Diagnostic Accuracy of Serology Testing for Helicobacter Pylori in Perforated Peptic Ulcer


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Abstract

Background: To compare H. pylori serology testing with histopathological biopsy, in patients with perforated peptic ulcer.

Methods: This cross sectional study was performed to document the diagnostic accuracy of H. pylori serology in patients with perforated duodenal ulcer. Perforated peptic ulcer was diagnosed on the basis of history, clinical findings and presence of free gas under the diaphragm on an x-ray. All individuals fulfilling inclusion criteria underwent laparotomy. Blood samples were sent for H Pylori serology by enzyme linked immunosorbent assay. At the same time, gastric antral biopsies were taken for histopathology for H. pylori.

Results: Sstudy included 191 patients with diagnosed perforated peptic ulcer at laparotomy. Majority (70.2%) were males. The mean age was 41.72 years. In majority the perforation was seen along the first part of duodenum (76.4%). H. pylori serology was positive in 69.1% patients. One hundred and thirty nine (72.8%) patients had positive histopathology for H pylori. Out of these, 92.8% also had a positive H. pylori serology. Fifty two (27.2%) patients had negative histopathology for H. pylori. Out of these 5.8% had a positive serology test. One hundred and twenty nine were true positive i.e both had positive histopathology and serology and 52 were true negative i.e., they had both a negative serology and a negative histopathology. The results were therefore correct in 94.76%. The overall accuracy of serology in comparison to histopathology was 94.76%. The calculated sensitivity, specificity, positive predictive value and negative predictive value of serology in comparison to histopathology was 92.8%, 94.5%, 97.7% and 83.9% respectively.

Conclusion: A high prevalence of H. pylori infection was found in patients with perforated peptic ulcers. Overall accuracy of serology in comparison to histopathology was 94.76%.

Serological testing is a useful noninvasive method for the diagnosis of Helicobacter pylori infection.

Key Words: Helicobacter pylori; perforated peptic ulcer;

Introduction

Erosive injury of the gastro duodenal mucosa leading to peptic ulcer occurs secondary to destruction of the protective layers of the gastric mucosa by a variety of etiologies. The most important factor is Helicobacter pylori, commonly presenting as a chronic infection found worldwide in patients of perforated peptic ulcer. Other causes include prolonged use of non-steroidal anti-inflammatory drugs, immunosuppressive medications, smoking, alcohol abuse and psychological stress. In the East, infection with H.pylori occurs at a younger age while in the West, peak incidence is more in the adult age groups. Reported incidence varies from 90% of cases of duodenal ulcer (DU) and over 70% of gastric ulcers.

Among its several complications, including chronic gastritis, atrophic gastritis, MALT lymphoma, the commonest complication in the surgical emergency is perforation of a peptic ulcer. After resuscitation and initial investigations, the patient is prepared for emergency laparotomy. Once the diagnosis is confirmed, an omental patch repair (Graham’s patch) followed by H.pylori eradication therapy is suggested.

Considering the significant mortality and morbidity associated with perforated peptic ulcer disease and the obvious role of H Pylori infestation in its pathogenesis, investigations to detect H.pylori are gaining obvious and due attention. These can be either invasive or endoscopic, including the rapid urease test, histology, smear cytology and culture; or non-invasive tests, like C14 or C13 urea breath test and serology. ‘Test and treat’ strategy that involves non-invasive testing without endoscopy and eradication therapy in patients is effective in management of peptic ulcers.
This calls for a simple, reliable and non-invasive diagnostic test for H. pylori infection in clinical practice. At present histopathology and biopsy however requires endoscopic biopsy of gastric mucosal tissue that is expensive, inconvenient for the patient and available only at specialized centers. Moreover, because of a patchy distribution of H. pylori in the gastric mucosa, biopsy tissue examination may yield false negative results. Serological tests that detect anti-H. pylori IgG antibodies are non-invasive, less expensive, not influenced by sampling error, and less likely to be confounded by suppression of H. pylori infection by colloidal bismuth, proton pump inhibitors, or antibiotics. The availability of such a diagnostic test that is non-invasive, cheap and rapid for detection of H. pylori in peptic ulcers is important since early institution of eradication therapy can help in curing this potentially disastrous condition. If serology is found to be as valid and accurate as histopathology in diagnosis of H. pylori, this can replace biopsy and histopathology since it is a rapid, cheap and non-invasive alternative.

