The Open Biomarkers Journal

Content list available at: https://openbiomarkerjournal.com

RESEARCH ARTICLE

Association between Human Leukocyte Antigen-DQ Polymorphisms and Treatment Response in Chronic Hepatitis B Egyptian Population: A Prospective Study

Amal A. Mohamed[1], Sherief Abd-Elsalam[2,*], Mariam Zaghloul[3], Mohamed Attala[4], Rania A. Khattab[5], Amir Khater[4], Dalia A. El-damasy[6], Eman El-Sayed[7], Soha Hassanin[8], Nehad Hawash[2] and Mohmoud R. Mohamed[9]

[1]*Department of Biochemistry, National Hepatology & Tropical Medicine Research Institute, Cairo, Egypt*
[2]*Department of Tropical Medicine, Tanta University, Tanta, Egypt*
[3]*Hepatology, Gastroenterology and Infectious diseases, Kafr-Elsheikh University, Kafr el-Sheikh, Egypt*
[4]*Department of Tropical , National Hepatology & Tropical Medicine Research Institute, Egypt*
[5]*Department of Microbiology and Immunology, Faculty of Pharmacy, Cairo University, Kasr Al-Aini 11562, Cairo, Egypt*
[6]*Department of Microbiology, Faculty of Pharmacy, Egyptian Russian University, Egypt*
[7]*Department of Clinical Pathology, Faculty of Medicine, Minia University, Minia, Egypt*
[8]*Department of Biochemistry, Modern University for Technology and Information, Egypt*
[9]*Internal Medicine, Faculty of Medicine, Minia University, Minia, Egypt*

Abstract:

Background & Aims:

Several studies, in different populations, have focused on the role of HLA-DQ gene polymorphism in the pathogenesis of HBV infection. However, these findings are still controversial. This study aimed to determine HLA-DQ polymorphism in Chronic HBV patients and its impact on the response to antiviral therapy.

Methods:

This study was carried out on a total number of 188 participants, they were subdivided as follows: Group I (patients' group): included 97 patients with chronic hepatitis B viral infection that was further subdivided according to response to treatment into responder and non-responder subgroups, Group II (Control group): included 91 normal healthy subjects who were matched to the patient group by sex and age. PCR (Polymerase Chain Reaction) testing, for HBV-DNA, was done for all participants enrolled in the study to measure the viral virus load before and after treatment. HLA- DQ polymorphism allelic discrimination assay was assayed using the Real-time equipment.

Results:

In a general analysis for the SNP rs7453920, the overall genotypes frequencies were 37% for A/A, 60.6% for A/G, and 37% for G/G. The G alleles of HLA-DQ rs7453920 were significantly increased in chronic HBV infection patients. A total of 77 (79.4%) patients were responders. Among this group, 72.7% were male, and the average age was 38.59 ±9.15 years. On evaluation of the association between polymorphisms in HLA-DQ gene and treatment response, the results indicated that response to treatment declined when patients were carrying the more unfavorable rs 7453920 GG with a response rate of 64%. Patients carrying the mutant allele AG, or the wild type allele AA were more likely to achieve a higher rate of response (84.8% and 83.3%, respectively).

Conclusion:

The presence of HLA-DQB2 rs 7453920-G serves as a risk factor for chronic HBV infection and treatment failure in the Egyptian population.

Keywords: Hepatitis B virus, HLA-DQ, Antiviral therapy, Polymorphism, Response, PCR.

