Sudan University of Science and Technology
College of Graduate Studies

Comparative Study between Antigen and Antibody Tests in   the Diagnosis of *Helicobacter pylori* in Symptomatic and Asymptomatic Patients in Relation to Selected Risk Factors

مقارنة إختبارات كشف الاجسام المضادة والمستضدات المستخدمة في تشخيص الاصابة بالبكتيريا الملوية البابية مصحوبة وغير مصحوبة الأعراض وعلاقتها ببعض عوامل الخطر

A dissertation Submitted in Partial Fulfillment for the Requirements of M.Sc in Medical Laboratory Science (Microbiology)

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January 2017

الأيـة

بسم الله الرحمن الرحيم

(وَإِن يَمْسَسْكَ اللَّهُ بِضُرٍّ فَلاَ كَاشِفَ لَهُ إِلاَّ هُوَ وَإِن يَمْسَسْكَ بِخَيرٍ فَهُوَ عَلَى كُلِّ شَيْءٍ قَدِيرٌ)

صدق الله العظيم

سورة الانعام الأية(17)

Dedication

To my parents

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To my teachers

To my colleagues and dear friends

Acknowledgement

By the grace of Allah and his help I completed this study. Special thanks to **Dr. Ahmed Ibrahim Hashim** my supervisor for his guidance, and understanding throughout this study. My thanks and appreciation are extended to all my colleagues.

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Abstract

Helicobacter pylori pathogen is an organism that cause a major health problem worldwide. More than half of the world populations are infected with this pathogen. This study was conducted in Medical Military Hospital, Khartoum State between March and January 2016 to compare between antigen and antibody tests in the diagnosis of *H. pylori* infection among symptomatic and asymptomatic Sudanese patients. In this cross sectional study one hundred patients (n=100) were enrolled 50 symptomatic and recruits were asymptomatic. In the symptomatic group 24(48%) were males and 26(52%) were females and their ages were between 13-84 years. Asymptomatic group18(36%) were males and 32(64%) were females and their ages were between 5-70 years. Data on smoking and other demographic information were obtained by questionnaire and the ABO blood group phenotypes were determined. The stool and blood samples were collected and analyzed for antigen and antibody respectively by immunochromatography test (ICT) cards. Symptomatic and asymptomatic patients 58(58%) showed positive results for *H. pylori* antibody while 11(11%) showed positive results for *H. pylori* stool antigen.

The sensitivity and specificity of stool antigen was 48% and the specificity and sensitivity of serum antibody was 47%. Smoking, blood group and age showed insignificant relationship with *H. pylori* infection (P>0.05). In conclusion, the frequency of *H. pylori* antibody was higher than the *H. pylori* antigen. The study showed insignificant relationship between age, blood group and smoking. Future studies should be conducted with larger sample size, wider area and advanced techniques to confirm the present results.

المستخلص

الملوية البابية هي بكتريا سالبة الجرام لولبية الشكل وهي تسبب مشكلة صحية كبيرة في جميع أنحاء العالم . أكثر من نصف سكان العالم مصابين بهذا الممرض. أجريت هذه الدراسة في مستشفي السلاح الطبي العسكري في ولاية الخرطوم خلال الفترة من مارس حتى نوفمبر 2016م مقارنة إختبارات كشف الاجسام المضادة والمستضدات المستخدمة في تشخيص الا صابة بالبكتيريا الملوية البابية مصحوبة وغير مصحوبة الأعراض تضمنت هذه الدراسة 100 مريض 50 منهم مصحوبي اعراض الاصابة بالبكتريا الملوية البابية وقد كان عدد الذكور (48%) 24 والاناث (52%) 26 وأعمارهم تتراوح بين13-84سنة 50 منهم غير مصحوبي الأعراض وقد كان عدد الذكور (35%) 18 والاناث (64%) 32 وأعمارهم تتراوح بين 5-70 سنة. وقد تم الحصول على معلومات عن التدخين وبيانات أخرى عن طريق الاستبيان وتم الكشف عن نتائج فصائل الدم ABO في المرضى المختارين. تم جمع عينات البراز والدم وتحليل المستضد والأجسام المضادة بواسطة اختبار المناعة اللوني (ICT) خلصت النتائج الى أن (58%) 58 من المرضى أظهرت نتائج ايجابية للأجسام المضادة للبكتريا الملوية البابية و (11%) 11 أظهرت نتائج ايجابية للمستضد في البراز وكانت نسبة حساسية ودقة اختبار الكشف المستضد كانت (48%) ونسبة حساسية ودقة الاجسام المضادة (47%). افادت هذه الدراسة بعدم وجود علاقة بين العمر، التدخين ، فصيلة الدم ، والاصابة بالبكتريا الملوية البابية حيث كانت القيمة الاحتمالية > 0.05أوصت الدراسة باجراء مزيد من الدراسات تتضمن أكبر حجما للعينة و مساحة أوسع وتقنيات حديثة.

