Original Research Article

Comparison of stool antigen and blood antibody test methods for detection of Helicobacter pylori infection and the risk factors

Abdulrahman S. Naji1, Gamal A. A-Ameri1, Mawhoob N. Alkadasi2, Samira Hanash3, Wahid A.M. Ali3* and Abdusaset A. Zaid4

1Department of Biology, Zabid Education College, Hudaiadah University, Yemen
2Department of Chemistry, Zabid Education College, Hudaiadah University, Yemen
3Department of Microbiology Medical College Taiz University, Yemen
4Post Graduate and Research Centre, Maulana Azad College, Aurangabad, India

*Corresponding author

ABSTRACT

The objective of the study is to compare between stool antigen and blood antibody test method for detection of Helicobacter pylori which associated with several upper gastrointestinal disorders. This study included 100 serum and stool specimens were collected from patients that chosen from different hospitals (Al-Yemen International, Cooperation, Al-Amal, Praise) and the laboratories in city (Abadan, Fidelity) and Countryside (Jabal Habashi - Laboratory friend, Laboratories Sharhab peace, Laboratory Dimna Khadder) containing 68 females and 32 males, their aged ranged from 10 to 80 years. The data were obtained by questionnaire. The stool samples were analysed for H. pylori antigen using Premier Platinum H. pylori stool antigen, enzyme immunoassay kit, while the serum were analysed for IgG antibody using Premier enzyme immunoassay. Results of this study showed that prevalence of the infection increased with age greater than 40 years. Drinking water resources, tea drinking status were asked by a self-administered questionnaire. Results of this study showed that there was a significant correlation between drinking tea also the type of drinking water consumed and H. pylori infection. H. pylori infection showed no significant correlation with sex. In the present study stools specimens and serum samples were subjected to examination for detection of Antigen and Antibody respectively. Antibody was detected in 72 out of the 100 samples tested (72%) whereas Stool Antigen was positive in 49 (49%) out of 100 samples tested. Majority of cases were in the age group of 40-49 years of female preponderance. The present study reveals that serology showed a slightly greater number of positive cases.

Keywords
Stool antigen and Blood antibody, H. Pylori, Risk factor

Introduction

Helicobacter pylori (H. pylori), a microaerophilic, flagellated, curved or spiral, gram-negative bacterium, selectively colonizes the human stomach. Its infection is widespread throughout the world, and is present in about 50% of the global human population; with 80% in developing countries and 20 - 50% in industrialized countries. It is the major cause of gastritis, that plays a key role in the etiology of peptic ulcer and is a risk factor for gastric carcinoma[1]. In view of the excitement and
interest generated by the link between H. pylori and gastric abnormalities, different investigators have thought to determine a role for the infection in a variety of non-gastrointestinal tract disorders. This is despite our current understanding that H. pylori infection is confined to gastric mucosa [2, 3].

Several studies have pointed to a possible role of H. pylori infection in the pathogenesis of various extra gastric diseases involving dermatologic conditions [4-8]. However, dermatologists seem to be unaware of the impact H. pylori may have on cutaneous pathology [9, 10]. Presently, its role has been established in chronic antral gastritis, duodenal ulcer, chronic gastric ulcer, dyspepsia, gastric cancer and gastric lymphoma. World Health Organization added H. pylori to its list of known carcinogens [11].

The diagnosis of H. Pylori gastritis can be made through many laboratory tests. The techniques are divided into two groups the invasive and non-invasive tests [12]. All invasive test methods are based on endoscopic examination during which biopsy specimens are obtained for direct (histological analysis, isolation) or indirect (urease test) diagnosis of H. pylori infection.

Noninvasive methods reveal the presence of H. pylori by measuring the activity of urease (ureabreath test), then by confirming the presence of antibodies in the serum. Meanwhile, other noninvasive tests were also evaluated in several studies including, detection of H. pylori antigens in stool and presence of H. pylori in the saliva. Stool antigen tests have recently been welcomed with great expectations as they are convenient to the patients and can be easily performed even in small laboratories [13, 14]. However, the accuracy of stool antigen tests in different clinical situations and outside of controlled studies is a matter of concern [15-17].