Article History	Received: March 30, 2020	Revised: May 03, 2020	Accepted: May 12, 2020

1. 1INTRODUCTION

Hepatitis B Virus (HBV) is one of the most common causes of liver diseases. It is estimated by the World Health Organization to infect 257 million people worldwide [1]. As a consequence, it results in an estimated 600,000 deaths every year through complications of end-stage liver disease and Hepatocellular Carcinoma (HCC) [2]. Clinical outcomes, after Hepatitis B virus (HBV) exposure, vary extremely from

spontaneous clearance to chronic hepatitis B and often progresses to Liver Cirrhosis (LC) and Hepatocellular Carcinoma (HCC) [3]. However, factors determining the chronicity of primary HBV infection and its disease progression to LC and HCC are not clearly understood. It has been shown that the outcomes of the HBV infection are not because of the variation in the virulence of viral strains [4], but rather due to host-immune response [5]. Human Leukocyte Antigen (HLA)-DQ genetic polymorphisms have been associated with chronic Hepatitis B Virus (HBV) outcomes. Several studies on different populations have focused on the role of HLA-DQ gene polymorphism in the pathogenesis of chronic HBV infection. However, these findings are still controversial [6 - 10]. This study aimed to determine the impact of HLA-DQ polymorphism on chronic HBV patients and their response to antiviral therapy.

2. MATERIALS AND METHODS

2.1. Patients

This study was carried out on a total number of 188 participants, they were subdivided as follows: Group I (patients' group): included 97 patients with chronic hepatitis B viral infection that was further subdivided according to response to treatment into responder and non-responder subgroups, Group II (Control group): included 91 normal healthy subjects who were matched to the patient group by sex and age. The patients were admitted to Tanta and Kafr-el Sheikh University hospital, Egypt, from April 2016 to January 2018. All patients with HBV infection, selected for this study, were further confirmed by being HBsAg (hepatitis B surface antigen) positive, HbcAb (hepatitis B virus core antibody) positive and HBeAg (hepatitis B e antigen) or HBeAb (hepatitis B e antibody) positive for at least 6 months. Patients were excluded from the study if they were known to have a disease other than HBV, such as HCV co-infection, renal insufficiency, proteinuria, suspected infections, clinically overt diabetes mellitus, thyroid dysfunction, other endocrine disorder or history of alcohol abuse.

Informed consent was obtained from all participants before the study. This study was approved by the Tanta University Faculty of Medicine research ethical committee. All patients were naïve and received lamivudine treatment.

2.2. Blood Sampling and Biochemical Assays

Fasting venous blood samples (10 ml) were collected by trained laboratory technicians. A portion of blood was allowed to clot and then centrifuged at 3500g for 5 min to separate the serum used for the assessment of Aspartate Aminotransferase (AST), Alanine Aminotransferase (ALT), total bilirubin and Albumin by Beckman Automatic Chemistry Analyzer. Another portion of blood was collected in vacutainer tubes containing EDTA for Complete Blood picture (CBC), which was done by phoenix machine.

AFP was measured using Abbott, Axyam (USA, Supplied by Al-Kamal company Cairo, Egypt).

* Address correspondence to this author at the Departmentof Tropical Medicine, Tanta University, Tanta, Egypt; Tel: 201063319696;
E-mail: sherif.abdelbaky@med.tanta.edu.eg

2.3. Serological Analysis

The serological HCV, HBsAg, HBsAb, HbcAb, HBeAg, and HBeAb were detected by using Abbott, Axyam (USA, Supplied by Al-Kamal company Cairo, Egypt).

2.4. PCR Testing

HBV-DNA was extracted from serum using a kit (Cobas TaqMan HBV Test Rouche Diagnostics, Manheim, Germany) and was used as a confirmatory approach for HBV positivity for all subjects under the present study.

Blood aliquots were stored at -80^{o}C until assayed and thawed immediately before the gene polymorphism measurement of HLA DQ.

2.5. Genotyping Assay

HLA DQB2 polymorphism or Single Nucleotide Polymorphism (SNP) was assayed using the ABI Taq Man allelic discrimination assay. Genomic DNA was extracted from peripheral blood samples by protease K digestion, phenol-chloroform extraction, ethanol precipitation, and DNA concentration was tested on Nano Drop 2000 spectrophotometer (Thermo Scientific, DE). According to the relevant HLA Class II SNP literature, HLA-DQ rs 7453920 SNP was selected as candidate site, as it is an HLA-DQ which shows minor allele frequency AA, AG, and GG. The SNP was genotyped using the Taq-Man allele identification assay on the ABI PRISM 7900HT system (Applied Biosystem, Foster City, CA, USA).

