Association of Hepatitis B and Hepatitis C Virus (HCV) Infection with Human Leukocyte Antigens (HLA)

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Abstract: Back ground: Hepatitis B (HBV) and hepatitis C (HCV) viral infection or co-infection leads to risk of development of chronic infection, cirrhosis and hepatocellular carcinoma (HCC). Immigration and globalization have added to the challenges of public health concerns regarding chronic HBV and HCV infections worldwide. Different human leukocycle antigen (HLA) types related to chronicity has been reported from different countries. **Objectives:** The objective of this study is to investigate the association between the frequencies of HLA Class I and chronic HCV and HBV infections. Also these will revealed the most susceptible and protective alleles. **Subjects and methods:** Thirty patients with chronic HCV infection as well as 30 patients with HBV, also 40 apparently healthy individuals as controls were included in this study.HCV and HBV infections were diagnosed by detecting the anti-HCV antibody by enzyme-linked immunosorbant assay (ELISA) and Hbs antigen. HLA-A and HLA-B typing by complement-dependent micro-lympho-cytotoxicity assay was performed for both groups among chronic hepatitis patients. **Results:** HLA-A2 (OR 5.6, CI=2.03-15.87, P=0001), A9andA19, B12, B13, B35 and B40 alleles were most prevalent among patients versus control group they were considered as susceptible alleles. On the other hand HLA-29, A32, and B8 were highly significant among controls, they were considered as protective alleles. Among patients with hepatitis (B), A2 (OR=11.0, CI =3.17-40.3, P=0001), A9, A24, B5, B13 and Bw35 alleles were highly significant among patients versus controls. On the other hand HLA-A29, A32, and B8 alleles were significantly higher among controls, they considered as protective alleles. On the other hand, A2 (OR= 3.06, CI =1.0-10.1, P=0.01), A3, A9, A19, B12, B13, B14 and B40 alleles were significantly prevalent among hepatitis C patients. While HLA- A32, B5 alleles were higher among controls. In comparison between different detected alleles among patients with hepatitis B and C, the results revealed that, HLA- A2 allele was more significant among hepatitis (B) patients. On the other hand A3, A9, B12, B13, and B40 alleles were most significant an among hepatitis C patient. **Conclusion:** Genetic predisposition may play a role in chronic hepatitis.HLA-2 was most significant among hepatitis B, while HLA-A9, B12, B13, and B40 were prevalent among hepatitis C patients. These alleles were considered as risk factors. Meanwhile A32 was considered as protective alleles. Racial diversity, variations in the study design, methodology and complex immune-regulatory mechanisms make it difficult to find consistent association of HLA alleles with a given HBV or HCV disease even in the same ethnic group of the global population.

Key words: Hepatitis C virus, Hepatitis B, Human leukocyte antigens.

1.Introduction:

Hepatitis B (HBV) together with hepatitis C (HCV) accounts for 75% of liver diseases and are regarded a major threat to public health worldwide **(1,2)**. Hepatitis B and C co-infections have raised major concern in HIV, transplant and other immunosuppressed patients. Intravenous drug abuse is currently the main risk but nosocomial infection is also of concern for HBV and HCV infection **(3,4,5)**. Three independent factors seem to be associated with fibrosis: age, daily alcohol consumption and male gender. Over two billion people are expected to be infected with HBV during their lifetime and about 350 million are estimated to be chronic carriers **(6)**.

Chronic carriers develop life-threatening liver cirrhosis or hepatitis carcinoma. Hepatitis C infected patients have greater chances to develop chronic hepatitis and liver cancer. About 54% to 86% of infected individuals develop chronic manifestation, and females and children tend to have lower rate of chronicity **(7,8)**. It is estimated that about 200 million people around the world are HCV infected. 4.1 million (1.6%) Americans have been infected with HCV, of whom 80% are chronically infected The mechanism of HBV and HCV pathogenesis remains elusive. Host genetic factors are proposed to be governing the pathology of disease progression**(9)**.

HBV infection can be determined by several factors like virulence of the viral strains and the host immune response. Variation in immune response is often associated with polymorphism of the major histocompatibility complex (MHC) **(10)**. Class I molecules of MHC include antigens encoded by genes in the HLA A, B and C loci, whereas class II

molecules include those encoded by genes in the HLA D regions **(11)**.

