Association of Human Leukocyte Antigen (HLA)-B5 and -BW22-J with Hepatic Cirrhosis of Apparently Unknown Etiology

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OHKUBO, A, OKIMOTO, K, IWASAKI, Y, KOSAKA, K, JUJI, T. and TOHYAMA, H. Association of Human Leukocyte Antigen (HLA)-B5 and -BW22-J with Hepatic Cirrhosis of Apparently Unknown Etiology. Tohoku J. exp. Med, 1977, 123 (3), 257-263 — The majority of patients with hepatic cirrhosis in Japan do not have a history of acute hepatitis or alcoholism. Twenty-nine patients with hepatic cirrhosis which were thought as random samples from cirrhotic patients without a history of acute hepatitis or alcoholism were Human Leukocyte Antigen (HLA) typed. Association of HLA-B5 and -BW22-J with hepatic cirrhosis of apparently unknown etiology was found in $p < 0.003$, and $p < 0.01$, respectively (128 controls). A predisposition linked to HLA-B5 or -BW22-J related gene or genes seems to play a role in the development of hepatic cirrhosis, at least in a substantial part of patients with hepatic cirrhosis of apparently unknown etiology in Japan. ——— hepatic cirrhosis; HLA-B5; HLA-BW22-J

In Japan where alcoholism is not so prevalent as in Europe and the United States of America (Takata 1976), hepatic cirrhosis develops insidiously without a history of acute hepatitis or alcoholism in the majority of the patients (Matsushita 1975). In cases of chronic hepatitis following acute viral hepatitis, abnormal immune response has been assumed to play a role in the development of chronic hepatitis (Dudley et al. 1972; Eddleston and Williams 1974).

In 1972, Mackay and Morris reported association of certain specificities of the human major histocompatibility system, HLA-A1 and -B8, with autoimmune active chronic hepatitis. Since then, several investigations on the association of certain specificities of HLA with chronic liver diseases have been reported on Caucasians (Freudenberg et al. 1973; Galbraith et al. 1974; Lindberg et al. 1975; Freudenberg et al. 1976).

These reports confirmed an association of HLA-B8 with chronic active hepatitis of apparently unknown etiology, but not with chronic active hepatitis with HBs antigenemia (Galbraith et al. 1974; Bertrams et al. 1974; Lindberg et al. 1975; Freudenberg et al. 1976). In contrast to chronic active hepatitis, a part of which

is expected to develop cirrhosis, few attempts have been reported to study association of certain specificities of HLA with hepatic cirrhosis. Mackay and Morris (1972) could not find association of certain HLA specificities with hepatic cirrhosis. Freudenberg et al. (1973) could not either find association of certain HLA specificities with hepatic cirrhosis as a whole. Bailey et al. (1976) reported a relatively high incidence of HLA-B8 in patients with alcoholic cirrhosis. However, after correction of p value by the number of HLA antigens tested, the difference became insignificant.

This paper is to report association of HLA-BW22-J, a Japanese specific HLA antigen, and HLA-B5 with hepatic cirrhosis of apparently unknown etiology in Japan.

PATIENTS AND METHODS

HLA types were determined for this first (A) and the second (B) loci with the lymphocytotoxicity technique (Terasaki and McClelland 1964). Some of the anti-HLA sera were favourably supplied from Drs. van Rood, Grumet, and Perkins. The remaining was prepared from multiparous women in our laboratory. In them, antisera by which two split antigens of BW22 were identified were included. In 1973 we had identified Japanese specific split antigen of BW22 and reported it as J-1 (Juji et al. 1973). This split antigen, which was named BW22-J by Japanese HLA Workshop Committee in 1977, had not been detected in 260 Caucasian lymphocyte samples (Juji et al. 1975). The other part of BW22 split antigens showed almost the same specificity as Caucasian BW22. All the HLA typed patients as well as the control 128 healthy subject were unrelated Japanese. Twenty-nine patients with hepatic cirrhosis who had visited our clinic in 1976 were HLA typed serially. The patients with hepatic cirrhosis accompanying congenital metabolic disorders, chronic circulatory failure, longstanding cholestasis, or alcoholism, and the patients with a history of acute hepatitis were excluded from HLA study. The diagnosis of hepatic cirrhosis was established histologically by needle biopsy. In 19 out of 29 cases, needle biopsy was carried out under laparoscopy. All the classifiable cases were those of posthepatic cirrhosis according to the criteria of Gall (1960). Statistical analysis was done by the χ^2 test with Yates' correction. The p values given here were corrected for the number of antigens tested.

RESULTS

As shown in Table 1, any manifestations suggestive of acute hepatitis or alcoholism were not found in the early history of 70% of the patients with hepatic cirrhosis admitted to our department during 1972-1976.

Fig. 1 shows that the age incidence of the HLA typed with hepatic cirrhosis admitted to our department during 1972-1976. The distribution pattern of the age incidence of patients in this study was similar to that of 2421 cases of hepatic cirrhosis observed in a Japanese nation-wide epidemiological survey of chronic liver diseases in 1972 (Matsushita 1975). In that survey, congenital metabolic disorders, cardiac cirrhosis, and biliary cirrhosis were excluded. The male to female ratio of the HLA typed patients was 2.6 to 1, and that of the inpatients was 2.5 to 1. Thus, the HLA typed patients were considered as random samples out of the majority of the patients with hepatic cirrhosis of apparently unknown etiology in Japan.

