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Association of HLA class I -A and B alleles with hydatidosis In Iraqi patients

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Abstract:

Hydatidosis is a parasitic disease caused by infection with larval stage of cestode worm Echinococcus granulosus ,it is a worle_wide disease results in serious health and financial loss. Iraq considered as an endemic region for this disease. the association of HLA –alleles frequency with the occurrence of various diseases were studied . and significant associations were found, no sufficient studies on hydatidosis and possible role of HLA typing and the immune status of the patients and association of them with cyst characteristics ,and cytokines profile and their effect on production of immunoglobulin . blood samples were collected for extraction of DNA and amplification by (PCR-SSO) and HLA typing by Automatic Line Probe Assay (Auto-Lipa) for HLA typing of both patients and healthy control groups, to study the association of certain HLA alleles. With occurrence of Hydatid disease. The present study reported increase in frequency of HLA-A*0273 in hydatid patients compare to healthy control .

Keywords: HLA, alleles , Hydatidosis , PCR

Introduction;

The pattern and relationships between Various diseases and HLA-alleles typing are the current areas of great interest throughout the world..many studies accomplished in this field and reveal a significant

association between certain alleles with many (including infectious) diseases. the T-helper I and II lymphocytes' profile seems to play an important role in susceptibility or resistance to the disease.(Guttstein2003) The most important determinants of genetic susceptibility to CE are located in the major histocompatibility complex (MHC) or human leukocytes antigen (HLA)gene area on shout arm of chromosome number 6. The HLA is a kind of of genetic marker of human beings (Kindit. et al;, 2007).The HLA component of the immune system ,encoded by highly polymorphic genes that vary across racial/ethnic groups ,has been suggested to be biologically based risk factor for CE and thus may explain some of its variation by race/ethnicity (Al-Ghoury.et al;2010).Moreover ,a number of HLA alleles have been reported to be associated with the occurrence of CE(Shcherbakov 1993;Azab et al 2004)while others were reported to be associated with protection against the parasite (Shcherbakov and Monje-Barredo 1989;Shcherbakov 1993).during the last decade ,molecular typing techniques have been developed to identify HLA alleles at DNA level, and DNA mutation now allow the identification of a high number of alleles according to the loci, and the resulting new nomenclature, have contributed to a better understanding of this system(Alves et al,2006).

In the present study we investigate association of HLA class I A and B alleles and CE in Iraqi patients, as well as to provide information about genotypes that confer susceptibility or resistance to develop the disease.

2-Material and Methods;

A total 33 Iraqi Arab patients with CE enrolled in this case control study and 20 healthy individuals as control groups. 5 ml of venous blood were collected and divided into 2 ml in (EDTA)anticoaguated tubes for DNA extraction by (Kia-gene Kit) ,and three ml in plain tube for serum extraction .Amplification by PCR-SSO (Saiki et al.1988) using an (HLA-DRBI,DQ) amplification kit developed and

manufactured by Innogenetics for Murex Biotech. Molecular typing of HLA alleles was performed using a reverse hybridization Automatic Line probe assay (Auto-Lipa) also supplied by Innogenetics for Murex Biotech, in which typing tests are based on the (reverse dot blot hybridization) (Buyse et al 1993).positive probes on each strips were recognized by typing table (provided with the kit) Statistical analysis; Data were analyzed with Statistical Package of Social Science (SPSS) Version 19 for windows. To assess the statistical significance of association of different allele frequencies with the disease susceptibility or resistance.

