

HLA-DQA1 & HLA-DQB1 Genotyping in *H. pylori* seropositive Individuals

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To study HLA-DQA1 and HLA-DQB1 genotyping in gastritis patients with positive RUT, this study was carried out in College of Medicine, University of Basrah. HLA-DQA1 and HLA-DQB1 genotyping was done in College of Medicine, University of Manitoba, Winnipeg, Canada during the period from 17th of April 2009 to 15th of July 2010. A total of 70 gastritis patients (29 males and 41 females) and 30 controls were included in this study. A significant increased frequency of DQA1*050101 and DQB1*020101 alleles was found in individuals (patients + controls) who showed (+RDT), but the association was weak (odds ratio = 0.39 & 0.33), as compared with individuals (patients + controls) with (- RDT). A significant decreased frequency of DQB1*050201 allele was found in individuals (patients + controls) with (+RDT). The association was very strong (odds ratio = 5.31), as compared with individuals (patients + controls) with (- RDT). A significant decreased frequencies of DQA1*0201 and DQB1*020101 alleles were found in gastritis patients with (+RDT). The association in DQA1*0201 was very strong (odds ratio = 6.38) and for -DQB1*020101 allele, the association was weak (odds ratio = 0.08), as compared with controls with (+ RDT). A significant increased frequency of DQA1*0201 allele was found in controls with (+ RDT) and the association was very strong (odds ratio = 6.18), as compared with controls with (- RDT). A significant increased frequency of DQB1*020101 allele was found in controls with (+RDT) but with weak association (odds ratio = 0.09), as compared with controls with (- RDT).

Keywords: *H. pylori*, seropositive, HLA-DQA1 & HLA-DQB1

INTRODUCTION

Human leukocyte antigens (HLA) are an inherent system of alloantigens, which are the products of genes of the major histocompatibility complex (MHC). These genes span a region of approximately 4 centimorgans on the short arm of human chromosome 6 at band p21.3 and encode the HLA class I and class II antigens, which play a central role in cell-to-cell-interaction in the immune system (Conrad *et al*, 2006). They encode peptides involved in host immune response; also they are important in tissue transplantation and are associated with a variety of infectious, autoimmune, and inflammatory diseases (Gregersen *et al*, 2006; Nair *et al*, 2006). Moreover, the HLA loci display an unprecedented

degree of diversity and the distribution of HLA alleles and haplotypes among different populations is considerably variable (Shao *et al*, 2004; Blomhoff *et al*, 2006). The expression of particular HLA alleles may be associated with the susceptibility or resistance to some diseases (Wang *et al*, 2006). Heterozygosity within the MHC genomic region provides the immune system with a selective advantage of pathogens (Fu *et al*, 2003; Kumar *et al*, 2007). *H. pylori* infection is, in addition to being the main etiologic agent for chronic gastritis, a major cause of peptic ulcer and gastric cancer (Suerbaum and Michetti, 2002). Many studies performed in Iraq about bacteriological and immunological aspects of *H. pylori* (Al- Janabi, 1992; Al-Jalili, 1996; Al-Baldawi, 2001; Al-Dhaher, 2001; Al-Saimary, 2008), but no study was performed yet on HLA genotyping, so results of the present study compared with studies done in other

countries. In developing countries, prevalence of *H. pylori* infection is > 80% among middle-aged adults, whereas in developed countries prevalence ranges from 20%-50%. Approximately 10%-15% of infected individuals will develop peptic disease and 3% a gastric neoplasm (Torres *et al*, 2005). Therefore, *H. pylori* infection is a necessary but not a sufficient cause of severe forms of gastric disease. *H. pylori* induce a host immune response, but the persistence of the infection suggests that the response is not effective in eliminating the infection. Furthermore, multiple lines of evidence suggest that the immune response contributes to the pathogenesis associated with the infection. As a result, the immune response induced by *H. pylori* is a subject of continuous study that has encouraged numerous questions (Azem *et al*, 2006). The inability of the host response to clear infections with *H. pylori* could reflect down-regulatory mechanisms that limit the resulting immune responses to prevent harmful inflammation as a means to protect the host (Yoshikawa and Naito, 2000).

The aims of the present study were to study HLA-DQA1 and HLA-DQB1 genotyping in gastritis patients with positive rut.

