Possible Association of HLA Class-I Molecules with Colorectal Cancer in Iraqi Patients

Batool H. Al-Ghurabi* Ph.D
Ahmed A. Al-Hassan**Ph.D
Sana'a A. Al-Naseri*** Ph.D
Khalida M. Mousawy**** Ph.D

Summary:

Background: *Genetic factors were found to play a crucial role in the development of colorectal cancer.*

Objectives: *This study was established to shed light on the possible association of HLA class-I antigens and CRC patients, and to correlate the findings with both family history and tumor location.*

Patients and Methodes: ***Lymphocytotxicity assay has been used to assess HLA-typing of 150 blood samples of 100 CRC patients and 50 healthy normal controls in*** *College of Dentistry/ University of Baghdad.*

Results: *Comparison between CRC patients and healthy controls showed several antigens deviations in their frequencies. HLA-A2, A28 and B39 antigens were observed with increased frequencies in patient's group with significant differences (P< 0.008, 0.011 and 0.023 respectively; moreover, yet however statistical analysis showed non significant correlation of these specific HLA-Ags with both family history of CRC and tumor location.*

Conclusions: *This finding demonstrated that HLA A2, A28 and B39 might play a role in CRC susceptibility.*

Key words: *Colorectal cancer, HLA, Family histor of CRC, Tumor location.*

Introduction:

Colorectal cancer (CRC) is the fourth most common malignancy in man preceded by lung, prostatic, and breast cancers (1). According to several observations, attention has been directed to the belief that a genetic background plays a crucial role in determining susceptibility to CRC.

The importance of participating genetic factors in this disease is currently based on the increased risk of cancer in first-degree relatives of patients with CRC: 15% of siblings and 10% of offspring (2,3), and increased frequency of specific genetic markers as certain human leukocyte antigen (HLA) in patients group than in general population (4). Therefore; several studies in different areas were undertaken to test the possibility of association between this disease and one or several of the HLA-Ags, as suggested by Alcaly and associates (5). Positive association with the antigen (A1) was reported in Caucasians patients (6), while another study reported by Gainullina et al., revealed an increased frequency of HLA-A25 and HLA-A28 and decreased HLA-A11 in Uzbek patients (7). Moreover, Hiwatashi and associates reported an increased frequency of HLA-B35 phenotype in his patients in Japan (8).

*Depr. Of Immunology, College of Dentistry/ University of Baghdad.
**Depr. Of Immunology , Al-Mussyaib Technical College.
***Depr. Of Pathology / MD, , Medical College / University of Baghdad.
****Depr. Of Immunology/ MD, Medical College / University of Baghdad.

Patients and Methods:

Patients:

The present study included 100 Arab, Iraqi CRC patients (42 females and 58 males; mean age 51.4 years, ranged between 21-81). Duke's classification and degree of differentiation are presented in Table-1, compared with 50 healthy age and sex matched control group.

HLA-Typing:

Microlymphoctotoxicity assay has been applied for HLA-typing as described by Terasaki and McClelland (9) and modified by Dick, et al., and Bender (10, 11).

Statistical Analysis:

Univariate analysis has been applied for the data depending on logistic regression and the results were reported as odds ratio (ORs), which represented the increased or decreased risk for CRC.

	Well Differentiated N=8	Moderately Differentiated N=18	Poorly Differentiated N=74
Duke's A	2	4	0
Duke's B	3	43	8
Duke's C	1	15	3
Duke's D	2	12	7

Table-1: Degree of differentiation and stage classification (Duke's A-D) of the tumor.

Results:

Tumors of the colon and rectum of 100 patients were localized as shown in Table-2, where 25 patients had the tumors in ascending colon, 6 in the transverse colon, 32 in the descending and sigmoid colon and 37 in the rectum.

Table-2:Localization of the CRC

Location	NO	Femal	Male
Cecum, ascending colon	25	11	14
Transverse colon	6	1	5
Descending colon, sigmoid colon	32	13	19
Rectum	37	16	21

A total of 100 Iraqi patients with CRC were typed for HLA-class I (A, B & C) antigens.The frequency distribution of various class I HLA-Ags for the studied groups are presented in Tables (3, 4 & 5). Comparison between CRC patients and healthy controls showed several antigens deviations in their frequencies. For instance HLA-A2, A28 and B39 antigens were observed with increased frequencies in patient's group (35, 27 and 18% respectively) versus healthy controls (58, 8 and 2% respectively), with P-values of (0.008, 0.011 and 0.023 respectively).