An efficient T-cell response to viral component is critical for protective immunity. The HBV is not a cytopathic agent. Inflammation and hepatic necrosis in HBV infection are assumed to result from the T lymphocyte response of host to viral antigens **(12)**. Antigen-specific T-cell receptors recognize peptides bound to HLA molecules and the repertoire of peptides bound, depends on the HLA type of the individual **(13)**. HLA tissue typing has become a useful system for studying the role of genetic factors in various diseases. The role of the HLA types in the outcome of HBV infection has been previously investigated, but they were unable to reach a definite conclusion **(14)**.

Different HLA types related to chronicity has been reported from different countries. It has been assumed that this difference could be due to racial characteristics **(15,16)**. The immune response is coordinated by the human leukocyte antigen (HLA) class I and class II molecules, which present foreign antigens to CD8$^+$ cytolytic T cells and CD4$^+$ helper T cells, respectively. The genes encoding these molecules are the most polymorphic in the human genome and are ideal candidates for the investigation of association with HBV outcomes.

Multiple factors may influence the host-virus interaction in patients infected with HCV infection **(17)**. Among host actors that have potential role in HCV infection and progression earlier studies suggest that there may be an association between HLA such as HLA-DR3 and Italian HCV positive patients **(18)**. Class I and Class II HLA are encoded by the most polymorphic genes that present antigens to CD8+ Certain HLA alleles have been shown to influence the outcome of chronic HCV infections **(19)**Various HLA alleles have been linked with either persistence or clearance of the virus. several studies have aimed to identify the involvement of HLA with different outcomes of HCV infection, but the results have not been consistent **(20)**. Moreover literature review revealed that the prevalence of HCV infection is significantly low in Indian population12-14. So far, no data on HLA association with HCV infection have been reported from western India **(21)**. A critical step for the understanding of the immunopathogenesis of HCV infection and HCV clearance is the presentation of viral epitopes on MHC class I molecules from infected cells **(22)**.

The purpose of this study was to determine HLA tissue typing of the patients with chronic hepatitis B and C virus infection, to show the possible role of HLA tissue types on the chronicity of HBV and HCV infections and to assess which HLA types were protective against chronicity.

2. Subjects and Methods

Sixty patients were diagnosed as having chronic hepatitis and were attending the out patient clinic of Internal Medicin King abdulaziz University Hospital; also fourty normal subjects were examined as a control group. The patients were 22 females and 38 males and their mean age was 45.5± 3.5 years.

All patients and controls were subjected to the following:

1- History taking: personal history, family history, history of previous liver disease, and gastrointestinal disturbance.

2- Complete clinical and abdominal examination for liver, spleen and gastrointestinal tract.

3- Ultrasound of abdomen to investigate the clinical condition of liver and spleen.

4- Diagnosis for chronic hepatitis B (a) seropositivity for HbsAg (b) High levels of serum transaminases activity (over 5 times normal). Liver transaminases (AST & ALT) were detected using Boehringer Manheuim kits.

5- Detection of hepatitis B markers by using the enzyme immunoassay kits (Behring-Germany) which included hepatitis surface antigen (HbsAg) and immunoglobulin M to hepatitis B core antigen, manufactures instructions were followed.

6-Detection of hepatitis C virus by positive HBC antibody.

7-Study of different human leukocycle antigen (HLA):

HLA typing was performed using the lymphocyte cytotoxicity method. The method was briefly using 4-6 ml of heparinised blood which layered over 2-3 ml of Ficoll Hipaque gradient (Biotest) in tubes. The tubes were centrifuged at 27.000 rpm for 30 minutes. The lymphocytes were washed with Hank solution and centrifuged at 3000 rpm for 10 minutes. The supernatant was removed and pellet was suspended in Hank and washed second time. The lymphocyte were counted and suspension adjusted with media to contain 2-2.5/10^6 lymphocyte suspension was added to every well of micro test plate, 72 A, B,C (Biotest)then complement was added and incubated for 1 hour, then Eosin B stain added and left 18-24 hours. The plates were examined and cell with cytotoxicity reactions were determined **(23)**