METHODS

A total of 70 (70%) showed abnormal endoscopic findings; 29 (41%) males and 41 (58.57%) females, with age groups from (15-66) years, with various gastritis symptoms attending endoscopy unit at Al-Sadder Teaching Hospital in Basrah and a total of 30 controls (18 males and 12 females), with age groups from (15-61) years, without any symptoms of gastritis, were selected randomly. 2ml of venous blood was drawn for serological test (rapid diagnostic test) was collected in plain tube and centrifuged (Janetzki T24, Germany) for 10 minutes (1500 rpm/min), then serum used for rapid diagnostic kit in screening for the presence of antibodies against *H. pylori*. 3ml of the blood was collected in tubes containing EDTA, kept under -18 °C and later used for HLA-DQ genotyping. The study was carried out during the period from (17th of April 2009 to 15th of July 2010).

Rapid Diagnostic test for *H. pylori* (RDT) (Maysiak–Budnik and Megraud, 1994)

A rapid one step test was done for the qualitative detection of IgG antibodies to *H. pylori* in human serum by using Rapid Diagnostic Test Kit, ACON Laboratories, Inc, USA.

DNA Isolated from the Blood Samples by using Wizard Genomic DNA purification Kit, Promega Corporation, USA; Protocol (Beutler *et al*, 1990)

HLA-DQA1 and –DQB1 Genotyping

HLA-DQA1 and –DQB1 genotyping protocol had done according to Sequence-Based-Typing (SBT), which had been developed in National Microbiology Laboratories (NML), Winnipeg, Canada (Luo *et al*, 1999). All the steps of HLA-DQA1 and –DQB1 genotyping were done under supervision of Dr. Ma Luo in Medical Microbiology Laboratory, College of Medicine, Manitoba University in Dr. Ma Luo Laboratory in National Microbiology Laboratories (NML).

Loading and Running DNA in Agarose Gel (Brody and Kern, 2004)

- 2µl loading buffer was mixed with 5ul DNA on paraffin paper, then added to its place in the gel
- 5µl of ladder DNA was added to its place in the gel
- Electrophoresis instrument set on 100 V.
- After 30 minutes, the gel was visualized under U.V. transilluminator, Vilber lourmant, EEC.

PCR Amplification of HLA-DQA1 and –DQB1 Gene

The PCR Amplification of HLA-DQA1 and –DQB1 gene was done in Medical Microbiology laboratories in College of Medicine, Manitoba University, Winnipeg, Canada.

DNA Purification

The Purification of the amplified HLA-DQA1 and –DQB1 gene was done in National Microbiology laboratories (NML), in Dr. Ma Luo Lab, in College of Medicine, Manitoba University, Winnipeg, Canada.

Three methods had been used for purification of the amplified PCR DNA samples:

1. DNA Purification by Using Vacuum

- Amplified PCR DNA samples thawed, then quick spin
- Then transferred into a 96-well Millipore plate (SV 96-well plate)
- Plate placed on vacuum (Vac-Man 96 Vacuum Main Fold) and turn on (the pressure read at approx. 15-25 psi (for 5-10 minutes)
- 100ul Of TE buffer (pH 8.0) added and vacuum again (5-10 minutes)
- Plate removed, blotted lightly on Kim-wipe, vacuum again for 1 minute
- 30ul water added, then placed on shaker for 10 minutes
- Samples transferred into a new 96-well plate, sealed with foil.

2. DNA purification by using: GenElute™ PCR Clean-Up Kit (Sigma- Aldrich, Inc. USA). GenElute™ PCR Clean-Up Kit

3. Purification in DNA Core Section in NML (NML, Canada)

The amplified PCR DNA was purified in DNA Core laboratory in National Microbiology Laboratories (NML) in Winnipeg, Canada.

Sequencing – PCR

Sequencing–PCR was done in National Microbiology laboratories (NML), under supervision of Dr. Ma Luo.

Ethanol Precipitation

Ethanol Precipitation was done under supervision of Dr. Ma Luo in National Microbiology laboratories (NML) in College of Medicine, Manitoba University, Winnipeg, Canada.

