Helicobacter pylori Specific IgE In gastric biopsies versus serum IgM and IgG Specific antibodies in dyspeptic patients

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Abstract

Background: The immune response to *Helicobacter pylori* is a versatile group of mechanisms involving responses that are both protective and damaging to the host. *H. pylori* can directly bind to mast cells which produce pro-inflammatory factors which migrate and accumulate in the gastric mucosa. It was found that infection with *H. pylori* is accompanied with increased total IgE and positive IgE specific for *H. pylori* in sera of some dyspeptic patients.

Aims of the study: This study aimed to investigate the presence of IgE specific for *H. pylori* in gastric biopsies from dyspeptic patients regarding their gender.

Patients and methods: Seventy six (76) adult patients from both genders were included in this study. Inclusion criteria included dyspeptic patients require upper gastrointestinal endoscopic examination. Patients were attending Gastrointestinal Endoscopy Unit at Baghdad Teaching Hospital during the period from December 2014 to May 2015. Gastric biopsies from patients with dyspepsia were investigated for IgE specific for *Helicobacter pylori* using ELISA test. Sera from the patients were tested for IgM and IgG specific for *H. pylori* using ELISA test.

Results: Study revealed that 75% of patients with serum positive IgM specific for *H. pylori* were showing positive IgE specific for *H. pylori* in their homogenates of their biopsies. While Sixteen (16) out of 60 (26.7%) positive *H. pylori* IgG patients were showing positive IgE specific for *H. pylori* in homogenates of their biopsies.

Conclusion: The current study concluded that IgE specific for *H. pylori* undergo release locally in gastric mucosa of dyspeptic patients infected with *H. pylori*.

Key Words: *H. pylori*, IgE, immunoglobulin.

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Introduction

*Helicobacter pylori* colonize the stomach, survive acidic pH of the lumen and burrow into the mucus to reach stomach's epithelial cell layer. The survival of *H. pylori* in the acidic stomach is dependent on urease, and it would eventually die without this enzyme. The innate and the adaptive immune responses lead to damaging leukocytes (PMN), T cells, macrophages, and plasma cells. Th1 and Th2 cells cross-regulate one another through their mediators. Among cells that infiltrates the gastric mucosa is Th17 which are CD4+ T cells associated with infections and inflammation. IgE-positive plasma cells show significant growth in gastritis patients positive for *H. pylori*, and *H. pylori* induced gastritis is accompanied by an increase in IgE production.
It was found that infection with *H. pylori* is accompanied with increased Total IgE and positive IgE specific for *H. pylori* in sera of some dyspeptic patients. This study aimed to study IgE specific for *H. pylori* in gastric biopsies from patients with dyspepsia in contrast to serum IgM and IgG specific for *H. pylori* using ELISA test.

**Patients and Methods:**

1. **Patients:** Seventy six (76) dyspeptic adult patients from both genders were included in this study. Patients were attending Gastrointestinal Endoscopy Unit at Baghdad Teaching Hospital during the period from December 2014 to May 2015. All patients were examined by senior physician and the endoscopic diagnosis was done by the consultant. Inclusion criteria included dyspeptic patients require upper gastro-intestinal endoscopic examination and a data sheets were reported according to a questionnaire for each patient including name, gender, age, smoking, alcohol intake and family history of chronic dyspepsia or peptic ulceration.

2. **Specimens:**

   A. **Biopsies:** Tissue biopsy specimen was placed in 1 ml of sterile normal saline and preserved at -20°C for homogenate preparation to be used for ELISA tests.

   B. **Blood:** Three ml of venous blood were taken in a dry sterile tube, serum was pooled and kept at -20 °C to be used for ELISA tests.

3. **Lab investigations:**

   A. **Homogenate preparation:** Homogenate suspension was prepared from each biopsy specimen using special kit (Bio vision, USA) following instructions of the manufacturing company.

   B. **Investigations for homogenate:** Each homogenate was tested for *H. pylori* specific IgE using ELISA test.

**H. pylori specific IgE:**

1. **Preparation of bacterial antigen discs:** *H. pylori* specific protein antigen from ( Mybiosourse, USA ) was employed to prepare antigen discs to be used for ELISA test as below:

   Whatman blotting papers number 1( NO 1) were used to get 0.5 cm diameter paper discs using paper puncher. These discs were sterilized by UV light illuminator for overnight. sterility test was done for sample from these disc. One hundred (100) sterile discs were impregnated with 1.5 ml from above mentioned *H. pylori* antigen to get antigen concentration 17 microgram for each disc. Discs were dried in incubator at 37°C for 40 minutes and kept in sterile test tube at 4°C to be used for IgE specific ELISA test within 3 days.

2. **Helicobacter pylori specific IgE ELISA test:**

   An enzyme linked immunosorbant assay (ELISA) method was used for the specific IgE determination in homogenate of each biopsy from each patients using *H. pylori* antigen impregnated discs as described by.

3. **Serological tests:** Serum IgM and IgG specific for *H. pylori* tests were done for each serum specimen using special ELISA kit for each test (Diagnostic Automation, incu, USA).

4. **Statistical analysis:** Statistical Descriptive analysis (Mean, SD, SE and percentage) was done to describe data, two independent samples T test, (F-test), correlation analysis were applied.

**Results:**

1. **H. pylori IgM:** Forty two (42), (55.26%) patients were showing positive *H pylori* IgM in their sera while (34) , (44.47%) patients showed negative IgM results.
2-H. pylori IgG:
Positive IgG for *H. pylori* was found in (51)(67.10%) patients while (25)(32.90%) patients showed negative results.