Statistical analysis:

The results obtained were analysed to detect of fisher exact test and Odd's ratio (Armitage) **(18)** Carrying the antigen relative to a group lacking it. The chi –square (X^2) test was used to evaluate differences between subgroups in the material. The phenotype frequencies, odds ratios (OR), probability value, Chi-square with Yates correction **(24)**

3. Results

Table (2) revealed that, among chronic hepatitis patients HLA-A2 (OR 5.6, CI=2.03-15.87, P=0001),

A9 and A19, HLA- B12, B13, and B35 were higher among patients versus control group they were considered as susceptible alleles. On the other hand HLA-29, A32, and HLA-B8 were highly significant among control, they were considered as protective alleles.

Table (3) showed that A2 (OR=11.0, CI =3.17-40.3, P=0001), A9, A24, B5, B13 and Bw35 were highly significant among hepatitis B patients versus controls. On the other hand HLA-HLA- A29, A32, HLA-B8 were significantly higher among controls.

Table (1); Different data of patients with hepatitis

Data of patients	Patients with Chronic hepatitis (60)	
	No.	**%**
Sex		
Female	22	36. 7
Male	38	63.3
Age: 20- <40 40-<50 >50 Mean S.D.	12 21 27 45.5± 3.5	20 35 45
History of alcohol		
Present	8	13.3
Absent	52	86.7
AST (mean±SD)	580±315ul/ml	AST (mean±SD)
ALT (mean ±SD)	480±180 ul/ml	ALT (mean ±SD)
Bilirubin (mean ±SD)	.1.20 3.5±	Bilirubin (mean ±SD)

Table (2); HLA among l investigated chronic hepatitis patients

Different alleles	Chronic hepatitis patients(60)		Controls (40)		OR(CI)	P value	Significant
	No.	%	No.	%			
A locus							
A1	10	16.7	3	7.5	2.47(0.57-1.2)	1.78	0.08
A2	35	58.3	8	20	5.6(2.03-15.87)	14.4	0.0001**
A3	25	41.2	10	25	2.14(0.82-5.7)	2.93	0.08
A9	30	50	2	5	19.0(3.92-125.2)	22.33	0.0001**
A10	12	20	4	10	2.25(0.6-9.09)	1.79	0.18
A11	6	10	3	7.5	1.37(0.28-7.46)	Fisher	0.73
A19	28	46.7	8	20	3.5(1.28-9.86)	7.41	0.006**
A23	6	10	1	2.5	2.49(0.9-7.05	Fisher	0.23
A24	23	38.2	8	20	0.01(0.0-0.09)	3.77	0.052
A25	7	11.7	2	5	2.51(0.44-18.6)	Fisher	0.31
A26	7	11.7	3	7.5	1.63(0.35-8.58)	Fisher	0.73
A28	3	5	3	7.5	0.62(0.1-4.32)	Fisher	0.68
A29	2	3.3	25	62.5	0.02(0.0-0.11)	42.6	0.0001**
A32	0	0	20	50	2.49(0.9-7.05)	37.5	0.0001**
B locus							
B5	23	38.3	8	20	0.75(0.2-2.79)	3.77	0.052
B7	7	11.7	6	15	0.14(0.02-0.77)	0.24	0.62
B8	2	3.3	8	20	2.67(1.0-7.5)	Fisher	0.01**
B12	24	40	8	20	8.22)2.09-37.5)	4.41	0.03*
B13	24	40	3	7.5	2.14(0.82-57.0)	12.8	0.0001**
B14	25	41.2	10	2.5	0.4(0.09-1.78)	2.93	0.08
B21	4	6.7	6	15	.5(0.1-2.35)	Fisher	0.19
Bw21	4	6.7	5	12.5	0.24(0.03-1.52)	Fisher	0.47
Bw22	2	3.3	5	12.5	0.0(0.0-1.47)	Fisher	0.11
B27	0	0	3	7.5	3.5(1.08-11.9)	Fisher	0.06
B35	20	33.3	5	12.5	15.5(3.2-102.5)	5.6	0.01**
B40	27	45	2	5	3.55(0.38-83.4)	18.65	0.0001**
B51	5	8.3	1	2.5	2.47(0.57-12.2)	Fisher	0.39
B53	10	16.7	3	7.5	0.14(0.2-2.97)	1.78	0.18

