



ISSN: 0975-833X

Available online at <http://www.journalcra.com>

INTERNATIONAL JOURNAL  
OF CURRENT RESEARCH

International Journal of Current Research  
Vol. 14, Issue, 10, pp.22412-22415, October, 2022  
DOI: <https://doi.org/10.24941/ijcr.44068.10.2022>

## REVIEW ARTICLE

# DETECTION OF IGG ANTIBODIES TO H. PYLORI IN PATIENTS WITH GASTRO-INTESTINAL INFECTIONS IN BAQUBA CITY

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### ARTICLE INFO

#### Article History:

Received 10<sup>th</sup> July, 2022  
Received in revised form  
27<sup>th</sup> August, 2022  
Accepted 19<sup>th</sup> September, 2022  
Published online 19<sup>th</sup> October, 2022

#### Key words:

H.pylori, Gastro-Intestinal, Baquba City.

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Citation: Ziyad Tariq Ibrahim. 2022. "Detection of IGg antibodies to H. pylori in patients with gastro-intestinal infections in Baquba city". *International Journal of Current Research*, 14, (10), 22412-22415.

### ABSTRACT

**Objective:** A lot of Recent This study was conducted for the detection of IgG antibodies using a rapid serological diagnosis technology (IgG) rapid immunochromatography (IgG) test for Helicobacter pylori to detect IgG antibodies, 50 venous blood samples were collected from patients with stomach and colon diseases in the endoscopy unit of Hospital Baquba Education, 10 venous blood samples were collected for the control group for the period from 15/11/2016 to 15/3/2017 the study showed that the infection rate in males is higher than females, as the percentage of infection in males constituted 58.33% (30 samples), while the percentage of females constituted 41.7% (20 samples), and the test group was divided into different age groups ranging from Between (1525-), which constituted 32% (16 samples), and the second age group (2646-), which constituted 68% (34 samples).

## INTRODUCTION

Helicobacter pylori is considered the main causative agent of infectious diseases such as chronic gastritis, peptic ulcer associated disorders, and gastric and duodenal carcinomas, which lead to fatalities in humans. The International Research Agency on cancer) These bacteria are in the first class as a carcinogen. is a bacterium (which used to be called sigmoid- negative and has a low affinity for air) found in the stomach and infects nearly half of the scientific population. The infection occurs most often in childhood and lasts for life if not treated. This human pathogen is known to stimulate many infectious disorders. gastric disorders, and it may be a accompaniment to other diseases outside the infectious etal, 2014; Symk *et al* 2014 There are many methods currently used to detect H.pylori, and these are not using the endoscopy . Tests are divided based on the use of biopsy, including histological tests, bacterial culture, and detection by polymerase chain reaction (PCR) ., and test Rapid urease (RUT) . All these tests are performed on the tissue biopsy obtained by the endoscope. In contrast, there are tests that are done without the use of the endoscope and include a urea breath test . serological tests and stool antigen tests, all of which are considered inappropriate

**The aim of the study:** Serological diagnosis of the level of IgG antibodies to H.pylori antigens in the blood of patients with gastro-intestinal infections using a one-step kit to detect the type of antibody (One step H.pylori Test Kit) (IgG).

2- Calculating the percentage of H. pylori prevalence in patients with gastro-intestinal infections and its relationship with age and gender.

### Literature review

11- General characteristics of H.pylori Spiral -shaped bacteria (classified as curved, not helical bacilli) that are Gram- negative, about 255.4- μm in length, and about 01- μm in diameter (Mandell eral 2010.). have little affinity for air (need low concentrations of oxygen) . These bacteria possess the enzyme hydrogen peroxide and can use it to obtain energy by oxidizing the hydrogen molecules produced by the intestinal bacteria (2002, Olson and Maier) These bacteria have 26- flagella that give them enough flexibility to withstand contractile and rhythmic movement in the stomach and to penetrate into the mucous layer of the stomach.can bacteriaH. pylori growth at a pH value (7-6) and at a temperature of 37 annually on a medium equipped with blood, heme and carbon sources, and a sugar medium (Skirrow), which is an optional medium, which is positive for the oxidase test. And urease, which are characteristics that distinguish it from the rest of the species. Patrick *et al* ,2007) Mandell er al: 2005) The ability of H.pylorito survive and thrive is due to several motility factors that enable it to penetrate the mucous membrane of the stomach to reach the surface of the epithelium, and it also secretes the wind urea that breaks down urea . It converts to carbon dioxide and ammonia, so it surrounds itself with an alkaline environment that facilitates its survival in an acidic environment, and this was used as a diagnostic tool in tests (Eurogast, 1993) Urea breath test, Clotest

