Seroprevalence of anti-*Helicobacter pylori* antibodies in population of Sulaimani governorate/Kurdistan Region/Iraq

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Abstract:

***Helicobacter pylori* (*H. pylori*) infection is one of the most common chronic bacterial infection in the world. Objective: This study was done to assess the extent of *H. pylori* infection in Suleimani governorate by noninvasive ELISA technique for quantitative detection of anti-*H. pylori* IgM, IgG and IgA antibodies in serum. Methods: The study included 335 adult and children volunteers from Chamchamal and Sulaimani cities in Kurdistan region / Iraq. The participants answered a structured questionnaire and a blood sample was taken for serological analysis from each participant. The level of serum antibodies for IgM, IgG and IgA were detected using ELISA technique. Results: In the total sample 20.4% had positive IgM, 32.3% positive IgG and 58.2% positive IgA. Two hundred thirty four (74%) subjects had one or more of the antibodies, 138 subjects (41.1%) had at least one antibody positive (IgM 11 (3.2%), IgG 35 (10.4%), IgA 92 (27.4%). In this study 20% had at least two antibodies positive {IgM 25 (7.4%): IgG 45 (13.4%): IgA 64 (19.1%)}. Conclusions: *H. pylori* infection is common in population of Sulaimani governorate. IgA anti-*H. pylori* is the most prevalent antibody of population in Sulaimani governorate. The age group of 20-30 years old has the most seroprevalence of antibodies specific to *H. pylori*.**

Keywords: Antibodies, Helicobacter Pylori, IgA, IgG, IgM, seroprevalence.

Introduction

Previous seroepidemiological studies indicate that about 50% of adults in the developed countries and nearly 90% of the adults in developing countries were seropositive for *H. pylori* [1]. Chronic *H. pylori* infection may be associated with chronic gastritis [2], peptic ulcer disease [3], mucosal associated lymphoid tissue lymphoma [4], and gastric adenocarcinoma [5].

H. pylori infection, once acquired is believed to persist throughout life unless treated [6]. The prevalence of *H. pylori* in children of developing countries is higher while in developed countries seroprevalence is more at young age [7,8,9].

H. pylori is a pathogenic, Gram-negative, microaerophilic bacterium that can inhabit the stomach, esophagus and upper duodenum. It can cause parietal cell degradation and thus impairs the production of hydrochloric acid in the stomach leading to compromised protein digestion, gastritis, peptic ulcer, acid reflux, and stomach cancer [10]; the infection might also be asymptomatic [11].

The diagnosis of *H. pylori* infection is made by invasive and noninvasive techniques. The invasive techniques

involve endoscopy during which biopsy specimens of the stomach and duodenum are obtained, and the diagnosis of *H. pylori* can be made by urease testing, histology staining, bacterial DNA detection by polymerase chain reaction, and or bacterial culture. While the noninvasive technique include detection of *H. pylori* antigen in stool, serological assays measure specific anti-*H. pylori* antibodies by quantitative or qualitative serology (anti-*H. pylori* IgM, IgG or IgA), or urea breath test [12].

Serological testing is often used to determine the presence or absence of infection with this organism [13]. Moreover, serology may be useful in monitoring the effectiveness of treatment in infected individuals [14, 15]. Studies supporting the clinical utility of IgA serology have appeared has not been clarified till now [16]. In the presence of IgG, IgA has been shown to correlate with active infection in 95% and 74% of cases of duodenal and gastric ulcers, respectively [17].

The magnitude of *H. pylori* infection among Sulaimani population has not been estimated earlier. The aim of this study is to analyze the impact of *H. pylori* infection in Suleimani governorate by noninvasive ELISA technique using quantitative detection anti *H. pylori* IgM, IgG and/or IgA in serum.

Material and methods

The study included 335 adults and children volunteers who were chosen by systematic random sampling method from Chamchamal and Sulaimani cities in Sulaimani governorate in Kurdistan region/Iraq. The study extended between August 2008 and October 2009. The study group was subjected to a questionnaire that included demographical and clinical data. An ELISA technique (Monobind Inc., USA) was performed for detecting the presence of IgM, IgG and/or IgA anti *H. pylori* antibodies in the serum of each participant in the study group.