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CHAPTER ONE

INTRODUCTION

1.INTRODUCTION

1.1Background

Helicobacter pylori, previously *Campylobacter pylori*, is G gram-negative, microaerophilic bacterium found usually in the stomach. It was identified in 1982 by Australian scientists Barry Marshall and Robin Warren, who found that it was present in a person with chronic gastritis and gastric ulcers, conditions not previously believed to have a microbial cause. It is also linked to the development of duodenal ulcers and stomach cancer. However, over 80% of individuals infected with the bacterium are asymptomatic, and it may play an important role in the natural stomach ecology (Blaser, 2006). *Helicobacter pylori* is a Gram negative, spiral flagellated bacterium that infects approximately 50 percent or more of the world population It is one of the most common chronic bacterial infections in human (Brown, 2000). The prevalence of *H. pylori* is 30% in the United States and other developed countries as opposed to 80% in most developing countries (Atherton and Blaser, 2005). Prevalence of infection increases with age and correlates positively with a low socioeconomic status during childhood (Malaty and Graham, 1994). The infection is usually acquired in early childhood and persists throughout life unless treated. The seroprevalence rate in lower socioeconomic class in India was found to be 79%. There is a strong association between the presence of this organism and gastric inflammation. Host and bacterial factors with interaction of environment contribute pathogenicity. *H. pylori* cytotoxin-associated geneA (cagA), vacuolating toxinA (vacA) and adherence factors to gastric epithelium have been linked to enhanced pathogenicity of the bacterium. Host genetic polymorphism of cytokines, related legends, receptors and enzymes influence *H. pylori* infection (Das and Paul, 2007)

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The presence of IgG antibodies to *H. pylori* can be detected by use of biochemical assay (Lerang *et al.,* 1998). Other non invasive tests can be used including, detection of *H. pylori* antigen in stool and presence of *H. pylori* in saliva**.** Stool antigen tests have recently been welcomed with great expectations as they are convenient to patients and can be easily performed even in small laboratories (Malfertheiner *et al.,* 2002).

Helicobacter pylori is the most common cause of peptic ulcer and chronic gastritis and co- factor for stomach cancer (Black, 2004). Approximately two third of world population was infected with *H. pylori* any age can get infection and women were affected just as often as men. In February1994 the National Institutes of Health Consensus Development Conference concluded that *H. pylori* infection is the major cause of peptic ulcer disease, and all patients with confirmed peptic ulcer disease associated with *H. pylori* infection should receive treatment with antimicrobial agent. A combination of genetic and environmental factors exemplified by the type of food intake and infection with *H. pylori* involved in pathogenesis of gastric carcinomas. *H. pylori* present on gastric mucosa of less than 20% of persons under age 30years but increase in prevalence to 40-60 years age including who are asymptomatic. *H. pylori* is more prevalent among the elderly and more frequent in male than female *H. pylori* is capable to form biofilm (Yamoka, 2008). *H. pylori* is Gram negative bacterium S. shape curved rod, about 2-4 micrometer long with diameter 0.5-0.9 micrometer (Black, 2004).

1.2 Rationale

Helicobacter pylori   infects approximately 50 percent or more of the world population It is one of the most common chronic bacterial infections in human (Brown, 2000). Laboratory testing for *H. pylori* infection is very important part of the diagnostic process for gastric and inflammatory disease. A number of different diagnostic tests methods both invasive and non invasive are available. Invasive methods have high specificity and sensitivity in the detection of *H. pylori* infection, however they are expensive and need special gastroenterologist to be performed . In contrast, non-invasive methods such as antibody (Ab) detection is cheap, easy to perform but may give false positive result since antibody level falls slowly after eradication in addition to low specificity (Quartero *et al.,* 2000). A non invasive diagnostic tool which is specific and sensitive is through the detection of stool antigen in the faeces of infected patients detection of *H. pylori* antigen (Ag) in stool samples which indicates the current presence of the infection. This study was aimed to compare between antigen and antibody tests in the diagnosis of *H. pylori* infection in symptomatic and asymptomatic patients and associated risk factor.