Table (3): HLA among l investigated hepatitis B patients

Alleles	Chronic patients with hepatitis B(30) No. %		Controls (40) No. %		OR(CI)	P value	Significant
A locus							
A1	5	16.7	2	67.5	3.8(0.58-31.0)	Fisher	0.013
A2	22	73.3	8	20	11.0(3.17-40.3)	19.9	0.0001**
A3	8	26.7	10	5	1.09(0.32-3.65)	0.02	0.87
A9	8	26.7	2	5	6.94(1.18-52.2)	Fisher	0.01**
A10	4	13.3	4	10	1.38(0.26-7.4)	Fisher	0.77
A11	3	10	3	7.5	1.37(0.2-9.43)	Fisher	1.0
A19	12	40	8	20	2.67(0.82-8.8)	3.36	0.06
A23	2	6.7	1	2.5	2.79(0.18-81.9)	Fisher	0.57
A24	13	43.3	8	20	3.06(1.0-10.0)	4.41	0.03*
A25	4	13.3	2	5	2.92(0.41-25.1)	Fisher	0.39
A26	1	3.3	3	7.5	0.43(0.02-5.02)	Fisher	0.62
A28	2	6.7	3	7.5	0.88(0.09-7.14)	Fisher	1.0
A29	0	0	25	62.5	0.0(0.0-0.11)	29.1	0.0001**
A32	0	0	20	50	0.0(0.0-0.19)	21.0	0.0001**
B locus							
B5	13	43.3	8	20	3.06(1.0-10.1)	4.44	0.03*
B7	2	6.7	6	15	0.4(0.05-2.5)	Fisher	0.46
B8	0	0	8	20	0.0(0.0-0.8)	Fisher	0.008**
B12	5	16.7	8	20	0.8(0.2-3.17)	0.13	0.72
B13	8	26.7	3	7.5	4.48(1.0-24.2)	Fisher	0.04*
B14	8	26.7	10	25	1.09(0.32-3.65)	0.02	0.87
B21	2	6.7	6	15	0.4(0.5-2.5)	Fisher	0.45
Bw21	2	6.7	5	12.5	0.5(0.01-2.36)	Fisher	0.69
Bw22	1	3.3	5	12.5	0.24(0.01-2.36)	Fisher	0.22
B27	0	0	3	7.5	0.0(0.0-3.01)	Fisher	0.25
Bw35	12	40	5	12.5	4.67(1.25-18.3)	7.05	0.007**
B40	2	6.7	2	5	1.36(0.13-14.6)	Fisher	1.0
B51	1	3.3	5	7.5	0.24(0.01-2.36)	Fisher	0.22
B53	5	16.7	2	5	3.8(0.58-31.0)	Fisher	0.13

Table(4): HLA among l investigated hepatitis C patients

Alleles	Chronic hepatitis(30) No. %		Controls (40) No. %		OR(CI)	P value	Significance
A locus							
A1	5	16.6	3	7.5	2.47(0.47-14.5)	Fisher	0.27
A2	13	43.3	8	20	3.06(1.0-10.1)	4.44	0.03*
A3	17	56.7	10	25	3.92(1.27-12.4)	7.26	0.007**
A9	22	73.3	2	5	52.5(8.9-404.7)	35.5	0.0001**
A10	8	26.7	4	10	3.27(0.76-14.9)	3.35	0.06
A11	3	10	3	7.5	1.37(0.2-9.4)	Fisher	1.0
A19	16	53.3	8	20	5.43(1.72-17.7)	10.9	0.0001**
A23	4	13.3	1	2.5	6.0(0.57-149.4)	Fisher	0.15
A24	10	13.3	8	20	2.0(0.6-6.79)	1.6	0.21
A25	3	10	2	5	2.11(0.26-19.7)	Fisher	0.64
A26							
A28	6	20	3	7.5	3.08(0.6-17.5)	Fisher	0.15
A29	1	3.3	3	7.5	0.43(0.02-5.02)	Fisher	0.62
A32	2	0	25	62.5	0.0(0.0-0.11)	29.2	0.0001**
B locus	0	0	20	50	0.0(0.0-0.19)	21.0	0.001**
B5	10	33.3	8	20	2.0(0.6-6.79)	1.6	0.21
B7	5	16.7	6	15	1.13(0.26-4.9)	Fisher	1.0
B8	2	6.7	8	20	0.29(0.04-1.65)	Fisher	0.17
B12	19	63.3	8	20	6.9(2.1-23.62)	13.5	0.0001**
B13	16	53.3	3	7.5	14.1(3.1-72.7)	18.2	0.0001**