2.6. Statistical Analysis

All data analysis was operated with SPSS (V.20 for Windows). Genotype and allele frequencies were determined by direct counting and Hardy-Weinberg equilibrium was evaluated by the chi-square test. Comparisons between individual demographic characteristics were analyzed as appropriate with either a Student's t-test (for continuous variables) or a $\chi 2$ test or Fisher's Exact tests (for categorical variables) with a two-tailed p-value. Each SNP was analyzed using co-dominant, dominant, and recessive genetic models. The co-dominant model considers homozygous type versus wild type and hybrid type versus wild type, respectively. The dominant model considers the homozygous type and heterozygous type together versus the wild type, and the recessive model considers the homozygous type versus the heterozygous type and the wild type together.

3. RESULTS

Socio-demographic data of chronic HBV infection group and controls were obtained. The average age of the 97 HBV patients was 38.9 ± 9.04 years and there were a total of 70 (72.2%) males. The control group was matched for age (mean age 37.54±9.79) and sex [56(44.4%) were males].

In a general analysis for the SNP rs7453920, the overall genotypes frequencies were 37% for A/A, 60.6% for A/G, and 37% for G/G (Table **1**).

The frequencies of the genotypes and alleles were in agreement with Hardy-Weinberg equilibrium (X^2 = 0.77 and P = 0.69) for HLA-DQB2 (rs7453920) evaluated in the controls but the cases violated the role (X^2 = 16.95 and P = <0.001).

Table 1. Bivariate analysis of the distribution of genotypes and alleles in HLADQB2 (rs7453920) SNPs in the case and control groups.

-	Group I (Chronic HBV) (N = 97)	Group II (Control) (N = 91)	OR (95%CI)	P
Genotypes	HLA-DQB2 (rs7453920)		-	-
AA	6(6.2%)	31(34.1%)	1	-
AG	66(68%)	48(52.7%)	7.1 (2.75–18.37)	<0.001
GG	25(25.8%)	12(13.2)	10.76 (3.5–32.7)	<0.001
Alleles G: A	116: 78	72:110	2.27(1.5-3.43)	<0.001

Table 2. Characteristics of patients with chronic hepatitis B related in response to NA treatment.

Variables	N-responder (n=20)	Responder (n=77)	P values	OR (CI 95%)
Mean age, year	40.05(8.69)	38.59(9.15)	0.69	1.02(0.96-1.07)
Male (%)	14(70)	56(72.7)	0.8	0.87(0.29-2.57)
Baseline HBV DNA (IU)	$2.9x10^5 \pm 8.92x10^5$	$7.1x10^5 \pm 3.53x10^5$	0.64	1(1-1)
BMI	29.8±3.7	30.25±4.8	0.38	0.97(0.87-1.09)
Coffee drink(yes/no)	12/8	45/32	0.9	0.93(0.34-2.56)
Smoking (yes/no)	9/11	36/41	0.89	1.07(0.4-2.8)
Alcohol consumption(yes/no)	3/17	8/69	0.56	0.65(0.16-2.74)
Residence (Rural/Urban)	11/9	50/27	0.41	0.66(0.24-1.79)
Hb(g/dl)	11.8±1.67	11.81±1.68	0.96	0.99(0.74-1.34)
ALT(IU)	71.6±27.75	81.54±29.73	0.17	0.98(0.96-1)
kAST(IU)	65.15±32.19	66.85±31.29	0.82	0.99(0.98-1.01)
Total Bilirubin	1.07±0.43	1.1±0.38	0.73	0.79(0.22-2.9)
INR	1.2±0.34	10.14±0.27	0.1	3.5(0.72-17.5)
ALB (g/L)	3.7±0.58	3.6±0.6	0.73	1.15(0.5-2.6)
S. Creatinine(mg/dl)	1.1±0.42	1±0.34	0.28	2(0.56-7.23)
Glucose level(mg/dl)	94.45±12.37	97.4±11.93	0.32	0.97(0.93-1.02)
AFP (ng/mL)	10.1±5.16	11.18±5.23	0.41	0.95(0.85-1.06)
U/S(Normal/Bright/Coarse)	0/13/7	8/34/35	0.21	1.9(0.68-5.3)