**Patients and Methods**

Total 191 patients both male and female between 20 to 80 yrs of age (mean age was 41.72\(\pm\)14.96 years) were included in the study. Among them 134 (70.2\%) were males and 57 (29.8\%) were females. Patients who were on, or had already received eradication therapy before laparotomy, those with any other additional cause of ulcer disease including gastric and duodenal carcinomas diagnosed mainly by history taking, clinical examination and computed tomography if suspected with CA and patients on chronic NSAID use for treatments of the various arthritics, chronic alcoholism and other major known causes of peptic ulcer disease were not included in the study. Patients presenting with an acute abdominal condition were assessed by their history and clinically by the presence of guarding, rigidity on their abdominal examination. They were subjected to relevant radiology to confirm the diagnosis of perforation with the presence of free gas under the diaphragm. During this work up, relevant base line investigations were sent after which management was started and they were put on nasogastric aspiration, parenteral fluids and antibiotics and were investigated for fitness for general anesthesia and surgery. Consent for surgery and inclusion in the study, were taken. Laparotomy was performed under general anesthesia and once perforated peptic ulcer was confirmed per-operatively, the blood sample was sent for H. pylori serology. Serum samples were tested with a commercially available enzyme linked immunosorbent assay (ELISA) kit for H. pylori IgG.

At the same time, gastric antral biopsies were taken for histopathology for H. pylori with Yeoman Biopsy Forceps, 15" (38.1 cm) shaft, 3.5 X 10 mm bite, with basket (MX Surgical, Integra Miltex) through the perforation site for Giemsa staining. At least 6-8 biopsy samples were obtained. Histological examination of tissue biopsy samples (was taken from four different parts of the Antrum) permitted detection of the bacterium together with evaluation of tissue damage reported by histopathology.

**Results**

A total of 191 patients with diagnosed perforated peptic ulcer at laparotomy were included in the study which was conducted from April 2012 to Nov 2012 in Surgical Department, Pakistan Institute of Medical Sciences Islamabad. Among 191 patients, majority (70.2\%) were males. The age ranged from 20 to 80 years. The mean age was 41.72 years. Majority of the perforations were found along first part of duodenum (Table 1) Eighty three (43.3\%) patients presented within11 to 24 hours of perforation(Table 2) .H pylori serology was positive in 132 (69.1\%) patients. The mean age of the patients with a positive serology was 43.8±15.6 years whereas the mean age of the patients with a negative serology was 37.05±12.1 years; this difference was statistically significant; p=0.004. One hundred and thirty (72.8\%) patients had positive histopathology for H pylori, out of these 129 (92.8\%) also had a positive H pylori serology and 10 (7.2\%) had a negative serology. 52 (27.2\%) patients had negative histopathology for H pylori. Out of these 3 (5.8\%) had a positive serology test and 49 (94.2\%) had a negative serology. 129 were true positive i.e both had positive histopathology and serology and 52 were true negative i.e they had both a negative serology and a negative histopathology. The results were therefore correct in 181 (94.76\%) cases. Hence the overall accuracy of serology in comparison to histopathology was 94.76\%. The calculated sensitivity of serology in comparison to histopathology was 92.8\%. The calculated specificity of serology in comparison to histopathology was 94.5\%. The calculated positive predictive value (PPV) of serology in comparison to histopathology was 97.7\%. The calculated negative predictive value (NPV) of serology in comparison to histopathology was 83.9\%.
Pakistan is a developing country.

Performance of the ELISA

Serological tests depend on the

Table 1: Perforated peptic ulcer- Sites

<table>
<thead>
<tr>
<th>Site</th>
<th>No(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>First part of duodenum</td>
<td>146(76.4)</td>
</tr>
<tr>
<td>Pre-pyloric region</td>
<td>27(14.1)</td>
</tr>
<tr>
<td>Lesser curvature</td>
<td>18(9.4)</td>
</tr>
</tbody>
</table>

Table 2: Perforated peptic ulcer- Time of presentation

<table>
<thead>
<tr>
<th>Time (Hours)</th>
<th>No(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>11-24</td>
<td>83(43.3)</td>
</tr>
<tr>
<td>0-10</td>
<td>70(36.7)</td>
</tr>
<tr>
<td>25-60</td>
<td>32(16.7)</td>
</tr>
<tr>
<td>&gt;61</td>
<td>6(3.1)</td>
</tr>
</tbody>
</table>

Table 3: H. pylori in perforated Ulcer- Summary of Histopathology and Serology results

<table>
<thead>
<tr>
<th>Histopathology</th>
<th>Serology</th>
<th>No</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>Positive</td>
<td>129</td>
<td>92.8</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
<td>10</td>
<td>7.2</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>139</td>
<td>100.0</td>
</tr>
<tr>
<td>Negative</td>
<td>Positive</td>
<td>3</td>
<td>5.8</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
<td>49</td>
<td>94.2</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>52</td>
<td>100.0</td>
</tr>
</tbody>
</table>