Alleles	Chronic hepatitis(30)		Controls (40)		OR(CI)	P value	Significance
	No.	%	No.	%			
B14	17	56.7	10	25	3.92(1.27-12.4)	7.26	0.0007**
B21	2	6.7	6	15	0.4(0.05-2.5)	Fisher	0.45
Bw21	2	6.7	5	12.5	0.5(0.06-3.26)	Fisher	0.69
Bw22	1	3.3	5	12.5	0.24(0.01-2.36)	Fisheher	0.22
B27	3	10	3	7.5	1.37(0.2-9.43)	Fisher	1.0
B35	8	26.7	5	12.5	2.55(0.64-10.5)	2.28	0.13
B40	25	80.3	2	5	95(14.5-835.3)	44.4	0.0001**
B51	4	13.3	1	2.5	6.0(0.57-149.4)	Fisher	0.15
B53	5	16.6	3	7.5	2.47(0.45-14.5)	Fisher	0.27

Table (4) showed that A2 (OR= 3.06, CI =1.0-10.1, P=0.01),   A9,A19, HLA-B12,B13, B14 and B40 were significantly prevalent among hepatitis C patients. While HLA- A29, A32   were higher among controls.

Table (5): Comparison of different alleles among both investigated groups of hepatitis B and C patients
Table (5) Comparison of HLA among l investigated hepatitis B and C patients

Alleles	Chronic hepatitis(30)		Controls (40)		OR	P value	Significance
	No.	%	No	%			
A locus							
A1	9	30	11	36.7	0.74(0.22-2.48)	0.3	*0.58*
A2	22	73.3	13	43.3	3.6(1.07-12.4)	5.5	*0.01***
A3	8	26.7	17	56.7	0.28(0.08-0.93)	5.5	*0.01***
A9	8	26.7	22	73.3	0.13(0.04-0.47)	13.1	***0.0001***
A10	4	13.3	8	26.7	0.42(0.09-1.86)	1.67	*0.19*
A11	3	10	3	10	1.0(0.14-6.99)	Fisher	*1.0*
A19	12	40	16	53.3	0.58(0.18-1.83)	1.07	*0.30*
A23	2	6.7	4	13.3	0.46(0.05-3.35)	Fisher	*0.67*
A24	1	3.3	0	0	Undefined	Fisher	*1.0*
A25	4	13.3	3	10	1.38(0.22-8.8)	Fisher	*1.0*
A26	1	3.3	6	20	0.14(0.01-1.31)	Fisher	*0.1*
A28	2	6.7	1	3.3	2.07(0.13-61.3)	Fisher	*1.0*
A29	0	0	2	6.7	0.0(0.0-4.15)	Fisher	*0.49*
A32			0	0			
B locus							
B5	13	43.3	10	33.3	1.53(0.47-4.99)	0.63	0.42
B7	2	6.7	5	16.7	0.36(0.04-2.38)	Fisher	0.42
B8	0	0	2	6.7	0.0(0.0-4.15)	Fisher	0.49
B12	5	16.7	19	63.3	0.12(0.03-0.45)	13.6	0.000**
B13	8	26.7	16	53.3	0.32(0.09-1.06)	4.4	0.03*
B14	2	6.7	3	10	0.64(0.07-5.3)	Fisher	1.0
B21	2	6.7	2	6.7	1.0(0.09-10.9)	Fisher	1.0
Bw21	2	6.7	2	6.7	1.0(0.09-10.9)	Fisher	1.0
Bw22	1	3.3	1	3.3	1.0(0.0-38.8)	Fisher	1.0
B27	0	0	3	10	0.0(0.0-2.23)	Fisher	0.23
Bw35	12	40	8	26.7	1.83(0.54-6.3)	1.2	0.27
B40	2	6.7	25	80.3	0.01(0.0-0.10)	35.6	0.000**
B51	1	3.3	4	13.3	0.22(0.01-2.39)	Fisher	0.35
B53	5	16.7	5	16.7	1.0(0.21-4.68)	0.0	1.0