1.3 Objectives

1.3.1 General Objective

To compare between non invasive tests to diagnose *H. pylori* infection among symptomatic and asymptomatic patients and associated risk factor. .

1.3.2 Specific Objectives

1- To detect stool antigens of *H. pylori* in symptomatic and asymptomatic faeces.

2- To detect serum antibodies against *H. pylori* in symptomatic and asymptomatic sera.

3- To determine selected risk factors associated with *H. pylori* infection.

4- To compare between antigen and antibody tests in the diagnosis of *H. pylori* in symptomatic and asymptomatic.

CHAPTER TWO

LITERATURE REVIEW

2. LITERATURE REVIEW

The presence of *H. pylori* was first reported in 1893 by bizzozer who described spiral bacteria in canine stomach nine years later , spiral microorganisms in the stomach of patients with gastric duodenal ulceration were reported (Rosenow & Sanford, 1915). *H. pylori* was originally classified in the genus *Campylobacter* but is now know to be differ from the *Campylobacter* in 16s r RNA sequence its unique fatty acid s contents and possess of multiple flagella which are located at polar side as shown by electron micro scope. It is now believed to be phylogenetically closer to *Welinella* than to *Campylobacter*.

2.1 Characteristics of *H. pylori*

The most common ulcer symptom is gnawing or burning pain in the epigastrium. This pain typically occurs when the stomach is empty, between meals and in the early morning hours, but it can also occur at other times. It may last from minutes to hours and may be relieved by eating or by taking antacids.

Less common ulcer symptoms include nausea, vomiting, and loss of appetite. Bleeding can also occur; prolonged bleeding may cause anemia leading to weakness and fatigue. If bleeding is heavy, hematemesis, hematochezia, or Melina may occur. *H. pylori* survive to the very acidic condition of the stomach by generating ammonia from urea the ammonia is believed to neutralize gastric acidity around *H. pylori* cell thereby allowing the organisms to survive and replicate (Black, 2004).

2.2 Mode of transmission

Mode of transmission is most likely from person to person, either by the oral-oral route (through vomitus or possibly saliva) or perhaps the fecal-oral route. The person-to-person transmission is supported by the higher incidence of infection among institutionalized children and adults and the clustering of *H. pylori* infection within families. Detection of *H .pylori* from faeces and dental plaque by culture and PCR suggesting a faecal-oral or oral- oral transmission Waterborne transmission due to fecal contamination may be an important source of infection especially in parts of the world in which untreated water is common (Goodman *et al*, 1996). Iatrogenic spread through contaminated endoscopes has been documented but can be prevented by using proper disinfection procedures (Fantry *et al*, 1995). It has been suggested that infection may be transmitted gastro-orally by gastroesophageal reflux or regurgitation of stomach contents (Stone, 1999).

2.3 Pathogenesis

The earliest descriptions of the organism classified it as predominately extracellular, Gram negative, flagellated, and motile. With the advancement of biochemical techniques, new information about the pathogenicity and virulence factors of   *Helicobacter pylori*   has emerged, indicating that infection by *Helicobacter pylori* requires a complex interaction of both bacterial and host factors. Investigators have identified several bacterial proteins necessary for colonization of the gastric mucosa by *Helicobacter pylori*, including proteins active in the transport of the organism to the surface of the mucosa (eg, flagellin, which is encoded on genes flaA and flaB). Once in the presence of the gastric mucosa, bacteria induce a transient hypochlorhydria by unknown mechanism . The