Discussion

More than 95% of patients suffering from duodenal ulcers and about 70-80% of patients with gastric ulcers are H. pylori positive.11-13 ‘Test and treat’ strategy that involves non-invasive testing without endoscopy and eradication therapy in young patients is effective in management of dyspepsia.12,13 This calls for a simple, reliable and non-invasive diagnostic test for H. pylori infection in clinical practice. At present there is no single test for H. pylori that can be used as the ‘gold standard’.14 Culture, rapid urease test and histology require endoscopic biopsy of gastric mucosal tissue that is expensive, inconvenient for the patient and available only at specialized centers. Because of a patchy distribution of H. pylori in the gastric mucosa, biopsy tissue examination may yield false negative results.15 Serological tests that detect anti-H pylori IgG antibodies are non-invasive, less expensive, not influenced by sampling error, and less likely to be confounded by suppression of H. pylori infection by colloidal bismuth, proton pump inhibitors, or antibiotics.16

Serological tests though widely used, cannot differentiate a current infection from a past exposure.17 Performance of serological tests depends on the antigen preparation used, and as H pylori strains differ among geographic locations, local validation of the test is necessary. Pakistan is a developing country with a high prevalence of H pylori infection and peptic ulcer. Data regarding H. pylori infection in perforated peptic ulcers are conflicting.18-19

Since its discovery many tests have been designed for diagnosis of H. pylori. But no test is accurate enough to be the ‘gold standard’. Serological tests are widely used for non-invasive diagnosis but a positive serological test does not mean active infection.20 The commercial ELISA that we evaluated was very sensitive but less specific. Performance of the ELISA kit varies in different populations. Hung et al concluded that a quantitative ELISA test is suitable for the diagnosis of H. pylori infection in patients with atrophic gastritis because of its excellent sensitivity. In patients with atrophic gastritis, all invasive and noninvasive tests for the diagnosis of H. pylori infection have their restrictions because the bacterial load of H. pylori decreases gradually during the progression of gastric atrophy, and bacteria are unevenly distributed in the stomach. In cases with extensive intestinal metaplasia, H. pylori can disappear completely.21 If H. pylori infection is patchy or if the number of bacteria is low, invasive diagnostic tests based on gastric biopsies can yield inaccurate results because of sampling errors.

Serological testing is a useful noninvasive method for the diagnosis of Helicobacter pylori infection. It is easy for patients to accept the test because of its noninvasiveness, and the results can be quickly obtained. Furthermore, this assay is a global test that evaluates the entire stomach. Therefore, potential sampling errors can be avoided.22 The sensitivity of enzyme-linked immunosorbent assay (ELISA)-based serological tests ranges between 90% and 97%, and the specificity ranges between 50% and 96%.23,26 The sensitivity and specificity of serological tests mainly depend on the nature of the antigenic materials used. In addition to the antigens used, the presence of atrophic gastritis is also one of the important factors that influence the test’s accuracy.

A “gray zone” result is a significant limitation of serological tests. H. pylori infection induces mucosal inflammation in the stomach. Infected patients have shown a wide variety of systemic antibody responses, thereby leading to several indeterminate results in serological tests. In cases with indeterminate results, other tests should be performed to determine the status of H. pylori infection. In addition, the accuracy of serological tests might vary between different races.
and geographic regions, possibly due to different antigenic properties of local bacterial strains and antibodies of commercial kits used for the diagnosis of H. pylori infection. The usefulness of a serological assay should be assessed in a local setting. In conclusion, the sensitivity of biopsy-based tests for the diagnosis of H. pylori infection decreases with the progression of atrophic gastritis. Quantitative ELISA is a noninvasive and sensitive test for detection of H. pylori infection in subjects with atrophic gastritis, but this test is less specific in the presence of atrophic gastritis.

Conclusion
1. Serological testing is a useful noninvasive method for the diagnosis of Helicobacter pylori infection. It is easy for patients to accept the test because of its noninvasiveness, and the results can be quickly obtained. This assay is a global test that evaluates the entire stomach.
2. A combination of at least 2 tests (e.g. serology plus urea breath test) is necessary in clinical practice to diagnose H. pylori infection accurately in patients with atrophic gastritis or intestinal metaplasia.
3. If discordant results of the tests exist, another sensitive test (e.g. histology) can be performed to determine the status of H. pylori.

References