- Plates were spin quickly in plate centrifuge (Thermo Electron Corporation-IEC CL 30) following sequencing-PCR
- Plate racks were attached to each plate
- 5ml ethanol and 250µl sodium acetate (Gainland Chemical Co., UK) were added into a reservoir, 21µl of the mixture was added to all columns of 96-well plates by using multichannel electronic micropipette (BIOHIT-e 1200)
- Plate was sealed with silicon foil and vortex (Lincolnshire, IL) and spin quickly
- Plate was placed in –20°C for at least 1 hour then spin at 4000 rpm for at least 1 hour (long spin)

Table 1 HLA-DQA1 genotype frequency of individuals (patients+controls) with positive and negative RDT

HLA-DQA1 allele	Individuals with +ve RDT		Individuals with -ve RDT		χ^2	P	OR	95% CI
	n=61	%	n=26	%				
010101/010102/010401/010402/0105	8	13.11	6	23.08	1.34	NS	1.99	0.61-6.45
010201/010202/010203/010204	16	26.23	10	38.46	1.30	NS	1.76	0.66-4.66
0103	8	13.11	8	30.77	3.78	NS	2.94	0.96-8.99
0201	14	22.95	6	23.08	0.00	NS	1.01	0.34-2.99
030101/0302/0303	16	26.23	4	15.08	1.21	NS	0.51	0.15-1.71
040101/040102/0402/0404	3	4.91	3	11.54	1.24	NS	2.52	0.47-13.42
050101/0503/0505/0506/0507/0508/0509	42	68.85	12	46.15	3.98	<0.05	0.39	0.15-1.00

Table 2 HLA-DQB1 genotype frequency of individuals with positive and negative RDT

HLA-DQB1 allele	Individuals with +ve(RDT)		Individuals with -ve (RDT)		χ^2	P	OR	95% CI
	N=64	%	N=29	%				
020101/0202/0204	37	57.81	9	31.03	5.73	< 0.05	0.33	0.13-0.83
030101/030104/0309/0321/0322/0324/030302	26	40.63	11	37.93	0.06	NS	0.89	0.36-2.20
030201	10	15.63	2	6.90	1.35	NS	0.40	0.08-1.96
030302	1	1.56	0	0.00	1.12	NS	0.98	0.95-1.02
0402	3	4.69	4	13.79	2.38	NS	3.25	0.68-15.60
050101	6	9.38	6	20.69	2.28	NS	2.52	0.74-8.63
050201	5	7.81	9	31.03	8.42	< 0.05	5.31	1.59-17.72
050301	1	1.56	0	0.00	0.46	NS	0.98	0.95-1.02
060101/060103	3	4.69	4	13.79	2.38	NS	3.25	0.68-15.60
060201	4	6.25	1	3.45	0.31	NS	0.54	0.06-5.02
060301/060401	4	6.25	4	13.79	1.44	NS	2.40	0.56-10.36
060401/0634	6	9.38	1	3.45	1.01	NS	0.35	0.04-3.01
060801	1	1.56	0	0.00	0.46	NS	0.98	0.95-1.02
0609	1	1.56	0	0.00	0.46	NS	0.98	0.95-1.02

Sequencing-using the (3100 Genetic Analyzer, USA)

HLA-DQA1 and –DQB1 genotyping protocol had done according to Sequence-Based-Typing (SBT), which had been developed in National Microbiology Laboratories (NML), Winnipeg, Canada (Luo *et al*, 1999).

Statistical Analysis

For qualitative variables, frequency data were summarized as percentage. Statistical significant of differences between two groups was tested by Pearson Chi-square (χ^2) with Yates' continuity correction. Risk was estimated using Odds ratio (OR) and 95% confidence interval (95% CI). P-value was determined by Fisher's exact test, P- value of (< 0.05) was considered statistically significant. Data were analyzed using SPSS program for window (Version 10).

RESULTS**Genotype Frequency of HLA-DQ in Individuals (patients+controls) with Positive and Negative RDT**

Results shown in (Table 1) indicated that HLA-DQA1*050101 was present in 42 out of 61 individuals (patients + controls) with positive RDT and in 12 out of 26 individuals (patients+controls) with negative RDT with frequencies of 68.85 and 46.15 respectively. The increased allele frequency in individuals with positive RDT was statistically significant but the association was weak (χ^2 = 3.98, P<0.05, OR= 0.39, 95% CI= 0.15-1.00) when compared with individuals with negative RDT. Results shown in (Table 2) indicated that HLA-DQB1*020101 was present in 37 out of 64 individuals

Table 3 Homozygosity of HLA-DQ in individuals (patients+controls) with positive and negative RDT

HLA-DQ Homozygosity*		Cases			
		RDT +ve		RDT -ve	
		No	%	No	%
DQA1**	homozygous	15	24.59	3	11.54
	heterozygous	46	75.41	23	88.46
	Total	61	100	26	100
DQB1***	homozygous	17	26.52	5	17.24
	heterozygous	47	73.48	24	82.76
	Total	64	100	29	100
DQA1+DQB1****	homozygous	20	35.71	5	19.23
	heterozygous	36	64.29	21	80.77
	Total	56	100	26	100