Results

In this study 335 subjects participated. The mean age was 42 years and the median was 31 years and the age ranged from 5 to 85 years. The proportion of males to females was equal. The majority were urban (90%) but 46% were from Sulaimani and 54% were from Chemchamal.

The distribution of positive serology for IgM, IgG and IgA is shown in table 1. It is clear that only urbanization (district) was related to the prevalence of IgM which shows that living in Sulaimani is related to a higher prevalence (30.4%) compared to living in Chamchamal (12.8%). While for IgG occupational status and urbanisation were related to the prevalence of IgG. Being military officers and the other group "other occupational group" had the highest prevalence of IgG and also living in a large city like Sulaimani was related to a higher prevalence (52.2%) as compared to Chamchamal (20.6%). But for IgA marital status and urbanization were only related to the prevalence. Being widowed or divorced and living in Sulaimani were related to a higher prevalence.

In the total sample 20.4% had a positive IgM, 32.3% had a positive IgG and 58.2% had a positive IgA. 234 (74%) subjects had some of the antibodies. 138 subjects (41.1%) had at least one antibody positive. For IgM the figure was 11 (3.2%): IgG 35 (10.4%): IgA 92 (27.4%). Six out of the total sample 67 subjects (20%) had at least two antibodies positive {IgM 25 (7.4%): IgG 45 (13.4%): IgA 64 (19.1%)}. 29 subjects (8.6%) had all three antibodies positive.

The level of IgM varied with age (Table 1). The highest level was detected

among the age group 50+. The lowest level was found among those ≤19 years. In both sexes about 20% had positive IgM. Interesting is that the prevalence of positive IgM is highest among those working as officers in the military. Also divorced had the highest prevalence as compared to other groups. This is also true for those who are from urban areas or from Sulaimani City as compared to others.

The prevalence of IgG was highest among the age group 31-40 years and among males (Table 1). The prevalence was also high among officers as well as among other occupational groups but interesting is that the prevalence was highest among singles than among other groups like married, divorced or widowed. This prevalence of IgG is also highest among urban and subjects living in Sulaimani.

The prevalence of IgA is highest among the age group 31-40, females, officers, widowed, urban subjects and those living in Sulaimani as compared to other groups (Table 1).

Interesting to notice that only district was the only variable significantly related to the prevalence of IgM, IgG and IgA. Occupations and residency were significantly related to IgG and sex to IgA.

Among life style factors, complaint symptom sand drugs only pain was significantly related to IgM, Alcohol to IgG and smoking to IgA (Table 2). Having pain was related to lower prevalence of IgM, alcohol use to higher prevalence of IgG and smoking to lower prevalence of IgA.

In the logistic regression when the impact of confounders was taken into consideration none of the studied variables were related to increased risk for IgM (Table 3). Only occupation and marital status were related to IgG meaning that other working groups, hand workers and officers had the highest odds ratio for IgM than the reference group (students). Singles had the highest OR for IgA as compared to married subjects.

Table 1. The prevalence (%) of seropositivity by sociodemographic characteristics.

