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DETECTION OF ANTI-HELICOBACTER PYLORI IgG ANTIBODIES IN HEALTHY SUBJECTS

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ABSTRACT

Introduction: Helicobacter pylori (H. pylori) are Gram – negative microaerophilic, spiral organisms. Factors such as family history of gastric disease, source of drinking water, number of siblings, sharing beds, and level of hygiene have been linked to acquisition of H. pylori infection. Most of the infected people do not have clinical symptoms. The study was planed to determine the level of anti-H. pylori IgG antibodies in the serum of healthy individuals.

Material and Methods: The study included 80 healthy subjects and was conducted in the Department of Immunology, University of Health Sciences Lahore. The studied population was divided on the basis of (a) eating food from outside home daily, twice a week or once a week, (b) using filtered or tap water for drinking, and (c) having family history of gastric ulcer or without family history of gastric ulcer. Level of anti H. pylori IgG antibody was determined by ELISA technique.

Results: Among 80 asymptomatic healthy individuals anti-H pylori IgG antibody was detected in 28 (35%) subjects who did not have these antibodies (p-value < 0.001). Mean level of anti-H. pylori IgG antibodies was 43 ± 39.3 U/ml, 30.7 ± 37.3 U/ml and 14.9 ± 19.7 U/ml in subjects eating food from outside their homes once a week, twice a week and daily respectively. Statistically significant difference was observed in the level of H. pylori antibodies with different eating habits (p = 0.015). However no statistically significant difference was observed in the level of anti-H. pylori antiodies between two genders, individuals using tap water and filtered water for drinking and with family history of gastric ulcer.

Conclusion: H. pylori IgG antibodies were present in asymptomatic healthy individuals of both the genders.

Key Words: Helicobacter pylori, Antibody, ELISA.

INTRODUCTION

Helicobacter pylori (H. pylori) are Gram – negative microaerophilic, spiral or gull – wing shaped organisms that can survive in stomach and duodenum. In 2005 Marshall and Warren received Noble prize on discovering the role of *H. pylori* in patients of gastritis and peptic ulcer disease.[1] It is the most common chronic bacterial infection which occurs worldwide but its prevalence varies among countries and even in different populations of the same country. About 80% of the individuals infected with *H. pylori* in developing countries and about 50% in the developed countries are asymptomatic. Only 10% to 20% of *H. pylori* infected individuals develop gastric hyperacidity and peptic ulcer. *H. pylori* infection varies widely by geographical area, age, race, and socioeconomic status.[2,3]

Exact mode of transmission is not known but housing cluster, overcrowding, number of siblings, sharing beds, level of hygiene, family history of gastric disease, and source of drinking water; have been linked to acquisition of *H. pylori*. The organism has been cultured from stool samples of infected children and adults therefore, probably the organism is transmitted by fecal – oral route.[2,4,5]

Strong humoral and cellular immune response is mounted against *H. pylori*. Normal gastric acidity and its peristaltic movements inhibit bacterial colonisation but *H. pylori* can evade defense mechanisms and establish infection that may persist for lifetime in the host. Survival of *H. pylori* in acidic stomach is dependent on urease which is an important virulence factor for gastritis[2] along with cag pathogenicity island (cag PAI) vacuolation cytotoxin, and cytotoxin associated gene A (Cag A) protein.[6,7] *H. pylori* also evade immune system by producing enzyme arginase which prevents nitric oxide (NO) production and therefore, it can survive within macrophages by interfering with lysosomal proteins.[8] The organism also evades adaptive immune response by virulent factor (Vac A) which blocks antigen dependent proliferation of T – cells.[9] Lipopolysaccharides (LPS) an flagellin (Fla A) of Gram negative bacteria activate TLR 4 and TLRS respectively but

LPS and Fla A of *H. pylori* are 100 – 1000 TLR assist in evasion of immune system.[10]