IgM and IgG regarding gender of patients:
Higher positive IgM and IgG for *H. pylori* were found in males, (54.75%) and (64.71%) for each respectively (p<0.05).

Specific IgE and *H. pylori* IgM:
Positive IgE specific for *H. pylori* patients results were seen more in IgM positive patients (Fig.1). Positive correlation between IgM in serum and IgE specific for *H. pylori* in homogenate (R=0.255, P=0.028).

Specific IgE and *H. pylori* Serum IgG:
Sixteen(16) out of 60 positive *H. pylori* IgG patients were showing positive IgE specific for *H. pylori* in their homogenates. While eight (8) out of (16) patients with negative *H. pylori* IgG showed positive IgE specific for *H. pylori* (table-1).

![Fig.1 IgE specific for *H. pylori* result in homogenate regarding IgM](image-url)

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IgE specific for \textit{H. pylori} regarding gender of patients:

Although males showed more positive IgE specific for \textit{H. pylori} in their homogenates than females, 14 (28.6\%) and 10 (17 \%) for them respectively, but this difference was non-significant (P>0.05).

**Discussion:**

Higher IgM and IgG positive results were seen in sera of male patients than females, this was also observed by\cite{14}, while discrepant results were found by\cite{15} who found that non-significant difference was found between males and females. While \cite{16} found equal infection males and females. This variability in the rates among different studies might be attributed to the different demographic distribution of bacteria, previous antibiotic consumption and different socioeconomic status.\cite{17,18} Gender related factors may affect immune status of an individual due to immune modulation effect of sex hormones in both genders.\cite{19}

In this study, higher number of patients with positive \textit{H. pylori} Specific IgG than those with positive IgM results. This might be attributed to the late consultation of patients to their physicians and this delay gave enough time to advanced immune response leading to secondary immune type which is characterized by lower IgM and higher IgG titers.\cite{20} That is why a patient develops a detectable serum \textit{H. pylori}-specific IgG response to chronic infection. Mast cells have been reported to involve gastric mucosal damage in patients with gastritis, especially those infected with \textit{H. pylori}\cite{21}, specimens showed the increased density of mast cells after the eradication therapy.\cite{22} In the current study positive IgE specific for \textit{H. pylori} in biopsies from positive \textit{H. pylori} IgM and IgG patients. This study confirmed that some patients are able to develop IgE mediated reaction through \textit{H. pylori} infection.

**Table-1** Serum IgG versus IgE specific for \textit{H. pylori} in homogenate of each biopsy

<table>
<thead>
<tr>
<th>IgG serum</th>
<th></th>
<th>IgE</th>
<th></th>
<th>Total</th>
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<tbody>
<tr>
<td></td>
<td>Count</td>
<td>Positive</td>
<td>Negative</td>
<td>Total</td>
</tr>
<tr>
<td>Positive</td>
<td></td>
<td>16</td>
<td>44</td>
<td>60</td>
</tr>
<tr>
<td></td>
<td>% within IgG serum</td>
<td>26.70%</td>
<td>73.30%</td>
<td>100.00%</td>
</tr>
<tr>
<td></td>
<td>% within IgE homogenate</td>
<td>66.70%</td>
<td>33.30%</td>
<td>78.90%</td>
</tr>
<tr>
<td>Negative</td>
<td>Count</td>
<td>8</td>
<td>8</td>
<td>16</td>
</tr>
<tr>
<td></td>
<td>% within IgG serum</td>
<td>50.00%</td>
<td>50.00%</td>
<td>100.00%</td>
</tr>
<tr>
<td></td>
<td>% within IgE homogenate</td>
<td>33.30%</td>
<td>16.60%</td>
<td>41.10%</td>
</tr>
<tr>
<td>Total</td>
<td>Count</td>
<td>24</td>
<td>52</td>
<td>76</td>
</tr>
<tr>
<td></td>
<td>% within IgG serum</td>
<td>31.60%</td>
<td>68.40%</td>
<td>100.00%</td>
</tr>
<tr>
<td></td>
<td>% within IgE homogenate</td>
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Positive correlation was seen between serum positive IgM and IgE specific for *H. pylori* in homogenates. This finding was confirmed by a study carried out by 16 who showed Positive IgE specific for *H. pylori* patients in serum.9 also found that *H. pylori* induced gastritis is accompanied by an increase in IgE production. This antibody can continuously activate mast cells in mucosal tissue 23, this result was in consistent with the study by24. Males showed more positive IgE specific for *H. pylori* in their homogenates than females and this was compatible with the results of IgM and IgG in patients of this study. These findings were in accordance with that of16 who found more IgE positive results in sera of male dyspeptic patients. This can be explained as, females may be exposed to infection as males but bacteria have low chance to establish its infection due to different factors mostly antibiotics that the female had taken it during her life course, pregnancy, after delivery, abortion and during urogenital infection. Moreover, gender differences to *H. pylori* induced gastro-duodenal diseases and in disease susceptibility may reflect differences in immune responses, hormonal effects and gender linked genetic factors and differences in bacterial colonization.25,26

In conclusion, *H. pylori* Specific IgE mediated reaction can be induced in mucosa of dyspeptic patients and this reaction differs regarding stage of infection and gender of patients.

References:


16- Hussein, Assel kasim (2015)Th1 versus Th2 in *H.pylori* infected patients, thesis Submitted to the Collage of Medicine and Committee of Postgraduate studies, Al- Anbar University in Partial Fulfillment of the Requirements for the Degree of Master of Science in Medical Microbiology.