Serological studies performed in the Indian subcontinent indicate a prevalence of 22% to 57% in children under 5 years of age, and increasing to 80% to 90% by the age of 20 years and remaining constant thereafter [18-20]. However, serological tests are reported to be unreliable for the diagnosis of H. pylori since they may return false negative results up to 60 days after infection and remain positive for a considerable time after eradication, but serology for IgG, against H. pylori may play an important role in decreasing the need for endoscopy provided the cut-off values must be determined for easy assay based on the prevalence of antibodies in the population [21].

The present study was therefore planned for comparative evaluation of stool antigen test and blood antibody test methods detection by a commercially available kit for diagnosis of H. pylori infection in cases of dyspepsia and risk factors.

Materials and Methods

Study population

A cross-sectional sero epidemiologic study was carried out in Taiz city. The study period was April 1st, 2014 to June 29, 2014. The total study population was 100 patients that chosen from different hospitals (Al-Yemen International, Cooperation, Al-Amal, Praise) and the laboratories in city (Abadan, Fidelity) and Country side (Jabal Habashi - Laboratory friend, Laboratories Sharhabpeace, Laboratory DimnaKhadder). The samples have been collected randomly from patients and the total sample was 100. The history of the patients was recorded in a predesigned data collection sheet.
Questionnaire

A trained physician interviewed each volunteer and completed a detailed questionnaire. The questionnaire was designed to obtain demographic data and socioeconomic status was also assessed.

Sample size

One hundred from randomly selected eligible patients subjected to examination blood and stool sample. The stool samples were analysed for H. pylori antigen using Premier Platinum H. pylori a stool antigen, enzyme immunoassay kit, while the serum were analysed for IgG antibody using Premier enzyme immunoassay kit (Meridian Bioscience, Inc. Cincinnati, Ohio). Positive results are samples with spectrophotometric optical density (OD) of ≥0.160 at 450nm wavelength, while negative results are OD< 0.140 at same wavelength. Equivocal results are results with OD=0.140 and < 0.160.

Results and Discussion

The current investigation included 100 patients their ages ranged between 10 and 80 years, with mean age of 36.23 + 6.317 years. The prevalence of H. pylori infection was defined according to the different demographic data of the patients, including gender and age. Among the 100 patients who completed data, the highest positive result was found in the age group of 41-80yr (80%) and (60%) which detected by blood antibodies test and stool antigen test methods respectively. There were no significant statistical results related to age or sex to be considered as a risk factor (table1). The tested cases included 32 males (32%) and 68 females (68%). A female predominance of 68/100 (68%) was observed, as compared to that of 32/100 (32%) in male. Most of the patients presented in the active age group of 19 to 40 years (Fig.1).

Risk factors were assessed in the current study among the 100 patients with gastric affection including drinking tea and drinking water. The prevalence of infection among patients who usually consumed tap water or well water during livelihood 73.7% compared with 47.9% among those who usually consumed filtered water. The drink of tea had a strong effect on the prevalence of H. pylori infection. The prevalence of infection among patients who drink tea was 72.8% compared with 68.4% among those who not drink tea table3.

72 patients were found positive by testing their blood for H. pylori antibody representing 72% and 28% as negative (Fig.2) and (Table4). The blood antibody test had a slightly higher proportion of positives in the female patients 52(76.4%) compared with the male which represented 22 (68.7%). The differences observed in age and sex were however not statistically significant. (Table1).