Laboratory techniques for the detection of H. pylori include both invasive and non-invasive methods. Invasive methods include rapid urease test, histological identification and culture of the organism from gastric biopsy. Non-invasive methods are urease breath test, serological tests, detection of *H. pylori* stool antigen (HpSA) and *H. pylori* antibodies in urine and saliva.[11] Urease breath test has advantages over serological analysis, but is expensive and time – consuming whereas serological techniques offer high sensitivity and specificity. It has been suggested that serological diagnosis was sufficient to detect *H. pylori* infection and it should be replaced by endoscopy – based procedures. Generally non-invasive tests are used for screening *H. pylori* in healthy population.[12-14] Therefore, a study was planed to determine the level of *anti-H. pylori* IgG antibodies in the serum of healthy individuals and ELISA method was used for this purpose.

MATERIAL AND METHODS

It was an observational cross sectional study conducted at the Department of Immunology, University of Health Sciences Lahore during February 2010 – September 2010. It included 80 healthy subjects and the studied population was divided on the basis of (a) eating food from outside home daily, twice a week or once a week, (b) using filtered or tap water for drinking, and (c) having family history of gastric ulcer or without family history of gastric ulcer. Level of *H. pylori* IgG antibody was determined by ELISA technique using quantitative immunoassay kit from Bio check, Inc (U.S.A).

Statistical Analysis

The data was entered and analysed using Statistical Pacage for Social Sciences 16 (SPSS – 16). Mean ± SD was given for quantitative variables. Frequencies, percentages and graphs were given for qualitative variables. Two independent sample *t* test was applied to observe group mean differences. A *p-value* of equivalent to < 0.05 was considered statistically significant.

RESULTS

Eighty (80) healthy subjects included in this study comprised of 31 (38%) males and 49 (64%) females. *Anti-H. pylori* IgG antibodies were detected in 28 (35%) subjects while 52 (65%) subjects did not have these antibodies in the serum (Fig. 1). Subjects with *anti-H. pylori* IgG antibodies had their mean level of 71 ± 30 U/ml and it was 8.9 ± 4.1 U/ml in subjects who did have these antibodies. The difference in the level of *anti-H. pylori* IgG antibodies between these groups was statistically significant (p = 0.001) (Table 1). The level of *anti-H. pylori* IgG antibodies among males and females was 38.7 ± 40.5 U/ml and 25.7 *H. pylori* 31.3 U/ml respectively, the difference in the level of *anti-H. pylori* IgG antibodies between these groups was not statistically significant (p = 0.11) (Table 2).

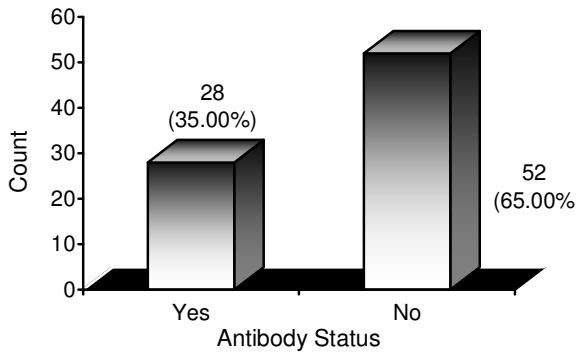

Fig. 1: *Frequency of anti-H. pylori IgG antibodies in healthy subjects.*

Table 1: *Number, mean and comparison of anti-H. pylori antibodies in healthy individuals.*

Antibody present Mean ± SD (n = 28)	*Antibody absent Mean ± SD (n = 52)*	*p-value*
71.4 ± 32 U/ml	8.90 ± 4.14 U/ml	< 0.001

n = number of patients; *p-value* < 0.05 = significant

Table 2: *Number, mean and comparison of anti-H. pylori antibodies in healthy individuals between two genders.*