urease enzyme produced by the bacteria alters the microenvironment of the organism to facilitate colonization. Adherence then occurs via interaction between cell-surface glycolipids and adhesins specific to *Helicobacter pylori*. There also appears to be a role played by proteins called cecropins, which are produced by *Helicobacter pylori* and inhibit the growth of competing organisms, as well as by a P-type adenosine triphosphatase, which helps prevent excessive alkalinization of the microenvironment by urease. Once attached to gastric mucosa, *Helicobacter pylori* causes tissue injury by a complex cascade of events that depends on both the organism and the host. *Helicobacter pylori,* like all gram negative bacteria, has in its cell wall lipopolysaccharide, which acts to disrupt mucosal integrity. Furthermore, *Helicobacter pylori* release several pathogenic proteins that induce cell injury. For example, the CagA protein, produced by cytotoxic-associated gene A (cagA), is a highly immunogenic protein that may be associated with more severe clinical syndromes, such as duodenal ulcer and gastric adenocarcinoma (although this question is far from settled). There is increasing evidence that CagA positivity is associated with an increased risk for distal, but not proximal, gastric adenocarcinoma. In addition, protein products of the vacuolating cytotoxin A gene (vacA) and the A gene induced by contact with epithelium (iceA) are known to be associated with mucosal injury. Once colonization of the gastric mucosa has taken place, the immunogenic properties of *Helicobacter pylori* induce an inflammatory reaction with neutrophilic gastritis that ultimately results in the clinical manifestations of the infection. This process is mediated by host factors, including interleukins 1, 2, 6, 8, and 12; interferon gamma, tumor necrosis factor, T and B lymphocytes and phagocytic cells. These factors mediate injury through release of reactive oxygen species and inflammatory cytokines. *Helicobacter pylori* additionally appear to increase the rate of mucosal-programmed cell death (also known as apoptosis

Seroprevalence studies demonstrate an acquisition rate in adults of 3% to4% per decade. *H. pylori* infection is a likely contributing factor in the development of gastric neoplastic diseases such as gastric adenocarcinoma and mucoassociated lymphoid tissue (MALT) lymphoma (Cullen *et al.*, 1993). *H. pylori infection* is considered to increase the risk of gastric adenocarcinoma with pooled odds ratios of 1.9 to 2.5 % (45, 54). The risks of ulcer recurrence and associated complications do not diminish unless *H. pylori* infection is cured (Cullen, 1993).

2.4 *H. pylori* infections

The infections caused by *H. pylori* include:

2.4.1 Peptic ulcer

The relationship between *Helicobacter pylori* infection and peptic ulcer disease has been studied exhaustively, and it is now accepted that the organism is the major cause, but not the only cause, of peptic ulcer disease worldwide. Eradicating the infection can alter the natural course of peptic ulcer disease by dramatically reducing its recurrence rate in treated patients, compared with untreated patients. This reduction occurs in patients with duodenal and gastric ulcers that have no history of non steroidal anti-inflammatory drug use. The mechanism by which *Helicobacter pylori* induces peptic ulcer disease is incompletely understood but most likely involves a combination of genetic predisposition of the host, virulence factors of the organism (eg, VacA and CagA proteins), mechanical damage to the mucosa, and alterations of gastric and duodenal secretions (Bardhan *et al.,* 1997).

2.4.2 Gastric cancer

Epidemiological studies link *H. pylori* infection both adenocarcinoma and lymphomas of stomach data from both American and European (Malfertheiner *et al*., 2007). Suggested mechanism is that chronic inflammation related to *H. pylori* infection lead to atrophic gastritis , with decrease in gastric acid a secondary in the number of bacteria which produce carcinogenic nitrosamine from dietary nitrate (Webb *et al*., 2004).

2.4.3 Non-ulcer dyspepsia

Non-ulcer dyspepsia comprises a constellation of varied symptoms, including dys motility-like, ulcer-like, and reflux-like symptoms. Many possible causes have been suggested for non-ulcer dyspepsia, including lifestyle factors, stress, altered visceral sensation, increased serotonin sensitivity, alterations in gastric acid secretion and gastric emptying, and *Helicobacter pylori* infection. A recent study also highlighted the role played by psychosocial impairment (eg, depression, somatization, anxiety) in patients with non-ulcer dyspepsia. In a study linking *Helicobacter pylori* infection to non-ulcer dyspepsia, patients with the latter condition were twice as likely to be positive for the organism. However, despite such epidemiologic evidence, treatment studies have failed to consistently show that eradication of *Helicobacter pylori* results in improvement of non-ulcer dyspepsia symptoms. Consequently, eradication of the organism cannot be considered the standard of care in all patients with non-ulcer dyspepsia, because *Helicobacter pylori* infection is only a single part of the multi-factorial etiology of the disease (Bardhan *et al.,* 1997).