Among patients, 49 of the 100 patients (49%) were diagnosed as positive for H. pylori infection, and 51 (51%) as negative, which determined by stool antigen test (table 4) and (Fig. 3). The stool antigen test had a slightly higher proportion of positives in the female patients 34(50%) compared with the male which represented 14 (43.7%). The differences observed in age and sex were however not statistically significant(Table1).
H. pylori have been designated as key organisms in the etiology of chronic gastritis, peptic ulcers and gastric cancer[22,23] and their suppression and elimination has been considered the gold standard therapy for infectious gastric diseases. Various diagnostic tests for H. pylori infection may have false negative results and the use of multiple tests may help to provide a more accurate diagnosis of H. pylori infection.1

The present study was undertaken to compare the different diagnostic methods, with non-invasive techniques (blood antibody and stool antigen test methods) for detection of H. pylori infection in patients with Acid peptic disease. The current study included 100 patients with upper GIT symptoms proven to be with gastric affection in the form of Acid peptic disease, that chosen from different hospitals (Al-Yemen international cooperation, Al-Yemen International, Cooperation, Al-Amal, Praise) and the laboratories in city (Abadan, Fidelity) and Countryside (Jabal Habashi - Laboratory friend, Laboratories Sharhabpeace, Laboratory Dimna Khadder). Ages of the study participants ranged between 10 and 80 years, with mean age of 36.23 ± 6.317 years. There was association between 41-80 years age group and infection density, though it did not reach statistical significance (Table 1). These results were comparable to European studies which reported correlation between age and gastric affection [24,25]; however, these studies found increased prevalence of gastritis and H. pylori colonization with increasing age. Furthermore Jones et al 9 observed that more complaints of dyspepsia in the older age group and suggested that the older subjects were probably more concerned about their health or were afraid of more serious underlying diseases. They further concluded that this may be of advantage, in that severe diseases could be detected early and management instituted promptly. Regrettably, there was no study on stool antigen test from developing countries of the world to compare our study with. Our study also agreed with Nulty which found that the more likely age of infection in patients over 50 years old (42%) than in younger patients (21%), another group of Liston cited by Nulty, found that (31.7%) of elderly patients with seropositive result had no evidence of active infection determined by endoscopy and urease test. Older patients are more likely to have developed atrophic gastritis and H. pylori can not readily colonize this type of gastric mucosa [26]. It was recognized that prevalence of H. pylori infection increase with age in a symptomatic persons in developed countries and this tend to plateau at around the age of 60 years, related to socioeconomic status and ethnicity [27-31].

In the present study higher level of H. pylori infection was observed among female patients in both blood antibody and stool antigen test methods (76.4% and 50%) rather than males (68.7% and 43.7%) respectively as shown in table 1. The slight preponderance of females in our study could reflect a greater consciousness in the issue of their health, or their ready presentation in the hospital could be due to emotional/psychosomatic disorders, which tend to be commoner among the female gender. In contrast to this study, Ihezuein the North-Central part of Nigeria, found that most of the dyspeptics (60%) were males and symptoms were commoner in those below the age of 40 years[32].

In the present study, found that tea consumption is considered as a risk factor for H. pylori infection. The frequency of H. pylori infection in the tea consumer (72.8%) and non-tea consumer (68.4%) was
significantly different in our study, this is compatible with results obtained by Endoh which have analyzed the relation between tea consumption and *H. pylori* infection suggested that *H. pylori* infection significantly rose with tea consumption [33].

In the current study, the *H. pylori* infection rate was higher in patients who drank well water (73.7%) as compared to patients who used bottled water (47.9%). This result is in agreement with other studies in developed and developing countries. They implicated the type of drinking water during childhood as the main risk factor for *H. pylori* infection. The microorganism is transmitted by the fecal-oral routs in the infected water to the child and persists through life and as the results showed that the type of drinking water during adulthood does not affect the infection rate.

In a study in Leipzig, Germany, which consisted of a self administered or parent-completed questionnaire (age-dependent), eliciting information on lifestyle habits and their use/drinking the well water as well as the *H. pylori* infection. A total of 91 subjects (44 users of *H. pylori* positively and 47 negatively tested wells) were screened for their *H. pylori* status. The group was comprised of 42 males and 49 females, i.e., 73 adults and 19 children under the age of 18 (mean age 39.5 years with a range between 3-80 years). Logistic regression analyses identified the drinking of well water as the significant risk factor for a positive colonization status [Odds Ratio (OR)=8.3; 95% confidence interval (95% CI) 2.4-29.0; P<0.001]. Water supplies have been identified as possible reservoirs to acquire the bacterium [34].