	N	IgM	Std Dev	IgG	Std Dev	IgA	Std Dev
Total sample	335	20.4	0.44	32.3	0.48	58.2	0.49
Age, *p-value*			n.s.		n.s.		n.s.
≤19	23	4.4	0.21	21.7	0.42	65.2	0.49
20-30	132	21.2	0.41	31.8	0.47	52.3	0.50
31-40	74	23	0.42	37.8	0.49	66.2	0.48
41-50	50	18	0.39	38	0.49	60	0.49
50-	39	25.6	0.44	39	0.49	56.4	0.50
Sex, *p-value*			n.s.		n.s.		<0.01
Male	167	20.7	0.41	36.8	0.48	50.3	0.50
Female	168	20.3	0.40	31.9	0.47	65.6	0.48
Occupation, *p-value*			n.s.		**<0.001**		n.s.
Student	24	8.3	0.28	16.70	0.38	54.2	0.51
Handworker	59	18.6	0.39	35.6	0.48	52.5	0.50
Officer	85	28.2	0.45	44.7	0.50	64.7	0.48
Housewife	87	12.6	0.33	19.5	0.40	64.4	0.48
Retired	6	33.3	0.52	16.7	0.41	50	0.55
Others	57	26.3	0.44	49.1	0.50	47.4	0.50
Marital status, *p-value*			n.s.		n.s.		<0.01
Married	208	18.8	0.39	30.3	0.46	51.4	0.50
Single	92	23.9	0.43	44.6	0.50	70	0.62
Divorced	7	28.6	0.49	28.6	0.49	71.4	0.49
Widow	11	18.2	0.41	27.2	0.47	81.8	0.40
Residency, *p-value*			n.s.		<0.05		n.s.
Urban	283	21.2	0.41	36.3	0.48	58.3	0.49
Rural	35	14.3	0.36	17.1	0.38	57.1	0.50
District, *p-value*			**<0.0001**		**<0.0001**		**<0.0001**
Sulaimani	138	30.4	0.46	52.2	0.50	61.6	0.49
Chamchamal	180	12.8	0.34	20.6	0.41	55.6	0.50

Table 2. The prevalence (%) of seropositivity by life style, complaints and drug use.

N		IgM	Std Dev	IgG	Std Dev	IgA	Std Dev
Chronic disease			n.s.		n.s.		n.s.
Yes	47	21	0.38	38.3	0.49	64	0.49
No	271	17	0.41	35.6	0.47	57	0.50
Family History			n.s.		n.s.		n.s.
Yes	29	17.2	0.38	27.6	0.45	52	0.51
No	289	20.8	0.41	35	0.48	59	0.49
Smoking			n.s.		n.s.		**<0.05**
Yes	42	14.3	0.35	40.5	0.50	43	0.50
No	276	21.4	0.41	33.3	0.47	61	0.49
Tea drinking (Cups/day)			n.s.		n.s.		n.s.
0	19	21.1	0.42	32	0.48	52.6	0.51
1-3	180	21.1	0.41	35	0.48	62.8	0.49
≥ 3	119	19.3	0.40	34	0.47	52.1	0.50
Alcohol			n.s.		<**0.01**		n.s.
Yes	21	23.8	0.44	62	0.50	42.9	0.51
No	297	20.2	0.40	32.3	0.49	52.3	0.49
Pain			**<0.05**		n.s		n.s.
Yes	86	12.8	0.34	32.6	0.47	58.1	0.50
No	232	23.3	0.42	34.9	0.48	58.2	0.49
Dyspepsia			n.s.		n.s.		n.s.
Yes	42	19	0.40	40.5	0.50	50	0.51
No	276	20.7	0.41	33.3	0.47	49.4	0.49
Others complain			n.s.		n.s.		n.s.
Yes	14	21.4	0.43	35.7	0.50	57.1	0.51
No	104	20.3	0.40	34.2	0.48	58.2	0.49
Non-steroidal anti-			n.s.		n.s.		n.s.
Yes	53	22.6	0.42	45.3	0.50	60.4	0.49
No	265	20	0.40	32.1	0.47	57.8	0.50

Table 3. Logistic regression analysis with odds ratios (OR) and 95% confidence interval for seropositivity in relation to possible determinants.