2.4.4 Gastroesophageal reflux

Reflux disease Much attention has been focused on the possible relationship between infection with *Helicobacter pylori* and gastroesophageal reflux disease (GERD) in its various manifestations (eg, esophagitis,Barrett's esophagus). Some investigators have suggested a link between the presence of *Helicobacter pylori* and a decreased risk for developing esophagitis and Barrett's esophagus ; although this inverse association is supported by many prevalence studies, others fail to show it. Studies have also indicated that certain strains of *Helicobacter pylori*, notably the CagA- positive strain, may be protective against the development of Barrett's esophagus. Moreover, Labenz and colleagues have shown that the incidence of esophagitis may in fact, increase after eradication of the organism. Treatment of *Helicobacter pylori* infection can lead to exacerbation of GERD in many patients, prompting many gastroenterologists to defer endoscopic antral biopsies in patients with significant GERD and absent ulcer. Conversely, other studies using endoscopic findings, pH probe measurements, and histology to determine the presence of *Helicobacter pylori* did not find any association between GERD (in any of its manifestations) and infection with *Helicobacter pylori*. Clearly, more definitive studies are necessary to define the relationship, if any, between these 2 entities (Deboer and Tytgat 1995).

2.4.5 Other disease associations

Investigators have further postulated a relationship between *Helicobacter pylori* infection and cardiovascular disease and iron-deficiency anemia. These associations, however, require much more study before a causal relationships is established Diagnostic testing (Anantharishnan, 1997)

2.5 Diagnosis of *H. pylori*

There are various methods for the diagnosis of *H. pylori* these methods are classified into two groups

2.5.1 Invasive Methods

Invasive methods include

2.5.1.1 Histopathology Technique

It s currently the gold standard for diagnosis of *H. pylori* infection small sample of biopsy is taken from stomach and small intestine during endoscopy   the organism can be identified on routine hematoxylin-eosin and other stain can be use.

2.5.1.2 Culture

Culturing of organism from biopsy is possible but the organism is fastidious and difficult to grow in the laboratory current recommendation are to culture specimen only when the procedure is necessary to determine antibiotic sensitivity (Logain, 1993).

2.5.1.3 Molecular Test

The P.C.R is able to isolate and replicate *H. pylori* DNA in biological material also to detect genetic variation between different strain of *H. pylori* (Greenwood *et al.,* 1997).

2.5.2 Non-Invasive Method

Non-Invasive Methods include

2.5.2.1 Serological Test

Antibody test do not work well after treatment. The reason of this is that Abs to *H. pylori* decline slowly and may remain elevated after *H. pylori* has been totally eradicated. However Abs tests are not affected much if you have taken antibiotics pepto-bismol , proton pump inhibitors, Cytoprotective, And H2receptor antagonist such as(zantac ,tagamet). Enzyme Linked Immunosorbent Assay Most extensive used in serological detection of *H. pylori* and produce reliable result. The Elisa test based on purified *H. pylori* Ag. is coated in service of micro well diluted patient serum is added, and if *H. pylori* Ab. When present bind with Ag. and non bound material washed away enzyme conjugate is added which bind to Ab –Ag complex the excess enzyme conjugate is washed off and solution of tetra methyl benzaldehyde substrate is added. The enzyme conjugate catalytic reaction is stopped at (15 minute) the intensity of color generated is proportional to the amount of DNA specific Abs in sample. The result is read by micro well reader compared in parallel manner with calibrator and control.

2.5.2.2 Urea Breath Test

Hydrolysis of c13- c14 labeled urea by *H. pylori* urease enzyme this test may be false negative in patient whom the level of *H. pylori* had been suppressed by recent antibiotics use or treatment with proton pump inhibitors Used in screening and epidemiological survey (Logain, 1993).

2.5.2.3 Stool antigen detection test

Stool Ag testing may be done to help support a diagnosis of *H. pylori* infection or to determine whether treatment for an *H. pylori* infection has been successful. Detection of *H. pylori* antigen indication of current infection.

2.5.2.4 Rapid urease Test

Simple test the principle of the test rest on powerful urease activity of *H. pylori* , urease activity is detected by the rise in the PH color change of the phenol red in dictator in culture medium from yellow to read the color change is obtained with in 30 minute (Buuer, 1996).