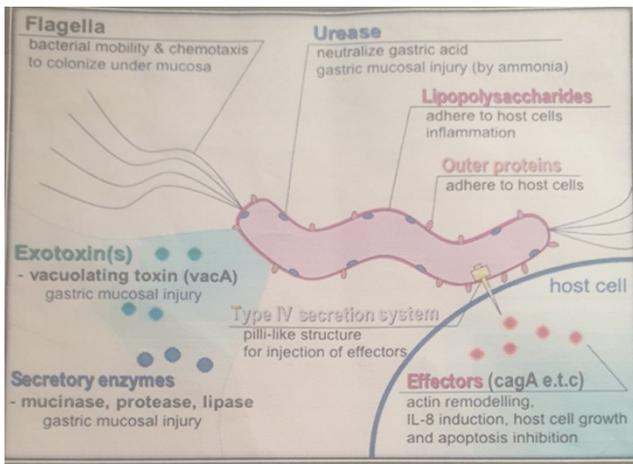


Figure 1. Shows the virulence factors of *H.pylori*

### H.pylori diagnosis

*H.pylori* bacteria colonies form on the surface of the renewed purity, and this bacteria colonization is not a disease in itself, but it is associated with a number of disorders of the upper gastrointestinal tract. Epithelial-associated lymphoma (MALT), after partial laparoscopic resection for early intestinal cancer and if there is a first degree association with intestinal cancer (Kusters *et al.*, 2006; Stenstrom *et al* 2008)

### There are several methods of diagnosis

**Serological tests:** Includes screening for antibodies to *H.pylori* in serum, serological tests remain positive for several years after successful eradication of the bacteria so it is used to ensure successful treatment because the antibodies in the blood decrease slowly (Rahman *et al.*, 2014; Lopes).

**Urea breath test with the parameter UBTS:** It is a simple, non-invasive test based on the enzyme *H.pylori* urease. Learn urea with a non-radioactive isotope C13 or with a low dose of radioactive C14 and is given to the patient if *H.pylori* is present in the stomach, the enzyme Urease strongly hydrolyzes urea to marked ammonia and carbon dioxide and can detect it in breath samples. This test is partly useful for "investigating the success of treatment, and is more accurate than serological tests, so it is used as a first diagnostic test when available. This must be done. "Test at least 2 weeks after stopping proton pump inhibitors and 4 weeks after stopping bismuth treatment and antibiotics to avoid getting false high results .

**Stool antigen test:** This test was recently developed as an "alternative to the Urea breath test, which needs to be presented higher at the level of clinical examinations as well as "to the damage caused . Debabrata\_eral 2007: Malferiner original

### Rapid urease examination on biopsy

After this, the most common examination "where the gastric endoscope is used to take Biopsy, the biopsy is placed in a medium solution of urea broth or urea agar with an indicator of the change in the acidity of the medium pH . Positive results, and to avoid negative results, the sample must be kept for 24 hours. Blood in the upper digestive tract may sometimes cause false negative results. The observance of the time to stop taking a PPI with antibiotics is similar to the UBTS examination and this is common for all gastric tissue biopsy tests ... Dirienzoetal) 2013; Lopes *et al.*, 2014)

**Histology:** *H. pylori* infection can be diagnosed by histological examination, as special dyes are used. The spread of infectious inflammation can give information on the severity of the disease if the biopsy is taken from the pyloric part of the stomach and the stomach is removed. Histological examinations can give other information for example intestinal atrophy or histological mutations that are signs of

an increased risk of gastric cancers (2014. Pourakbari) .eral ., 2013; Lopes eral