*Homozygous at one or both loci

** $\chi^2 = 1.89$, P =NS, OR= 0.40, 95% CI= 0.11-1.52*** $\chi^2 = 0.96$, P =NS, OR= 0.59, 95% CI= 0.19-1.75**** $\chi^2 = 2.28$, P =NS, OR= 0.43, 95% CI= 0.14-0.31

(patients + controls) with positive RDT and in 11 out of 29 individuals (patients + controls) with negative RDT with frequencies of 57.81 and 31.03 respectively. The increased allele frequency in individuals (patients + controls) with positive RDT was statistically significant but with weak association ($\chi^2 = 5.73$, $P < 0.05$, OR= 0.33, 95% CI= 0.13-0.83) when compared with individuals (patients + controls) with negative RDT. Results in (Table 2) showed that HLA-DQB1*050201 was present in 5 out of 64 individuals (patients + controls) with positive RDT and in 9 out of 29 individuals (patients + controls) with negative RDT with frequencies of 7.81 and 31.03. The decreased allele frequency in individuals (patients + controls) with positive RDT was statistically significant with a strong association ($\chi^2 = 8.42$, $P < 0.05$, OR= 5.31, 95% CI= 1.59-17.72) when compared with individuals (patients + controls) with negative RDT.

Homozygosity of HLA-DQ in Individuals (patients + controls) with Positive and Negative RDT

HLA-DQ homozygosity was studied in individuals (patients + controls) with positive and negative RDT. Results shown in (Table 3) indicated that for HLA-DQA1, out of 61 individuals (patients + controls) with positive RDT, 15 were homozygous in one or both loci and out of 26 individuals (patients + controls) with negative RDT, 3 were homozygous in one or both loci, with frequencies of 24.59 and 11.54 respectively. No significant differences were observed in frequency of homozygous HLA-DQA1 genotype between individuals (patients + controls) with positive and negative RDT ($\chi^2 = 1.89$, P =NS, OR= 0.40, 95% CI= 0.11-1.52). For HLA-DQB, out of 64 individuals (patients + controls) with positive RDT, 17 were homozygous in one or both loci, and out of 29 individuals

(patients + controls) with negative RDT, 5 were homozygous in one or both loci, with frequencies of 26.52 and 17.24 respectively. No significant differences were observed in frequency of homozygous HLA-DQB1 genotype between individuals (patients + controls) with positive and negative RDT $\chi^2 = 0.96$, P =NS, OR= 0.59, 95% CI= 0.19-1.75 (Table 3). For HLA-DQ- (A1+B1), out of 56 individuals (patients + controls) with positive RDT, 20 were homozygous in one or both loci, and out of 26 individuals (patients + controls) with negative RDT, 5 were homozygous in one or both loci with frequencies of 64.29 and 80.77 respectively. No significant differences were observed in frequency of homozygous HLA-(DQA1+DQB1) genotype between individuals (patients + controls) with positive and negative RDT $\chi^2 = 2.28$, P =NS, OR= 0.43, 95% CI= 0.14-0.31.

HLA-DQ Allele Frequencies in Gastritis Patients and Controls with Positive RDT

A comparison was done between gastritis patients with (+ve RDT) and controls with (+ve RDT). Results shown in (Table 4) indicated that HLA-DQA1*0201 allele was present in 11 out of 61 gastritis patients with (+ve RDT) and in 4 out of 7 controls with (+ve RDT) with frequencies of 17.74 and 57.14 respectively. The decreased allele frequency in gastritis patients with (+ve RDT) was statistically significant with strong association ($P < 0.05$, OR= 6.38, 95%CI= 1.52-7.48) when compared with controls with (+ve RDT). Results shown in (Table 5) indicated that HLA-DQB1*020101 allele was present in 33 out of 61 gastritis patients with (+ve RDT) and in 6 out of 7 controls with (+ve RDT) with frequencies of 50.76 and 85.71 respectively. The decreased allele frequency in gastritis patients with (+ve RDT) was statistically

Table 4 DQA1 alleles frequencies in gastritis patients & controls with +ve RDT

DQA1 allele	Patients (RDT+ve) (n=61)	Controls (RDT +ve) (n=7)
010101/010102/010401/010402/0105	10 (16.13%)	0 (0.00%)
010201/010202/010203/010204	20 (32.23%)	2 (28.57%)
0103	10 (16.13%)	0 (0.00%)
0201*	11 (17.74%)	4 (57.14%)
030101/0302/0303	16 (25.81%)	1 (14.29%)
040101/040102/0402/0404	4 (6.45%)	0 (0.00%)
050101/0503/0505/0506/0507/0508/0509	38 (61.29%)	6 (85.17%)