Regarding the correlation between the specific HLA-Ags (A2, A28, B39, and DR7) and patients with family history, the present study revealed strong correlation between HLA-A28 and CRC patients with positive family history (Inverse OR= 5.1) though statistically not significant, as shown in Table (6). However, no differences in the distribution of these specific antigens were found between colon and rectum (location of tumor), Table (7).

Table -3: Antigens frequency of the HLA-A (%, OR, inverse OR, P, EF, PF) of the CRC patients and healthy control.

HLA antigen	Healthy control		Colorectal cancer cases		OR	Inverse OR	P	EF	PF
	N	%	N	%					
HLA-A									
1	15	30.0	25	25.0	0.8	1.3	NS	**	0.067
2	29	58.0	35	35.0	0.4	2.6	0.008	**	0.354
3	6	12.0	11	11.0	0.9	1.1	NS	**	0.011
9	3	6.0	5	5.0	0.8	1.2	NS	**	0.011
10	1	2.0	3	3.0	1.5	**	NS	0.010	**
11	5	10.0	7	7.0	0.7	1.5	NS	**	0.032
23	2	4.0	5	5.0	1.3	**	NS	0.010	**
24	10	20.0	18	180	0.9	1.1	NS	**	0.024
25	1	2.0	3	3.0	1.5	**	NS	0.010	**
26	3	6.0	4	4.0	0.7	1.5	NS	**	0.021
28	4	8.0	27	27.0	4.3	**	0.011	0.207	**
29	3	6.0	2	2.0	0.3	3.1	NS	**	0.041

HLA antigen	Healthy control		Colorectal cancer cases		OR	Inverse OR	P	EF	PF
	N	%	N	%					
30	3	6	4	4.0	0.7	1.5	NS	**	0.021
31	2	4.0	4	4.0	1.0	**	NS	**	**
32	2	4.0	2	2.0	0.5	2.0	NS	**	0.020
33	2	4.0	3	3.0	0.7	1.3	NS	**	0.010
34	1	2.0	2	2.0	1.0	**	NS	**	**
36	1	2.0	4	4.0	2.0	**	NS	0.020	**
Blank	7		36						
Total	100		200						

OR= Odds ratio
EF= Etiologic fraction
PF= Preventive fraction

Table-4: Antigens frequency of the HLA-B (%, OR, inverse OR, P, EF, PF) of the CRC patients and healthy control

HLA antigen	Healthy control		Colorectal cancer cases		OR	Inverse OR	P	EF	PF
	N	%	N	%					
HLA-B									
5	3	6.0	4	4.0	0.7	1.5	NS	**	0.021
7	0	0	3	3.0	3.6	**	NS	0.022	**
8	7	14.0	9	9.0	0.6	1.6	NS	**	0.055
12	0	0.0	3	3.0	3.6	**	NS	0.022	**
13	0	0	0	0.0	0.5	2	NS	**	**
14	4	8.0	9	9.0	1.1	**	NS	0.011	**
15	1	2.0	3	3.0	1.5	**	NS	0.010	**
17	2	4	1	1.0	0.2	4.1	NS	**	0.030
18	5	10.0	5	5.0	0.5	2.1	NS	**	0.053
21	1	2.0	8	8.0	4.3	**	NS	0.061	**
22	0	0	0	0.0	0.5	2.0	NS	**	**
27	3	6.0	6	6.0	1.0	**	NS	**	**
35	4	8.0	2	2.0	0.2	4.3	NS	**	0.061
37	3	6	3	3.0	0.5	2.1	NS	**	0.031
38	5	10.0	7	7.0	0.7	1.5	NS	**	0.032
39	1	2.0	18	18.0	10.8	**	0.023	0.163	**
40	2	4.0	1	1.0	0.2	4.1	NS	**	0.030
41	3	6	3	3.0	0.5	2.1	NS	**	0.031
44	6	12.0	4	4.0	0.3	3.3	NS	**	0.083
45	1	2.0	0	0.0	0.2	6.1	NS	**	**
47	1	2.0	1	1.0	0.5	2.0	NS	**	0.010
49	1	2	0	0.0	0.2	6.1	NS	**	**