Males Mean ± SD (n = 31)	*Females Mean ± SD (n = 49)*	*p-value*
38.7 ± 40.5 U/ml	27.5 ± 31.3 U/ml	0.11

n = number of patients; *p-value* < 0.05 = significant

Mean level of *anti-H. pylori* IgG antibodies was 43 ±39.3 U/ml, 30.7 ± 37.3 U/ml and 14.9 ± 19.7 U/ml in participants who used to eat once a week, twice a week and daily fom outside home respectively. Statistically significant difference was observed in the level of *H. pylori* antibodies among individuals with different eating habits (*p* = 0.015) (Table 3). By applying post Hoc test statistically significant difference was observed in the level of *H. pylori* antibodies among participants who were eating from outside home once a week (*p* = 0.011). No statistically significant difference was observed bet-

ween individuals using tap water or filtered water for drinking (p = 0.701). Among participants with family history of gastric ulcer, mean level of *H. pylori* antibodies was 30.2 ± 31.9 U/ml and it was 30.9 ± 36.6 U/ml who did not have family history of gastric ulcer. The difference in the level of *anti-H. pylori* IgG antibodies between these two groups was not statistically significant (p = 0.948).

Table 3: *Number, mean and comparison of anti – H. pylori antibodies in healthy individuals with eating habits.*

Once a week Mean ± SD (n = 30)	*Twice a week Mean ± SD (n = 27)*	*Daily Mean ± SD (n = 23)*	*p-value*
43.05 ± 39.3 U/ml	30.7 ± 37.3 U/ml	14.90 ± 19.7 U/ml	< 0.001

n = number of patients; *p-value* < 0.05 = significant

DISCUSSION

In the present study, among 80 healthy subjects *Anti-H. pylori* IgG antibodies were detected in 28 (38%) subjects. The present study is not in agreement with Bakka *et al*[15] who documented *anti-H. pylori* IgG antibodies in 66% of healthy subjects in the same age group. Studies showed variation in the prevalence *of anti-H. pylori* IgG antibodies in different parts of the world such as Rodrigues *et al*[16] found 77% in Brazil, Gracia *et at*[16] documented 17% in Orense, and Kirn *et al*[18] showed 46% in South Korea among healthy individuals.

In the present study there was no statistical significant difference in the level of *anti-H. pylori* IgG antibodies in two genders. Our findings are similar to the studies of Bakka *ct al,*[15] Rodrigues *et al*[16] and Kirn *et al*[18] who also could not find statistically significant difference in the level *of H. pylori* IgG antibodies in both the genders. In the present study the difference in the level of *anti-H. pylori* IgG antibodies between the participants who were using tap water or filtered water for drinking was not statistically significant (p = 0.701). The findings of Lin et *al*[19] was similar to the present study because he also could not find statistically significant difference in the level of *anti-H. pylori* IgG antibodies among people who used tap water and river or well water for drinking but Nurgalieva *el al*[20] linked drinking river water with high risk of *H. pylori* infection as compared to tap water.

In the present study, mean antibody level of *anti-H. pylori* IgG antibodies was 30.17 ± 31.9 U/ml and 30.9 ± 36.6 U/ml in subjects having family history of gastric ulcer and those who did not have such family history respectively and the difference in the level of *anti-H. pylori* IgG antibodies between these groups was not statistically significant. Escobar[22] *et al* reported high level of *anti-H. pylori* antibody in subjects with family history of peptic ulcer disease as compared to those who did not have such family history. Salih[21] *et al* found that children of mothers with gastric ulcer were at a greater risk of acquiring *H. pylori* infection and he suggested through physical contact mothers transmit *H. pylori* to their children.

In the present study, statistically significant difference was observed in the level of *H. pylori* antibodies among individuals with different eating habits (p = 0.015). Beguc *el al*[23] documented increased prevalence of *H. pylori* infection in individuals who consumed food from street vendors and he supported that food prepared under unhygienic conditions as a source of transmission of this infection. On the other hand the study of Duynhove *et al*[24] documented no direct evidence of food in the transmission of *H. pylori.*

It is *concluded* that *anti-H. pylori* IgG antibodies were found in asymptomatic healthy individuals of both genders.

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