*p<0.05(significant)   **p<0.01(highly significant)   p>0.05(non-significant)

Table (5) In comparison between different detected C, the results revealed that, HLA- A2 allele was more significant among hepatitis B patients. On the other hand   A3, A9, HLA- B12, B13, and B40 were most significant alleles among hepatitis C patient.

4. Discussion

Hepatitis B (HBV) and hepatitis C (HCV) viral infection or co-infection leads to risk of development of chronic infection, cirrhosis and hepatocellular carcinoma (HCC). Immigration and globalization have added to the challenges of public health

concerns regarding chronic HBV and HCV infections worldwide Extensive literature search was conducted to explore the HLA associations in HBV and HCV infections reported across global populations over the past decade to understand the knowledge status, weaknesses and strengths of this information in different ethnic populations **(25)**.

HLA loci diversity due to racial admixture, environment and selection pressure and by inherent polymorphic nature results in allelic variation in different ethnic groups, correspondingly different HLA associations were detected with disease in different populations. Thus association of disease outcome with HLA alleles appears to depend upon the ethnicity of the infected individual and therefore is inconsistent across the populations despite being robust within an ethnic group **(13)**.

Our findings declared that, HLA-A2 (OR 5.6, CI=2.03-15.87, P=0001), A9 (OR 19.0, CI=3.92-125.2, P=0.0001) and A19(OR 3.5, CI=1.28-9.86, P=0.0001), B12 (OR 8.22, CI=2.09-37.5, P=0.03), B13 (OR 2.14, CI=0.82-57.0, P=0.0001), and B35(OR 15.5, CI=3.2-102.5, P=0.01) alleles were higher among patients versus control group they were considered as susceptible alleles. On the other hand HLA-A29 (OR 0.02, CI=0.0-0.11, P=0.0001), A32 (OR 2.49, CI=0.9-7.05, P=0.0001), and B8 (OR 2.67, CI=1.0-7.5, P=0.01) alleles were highly significant among control, they were considered as protective alleles. Our results were agreement with that detected by **(26)**.

Abbott et al. (26) explained that, Cytotoxic T lymphocytes (CTL) were important to the control of viral replication and their presence may be important to disease outcome. An understanding of the spectrum of proteins recognized by hepatitis C virus (HCV)-specific CTL and the functional properties of these cells were important step in understanding the disease process and the mechanisms of persistent infection, which occurred in the majority of HCV-infected individuals. The authors reported that, HCV-specific CTL responses restricted by the HLA class I molecules A2, A3, A11, A23, B7, B8, and B53..

Our finding were in agreement with that detected by **(11)**, **Chloe et al (11)** revealed that single class I allele, A 0301 (odds ratio [OR], 0.47; 95% confidence interval [CI], 0.30 to 0.72; P = 0.0005) was associated with viral clearance. B 08 was associated with viral persistence both independently (OR, 1.59; 95% CI, 1.04 to 2.43; P = 0.03) and as part of the conserved Caucasian haplotype.

Regarding to hepatitis B patients, our results declared that, HLA-A2 (OR=11.0, CI =3.17-40.3,P=0001), A9 (OR 6.94, CI=1.18-52.2 P=0.01), A24 (OR 3.06, CI=1.0-10.0, P=0.03), B5 (OR 3.06, CI=1.0-10.1, P=0.03), B13 (OR 4.48, CI=1.0-

24.2,P=0.04) and Bw35 (OR 4.67, CI=1.25-18.3, P=0.007) alleles were highly significant among versus controls. On the other hand HLA-A29, A32, HLA-B8 were significantly higher among controls.