HLA antigen	Healthy control		Colorectal cancer cases		OR	Inverse OR	P	EF	PF
	N	%	N	%					
50	1	2	1	1.0	0.5	2	NS	**	0.010
51	7	14.0	8	8.0	0.5	1.9	NS	**	0.065
52	1	2	0	0.0	0.2	6.1	NS	**	**
53	3	6.0	2	2.0	0.3	3.1	NS	**	0.041
54	0	0.0	1	1.0	1.5	**	NS	0.003	**
55	1	2.0	6	6.0	3.1	**	NS	0.041	**
56	1	2.0	2	2.0	1.0	**	NS	**	**
57	1	2.0	1	1.0	0.5	2.0	NS	**	0.010
60	0	0.0	1	1.0	1.5	**	NS	0.003	**
62	1	2.0	3	3.0	1.5	**	NS	0.010	**
63	1	2.0	2	2.0	1.0	**	NS	**	**
70	1	2.0	1	1.0	0.5	2.0	NS	**	0.010
73	0	0.0	2	2.0	2.6	**	NS	0.012	**
Blank			80						
Total			200						

OR= Odds ratio
EF= Etiologic fraction
PF= Preventive fraction

Table-5: Antigens frequency of the HLA-C (%, OR, inverse OR, P, EF, PF) of the CRC patients and healthy control.

HLA antigen	Healthy control		Colorectal cancer cases		OR	Inverse OR	P	EF	PF
	N	%	N	%					
HLA-C									
1	2	4.0	4	4.0	1.0	**	NS	**	**
2	5	10.0	7	7.0	0.7	1.5	NS	**	0.032
3	4	8.0	6	6.0	0.7	1.4	NS	**	0.021
4	10	20	18	180	0.9	1.1	NS	**	0.024
5	2	4.0	3	3.0	0.7	1.3	NS	**	0.010
6	8	16	13	13.0	0.8	1.3	NS	**	0.034
7	8	16	13	13.0	0.8	1.3	NS	**	0.034
8	1	2.0	3	3.0	1.5	**	NS	0.010	**
Blank	60		133						
Total	100		200						

OR= Odds ratio
EF= Etiologic fraction
PF= Preventive fractio

Table-6: Antigens frequency of the specific HLA-Ags in CRC patients according to family history.

	Family history of colorectal cancer						
	Negative		Positive				
HLA antigen	N	%	N	%	OR	Inverse OR	P
HLA-A2							
Negative	58		7				
Positive	29	33.3	6	46.2	1.7	**	NS
HLA-A28							
Negative	61		12				
Positive	26	29.9	1	7.7	0.2	5.1	NS
HLA- B39							
Negative	73		9				
Positive	14	16.1	4	30.8	2.3	**	NS

Table-7: Antigens frequency of the specific HLA-Ags in CRC patients according to tumor location.

	Tumor location						
	Rectum		Colon				
HLA antigen	No.	%	No.	%	OR	Inverse OR	P
HLA-A2							
Negative	23		42				
Positive	14	37.8	21	33.3	0.8	1.2	NS
HLA-A28							
Negative	26		47				
Positive	11	29.7	16	25.4	0.8	1.2	NS
HLA- B39							
Negative	30		52				
Positive	7	18.9	11	17.5	0.9	1.1	NS

Discussion:

The role of genetic factors in the etiology of CRC was documented many decades ago. Sivak and colleagues have reported a family study compatible with the linkage of adenocarcinoma of the colon with the MHC (12).

In the present work, there was a significant association of HLA-A2 and A28 with CRC patients [p= (0.008), (0.011)] as compared with healthy group.This result is in agreement with that of Gainullina et al.(7) regarding significant statistical association of A2 and A28 in comparison with control group. Meanwhile many authors reported that the HLA-haplotypes A2/B22 and A28/B27 were found to be significant in patients with CRC whereas only A28/B27 was significant in familial adenomatous polyposis (FAP) patients.

Regarding the presence of HLA-B Ag and its proposed association with CRC, extensive studies were conducted on HLA-B locus to give a contradicting results. In this study the increasing frequency of HLA-B39 was statistically significant in CRC patients (p=0.02) compared to control group. This result is at variance with some other studies (5, 7, 8), in which there was no significant association between B39 and this cancer, but there was positive association with other antigens of HLA-B.

Hiwatashi and colleagues (8) who studied the differences between CRC and colorectal adenoma in comparison with controls using HLA markers in Japanese individuals, reported that B40 was increased while B5 was decreased in the colorectal adenoma cases, whereas in the CRC cases, B35 was increased.

Interestingly, the present study failed to demonstrate a significant association of these specific HLA-Ags (A2, A28 andB39) with both family history of CRC and tumor location. This might, in part, result from the limited number of investigated patients. However; another study reported a strong positive association of some HLA-Ags such as A1, B18 and DQ5 with a family history of CRC in Greece population (4).

In conclusion, this finding demonstrated that HLA A2, A28 and B39 might play a role in CRC susceptibility.

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