		IgM		IgG		IgA	
		OR	95%CI	OR	95%CI	OR	95%CI
Age							
	<19	1.00		1.00		1.00	
	20-30	3.94	0.42-37.3	0.87	0.23-3.30	0.56	0.18-1.79
	31-40	4.38	0.45-44.1	1.10	0.27-4.45	1.13	0.32-3.97
	41-50	3.21	0.30-34.5	1.12	0.26-4.85	0.87	0.24-3.22
	50-	6.67	0.62-72.3	1.88	0.42-8.44	0.68	0.18-2.60
Sex							
	Male	1.00		1.00		1.00	
	Female	1.34	0.67-2.70	1.22	0.66-2.50	1.45	0.80-2.64
Occupation							
	Student	1.00		1.00		1.00	
	Handworker	1.79	0.30-10.8	4.83	1.10-21.18	2.38	0.69-8.17
	Officer	2.80	0.50-15.6	5.37	1.28-22.56	2.69	0.81-8.90
	Housewife	0.91	0.14-5.71	1.75	0.40-22.56	2.70	0.77-9.43
	Retired	3.13	0.30-36.4	1.08	0.08-14.4	2.14	0.28-16.4
	Others	2.67	0.46-15.6	**7.70**	1.78-33.27	1.33	0.39-4.75
Marital status							
	Married	1.00		1.00		1.00	
	Single	1.35	0.68-2.69	2.07	1.18-3.82	3.00	1.58-5.71
	divorced	1.45	0.25-8.42	0.59	0.10-3.32	3.74	0.66-21.29
	Widow	1.28	0.22-7.48	1.25	0.27-5.80	3.03	0.57-15.96
Residency							
	Urban	1.00		1.00		1.00	
	Rural	0.85	0.39-2.40	0.45	0.17-1.22	1.08	0.50-2.36
Chronic disease							
	Yes	1.00		1.00		1.00	
	No	1.63	0.69-3.83	0.90	0.45-1.80	0.83	0.41-1.66

Discussion

In this study, the seroprevalence of IgM, IgG, and IgA antibodies specific to *H. pylori* in the sera of adult population in Sulaimani governorate was investigated. The prevalence of all the three types together was 74%. The prevalence of *H. pylori* infection was not studied before in this city but the prevalence was estimated before in other parts of Iraq like that which is done in Al-Anbar city in 2000 and it was 77% [18], a result which is near to the results presented in this study. However, in Al-Anbar city the IgG antibodies was the only antibody studied and this might underestimate the real prevalence.

The seroprevalence of IgM antibodies specific to *H. pylori* was 20.4% and this indicates that one fifth of the population is with seroconversion and are possibly recently infected with *H. pylori* which is relatively a high percentage when compared to other studies [19]. The positive IgM test result indicates an active infection and colonization by *H. pylori*. It

does not necessarily indicate that gastrointestinal disease is present.

The higher prevalence of IgM anti- *H. pylori* antibodies among the elderly reflects infection at later ages, though some of them (7.4%) might represent re-infections as they have another antibody (IgG or IgA) in addition to IgM or all the three types of antibodies together (8.6%); these results might also be explained as conversion of acute infections from IgM to IgG and or IgA types.

The retired subjects and officers had the highest prevalence of IgM anti-*H. pylori* antibodies among different occupational groups and this might be related to their age group. The prevalence of IgM anti-*H. pylori* antibodies in Suleimani city are higher than in Chamchamal city and the difference was statistically significant. Chamchamal city is a small city located in the west part of Suleimani city. Chamchamal city has lower socioeconomic status and lower population than Suleimani city. Thus the crowding and style of life in Suleimani city might cause the transmission of H. pylori to be easier and reflects higher urbanization than Chamchaml.

The study showed that epigastric pain is not a significant feature in those who are IgM anti-*H. pylori* positive, and paradoxically those who are free of pain were more and their number was statistically significant. This indicates that pain is not an essential clinical feature of *H. pylori* infection and asymptomatically infected people are more than those with epigastric pain and the development of peptic ulcer might follow the infection after long duration and other factors might be important in development of peptic ulcer. Sex, marital status, resident in urban or rural areas, presence of chronic diseases especially history of chronic peptic ulcer, presence of family history of peptic ulcer, cigarette smoking, alcohol drinking, tea drinking, presence of dyspepsia or other complaints, and the use of non-steroidal anti-inflammatory drugs are not significant parameters associated with presence of IgM anti-*H. pylori.* Abdulrauof and his colleagues found similar results but they conclude that tea drinking is protective against *H. pylori* infection, a finding which is contradictory to the findings in the present study [20].