The G alleles of HLA-DQ rs7453920 were significantly increased in chronic HBV infection patients (AG + GG versus AA: OR = 7.84, 95% CI: (3.08–19.92), $P = < 0.001$; GG versus AG + AA: OR = 2.29, 95% CI: (1.07–4.88), $P = 0.03$; GG versus AA: OR = 10.76, 95% CI: 3.5–32.7, $P < 0.001$; AG versus AA: OR = 7.1, 95% CI: 2.75–18.37, $P < 0.001$; G versus A: OR = 2.27, 95% CI: 1.5-3.43, $P < 0.001$), as compared to controls.

The participating patients were further subdivided into two subgroups according to the response to treatment. The baseline demographic and laboratory characteristics of the 97 enrolled patients are shown in Table **2**.

A total of 77 (79.4%) patients were responders. Among this group, 72.7% were male, and the average age was 38.59 ±9.15 years.

There was no difference in gender and age between the responder group and non-responder group (p>0.05). Moreover, smoking, alcohol consumption, BMI, drinking coffee, or residency showed no significant difference between the two groups (p>0.05). In addition, the baseline levels of hemoglobin, ALT, AST, total bilirubin, INR, serum albumin, serum creatinine, blood glucose, and alpha-fetoprotein were similar in the two groups (p>0.05) (Table **2**).

Association between polymorphisms in *HLA-DQ* gene and treatment response.

SNP was in Hardy-Weinberg equilibrium in allele frequency in the non-responder group. When the association between polymorphisms in HLA-DQ gene and treatment response was evaluated, the results indicated that response to treatment declined when patients were carrying more unfavorable rs 7453920 GG with a response rate of 64%. Patients carrying the mutant allele AG, or the wild type allele AA were more likely to achieve a higher rate of response (84.8% and 83.3%, respectively) (Table **3**).

The G alleles of HLA-DQ rs7453920 were significantly increased in non-responders chronic HBV patients (AG+GG *vs* AA: OR 1.31, 95% CI 0.15 to 11.98, P=0.64; GG *vs* AG+AA: OR 3.12, 95% CI 1.11 to 8.82, P=**0.03***; GG versus AA: OR = 2.8, 95% CI: 0.28–27.97, $P = 0.63$; AG versus AA: OR = 0.89, 95% CI: 0.09–8.47, $P = 0.65$; G versus A: OR = 2.38, 95% CI:

1.15-1.15, $P = 0.01$)) as compared to responders.

Table 3. Association of single nucleotide polymorphisms in human leucocyte antigen-DQ with hepatitis B virus treatment response.

Genotype	N-responder (n=20)	Responder (n=77)	Response rate (%)	OR (95% CI)	P values
rs 7453920	-				
AA	1(5%)	5(6.5%)	83.3%	1	----
AG	10(50%)	56(72.7%)	84.8%	0.89(0.09-8.47)	0.65
GG	9(45%)	16(20.8%)	64%	2.8(0.28-27.97)	0.63
Alleles G *vs* A	38:12	88:66		2.38(1.15-4.9)	0.01*

4. DISCUSSION

The outcome of HBV infection is determined by host factors, including immunological and viral factors, as well as viral load and viral genotypes [11 - 15].