TABLE 1. *Classification of the previous history of the patients with hepatic cirrhosis admitted to our department during 1972-1976*

Previous history	Number of patients	%
Acute hepatitis	6	6
Alcoholism (alcohol intake ≥150 g/day)	24	24
None, attributable to	69	70
Total	99	100

Hepatic cirrhosis with congenital metabolic disorders, cardiac cirrhosis, and biliary cirrhosis were excluded.

Fig. 1. The age and sex distribution of the patients with hepatic cirrhosis admitted to our department during 1972-1976. The shaded parts represent cases of the HLA type.

Table 2 shows the results of HLA typing of the patients with hepatic cirrhosis which had developed insidiously without a history of acute hepatitis or alcoholism. Out of 29 patients, 5 were positive for HBs antigen. Four patients (14%) had cirrhotic patients in the family. In 2 out of these 4 cases of positive family history of liver diseases, HBs antigen was positive and was thought responsible for the familial occurrence of hepatic cirrhosis.

The incidence of hepatic cirrhosis in the family of the HLA typed patients was not different from the incidence of liver diseases in the family of the patients with hepatic cirrhosis reported in the nation-wide epidemiological survey (11.3%) (Matsushita 1975). The incidence of hepatic cirrhosis among the family of the cirrhotic patients was thought high in comparison with the incidence of hepatic cirrhosis in general population in Japan. This high incidence of hepatic cirrhosis among the family may be explained by two factors, an environmental factor such as a viral infection within the family, and a genetic factor, i.e., susceptibility to hepatic cirrhosis. This study was done to clarify inheritable susceptibility to hepatic cirrhosis. In view of scanty evidence of homogeneous etiology in cases of the HLA typed hepatic cirrhosis, it was astonishing to find that 21 out of 29 patients with hepatic cirrhosis (72%) were positive for HLA phenotype B5, and 11 (38%)

TABLE 2. *Data of the patients with hepatic cirrhosis*

No.	Name	Age	Sex	Abnormal GTT	HBs antigen	Cirrhosis in the family	Diabetes in the family	HLA locus	
								A	B
1	H.K.	55	F	+	-	-	-	2, 9	5, W16
2	M.I.	43	M	+	-	-	-	2, 9	5, W22-J
3	T.On.	47	M	+	+	-	-	9	5
4	T.Ok.	70	F	+	-	-	-	9, 11	5
5	Y.S.	51	M	+	-	-	-	9, W19	5, W35
6	M.T.	52	M	+	-	-	-	9	5, W22-J
7	M.M.	40	M	+	-	+	+	9, 11	5, 8
8	F.W.	42	M	+	-	-	-	2, 9	5, W35
9	T.T.	44	M	+	-	-	-	2, 10	W40, W22-J
10	N.T.	58	M	+	+	-	-	9, 10	W16, W40
11	H.Y.	66	F	+	-	-	-	9, W19	5, W22-J
12	K.Su.	56	F	-	-	-	+	2, W19	W15, W22-J
13	G.M.	66	M	-	-	-	-	9	7, W22-J
14	K.N.	41	M	-	-	-	-	2, 9	W15, W22-J
15	T.S.	39	M	-	-	-	-	10, 11	5, W22-J
16	F.T.	63	M	-	-	-	-	9, W19	12, W15
17	T.Oo.	73	M	-	+	-	-	9	5
18	T.K.	55	F	-	-	-	-	9	5
19	T.M.	37	M	-	-	-	-	2, 10	5, W15
20	S.I.	61	M	-	-	-	-	9, 10	5, W40
21	S.Oz.	44	M	-	-	-	-	2	5
22	S.Oo.	45	M	-	-	-	-	10, W19	W15, W40
23	N.S.	59	M	-	-	-	-	9	5, W22-J
24	K.K.	27	M	-	+	+	-	9, W19	5, W16
25	K.Se.	36	F	-	-	-	-	9, 10	5, W40
26	A.S.	63	F	-	-	-	-	2	5, W40
27	K.O.	40	M	-	+	+	-	9	5, W22-J
28	I.T.	52	F	-	-	+	-	9, 11	5, W22-J
29	T.S.	63	M	-	-	-	-	9	W15, W35

Glucose tolerance test (GTT) was diagnosed abnormal when the blood glucose at 60 min after oral administration of 100 g glucose was more than 180 mg/100 ml, the blood glucose at 120 min more than 160 mg/100 ml, and the ratio of the increase of whole blood insulin (IRI) in the first 30 min after glucose administration to the increase of blood glucose was less than 0.4.

positive for HLA phenotype BW22-J. In the control group, 41 out of 128 (32%) were positive for B5, and 13 (10%) for BW22-J. The differences in the frequencies of B5 and BW22-J between the patients and the controls were statistically significant ($p < 0.003$, and relative risk=5.57, for B5: $p < 0.01$, and relative risk=5.40, for BW 22-J) as shown in Table 3.