2.6 Treatment of *H. pylori*

The first-line treatment remains clarithromycin, amoxicillin or metronidazole and proton pump inhibitor twice daily, but a number of recent studies have shown low eradication rates with this treatment. Increased duration of therapy has been recommended to overcome the falling eradication rates. However, conflicting findings have been reported on the benefits of extending the length of traditional therapy. Sequential therapy may be an effective alternative to standard triple therapy in regions of increased antimicrobial resistance. Probiotics reduce side-effects from traditional regimens and may improve eradication rates. aquinolone-based second-line triple therapy appears to be effective and well tolerated. Bismuth-based quadruple therapy is also an effective alternative if available. In the future, regional antimicrobial resistance and eradication rates will determine the best treatment for *H. pylori* (Malfertheiner *et al* 2007).

CHAPTER THREE

MATERIALS AND METHODS

3. Materials and method

3.1 Study design:

This was a/cross- sectional study.

3.2 Study area

The study was conducted at Medical Military Hospital in Khartoum State,,Sudan.

3.3 Study population

Randomly one hundred patients from both genders 50 symptomatic and 50 asymptomatic attending Medical Military Hospital, were enrolled in this study

3.4 Study duration

This study was carried out between March and October 2016.

3.5 Ethical consideration

Approval was taken from the faculty and hospital administration and verbal consent was taken from all participants.

3.6 Data collection

Structured questionnaire was used for collection of the data which include blood group, smoking and age. .

3.7 Sample collection

3.7.1 Blood sample

5 ml of Blood samples were collected in plain blood container let to clot. Then the serum was separated by centrifugation to subsequent detection of *H. pylori* antibody by using commercial kits immunochromatography test cards (*H. pylori* Antibody Rapid Test Cassette All Test Biotech Company).

3.7.2 Stool sample

Fresh Stool samples were collected into wide mouth container outer labeled for detection of *H. pylori* antigen by Using wood stick small portion of the stool sample was transferred into the buffer, incubated for 2 minutes and then two to three drops of the mixture were poured in the hole of the ICT of *H. pylori* stool antigen detection. (*H. pylori* Antigen Rapid Test Cassette All Test Biotech Company).

3.8 laboratory methods

3.8.1 Immunochromatography test

3.8.1.1 Principle of the test

Rapid H. pylori antibody test employs chromatographic lateral flow test device in a cassette format. Colloidal gold conjugate *H. pylori* antigen are dry immobilized at the end of nitrocellulose membrane strip. *H. pylori* antigens are bound at the test zone (T). When the sample was added; it was migrates by capillary diffusion rehydration the gold conjugate. If anti *H. pylori* antibodies present in sample

antibodies will bind with the gold conjugated antigen forming particle . These particle will continue to migrate long the strip until the test zone (T) where they are captured by *H. pylori* antigens generating visible red line .

3.8.1.2 storage and stability

The sealed pouches in the kit may be stored between 3-4c0for the duration of the shelf life as indicated on the pouch

3.8.1.3 Procedure of the test

The kits components were brought to room temperature, the pouch and card were opened. Once opened the test card must be used immediately. The test card was labeled with patients identity, 2-3 drops of serum or stool mix with buffer were applied to the sample well marked as (S). At the end of 10 minutes the result were red.

3.8.1.4 Interpretation of the results

Negative :only control line appears

Positive:both control line and the test line appear

3.8.1.5 Invalid results

After 10 minutes no line is visible within the control zone the result is invalid . The test should be repeated with a new test card**.**

3.9 Statistical analysis

The data was analyzed using Statistical Package of Social (SPSS) version 20.0. frequencies, cross tabulation and p. values were calculated by Chi-Square and significant level were set at (P $<$.0.05).

3.10 sensitivity and specificity

Sensitivity and specificity were calculated using the formula:

$$\text{Sensitivity} = \frac{\text{True positive}}{\text{True positive} + \text{false negative}}$$

$$\text{Specificity} = \frac{\text{True negative}}{\text{False positive} + \text{true negative}}$$

3.11 ABO blood group sets.

Used for blood group detection.

CHAPTER FOUR

RESULTS

4. RESULTS

The distribution of symptomatic and asymptomatic patients are shown in table1.

Table (4.1) Gender distribution of the symptomatic and asymptomatic groups

Gender	Symptomatic	Asymptomatic	Total
Male	24(48%)	18(35%)	42(42%)
Female	26(52%)	32(64%)	58(58%)
Total	50(100%)	50(100%)	**100(100%)**

Smoker and non smoker were almost similar in symptomatic and a

symptomatic patient shown in table 2.

