating from Europe or America.\textsuperscript{12,13} This ecological anomaly is consistent with the apparently stronger association of \textit{H. pylori} seroprevalence with the incidence of peptic ulcer disease in young Israelis of Western origin in our study. However, the number of cases in our study was too small for adequate power to detect even strong ethnic interactions. The ethnic differences in the strength of the association suggested by our study require confirmation because they point to an increased prevalence of an unidentified cofactor or constellation of factors (including genetic determinants) associated with increased risk in Jews of Western origin.

In summary, in this prospective seroepidemiological study the presence of serum IgG antibodies to \textit{H. pylori} is related to an increased risk of developing peptic disease of the duodenum. About half of the incidence may be attributable to \textit{H. pylori} infection, indicating the important aetiological role of this bacterial infection in this young population and providing further support for the generalizability of a causal association to varied populations.

References