3-Results;

The majority of infected individuals were females in comparison to males. The majority of cysts were located in the liver (hepatic cysts)and then in the lunge (pulmonary) with few number of combined infections(liver +lunge). There were significant differences in frequency of HLA-A HLA-A*0273 alleles between patients and healthy control groups with P value less than 0.020

4-Discussion;

In the present study an increase frequency of HLA-A*0273 allele which is absent in Patients (0.0%) of patients while the frequency of this allele in the control groups was(20%)in hydatid patients .so this allele might confer protection from the disease, there were a significant P value 0.021 .the present study revealed alleles frequency in Iraqi hydatid patients different in their profile compare to other studies in various region in the world. Since various ethnic groups have been studied to determine HLA association with hydatid disease and different alleles have been reported as resistance alleles and disease susceptibility alleles ,HLA-A10,HLA-B5,and HLA-B18ted have been reported to be associate with susceptibility to Cystic echinococcosis(Shcherbakov 1993)

Shcherbakov and Monje -Barredo 1989 showed a statistically positive association between HLA-A1 and HLA-

B35 alleles and CE.HLA-A9 was found to be statistically negatively associated with AE(Shcherbakov,1993).

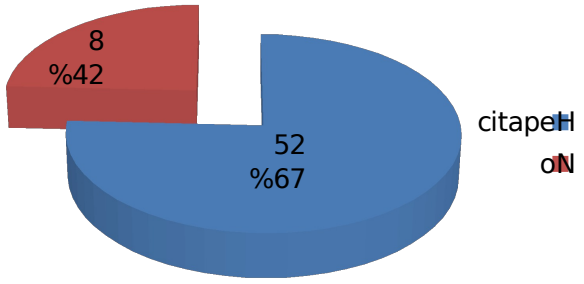

Figure 1; distribution of patients according to cyst location

Table 1; significant HLA-class I,A alleles in Hydatidosis patients compare to healthy control

P value	PF	EF	IOR	OR	%	Control	%	Hydatidosis patients	HLA-A allele
NS	0.81	4.22-	3.109	0.322	20.00%	4	6.67%	2	**0101***
NS	1.41	3.43	0.144	6.962	0.00%	0	13.33%	4	**0106***
NS	0.00	0.00	4.692	0.213	5.00%	1	0.00%	0	**0109***
NS	0.00	0.00	4.692	0.213	5.00%	1	0.00%	0	**0123***
NS	2.15	1.87	0.627	1.596	10.00%	2	16.67%	5	**0130***
NS	3.25	1.44	0.278	3.596	0.00%	0	6.67%	2	**0201***
NS	1.08-	0.52	0.480	2.085	0.00%	0	3.33%	1	**0205***
NS	0.00	0.00	8.243	0.121	10.00%	2	0.00%	0	**0236***
NS	3.25	1.44	0.278	3.596	0.00%	0	6.67%	2	**0247***
021.	0.00	0.00	16.636	0.060	20.00%	4	0.00%	0	**0273***

Table2 ; HLA class I,B alleles in Hydatidosis patients compare to healthy control

P value	PF	EF	IOR	OR	%	Control	%	Hydatidosis patients	HLA-B allele
NS	0.34	-0.51	1.513	0.661	5.00%	1	3.33%	1	0103*
NS	0.00	0.00	4.692	0.213	5.00%	1	0.00%	0	0193*
NS	-0.33	0.25	0.877	1.140	5.00%	1	6.67%	2	0205*
NS	-1.08	0.52	0.480	2.085	0.00%	0	3.33%	1	0206*
NS	-1.08	0.52	0.480	2.085	0.00%	0	3.33%	1	0309*
NS	-1.08	0.52	0.480	2.085	0.00%	0	3.33%	1	0412*
NS	-1.08	0.52	0.480	2.085	0.00%	0	3.33%	1	0708*
NS	1.45	3.22	0.357	2.804	5.00%	1	16.67%	5	0801*
NS	-0.57	0.36	0.927	1.078	15.00%	3	16.67%	5	0810*
NS	-1.08	0.52	0.480	2.085	0.00%	0	3.33%	1	0816*
NS	-1.08	0.52	0.480	2.085	0.00%	0	3.33%	1	1302*
NS	0.00	0.00	8.243	0.121	10.00%	2	0.00%	0	1517*
NS	-1.08	0.52	0.480	2.085	0.00%	0	3.33%	1	1529*
NS	-1.08	0.52	0.480	2.085	0.00%	0	3.33%	1	1561*

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