Table (4.2) Distribution of smoking among symptomatic and asymptomatic Groups

Smoking	Symptomatic	Asymptomatic	Total
Smoker	7(14%)	6(12%)	13(13%)
Non smoker	43(86%)	44(88%)	87(87%)
Total	50(100%)	50(100%)	**100(100%)**

Variation in blood grouping was found among both participants are shown in table 3.

Table (4.3) Distribution of blood group among symptomatic and asymptomatic groups

Blood group	Symptomatic	Asymptomatic	Total
O+ve	30(60%)	20(40%)	50(50%)
O-ve	3(6%)	2(4%)	5(5%)
A+ve	7(14%)	12(24%)	19(19%)
A-ve	0(0%)	6(12%)	6(12%)
B+ve	6(12%)	8(16%)	14(14%)
B-ve	4(8%)	2(4%)	6(6%)
Total	50(100%)	50(100%)	**100(100%)**

The prevalence of antigen and antibody in the stool and serum for symptomatic patients shown in table 4.

Table (4.4)Comparison between the results of stool antigen and serum antibody in the participants (symptomatic and asymptomatic).

The results	ICT	
	Antigen	Antibody
Positive	11(11%)	58(58%)
Negative	89(89%)	42(42%)
Total	100(100%)	100(100%)

(P. value 0.00) These findings showed significant results between antigen and antibody test result in diagnosis of *H. pylori* P value <0.05.

Sensitivity of antigen test 48%. Specificity of antigen test 48%.

Sensitivity of serum antibody test 47%. Specificity of serum antibody test 47%.

The relationship between smoking and and *H. pylori* antigen in the participants (symptomatic and asymptomatic) shown in table 5

Table (4.5) Comparison between the results of stool antigen detection of *H. pylori* and smoking among the participants (symptomatic and asymptomatic).

Smoking	Positive Antigen	Negative Antigen	Total
Yes	2(2%)	11(11%)	13(13%)
No	10(10%)	77(77%)	87(87%)
Total	12(12%)	88(88%)	100(100%)

P.value 0.08 the findings showed insignificant association between smoking and *H. pylori* antigen results P.value >0.05.

The relationship between smoking and *H. pylori* antibody among participants(symptomatic and asymptomatic)shown in table 6.

Table (4.6) Comparison between the results of serum antibody of *H. pylori* and smoking among the participants(symptomatic and a symptomatic).

Smoking	Positive Antibody	Negative Antibody	Total
Yes	8(8%)	5(5%)	13(13%)
No	49(49%)	38(38%)	87(87%)
Total	57(57%)	43(43%)	100(100%)

(P. value 0.06). These finding showed insignificant results between smoking and *H. pylori* antibody P.value >0.05

The relation between blood grouping and tests in the tested samples shown in table 7.

Table (4.7) Comparison between the results of the stool antigen detection of *H. pylori* and blood group among the participants (symptomatic and asymptomatic).

Blood group	Positive Antigen	Negative Antigen	Total
O	28(28%)	39(39%)	67(67%)
A	3(3%)	22(22%)	25(25%)
B	4(4%)	4(4%)	8(8%)
Total	35(35%)	65(65%)	100(100%)

P.value 0.8). These finding showed insignificant results between blood group and (*H. pylori* antigen P. value >0.05.

 The relation between blood grouping and tests in the tested samples shown in table 8.

Table (4.8) Comparison between the results of the serum antibody detection of *H. pylori* and blood group among the participants(symptomatic and asymptomatic).

Blood group	Positive Antibody	Negative Antibody	Total
O	32(32%)	24(24%)	56(56%)
A	14(14%)	12(12%)	26(26%)
B	11(11%)	7(7%)	18(18%)
Total	(57%)57	73(73%)	100(100%)

P.value 0.6). These finding showed insignificant results between *H. pylori* antibody and blood group P. value >0.05.