*(P < 0.05, OR= 6.38, 95%CI= 1.52-7.48)

Table 5 DQB1 alleles frequencies in gastritis patients & controls with +ve RDT

DQB1 allele	Patients (RDT +ve) (n=61)	Controls (RDT +ve) (n=7)
020101/0202/0204*	33 (50.76%)	6 (85.71%)
030101/030104/0309/0321/0322/0324/030302	25 (38.46%)	2 (28.57%)
030201	10 (15.38%)	0
030302	3 (4.61%)	0
0402	5 (7.69%)	0
050101	8 (12.31%)	0
050201	10 (15.38%)	1 (14.29%)
050301	1 (1.53%)	0
060101/060103	3 (4.61%)	0
060201	3 (4.61%)	1 (14.29%)
060301/061401	6 (9.23%)	0
060401/0634	6 (9.23%)	0
060801	1 (1.53%)	0
0609	1 (1.53%)	0

*(P < 0.05, OR= 0.08, 95%CI=0.01=0.83)

significant (P < 0.05, OR= 0.08, 95%CI=0.01=0.83) when compared with controls with (+ve RDT).

HLA-DQ Alleles Frequencies in Controls with +ve & -ve RDT

A comparison was done between controls with positive RDT and controls with negative RDT. Results shown in (Table 6) indicated that HLA-DQA1*0201 allele was present in 4 out of 7 controls with (+ ve RDT) and in 5 out of 18 controls with (- ve RDT) with frequencies of 57.14 and 27.78 respectively. The increased allele frequency in controls with (+ ve RDT) was statistically significant (P < 0.05, OR= 6.18, 95%CI= 1.21-6.18) when compared with controls with negative RDT. Results shown in (Table 7) indicated that HLA-DQB1*020101 allele was present in 4 out of 7 controls with (+ ve RDT) and in 7 out of 21 controls with (-ve RDT) with frequencies of 85.71 and

33.33 respectively. The increased allele frequency in controls with (+ ve RDT) was statistically significant (P < 0.05, OR= 0.09, 95%CI=0.01=0.63) when compared with controls with (- ve RDT).

DISCUSSION

Serological tests used for diagnosis of *H. pylori*, are fast and non invasive, also these tests are capable of reducing the endoscope workload. In the present study a rapid one step test was done for the qualitative detection of IgG antibodies to *H. pylori* in human serum by using (Rapid Diagnostic Test) Kit. Results of rapid diagnostic test showed that, out of 70 gastritis patients, 61 (87.14%) were positive rapid diagnostic test (RDT) and 9 (12.86%) were negative RDT, that was compatible with a study performed by Maysiak–Budnik and Megraud, (1994). The test has been tested for interference from visibly

Table 6 DQA1 alleles frequencies in controls with +ve & -ve RDT

DQA1 allele	RDT +ve Controls (n=7)	RDT -ve Controls (n=18)
010101/010102/010401/010402/0105	0 (0.00%)	4 (22.22%)
010201/010202/010203/010204	2 (28.57%)	4 (22.22%)
0103	0 (0.00%)	6 (33.33%)
0201*	4 (57.14%)	5 (27.78%)
030101/0302/0303	1 (14.29%)	3 (16.67%)
040101/040102/0402/0404	0 (0.00%)	2 (11.11%)
050101/0503/0505/0506/0507/0508/0509	6 (85.17%)	10 (55.56%)

*(P= 0.03, OR= 6.18, 95%CI= 1.21-6.18)

Table 7. DQB1 alleles frequencies in controls with +ve & -ve RDT

DQB1 allele	RDT +ve Controls (n=7)	RDT -ve Controls (n=21)
020101/0202/0204*	6 (85.71%)	7 (33.33%)
030101/030104/0309/0321/0322/0324/030302	2 (28.57%)	10 (47.62%)
030201	0	2 (9.52%)
030302	0	2 (9.52%)
0402	0	2 (9.52%)
050101	0	4 (19.05%)
050201	1 (14.29%)	3 (14.29%)
050301	0	0 (0.00%)
060101/060103	0	4 (19.05%)
060201	1 (14.29%)	1 (4.76%)
060301/061401	0	2 (9.52%)
060401/0634	0	1 (4.76%)
060801	0	0
0609	0	0