Our finding were in accordance with that detected by **Karan et al (7)** who investigated the mechanisms of underlying development of chronic hepatitis B virus infection (HBV) in Turkish population using HLA tissue typing. The frequencies of HLA- A2, B8, B13, CW3, were significantly higher in patients compared to the controls group. They concluded that, HLA-A24 and Cw1 were associated with low risk for HBV-related chronic liver disease and HLAB13, B8, DR7, DR13 and DQ3 were associated with high risk for chronic HBV infection in the Turkish population

Other studies were performed to investigate the HLA association with HBV there were many results variations as in Russian populations the investigators found that, chronic HBV and HCV infection were associated with HLA B35 and B40 **(27)**, and the authors suggested that, chronic hepatitis associated with HLA -B 35, independent of virus type. HLA B35 involvement was related with viral persistence of HCV in Koreans (**14**), and of HBV in Han Chinese **(28)** HLA B8 was associated with viral persistence and chronicity for both HBV and HCV infections in Caucasian populations. HLA Cw*07 was a risk factor for vertical infection in Italians (**29**).

Worldwide, chronic hepatitis B affects an estimated 350 million persons and is the leading cause of cirrhosis and hepatocellular carcinoma (Infection with hepatitis B virus (HBV) in adulthood results in viral persistence and development of chronic hepatitis in 5 to 10% of cases, but factors that determine viral persistence or clearance are not well understood **(11)**. Certain groups of individuals are known to be at increased risk of developing chronic hepatitis B, including those who are human immunodeficiency virus (HIV) seropositive, male, immunosuppressed, and elderly at the time of infection **(30)**. In addition, the host immune response plays a key role in determining the outcome, since a vigorous, polyclonal cytotoxic-T-lymphocyte (CTL) response correlates with viral clearance. These immune responses may be genetically determined, since twin and family studies have suggested an inherited component in the development of chronic hepatitis B **(31)**

The immune response is coordinated by the human leukocyte antigen (HLA) class I and class II molecules, which present foreign antigens to $CD8^+$ cytolytic T cells and $CD4^+$ helper T cells, respectively. The genes encoding these molecules are the most polymorphic in the human genome and are

ideal candidates for the investigation of association with HBV outcomes **(12,32)**

Systematic studies in compliance with global regulatory standards are required to identify the HLA specific viral epitope, stage specific T cell populations interacting with different HLA alleles during disease progression and viral clearance of chronic HBV or HCV infections among different ethnic populations. These studies would facilitate stage specific therapeutic strategies for clearance of HBV and HCV infections or co-infections across global populations and aid in identification of HBV-HCV combined vaccine. HLA associations of chronic HBV or HCV development with confounding host factors including alcohol, drug abuse, insulin resistance, age and gender are lacking and warrant detailed investigation across global populations **(32)**.

On detecting the prevalent alleles among hepatitis C patients our results showed that, A2 (OR= 3.06, CI =1.0-10.1, P=0.01), A9 (OR 52.5, CI= 8.9-404.7, P=0.0001),A19 (OR 5.43, CI= 1.72-17.7, P=0.0001),B13 (OR 6.9, CI=2.1-23.62, P=0.0001), B14 OR, 14.1, CI=3.1-72.7, P=0.0001), B21 (OR 3.92, CI= 1.27-12.4,P=0.0007) and B40 (OR 95, CI=14.5-835.3, P=0.0001) alleles were significantly prevalent among hepatitis C patients. While HLA- A29, A32 were higher among controls.

Our findings were confirmed by that detected by **Hadhoud et al.(33)** who linked the susceptibility to HCV infections with A-19 alleles in Saudi people.

Other studies declared many different associations of HLA with hepatitis C as that, detected by **Zekri et al (15)** who found prevalence of HLA A-28, A-9, B-14, DR7 in Egyptians᾿ in Irelan, Fanning et.al **(34)** detected that HLA A11, HLA B-35 and B8 were strongly associated with chronic HCV infection, In American whites **(27)**, HCV persistence was associated with HLA Cw*04 and HLA B53 **(35).**

Many studies were performed to detect the associated Class I alleles and hepatitis C, our results were correlated to that, performed with **Mosaad et al (23)** who investigated HLA alleles class I among patients with hepatitis C in Egypt, HLA-A11 antigen was significantly increased in patients with chronic HCV infection versus controls (OR 3.98; 95% CI = 1.85-8.89; P = 0.001; and Pc = 0.021). Also, HLA-B12, HLA-B13, HLA-B17 and HLA-B40 were higher in patients, and HLA-A32 and HLA-B14 were higher in controls.