The relationship between the age groups and *H. pylori* antigen in the participants (symptomatic and asymptomatic) shown in table

Table (4.9) Comparison between the results of the stool antigen detection of *H. pylori* and age groups among the participants

Age groups	Positive Antigen	Negative Antigen
5-20	5(5%)	13(13%)
21-36	12(12%)	22(22%)
37-51	2(2%)	18(18%)
52-67	5(5%)	9(9%)
68-83	4(4%)	10(10%)
Total	28(28%)	72(72%)

P. value 0.2). These finding showed insignificant results between *H. Pylori* antigen and age groups P. value>0.05.

The relationship between the age groups and *H. pylori* antibody in the participants (symptomatic and asymptomatic) shown in table10.s

Table (4.10) Comparison between the results of the serum antibody detection of *H. pylori* and age groups among the participants

Age groups	Positive Antibody	Negative Antibody
5-20	10(10%)	6(6%)
21-36	20(20%)	12(12%)
37-51	10(10%)	9(9%)
52-67	9(9%)	11(11%)
68-83	4(4%)	8(8%)
Total	53(53%)	47(47%)

P. value 0.2). These finding showed insignificant results between *H. Pylori* antibody and age groups P. value>0.05.

CHAPTER FIVE

DISCUSSION

DISCUSSION CONCLUSIONS& RECOMMENDATIONS

5.1 DISCUSSION

In this study 100 patients were enrolled 50 symptomatic and 50 asymptomatic with differents age group and gender characteristic . The results of *H. pylori* antibody detection in patients serum was 58(58%) positive and in stool detection was 11(11%) positive. These results were lower than those reported in eastern Sudan by (Abdallah *et al.,* 2014) where a seroprevalence of 65% was reported. The variation according to (Abdallah *et al*., 2014) may be due to the difference in sample size, study population, Different in diets intake as vitamin C and citrus fruit rich diet are effective in the prevention of *H .pylori* infection, socioeconomic status, water supply, educational level and behavior, ethnic background and age group all these have effect in the infection. The highest age groups showing positive results was found between 21-36 years (12 patients showed positive results for antigen whereas20 (20%) showed positive results for serum antibody).These results different from results reported by (Suerbaum and Michetti, 2002) in developing country who found *H. pylori* more common in middle age between 20-50years. in The study showed that *H. pylori* positivity decrease with age. These results disagree with results of (Kanby *et al.,* 2005).

In this study smoking does not have clear effect on patients infected with *H. pylori* as the P.value was >o.o5. This comes in total disagreement with results and suggestion of (Ogihara *et al.,* 2000) who suggested that smoking has positive effect in protection against *H. pylori* infection. Moreover, these results are similar to those reported by (Chodos, 1988). Who found there was no significant difference between the rate of infection among smoker and non smoker patients and disagree with (Martin, 1989). From USA who found there was significant difference between the infection and smoking. Differences in results may be due to difference in behaviors, sample size, study population and may be due to

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attributable to high levels of hydrochloric acid excretion which is stimulated by cigarette smoking. HCL protective barrier against *H. pylori* infections .

In the current study blood group O have higher susceptibility to *H. pylori* infection than other blood groups but there was statistically insignificant relationship between ABO blood group and *H. pylori* infection (P> 0.05). These results agree with (Mentis *et al.,* 1991) where there was no relationship between *H. pylori* infection and ABO blood group and this disagree with (Kanbay *et al*., 2005). Who reported relationship between *H. pylori* and blood group O. The different in the results may be due to different in sample size and study population, and blood group O individuals express higher inflammatory and socioeconomic level.

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1.2 Conclusions

1. Study explain that there is no correlation between, blood group, smoking and age.
2. Findings of positive results in serum antibody method was highly among (O) positive blood group individuals.
3. Blood group O have higher susceptibility to *H. pylori* infection than other blood group but there was statistically insignificant.
4. Antigen detection more sensitive and specific than antibody detection.

1.3 Recommendations

1) Further studies and using large sample size , and advanced techniques to ensure more accurate results.

2) Further studies might involve culture and sensitivity before and after treatment to confirm the eradication of bacteria.

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بسم الله الرحمن الرحيم

جامعة السودان للعلوم و التكنولوجيا

كليه الدراسات العليا

برنامج ماجستير العلوم في المختبرات الطبيه قسم الاحياء الدقيقه

استبيان لجمع العينات بغرض البحث

Questionnaire

Serial number ……………………………………………

Name……………………………………………

Sex [ ]   Age [ ]

Male   Female

BLOOD group

A [ ]   B [ ]   O [ ]   Other [ ]

Smoking [ ]

Type of tests used

BLOOD-Ab detection [ ]

STOOL Ag detections [ ]

ICT for *H. pylori* Ag

ICT for *H. pylori* Ab