**Culture:** A tissue biopsy of the mucous layer taken by laparoscopy can be cultured on special media to detect *H. pylori*, but this test is considered the least used among other diagnostic tests because of treatments that inhibit the growth of these bacteria and give a negative result for culture, and it is considered one of the important diagnostic tests to detect The sensitivity of these bacteria to certain antibiotics, and this is important in directing specific antibiotics in the event that previous treatments fail. 2014 Lopes eral

**Virulence Factors:** The high pathogenicity of *H. pylori* strains and their susceptibility to disease is due to the basic virulence factors of this bacteria, which include: 1-3-1 Cytotoxin associated gene A (Cag A), which is one of the most important virulence factors for *H. pylori* and encodes Numerous studies have confirmed that infection with gene-positive strains of bacteria is associated with cytotoxicity . (CagA) is associated with chronic gastritis, high prevalence of peptic ulcers and stomach cancer (Wang *et al*, 2013) . The time associated with cytotoxicity is an indicator of the pathogenicity CagA Island (PAI) sites in the Gram -stained bacterial DNA responsible for the bacterial local secretory system

**Vacuolating cytotoxin A VaeA:** Vacuole - forming cytotoxin (VacA) is produced by most pathogenic strains. This protein is yet one of the virulence factors that contribute to the pathology of gastric ulcers. The vacated gene encodes for this protein under normal conditions, as it consists of two different parts, which are the terminal part. Or a final part called (s-region) and the second part is the middle part and is called (Bagheri )M-region) *et al.*, 2013) 13-3- Flagella *pylori* has (2-6) flagella and is unipolar and thick and covered to protect it from high acidity of the stomach and to enable it to penetrate the mucous layer of the stomach cavity. The flagella thread consists of two proteins and there are about 40 genes involved in the formation and release of substances Flag \_ \_ \_ Penetration of the mucous layer and access to the epithelial cells of the stomach (Rust, 2008)

**Outer Membrane Protein (OM):** In *H. pylori* bacteria, the outer membrane proteins CONIP have clearing properties that enable them to colonize (Dumrese2009), the first protein is called (Bab A (Adhesion) and the second is Saba Adhesion, and they are one of the most important outer membrane proteins because they bind to the receptors on the host receptor cells God believes that the presence of these adhesive proteins increases the pathogenicity of these bacteria (Falasmazi, 2010)

**Lipopolysaccharides (LPS):** LPS (which is a component of the outer membrane) covers the surface of *H.pylori* and contains bacterial adhesives (Bacterial adhesion) . It is composed of polymeric units of liquid carbohydrates in the composition of the blood type antigens that are expressed in the body of the host, and that 8090-% of *H. pylori* strains have the ability to express these antigens that help the bacteria to evade the immune system (Immune Evasion) and it was included in the process of adhesion (Lundin., 2004).

**Urease enzyme Urease Enzyme:** *H.pylori* bacteria produce urease, which is one of the most important factors of virulence of this bacteria, as it enables it to survive in the stomach environment that is highly acidic, as it protects bacteria from stomach acidity by generating basic hydroxyl ions OH and thus the formation of ammonia that Reduces the acidity of the stomach, and the resulting ammonia is a toxic substance that causes tissue breakdown, and thus releases its metabolites, which encourages bacterial growth, as well as liberating the protons resulting from the decomposition of urea naturally present in the stomach into ammonia and carbon dioxide, where they combine with water molecules and thus Diammonium carbonate is produced, which has a clear role in neutralizing acidity and providing an alkaline medium that protects bacteria from stomach acidity (Gillespie and Bamford (2012) . Figure (11-) shows the virulence factors of *H. pylori* bacteria

## Materials and working methods

# MATERIALS

**The devices used Instrument:** The following fees were used to conduct the examinations mentioned in the study:

**Table 1. Devices, producing company and origin**

company and the origin	Device name Device
Gallenkamp(England)	centrifuge Centrifuge