*(P < 0.05, OR= 0.09, 95%CI=0.01=0.63)

hemolysed as well as specimens containing high bilirubin levels. In addition, no interference was observed in specimens containing hemoglobin up to 1.000 ug/ dL, bilirubin, and up to 2.000 um/ dL human serum albumin (Marshall *et al*, 1985; Faigel. *et al*, 2000). This evidence indicated that test is sensitive, fast and convenient and should be used in screening for anti *H. pylori* antibodies. Many factors such as socioeconomic status, ethnic group, different populations, geographical location and the type of the trouble associated with the infection also contribute to the observed variations in prevalence in the present study. Asymptomatic and untreated patients continue to test IgG seropositive as long as the *H. pylori* organisms are presents, even after histological resolution. These results showed that humoral immunity was present throughout the spectrum, but does not seem to provide protection in patients, Although controls showed (+RDT), but they have no symptoms of gastritis, this might be explained as in such individuals, the genetic factors may control host immune responses to the infectious agents and provide protection (Mattsson *et al*,

1998). Genotype frequency of HLA-DQ was studied in individuals (patients + Controls) with positive and negative RDT. Results shown in (Table 1) indicated significant increased frequency of HLA-DQA1*050101 allele in individuals with positive RDT but the association was weak ($\chi^2= 3.98$, $P<0.05$, OR= 0.39, 95% CI= 0.15-1.00) when compared with individuals with negative RDT. Results shown in Table 4.29 indicated significant increased frequency of HLA-DQB1*020101 allele in individuals (patients + controls) with positive RDT, but with weak association ($\chi^2= 5.73$, $P<0.05$, OR= 0.33, 95% CI= 0.13-0.83) when compared with individuals (patients + controls) with negative RDT. Results in Table 4.29 showed significant decreased frequency of HLA-DQB1*050201 allele in individuals (patients + controls) with positive RDT with a strong association ($\chi^2= 8.42$, $P<0.05$, OR= 5.31, 95% CI= 1.59-17.72) when compared with individuals (patients + controls) with negative RDT. HLA-DQ homozygosity was studied in individuals (patients + controls) with positive and negative RDT. Results shown in Table 4.30 indicated that there was no

significant differences were observed in frequency of homozygous HLA-DQ genotype between individuals (patients + controls) with positive and negative RDT. A comparison was done between gastritis patients with (+ve RDT) and controls with (+ve RDT). Results shown in Table 4.31 indicted a significant decreased frequency of HLA-DQA1*0201 allele in gastritis patients with (+ve RDT) with strong association ($P < 0.05$, OR= 6.38, 95%CI= 1.52-7.48) when compared with controls with (+ve RDT). Results shown in Table 4.32 indicated significant decreased frequency of HLA-DQB1*020101 allele in gastritis patients with (+ve RDT) ($P < 0.05$, OR= 0.08, 95%CI=0.01=0.83) when compared with controls with (+ve RDT). Wu *et al*, 2002 reported lower seropositivity of *H. pylori* and a higher ratio of diffuse/intestinal type carcinoma in Taiwanese patients carrying the HLA-DQB1*0301 allele. A comparison was done between controls with positive RDT and controls with negative RDT. Results shown in Table 4.33 indicated significant increased frequency of HLA-DQA1*0201 allele in controls with (+ve RDT) ($P < 0.05$, OR= 6.18, 95%CI= 1.21-6.18) when compared with controls with negative RDT. Results shown in Table 4.34 indicated a significant increased frequency of HLA-DQB1*020101 allele in controls with (+ve RDT) ($P < 0.05$, OR= 0.09, 95%CI=0.01=0.63) when compared with controls with (-ve RDT). These results were compatible with a study performed by Magnuson *et al*, (2001) who reported that HLA-DQA1*0102 was inversely associated with *H. pylori*-seropositivity. Current evidence indicates that the majority of individuals harboring *H. pylori* infection remain asymptomatic during their lifetime, with no clinical consequence from their infection. In a community-based seroepidemiologic study in Mexico (Torres *et al*, 1998) who reported that seropositivity for *H. pylori* infection was 66%, and > 80% of adults were infected by age 25 years; seroprevalence remained nearly unchanged after the third decade of life, with an increment in seropositivity of < 0.5% per year in persons between 30 and 69 years. Taken together, these data suggest that risk for gastric diseases depends on factors other than *H. pylori* infection and age.

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