DISCUSSION

Occurrence of the HLA antigens in such a high frequency in the cases which were considered as random samples from the patients with hepatic cirrhosis of unknown etiology strongly suggests that a predisposition linked to HLA associated genes plays an important role in development of hepatic cirrhosis of otherwise unknown etiology in Japan. Knowledge about the way of intervention of HLA associated genes in the development of hepatic cirrhosis in these patients is not

TABLE 3. Comparison between the patients with hepatic cirrhosis of unknown etiology and the healthy controls with regard to the frequency of HLA phenotypes in the first (A) and the second (B) loci

HLA	Cirrhosis (n=29)		Control (n=128)		$\chi^2_{(1)}$	p_{cor}	RR
	+	%	+	%			
1st locus							
A 1	0	0	3	2.3		n.s.	
A 2	9	31.0	49	38.3		n.s.	
A 9	22	75.9	83	64.8		n.s.	
A10	7	24.1	15	11.7		n.s.	
A11	4	13.8	21	16.4		n.s.	
AW19	6	20.7	25	19.5		n.s.	
2nd locus							
B 5	21	72.4	41	32.0	14.49	<0.003	5.57
B 7	1	3.4	21	16.4		n.s.	
B 8	1	3.4	0	0		n.s.	
B12	1	3.4	16	12.5		n.s.	
B18	0	0	0	0		n.s.	
BW15	6	20.7	15	11.7		n.s.	
BW16	3	10.3	8	6.3		n.s.	
BW17	0	0	1	0.8		n.s.	
BW22							
BW22-1	0	0	5	3.9		n.s.	
BW22-J	11	37.9	13	10.2	12.02	<0.01	5.40
BW35	3	10.3	24	18.8		n.s.	
BW40	6	20.7	46	35.9		n.s.	

p_{cor} , p value corrected for the number of antigens investigated ($\times 18$); RR, relative risk.

only interesting but also indispensable for prevention and treatment of hepatic cirrhosis. As for BW22-J, Ohtawa et al. (1974) reported association of this HLA specificity with thromboangiitis obliterans. According to Okimoto et al. (1977), BW22-J associates also with the juvenile-onset type of diabetes, but not with the adult-onset type of diabetes in Japanese (unpublished work). In our series of hepatic cirrhosis, 38% of the cases were accompanied with abnormal oral glucose tolerance test (GTT). But, the difference in the frequency of BW22-J between the patients with abnormal GTT and the patients without it was not statistically significant. The frequency of BW22-J is rare among Caucasians as mentioned before (Juji et al. 1975). Instead of BW22-J, B8, the frequency of which is reported to be rare among Japanese (Mittal et al. 1973; Yasuda et al. 1976) has been reported to associate with the juvenile-onset type of diabetes in Caucasians (Nerup et al. 1974; Thomsen et al. 1975). B8 has also been reported to associate with several autoimmune diseases such as myasthenia gravis (Pirskanen et al. 1972; Feltkamp et al. 1974), Graves' disease (Grumet et al. 1973) as well as autoimmune chronic hepatitis in Caucasians (Freudenberg et al. 1973; Galbraith et al. 1974; Lindberg et al. 1975; Freudenberg et al. 1976). But, from the age and sex distribution of the patients, it seems difficult to assume that most of these HLA typed patients suffered from autoimmune chronic hepatitis described by Mackay et al. (1965). LE cells were negative for all the cases investigated, and anti-thyroid antibody were

negative for most of these cases.

Up to date, only one attempt to find association of certain specificities of HLA with chronic liver diseases was reported in Japan (Ito et al. 1975). In that paper, hepatic cirrhosis was not described, and 15 cases of chronic active hepatitis and 8 cases of autoimmune chronic hepatitis in accord with the criteria of Mackay et al. (1965) were reported not to associate with any specificities of HLA. These results suggest that ethnic difference between Japanese and Caucasians must be taken into account in study of susceptibility to liver diseases in relation to HLA antigens. The way of intervention of BW22-J associated genes in the development of hepatic cirrhosis in Japanese seems to be similar to, but probably not the same as, that of B8 associated genes in the development of hepatic cirrhosis in Caucasians. The problem remains to be clarified.

B5 is one of the commonest HLA specificities in Japanese, and also common in Caucasians. Association of polycystic kidney (Dausset and Hors 1975), Behçet's disease (Ohno et al. 1973), and Buerger's disease (McLoughlin et al. 1976) with B5 were reported. From the consideration of these reports and our findings, the predisposition linked to B5 associated genes seems to intervene with the development of hepatic cirrhosis in ways different from the predisposition linked to BW22-J associated genes. In other words, the HLA typed patients in this study, and probably the majority of the patients with hepatic cirrhosis of apparently unknown etiology in Japan were thought to have a predisposition linked to HLA associated genes and divided into probably two different groups in terms of the way of intervention of HLA associated genes in the development of hepatic cirrhosis. Anyway, a substantial part of the patients with hepatic cirrhosis in Japan are expected to bear a predisposition linked to HLA associated genes.

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