## The Equipment and diagnostic Kits

**Table 2. Equipment, producing company and origin**

Company and Origin	Diagnostic supplies and kits
Racks (Czechoslovakia)	pipe holders Tubes Holders
Eppendorf(Germany)	micro pipette Micropipette 2-200 µTips
Biobasic(Canada)	Abendrov tubes Ependroff tubes
Diaspot (USA)	One step H.pylori Test Device Kit (serum plasma)

## Working methods

**Collection of samples:** 60 blood samples were collected from patients with gastric and gastric ends who attended Baquba Teaching Hospital for the period from 11/15/2016 to 3/1/2017, and were divided into two groups as follows

- A - The uninfected control group (10) people  
B - Patients group \_

Milliliters of venous blood was withdrawn from the infected patients and the healthy ones (control group) after placing the paws and sterilizing the area of blood withdrawal with ethyl alcohol (70%), after sterilizing the same area with iodine solution (2%). By using medical syringes, wine was drawn and placed in septic laboratory tubes, taking into account not to throw used syringes without returning the covers on them in special containers, which in turn were sent to the incinerator for disposal. Leave the blood for 1015- minutes at room temperature until clotting occurs. The serum was separated using a centrifuge at 3000 revolutions per minute for 5 minutes, after which the test was performed.

## Materials and working methods

**Using the One-Step Kit to Detect Antibodies in Serum One-step IH pylori Kit (IgG) Test Device:** A rapid one-step specific immunoassay kit was used to detect H.pylori antibodies. In the sera of patients and control according to the instructions of the company (Diaspot (USA), which is a strip with a hole in which 3 drops of the patient's serum (containing anti-bacterial antibodies) are placed, which interacts with the bacteria antigen molecules covering the surface of the lane, this interaction leads to a change in the color of the line that It appears in the examination area (Test), which indicates that the result is positive, while the absence of the red or burgundy line in this area indicates that the test is negative, and the tape contains the control indicator represented by the appearance of a red or burgundy line in the control line area, which Indicates the integrity of the test. In the event that the red or burgundy line does not appear after the test is unsuccessful, it must be repeated using a new strip, and according to the instructions of the producing company, the result is calculated within 10 minutes after which it will be neglected.

# RESULTS AND DISCUSSION

## Results and discussion

**Sample collection:** The current study included investigating the presence of IgG antibodies to H. pylori in the blood serum of patients with gastro-intestinal infections of different age groups and both sexes

at Baquba Teaching Hospital in Diyala Governorate. 50 blood samples were collected from these patients as a test group, 104 were stoned Blood cells from healthy people as a control group .

The selection group was divided into two groups, according to gender, and the proportion of males constituted 58.33% (30 samples), while the proportion of females constituted 41.7 % (201 samples), and the selection group was divided into different age groups that ranged between (1525-), which constituted 32% (16 samples) and the second age group (2646-), which constituted 48 percent (34 samples), as shown in Table No. (14-).

Age \ Gender	Patients		*Corresponding author:		
	No.	%	No.	%	
Age	15-25	16	32	5	50%
	26-46	34	68	5	50%
Gender	Total	50	100%	10	100%
	Male	30	58.33	5	50%
	Female	20	41.7	5	50%
	Total	50	100	10	100%

The results agreed with many researchers, as Malatyand his group (2002) indicated that the incidence of H. pylori infection in males is greater than in females, as the researcher Munaand her group (2016) noted that the percentage of presence of antibodies of a type is about 89% and an increase in the percentage was noted in The age group (4021- years), but indicated that the rate of infection in females is more than in males, as it was in females 55% and in males 45%)

## Conclusion

- The incidence of infection in males is more than in females
- The infection rate is in the age group (4266- years), as it constituted 68 % (34 samples) .
- Recommendations
- Conducting a study to investigate the presence of IgM in the blood serum of patients with gastro-intestinal infections
- Conducting a study to detect the presence of anti-bacterial antibodies in the stool

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