ABSTRACT • STATEMENT OF THE PROBLEM: Helicobacter pylori plays a major etiologic factor in the pathogenesis of chronic gastritis, peptic ulcer disease, gastric adenocarcinoma, and mucosa associated lymphoid tissue lymphoma. However, most of the infected subjects remain asymptomatic. The aim of this study is to establish fecoprevalence of Helicobacter pylori infection in a convenient non-probabilistic sample of asymptomatic Lebanese children.

METHODS: Four-hundred fourteen children aged between one month and 17 years of different socioeconomic standards were selected for Helicobacter pylori antigen testing in stool. Demographic characteristics, health and nutritional status were obtained through a questionnaire.

RESULTS: Fecoprevalence of Helicobacter pylori infection was 0.21 of whom 28.7% were between 0-3 years, 34.5% between 4-9 years and 36.8% between 10-17 years. Seventy-five (86.2%) of the fecopositive children were from low socioeconomic standards and 12 (13.8%) were from middle to high socioeconomic standards (p < 0.0001). Environmental variables demonstrated higher frequency of fecopositivity in children living in overcrowded houses, lower family income and poor parental education (p < 0.05).

CONCLUSIONS: Helicobacter pylori is prevalent in asymptomatic Lebanese children. Prevention is worthy by improving the levels of education and the standards of hygiene.

INTRODUCTION

Helicobacter pylori (H. pylori) infection is one of the most common chronic bacterial infections among humans worldwide [1-5]. It plays a major etiologic factor in the pathogenesis of chronic gastritis, peptic ulcer disease, gastric adenocarcinoma, and mucosa associated lymphoid tissue lymphoma. However, most of the infected subjects remain asymptomatic [6-9].

World Health Organization estimates indicated that approximately 50% of the world’s population is infected with H. pylori [10]. As high rates of H. pylori infection are associated with low socioeconomic status and educational levels, poor housing and personal hygiene, overcrowding and unhygienic sources of drinking water [11-12], its prevalence is significantly higher in the developing countries than in the developed countries [13].

H. pylori infection is predominantly acquired in early childhood, with the prevalence curve rising with increasing age, and persists throughout life, unless specific treatment is applied [14-15].
Early detection and eradication of the organism can lead to long-term healing of all \textit{H. pylori}-related diseases [16-17].

Many modes of transmission have been discussed. However, person-person transmission including both fecal-oral and oral-oral routes of transmission early in life have been suggested [18-20].

As childhood appears to be the principal period of acquisition of \textit{H. pylori} infection, particularly in developing countries, rational approaches to the prevention and control of active \textit{H. pylori} infection are critically needed [5].

No previous studies are known to have been conducted on the epidemiology of \textit{H. pylori} infection among Lebanese children. The purpose of this cross-sectional survey was to determine the prevalence of \textit{H. pylori} infection, using the non invasive monoclonal \textit{H. pylori} stool antigen test, among different pediatric age groups of a convenient sample of asymptomatic Lebanese children in order to identify the most common determinants for this infection.

\section*{MATERIALS AND METHODS}

The study was conducted between June 1\textsuperscript{st}, 2005 and April 30\textsuperscript{th}, 2006, following approval obtained from the Research and Ethics Committee at Makassed General Hospital.

Initially, a cross-sectional study was carried out, which allowed characterizing the study subjects and establishing the infection fecoprevalence according to gender and age group; later, a comparison was performed between the group of children with positive \textit{H. pylori} stool antigen test and that with negative \textit{H. pylori} stool antigen test.

A convenient non-probabilistic sample was used, i.e., from the day established for the beginning of the research, children aged between one month and 17 years from several Lebanese schools of poor and middle to high socioeconomic standards, along with their day care centers were enrolled in the study. Schools of Lebanese orphanages mainly include children whose families are of poor socioeconomic status. However, schools selected with middle and high socioeconomic status have an annual tuition ranging from 2000 US$ to 4000 US$.

Written approval was obtained from the directors of schools after making several phone calls, sending letters and arranging meetings explaining the aims of this screening survey.

About nine hundred airtight clean stool containers together with questionnaires and informed consents were distributed among the private schools, their day care centers (Figure 1). The children’s parents signed a free consent form after being informed about the objectives of the study.

Children were excluded at enrollment if they had taken antibiotics or acid suppressive agents (proton pump inhibitors, H\textsubscript{2} receptor antagonists or antacids) within 4 weeks prior to testing.