In the present study the positive *H. pylori* infection which detected by blood antibodies test was seen in 72% patients. These results were comparable with Luthra GK [35] who found that the positive result was 63%, whereas another study conducted by Satti SA et al. [36] who showed 87.7%. This difference is because of unawareness of Hygiene and old study.

We also found that the positive *H. Pylori* infection which detected by stool antigen test was seen in 48% patients which is lower than Chisholm SA et al. [37,38] and Gisberg JP [39], this was due to more antigens present in stool in above study and they have more advance than us in diagnostic methods. Our study showed lower number of positive cases which may be due to insufficient amount of antigen in the stools. Difference of *Pylori* infection which detected by stool antigen test from other study is due to the difference of climate and may be quality of kit. This is also comparable to study by MahirGulcan et al [40] who reported positive result in 37 out of 80 children (Table 4) which was comparatively lower than this study.

The blood antibodies test method showed greater number of positive cases (72%) than the stool antigen tests method (48%) which may be due to past infection. This is comparable to the study by Arora et al, who reported greater case detection by serology than by conventional tests. The patchy distribution of organism in the gastric mucosa may have resulted in a lower value for biopsy based test. Another factor could be the presence of gastric atrophy and intestinal metaplasia that are hostile to *H. pylori* [41].
Table 1: Show the prevalence of infection in different age groups.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Blood test</th>
<th>Stool test</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Positive</td>
<td>Negative</td>
</tr>
<tr>
<td></td>
<td>No.</td>
<td>%</td>
</tr>
<tr>
<td>Age</td>
<td></td>
<td></td>
</tr>
<tr>
<td>10-18</td>
<td>9</td>
<td>75%</td>
</tr>
<tr>
<td>19-40</td>
<td>47</td>
<td>69.1%</td>
</tr>
<tr>
<td>41-80</td>
<td>16</td>
<td>80%</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>22</td>
<td>68.7%</td>
</tr>
<tr>
<td>Female</td>
<td>52</td>
<td>76.4%</td>
</tr>
</tbody>
</table>

Fig.1 Sex distribution among patients

Table 2: Life style variables

<table>
<thead>
<tr>
<th>Variable</th>
<th>Positive</th>
<th>Negative</th>
<th>Positive</th>
<th>Negative</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No.</td>
<td>%</td>
<td>No.</td>
<td>%</td>
</tr>
<tr>
<td>Drinking tea</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>59</td>
<td>72.8%</td>
<td>22</td>
<td>27.2%</td>
</tr>
<tr>
<td>No</td>
<td>13</td>
<td>68.4%</td>
<td>6</td>
<td>31.6%</td>
</tr>
<tr>
<td>Drinking water</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>well water or tap water</td>
<td>28</td>
<td>73.7%</td>
<td>10</td>
<td>26.3%</td>
</tr>
<tr>
<td>Filtered water</td>
<td>34</td>
<td>47.9%</td>
<td>38</td>
<td>52.1%</td>
</tr>
</tbody>
</table>

Stool test                  | Blood test |          |          |          |
| Positive | Negative | Positive | Negative | Positive |
| %       | No.      | %        | No.      | %        | No.      | %        | No.      | 51%  | 51 | 49% | 49 | 28% | 28 | 72% | 72 |
Table 3: Comparative evaluation of Blood test and Stool Test for diagnosis

<table>
<thead>
<tr>
<th></th>
<th>Stool test</th>
<th>Blood test</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Negative</td>
<td>Positive</td>
</tr>
<tr>
<td></td>
<td>%</td>
<td>No.</td>
</tr>
<tr>
<td>No.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>51%</td>
<td>51</td>
<td>49%</td>
</tr>
</tbody>
</table>

Fig. 2: Show the percentage of positive and negative *H. pylori* infection diagnosed by blood test.

Fig(3): Show the percentage of positive and negative *H. pylori* infection diagnosed by Stool antibody test.
References


[33] Iliezue CH, Oluwole FS, Onuminya JE, Okoronkwo MO. Dyspepsia