The seroprevalence of IgG antibodies specific to *H. pylori* was 32.3% which is less than the seroprevalence or the same antibody in Duhok city/Iraq in 2008 which was 79% [21]. Many other studies done in neighboring countries: in Eastern Turkey, 64% of the population was found to be seropositive for *H. pylori* [22] whereas in Malayer a city in Iran, *H. pylori* seroprevalence was only 38.83% [23-25]. In another study from Iran, *H. pylori* sero-positivity was 48% in Ardebil province and 31% in Yazd province [26]. The Iranian results were close to the results from this study. High infection rates have been reported from other developing countries, e.g., Thailand [27], Mexico [28], India [29], Japan and Korea [30], or parts of countries, e.g., Siberia [31]. Also, a similarly high prevalence has been reported from an early, smaller study investigating the seroprevalence of *H. pylori* infection in Vietnam and other countries [32]. Somewhat lower prevalence rates, 40 to 60%, have been reported from Mexico [33], Korea [34] and from parts of a multiracial population in Malaysia. Seroprevalence rates around or below 40% have been reported for ethnic Malay [35] and Seoul (28) populations.

The relatively lower seroprevalence of IgG antibodies specific to *H. pylori* in Sulaimani city might be related to type of personal habit and cofactors related to bacterial transmission. The seroprevalence of IgG antibodies specific to *H. pylori*

among population is higher in age group above 50 years and this might reflect the cumulative effect of previous infections among people. This result is in accordance to that done in Al-Anbar city (18) but some what differs from results in Duhok city (21). Concerning sex, there is higher seroprevalence in male gender and the result was statistically significant although such differences are not seen in IgM antibodies and this might reflect the presence of other cofactor affecting the susceptibility to infection like the chronicity of some *H. pylori* infections and alcohol consumption which was also significantly associated with sero-positivity. The urban, those live in Suleimani city, and those who are singles were more prone to develop sero-positivity. This result is in agreement with a study done in Vietnam [36]. The handworkers and officers were significantly associated with higher prevalence of *H. pylori* infections when compared to students and housewives, this result might reflect higher exposure rate or some other factors responsible for differences in seroprevalence in occupational groups like marital status which when taken in account there will high higher frequency of IgG seroprevalence especially in singles. All other parameters showed no significant differences like personal or family history of peptic ulcer, presence of gastric pain or dyspepsia, cigarette smoking, tea drinking, and chronic use of non-steroidal anti-inflammatory drugs.

The seroprevalence of IgA antibodies specific to *H. pylori* was 58.2% which is the highest percentage among all the three types of antibodies specific to *H. pylori.* The distribution of IgA antibodies among different age groups showed no significant differences, yet variations in prevalence are present with the age group 31-40 years with highest prevalence. For each age group, the sum of prevalence for IgM IgG, and IgA antibodies is highest in this age group (31-40 years), a result might reveal the contribution of all humoral immune response arms in protection against *H. pylori*, a feature which might be due to microbial and host factors. Glassman and his colleagues compared the serological methods by measuring anti-*H. pylori* IgA and G levels are useful serological methods in diagnosing the infection [37].

There no sex difference in IgA seroprevalence, and the same result was seen for residency (urban and rural), presence or absence of chronic diseases, family history of peptic ulcer, alcohol consumption, presence or absence of gastric pain or dyspepsia, and the consumption of non-steroidal anti-inflammatory drugs. Regarding martial status widows have higher IgA seroprevalence than other groups and this might be due to the small number of widows when compared to other groups (singles, married, and divorced). The same explanation can be concluded for higher seroprevalence in non-smokers when compared to smokers.

Those who are living in Suleimani city have more frequent seroprevalence when compared to Chemchamal city which has lower standard of living. This result is contradictory to the results from a study from United Arab Emirate which showed no differences among different standard of living [38]. Ertem and colleagues reported that *H. pylori* and people with low socioeconomic status are more frequently infected [39]. Patel and colleagues could not determine an association between *H. pylori* infection and socioeconomic level of the family [40]. The report from Brazil establishes that the most important factor to prevent *H. pylori* infection is a high education level [41].

Conclusion:

This study revealed that *H. pylori* is a common infection in Sulaimani Governorate/Iraq, and IgA anti-*H. pylori* antibodies is the most common type of antibodies prevalent among population in Sulaimani governorate, and some people have IgA and/or IgM positive but IgG negative results, therefore estimation of *H. pylori* seroprevalence should take in consideration IgM as well as IgA in addition to IgG immunoglobulin, also people that are 20-30 years old have the most seroprevalence of antibodies specific to *H. pylori*.

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