Initially, the parents filled in a questionnaire, designed to obtain demographic characteristics such as: age and gender of the child, educational level of the parents, family income, housing and living conditions [residence, number of rooms in the house, number of household members sharing a bedroom and the source of drinking water]. Toilet practices, feeding habits, any medical history of abdominal pain, persistent vomiting, anemia, anorexia, abdominal distention or any other illnesses, and any family history of peptic ulcer disease, persistent abdominal pain or any other gastrointestinal diseases were assessed as present or absent. Weight and height were measured by the principal investigator and the nutritional status was evaluated according to weight/age, height/age, weight/height ratios, using the National Center of Health Statistics/NCHS curve 2000 as reference; a child was considered to be malnourished when he/she was below the third percentile.

A stool sample was collected from each child, either at home or at school, in the airtight clean container and was preserved in a special place in the fridge at a temperature between 2 and 8 °C. A bus driver brought up the stool samples from the diverse study centers, either on the same day of collection or within 48 hours at the maximum, in a portable fridge at a temperature between 2 and 8 °C to Makassed General Hospital.
**Helicobacter pylori stool antigen test**

*H. pylori* antigen testing in stools was done immediately upon arrival of the samples to the hospital. Therefore, sample handling was in accordance with that recommended by the assay manufacturer (Meridian Diagnostics, Inc., Milano, Italy, Europe).

Each stool sample was tested for the presence of *H. pylori* antigen using Immunocard Stat! HpSA.

The stool antigen test is a rapid immunoassay, based on a lateral flow chromatography technique using monoclonal antibodies for the qualitative detection of *H. pylori* antigens in human stool at a minimum concentration of 64 ng/ml. The sample diluent prevents nonspecific reactions. The strip in the cassette contains, above the sample port, red latex particles bound to the *H. pylori* monoclonal antibody and blue latex particles bound to a control protein. In the presence of *H. pylori* antigens, the red particles will be bound to the antigen during the flow and captured by a line of the same monoclonal antibodies, while the blue particles will be bound by a line of control antibody. The tests were performed at the two local sites according to manufacturer’s recommendations. In brief, a small portion (5 to 6 mm diameter) of stool specimen was transferred into the sample diluent using an applicator stick and mixed properly for 15 to 20 seconds. Thereafter, the tip of the vial was broken off and 4 drops were vertically dispensed into the round window at the lower end of the device. Results were read exactly after 5 minutes. A sample was considered as negative, when only one blue control band appeared, and as positive, when a distinguishable pink-red line was seen in addition to the blue control band. All tests were performed and read by the principal investigator and rechecked by a specialized laboratory technician.

**Statistical analysis**

Chi² test and the Mann-Whitney U test were used for categorical (parental level of education, monthly income, source of drinking water) and binary variables (age, gender, feeding habits, toilet habits). T-tests were applied to test significant differences between *H. pylori* positive and negative children in regard of variables such as family members, number of persons sharing the same room, residential crowding, monthly income. \( P \leq 0.05 \) was considered significant.

**RESULTS**

A total of 414 children were entered into the final data analysis, of whom 212 (51%) were from middle to high socioeconomic schools and 202 (49%) were from poor schools (Figure 1).

The age of the children varied between one month and 17 years. Ninety-five children (23%) were between 0-3 years, 228 children (55%) were between 4-9 years and 91 children (22%) were between 10-17 years.

In regard of gender distribution, 235 children (56.8%) were males; and 179 (43.2%) were females.

Concerning the place of origin, 32.4% lived in Beirut, 38.6% lived in the North, 12.6% lived in the Bekaa, 7.4% lived in the South and 8.9% lived in Mount Lebanon.

Among the 414 children tested, 87 were *H. pylori* positive, with a total fecoprevalence of 21%. There was a statistically significant difference concerning *H. pylori* positive and negative children in regard of variables such as family members, number of persons sharing the same room, residential crowding, monthly income. \( P \leq 0.05 \) was considered significant.

![Figure 2. Distribution of *H. pylori* fecopositivity between low socioeconomic status children and middle to high socioeconomic status children.](image-url)
fecoprevalence between the low socioeconomic standards and middle to high socioeconomic standards (p < 0.0001).

The fecoprevalence was similar in males (21.3%) and females (20.7%) (p = 0.49).

It was seen that the prevalence of H. pylori infection increased with age and the highest prevalence was in children above 10 years of age (28.7% between 0-3 years, 34.5% between 4-9 years and 36.8% between 10-17 years).

H. pylori fecopositivity among different age groups showed statistically significant difference between children from low socioeconomic standards and those from middle to high socioeconomic standards (p < 0.001) (Figure 2).

No significant differences were found between the two groups regarding feeding, toilet and sleeping habits. In relation to housing characteristics, there was significant statistical association between source of drinking water and H. pylori fecopositivity (p < 0.0001).

Table I showed significant differences between positive and negative fecoprevalence in regards of residential crowding, family members, number of persons sharing the same bedroom, monthly income and parental level of education. The highest proportions of infected children were found in houses where two or more persons were sharing the same room (p < 0.001). Prevalence of the infection was inversely related to the education of both parents (p < 0.0001).