Variations in HLA class II strongly correlated with persistence or spontaneous clearance of HBV infection [16, 17]. In addition, these variations may influence treatment efficacy and response to vaccination [18]. The host genetic background influences the immune processing of APC cells, which is necessary for an effective immune response. SNP rs7453920 affects the gene expression of the HLA-DQB2 molecule and it was found to be pivotal in the recognition and processing of viral particles [19]. However, these findings are still controversial.

This study was conducted to investigate the association between HBV chronic infection and HLA-DQB2 SNP (rs7453920) and clarify its effect on the treatment response. It was found that the G alleles of HLA-DQ rs7453920 significantly increased in chronic HBV infection patients as compared to controls. This was in accordance with Tao Xu *et al.* [11] who found that HLA-DQ rs7453920-A polymorphism was associated with decreased risk of HBV infection. All patients were treatment-naive and all received Lamivudine as treatment, then they were divided into two subgroups according to their response to treatment. No significant difference was found in basic characteristics including (age, gender, smoking, alcohol consumption, BMI, drinking coffee, PCR of HBV DNA) between two groups although these factors were proved to influence the risk of HBV infection persistence in previous studies [20, 21].

Our study showed that patients carrying the more favorable rs 7453920 genotypes AG and AA had a significantly higher rate of response to treatment. Several previous clinical trials attempted to clarify the effect of the HLA gene variant on HBV treatment response. Chang *et al*., who studied the effect of HLA variants and response to interferon therapy in male Han Taiwanese patients with CHB, concluded that "G-C" haplotype of the five SNPs, including rs9277535 (HLADPB1), rs9276370 (HLA-DQA2), rs7756516 and rs7453920 (HLA-DQB2), and rs9366816 near HLA-DPA3, is associated with sustained therapeutic response to IFN-α treatment [22]. In addition, results of a meta-analysis performed by Xu *et al*. concluded that the HLA-DQ rs7453920-A allele was a protecting factor against chronic HBV infection, while rs 7453920-G serves as a risk factor for HBV infection [23].

On the other hand, Zhang *et al*. studied a different HLA-DQ locus and concluded that rs9275572 can be considered as a predictor of response to Lamivudine therapy [24].

To the best of our knowledge, this the first study to investigate HLA-DQB2 rs 7453920 polymorphism in chronic HBV Egyptian patients, but this study has its own limitation due to the small sample size. Therefore, further studies on a larger cohort are needed to validate these results.

CONCLUSION

The presence of HLA-DQB2 rs 7453920-G serves as a risk factor for chronic HBV infection and treatment failure in the Egyptian population.

LIST OF ABBREVIATIONS

AFP	=	Alfa fetoprotein
ALB	=	Albumin
ALT	=	Alanine Aminotransferase.
AST	=	Aminotransferase
BMI	=	Body Mass Index
CBC	=	Complete Blood Picture
FDR	=	False discovery rate.
HBV	=	Hepatitis B Virus.
HBsAg	=	Hepatitis B Surface Antigen
HbcAb	=	Hepatitis B Virus Core Antibody
HBeAg	=	Hepatitis B e Antigen
HBeAb	=	Hepatitis B e Antibody
HCC	=	Hepatocellular Carcinoma
(HLA)-DQ	=	Human Leukocyte Antigen
LC	=	Liver Cirrhosis
PCR	=	Polymerase Chain Reaction
SNP	=	Single Nucleotide Polymorphism
SVR	=	Sustained Virological Response
T. BIL	=	Total Bilirubin

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

The research was approved by the Tanta University Faculty of Medicine research ethical committee, 31412/03/16 Egypt .

HUMAN AND ANIMAL RIGHTS

No animals were used in this research. All human research procedures were followed in accordance with the ethical standards of the committee responsible for human experimentation (institutional and national), and with the Helsinki Declaration of 1975, as revised in 2013.

CONSENT FOR PUBLICATION

Written informed consent was obtained from all the participants.