One of the striking features of HCV infection is the very high rate of development of chronicity. Approximately 15 per cent of infected patients successfully eliminate the virus, while others develop chronic infection with a wide spectrum of disease. Some will remain asymptomatic whereas others may have a more severe course leading to cirrhosis or hepatocellular carcinoma **(36)**.

There are evidences that immune mechanisms contribute to control the HCV

Infection in the host immune reaction against viral infections, HLA alleles play vital role in modulating the immune responses **(37)**. Hence, many studies were designed to examine the frequencies of HLA class I and class II genotype profiles in HCV infected western Indian individuals **(38, 39)**. **Yoon et al (14)** reported that, the frequencies of HLA-A3, HLA-B35 and HLA-B46 significantly increased in chronic HCV carriers

Anuradha et al (21) in India reported that, Host genetic diversity is believed to contribute to the spectrum of clinical outcomes in hepatitis C virus (HCV) infection. The authors' found out the frequencies of HLA class I and class II alleles of HCV infected individuals, they revealed an association of HLA alleles HLA A*03 (OR= 16.69, EF, 0.44, *P*=7.9E-12), A*32 (OR= 1474, EF 0.21, *P*=1.8E-9), HLA B*15 (OR=14.11, EF 0.39, *P*=2.18E-10), B*55 (OR= 12.09, EF 0.07, *P*=0.005),

Our results showed that, HLA- A29 and A32 were higher among controls this results were in agreement with others **who found that** Protection from HCV infection is associated with HLA-B51, -B52, -B55, -B56, -B61, B70, -Cw1, -Cw3, and -Cw4 in Japanese population **(40)**. In Ireland HLA B27 and A 3 had strong association with self-resolving HCV infection**(19)** Spontaneous HCV clearance was associated with HLA B-27 in German women cohorts **(41)** and HLA A*1101, B57 and Cw0102 were reported to be associated with viral clearance in American white**s (35)**.

NK cells mediate direct killing of infected and transformed cells contributing to viral clearance and protection from tumor development. This killer activity is under the control of HLA class I molecule interaction with inhibitory and activation receptors-killer cell immunoglobulin like receptors (KIR) of NK cells. HLA C molecules interact with KIRs of NK cells and modulate their cell killer activity. Activation receptor KIRDL3 and HLA-C1C1 ligand interact directly to influence viral clearance in HCV infection **(42).**

In comparison between different detected C, the results revealed that,st HLA- A2 allele was more significant among hepatitis B patients. On the other hand A3, A9,HLA- B12, B13, and B40 were most significant alleles among hepatitis C patient.

The specific HLA associations with HBV and HCV infections are different, agreeing to their differences in viral properties and disease pathogenesis, with few exceptions where they share few HLA loci **(42)**. This could be due to linkage

disequilibrium of HLA alleles with disease-associated genes or shared HLA restricted HCV and HBV T cell viral epitopes. It is intriguing that HLA associations are oppositely directed and indicate interactions with other unidentified factors in influencing the HLA mediated immune signaling **(43)**. The knowledge of HLA gene marker association with risk for HBV/HCV related disease progression/cirrhosis may be helpful while seeking donor-recipient HLA match before transplantation **(44, 45)**. It is also observed that HBV and HCV infections tend to suppress each other. HBV and HDV can suppress HCV infection and HCV and HDV can have negative effect on HBV. HCV super infection is seen to reduce HBsAg expression and promote its clearance **(5)**.

In conclusion

Extensive allele diversity is observed in HLA associations with susceptibility and protection regarding HCV and HBV infections and disease progression in different global ethnic populations HLA loci diversity due to racial admixture, environment and selection pressure and by inherent polymorphic nature results in allelic variation in different groups, correspondingly we get different HLA associations with disease in different populations. Thus association of disease outcome with HLA alleles appears to depend upon the ethnicity of the infected individual and therefore is inconsistent across the populations despite being robust within an ethnic group**.** The knowledge of HLA gene marker association with risk for HBV/HCV related disease progression/cirrhosis may be helpful while seeking donor-recipient HLA match before transplantation

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