There was statistically significant association between nutritional status particularly weight/age and height/age and positive stool antigen test for H. pylori infection.

40.6% (37/91) children with height/age ≤ 10th percentile were H. pylori positive versus 15% (50/323) children with height/age > 10th percentile (p < 0.01). On the other hand, 30% (27/89) children with weight/age ≤ 10th percentile were H. pylori positive versus 18% (60/325) children with weight/age > 10th percentile (p < 0.001).

No statistical association was demonstrated between H. pylori fecopositivity and child’s medical history of abdominal pain, persistent vomiting, anorexia, abdominal distention, anemia, family history of peptic ulcer disease, abdominal pain and any gastrointestinal disease.

<table>
<thead>
<tr>
<th>TABLE I</th>
<th>DETERMINANTS OF HELICOBACTER PYLORI INFECTION AMONG ASYMPTOMATIC CHILDREN</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>H. PYLORI STOOL ANTIGEN TEST</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Positive</strong></td>
<td><strong>Negative</strong></td>
</tr>
<tr>
<td>Number (%)</td>
<td>Number (%)</td>
</tr>
<tr>
<td><strong>SIBLINGS</strong></td>
<td></td>
</tr>
<tr>
<td>≤ 3</td>
<td>5 (13.9%)</td>
</tr>
<tr>
<td>4-6</td>
<td>30 (13.2%)</td>
</tr>
<tr>
<td>≥ 7</td>
<td>23 (28.4%)</td>
</tr>
<tr>
<td><strong>NUMBER OF PERSONS SLEEPING IN ONE ROOM</strong></td>
<td></td>
</tr>
<tr>
<td>1 person / room</td>
<td>2 (8.3%)</td>
</tr>
<tr>
<td>2 persons / room</td>
<td>9 (6.1%)</td>
</tr>
<tr>
<td>3 persons / room</td>
<td>4 (7.0%)</td>
</tr>
<tr>
<td>≥ 4 persons / room</td>
<td>72 (38.7%)</td>
</tr>
<tr>
<td><strong>MONTHLY INCOME</strong></td>
<td></td>
</tr>
<tr>
<td>&lt; 1 million</td>
<td>69 (39.9%)</td>
</tr>
<tr>
<td>1-2 million</td>
<td>6 (15.4%)</td>
</tr>
<tr>
<td>&gt; 2 million</td>
<td>12 (5.9%)</td>
</tr>
<tr>
<td><strong>FATHER’S EDUCATIONAL LEVEL</strong></td>
<td></td>
</tr>
<tr>
<td>Illiterate</td>
<td>38 (413%)</td>
</tr>
<tr>
<td>Elementary</td>
<td>30 (28.3%)</td>
</tr>
<tr>
<td>Intermediate</td>
<td>8 (19.0%)</td>
</tr>
<tr>
<td>Secondary</td>
<td>7 (13.2%)</td>
</tr>
<tr>
<td>University</td>
<td>4 (3.3%)</td>
</tr>
<tr>
<td><strong>MOTHER’S EDUCATIONAL LEVEL</strong></td>
<td></td>
</tr>
<tr>
<td>Illiterate</td>
<td>39 (43.3%)</td>
</tr>
<tr>
<td>Elementary</td>
<td>26 (30.6%)</td>
</tr>
<tr>
<td>Intermediate</td>
<td>13 (25.5%)</td>
</tr>
<tr>
<td>Secondary</td>
<td>4 (7.4%)</td>
</tr>
<tr>
<td>University</td>
<td>5 (3.7%)</td>
</tr>
</tbody>
</table>

* p < 0.05 is considered statistically significant
DISCUSSION

Great variation exists in the prevalence of *H. pylori* infection among different countries and age groups [21-24]. The infection is rare in developed countries once compared to developing countries [25-26]. In the developed world, *H. pylori* infection rate in children ranged from 5 to 15%, whereas in developing countries, it varies between 36 and 82% [9]. In our study, we have detected an overall fecoprevalence of 21% *H. pylori* infection among the asymptomatic children examined. This is in accordance with a study performed in Turkey detecting fecoprevalence of *H. pylori* infection in 25% of asymptomatic children aged from 2-15 years [27]. Sykora et al. also determined a similar rate of *H. pylori* infection in 31% of 91 symptomatic children using endoscopy in Slovakia [28]. However, Cheng et al. observed an *H. pylori* infection prevalence of 13.7% among children and young adults in Taiwan [29].

Endoscopy with concordant results of biopsy based methods (culture, histology and rapid urease test) is considered to be the gold standard for the diagnosis of *H. pylori* infection. However, we choose the stool antigen assay, rather than a number of other possible non-invasive tests for reasons given below.