AVAILABILITY OF DATA AND MATERIALS

The authors' institution does not allow public data access.

FUNDING

None.

CONFLICT OF INTEREST

The author declares no conflict of interest, financial or otherwise.

ACKNOWLEDGEMENTS

Declared none.

REFERENCES

[1] Todo S, Demetris AJ, Van Thiel D, Teperman L, Fung JJ, Starzl TE. Orthotopic liver transplantation for patients with hepatitis B virus-related liver disease. Hepatology 1991; 13(4): 619-26. [PMID: 2010156]

[2] Lerut JP, Donataccio M, Ciccarelli O, *et al.* Liver transplantation and HBsAg-positive postnecrotic cirrhosis: adequate immunoprophylaxis and delta virus co-infection as the significant determinants of long-term prognosis. J Hepatol 1999; 30(4): 706-14. [http://dx.doi.org/10.1016/S0168-8278(99)80203-7] [PMID: 10207814]

[3] Lavanchy D. Hepatitis B virus epidemiology, disease burden, treatment, and current and emerging prevention and control measures. J Viral Hepat 2004; 11(2): 97-107. [http://dx.doi.org/10.1046/j.1365-2893.2003.00487.x] [PMID: 14996343]

[4] Cacciola I, Cerenzia G, Pollicino T, *et al.* Genomic heterogeneity of hepatitis B virus (HBV) and outcome of perinatal HBV infection. J Hepatol 2002; 36(3): 426-32. [http://dx.doi.org/10.1016/S0168-8278(01)00295-1] [PMID: 11867188]

[5] Chisari FV, Ferrari C. Hepatitis B virus immunopathogenesis. Annu Rev Immunol 1995; 13: 29-60. [http://dx.doi.org/10.1146/annurev.iy.13.040195.000333] [PMID: 7612225]

[6] Lu Y, Wu Z, Peng Q, *et al.* Role of IL-4 gene polymorphisms in HBV-related hepatocellular carcinoma in a Chinese population. PLoS One 2014; 9(10): e110061. [http://dx.doi.org/10.1371/journal.pone.0110061] [PMID: 25295591]

[7] Azar SS, Mansoori M, Attar M, Shahbazi M. Tumor necrosis factor alpha 308 g/a single nucleotide polymorphism and susceptibility to hepatocellular carcinoma *Via* hepatitis B italicnfection. Asian Pac J Cancer Prev 2016; 17(7): 3381-4. [PMID: 27509979]

[8] Huang X, Li H, Wang J, *et al.* Genetic polymorphisms in Toll-like receptor 3 gene are associated with the risk of hepatitis B virus-related liver diseases in a Chinese population. Gene 2015; 569(2): 218-24. [http://dx.doi.org/10.1016/j.gene.2015.05.054] [PMID: 26024592]

[9] Ratnasari N, Nurdjanah S, Sadewa AH, Hakimi M. The role of vascular endothelial growth factor -634 G/C and its soluble receptor on chronic liver disease and hepatocellular carcinoma. Arab J Gastroenterol 2016; 17(2): 61-6. [http://dx.doi.org/10.1016/j.ajg.2016.06.005] [PMID: 27426958]

[10] Huang R, Hao Y, Fan Y, *et al.* Association between cytotoxic T-lymphocyte-associated antigen 4 +49A/G polymorphism and persistent hepatitis B virus infection in the Asian population: Evidence from the current studies. Genet Test Mol Biomarkers 2013; 17(8): 601-6. [http://dx.doi.org/10.1089/gtmb.2013.0069] [PMID: 23725008]

[11] Pan CQ, Zhang JX. Natural history and clinical consequences of hepatitis B virus infection. Int J Med Sci 2005; 2(1): 36-40. [http://dx.doi.org/10.7150/ijms.2.36] [PMID: 15968338]