The 14C-urea breath test gives an excellent performance in both adults and children, but specificity decreases in very young children and collection of exhaled air is difficult in this age group.

Stool samples can be obtained from children without their active collaboration. Another advantage of the test is its possible performance by individual care givers without the need of a laboratory or any equipment. In addition, the test is less costly and time consuming. The result is available within 5-10 minutes [30]. Recently, an enzyme immunoassay based on polyclonal antibodies was developed for detection of *H. pylori* antigen in stool showed large conflicting results [31-34]. In contrast, stool enzyme immunoassay based on monoclonal antibodies showed excellent results with very high sensitivity and specificity. The inter observer agreement was excellent : in 95% of all tests performed in Munich where both independent observers judged either positive or negative [30]. In our study, two investigators independently judged the intensity of the bands with a better inter observer agreement (100%). In order to reduce the false negative results induced by acid suppressive drugs or antibiotics, all children taking such medications during the 4 weeks period prior to testing were excluded [35].

The fecoprevalence of *H. pylori* infection among children in our study showed a meaningful correlation between fecopositivity and age, where older children had a higher prevalence of *H. pylori* infection i.e. the highest prevalence was among the 10-17 years age group (36.8%). On the other hand, prevalence rates in age group younger than 3 years were the lowest (28.7%). This indicates that the prevalence curve of acquisition of *H. pylori* infection occurs during the first 3 years of life then rises progressively with age and seems to be related to the continuous risk of infection throughout life ; thus, determining a cumulative effect on the fecoprevalence curve [36]. The high rate of acquisition in older children might be due to outdoor activities and exposure to potential external sources [21]. Ertem et al. showed that *H. pylori* infection progressively increased from 18.2% under 4 years of age to 63% at 10-12 years of age [16].

In accordance with many previous surveys [2, 5, 15, 27], gender appears to have no effect on the acquisition of *H. pylori* in our subjects.

The acquisition of *H. pylori* infection varies remarkably between and within populations. The age specific prevalence of *H. pylori* is higher in those of lower socioeconomic status whether accessed by income, housing and education. Our results confirmed literature reviews suggesting that the transmission of *H. pylori* microorganism may be facilitated by precarious hygiene conditions and crowded homes in low income families [11, 37-40].

Our results confirmed many studies findings that *H. pylori* infection in childhood was mostly asymptomatic and not associated with specific gastrointestinal symptoms [41-43]. This first cross-sectional survey in Lebanon showed that *H. pylori* is prevalent in asymptomatic children. Low socioeconomic status, poor parental education and tap water drinking source were significant determinants of this *H. pylori* fecoprevalence. As this infection starts early in childhood and increases with age, prevention is worthy by improving the levels of education and the standards of hygiene among orphanages and families of low socioeconomic standards.

ACKNOWLEDGEMENTS

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انتشار البرازوي حاسم للالتهاب باللمحنيات البوابية بدون اعراض عند اطفال لبنان

الموضوع: تعلم المنحنیات البوابية دورا مسيبا هاما في الاعتدال الفعلي المزمن في الترحة الضخمة ومع ذلك فإن اغلب الفضائيين لا تظهر اعراض عليهم. غاية هذه الدراسة انتشار البرازوي للالتهاب باللمحنيات البوابية عند الأطفال اللبنانيين دون اعراض.

المETHOD: دراسة سيربية استقصائية عند 244 طفلا عمرهم أقل من 17 عاما من عدة مناطق لبنانية ومن مستويات اجتماعية مختلفة أجري لهم اختبار مولد الضد لللمحنيات البوابية. النتائج المتعلقة بالعصابات البشرية أخذت بواسطة استبان ملاء الأهل مع موافقتهم لهذه الدراسة.

النتائج: انتشر البرازوي لللمحنيات البوابية التي حصلنا عليها كانت 42.1% منها 66.7% للفتيان عمرهم صغر -3 سنوات و 24.0% للفتيان عمرهم 4-9 سنوات و 21.8% للفتيان عمرهم 10-17 عاما. 87.2% من الأطفال كان الاختيار ايجابيا لللمحنيات البوابية ونسبة اجتماعي متقدم ولكن 21.8% كان النتائج الاجتماعي متوسط أو عالية (احتمال الخطا أقل من 0.01). أظهرت العوامل المتوسطة ارتفاع نسبة الانتشار البرازوي عند القاطنين في منازل مزدحمة بالسكان ومن عائلات حالتهم المادية ضعيفة وثقافة الأهل متدينة (احتمال أقل من 0.05).

الخلاصة: المنحنیات البوابية مزدادة عند الأطفال اللبنانيين بدون اعراض. الوقاية ممكنة بتخصيص ثقافة الأهل وشروط الحياة الصحية.