[12] Kacprzak-Bergman I, Nowakowska B. Influence of genetic factors on the susceptibility to HBV infection, its clinical pictures, and responsiveness to HBV vaccination. Arch Immunol Ther Exp (Warsz) 2005; 53(2): 139-42. [PMID: 15928582]

[13] Tawfik AK, Amin AM, Yousef M, *et al.* IL-1α correlates with severity of hepatitis C virus-related liver diseases. J Inflamm Res 2018; 11: 289-95. [http://dx.doi.org/10.2147/JIR.S166564] [PMID: 30022847]

[14] Abd-Elsalam S, Habba E, Elkhalawany W, *et al.* Correlation of platelets count with endoscopic findings in a cohort of Egyptian patients with liver cirrhosis. Medicine (Baltimore) 2016; 95(23): e3853. [http://dx.doi.org/10.1097/MD.0000000000003853] [PMID: 27281094]

[15] Abd-Elsalam S, Kobtan A, El-Kalla F, *et al.* A 2-week Nitazoxanide-based quadruple treatment as a rescue therapy for *Helicobacter pylori* eradication: A single center experience. Medicine (Baltimore) 2016; 95(24): e3879. [http://dx.doi.org/10.1097/MD.0000000000003879] [PMID: 27310977]

[16] Hu L, Zhai X, Liu J, *et al.* Genetic variants in human leukocyte antigen/DP-DQ influence both hepatitis B virus clearance and hepatocellular carcinoma development. Hepatology 2012; 55(5): 1426-31. [http://dx.doi.org/10.1002/hep.24799] [PMID: 22105689]

[17] Seto WK, Wong DKH, Kopaniszen M, *et al.* HLA-DP and IL28B polymorphisms: Influence of host genome on hepatitis B surface antigen seroclearance in chronic hepatitis B. Clin Infect Dis 2013; 56(12): 1695-703. [http://dx.doi.org/10.1093/cid/cit121] [PMID: 23449268]

[18] Singh R, Kaul R, Kaul A, Khan K. A comparative review of HLA associations with hepatitis B and C viral infections across global populations. World J Gastroenterol 2007; 13(12): 1770-87. [http://dx.doi.org/10.3748/wjg.v13.i12.1770] [PMID: 17465466]

[19] Mohamadkhani A, Katoonizadeh A, Poustchi H. Immune-Regulatory Events in the Clearance of HBsAg in Chronic Hepatitis B: Focuses on HLA-DP. Middle East J Dig Dis 2015; 7(1): 5-13. [PMID: 25628847]

[20] Wang FS. Current status and prospects of studies on human genetic alleles associated with hepatitis B virus infection. World J Gastroenterol 2003; 9(4): 641-4. [http://dx.doi.org/10.3748/wjg.v9.i4.641] [PMID: 12679901]

[21] Alatrakchi N, Koziel MJ. Antiviral T-cell responses and therapy in chronic hepatitis B. J Hepatol 2003; 39(4): 631-4. [http://dx.doi.org/10.1016/S0168-8278(03)00398-2] [PMID: 12971976]

[22] Chang SW, Fann CSJ, Su WH, *et al.* A genome-wide association study on chronic HBV infection and its clinical progression in male Han-Taiwanese. PLoS One 2014; 9(6): e99724. [http://dx.doi.org/10.1371/journal.pone.0099724] [PMID: 24940741]

[23] Xu T, Sun M, Wang H. Relationship between HLA-DQ Gene Polymorphism and Hepatitis B Virus Infection. BioMed Res Int 2017; 2017: 9679843. [http://dx.doi.org/10.1155/2017/9679843] [PMID: 28512640]

[24] Zhang X, Jia J, Dong J, *et al.* HLA-DQ polymorphisms with HBV infection: different outcomes upon infection and prognosis to lamivudine therapy. J Viral Hepat 2014; 21(7): 491-8. [http://dx.doi.org/10.1111/jvh.12159] [PMID: 24750255]