HLA-DRw Antigens in Mexican-American and Black-American Diabetic Patients

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SUMMARY

HLA-A, -B, and -C antigens were studied in 67 Mexican-American and 38 black-American diabetic patients who had the onset of their disease before age 31 yr. Control populations consisted of 322 Mexican-American and 367 black-American subjects for HLA-A, -B, and -C antigens. In addition, HLA-DRw antigens were studied in 60 Mexican-American and 34 black-American diabetic patients. Control populations for HLA-DRw antigens consisted of 189 Mexican-American and 145 black-American subjects.

We found that juvenile-onset–diabetic patients of Mexican-American origin who had the onset of their disease before age 19 demonstrated a significant increase in HLA-DRw4. HLA-DRw4 was also significantly increased in black-American patients with juvenile-onset diabetes mellitus. HLA-DRw2 was not detected in any patient with juvenile-onset diabetes in either ethnic group. A significant association was found between HLA-B18 and HLA-DRw3 in Mexican-American juvenile-diabetic patients.

These findings, which are comparable to those in similar Caucasian patients, provide additional information to support the hypothesis that HLA-DRw antigens play a major role in determining the susceptibility to juvenile-onset diabetes mellitus. DIABETES 29:247–250, April 1980.

The genes that code for the histocompatibility complex of man occupy four separate loci on the short arm of chromosome 6.[1] Several alleles have been recognized for each locus. This human gene complex appears to be analogous to the H2 locus of the mouse, an area referred to as the immune region, which influences immunologic responses to a single synthetic antigen.[2] Additional evidence has accumulated to imply that the immune response and the susceptibility of laboratory animals to experimentally induced diseases such as autoimmune thyroiditis[3] and allergic encephalomyelitis[4] may be under the control of genes linked with the histocompatibility complex. These genes are thought to control the immune response and are termed Ir genes. Their products are referred to as immune region–associated, or Ia, antigens.[5]

The HLA-DRw locus is part of the human histocompatibility complex and is distinct from other HLA loci (A, B, C).[1] This locus has seven alleles, which are termed DRw1–DRw7.[6] The gene products of these alleles have been serologically detected on B lymphocytes, macrophages, sperm, and epithelial cells.[7–9] The HLA-A, -B, and -C loci code for glycoproteins, which are also serologically detectable by cytotoxicity assay methods.[10] The D locus has also been described, and the cell surface antigens coded at this locus have been detected by mixed lymphocyte culture (MLC) techniques.[11] There is controversy as to whether the DRw locus and D locus are the same or whether they represent separate genes coding for distinct cell surface antigens.[12]

Recent studies have clearly demonstrated an association between insulin-dependent diabetes mellitus and the HLA antigens B8, B15, B18, and Cw3 in Caucasians[13–18] and Bw54 in Japanese.[19] In Caucasian diabetic patients, a combination of B8 and B15 or B8 and B18 has been found to increase the susceptibility to the disease.[20] Juvenile-onset diabetes mellitus has also been found to be strongly associated with HLA-DRw3 and -DRw4.[12,21] In sharp contrast, the frequency of DRw2 is decreased or absent in Caucasian patients with juvenile-onset diabetes mellitus.[22]

Since HLA frequencies vary significantly with ethnic background, it was of interest to study the association of HLA-A, -B, -C, and -DRw antigens in diabetic patients of Mexican-American and black-American origins.

PATIENTS AND METHODS

Sixty-seven unrelated Mexican-American and thirty-eight unrelated black-American patients whose diabetes began

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before age 31 yr were studied for HLA-A, -B, and -C antigens. Sixty Mexican-Americans and thirty-four black-Americans were also studied for HLA-DRw antigens. The diabetic patients, all of whom received insulin, were divided into two groups. Group 1 consisted of juvenile-onset, insulin-dependent, ketosis-prone diabetic patients whose disease began before age 19; none were overweight. Group 2 consisted of insulin-treated patients whose diabetes began between ages 19 and 31; they were not prone to ketosis and they were more than 20% above their ideal weight. Control populations consisted of 332 Mexican-Americans for HLA-A, -B, and -C and 189 Mexican-Americans for HLA-DRw. Control populations consisted of 367 black-Americans for HLA-A, -B, and -C and 145 black-Americans for HLA-DRw.

HLA-A, -B, and -C antigens were determined on peripheral blood lymphocytes by the microdroplet lymphocyte cytotoxicity test.[23] The following HLA antigens were determined: HLA-A1, -A2, -A3, -A11, -A25 (10), -A26 (10), -A28, -A29, -Aw23 (9), -Aw24 (9), -Aw30, -Aw31, -Aw33, -Aw34, and -Aw36; HLA-B7, -B8, -B13, -B14, -B15, -B17, -B18, -B27, -B37, -B40, -Bw22, -Bw35, -Bw38, -Bw39 (16), -Bw44 (12), -Bw45 (12), -Bw49 (21), -Bw50 (21), -Bw51 (5), -Bw52 (5), -Bw53, and -Bw54 (22); and HLA-Cwl, -Cw2, -Cw3, and -Cw4.

The HLA-DRw antigen typing (DRw 1–5 and DRw7) was performed on B-cell–enriched lymphocyte populations by microtoxicity testing as previously described.[10]

Statistical analysis. Chi-square tests for significance and relative risk for the disease were calculated as described by Svejgaard et al.[24–25]

RESULTS

The frequencies of HLA-A, -B, and -C antigens among Mexican-American patients in groups 1 and 2 were compared with those found in a comparable control population. The frequencies of HLA-B and -C antigens in diabetic patients, which were found to deviate from those in control subjects, are shown in Table 1. Among the patients in group 1, the frequency of HLA-B18 was higher than it was in the control population but the difference was not statistically significant. Among patients in group 2, the frequency of HLA-B7 and -B8 was greater than it was in control subjects, but neither of these increases was statistically significant. Several HLA antigens were present in decreased frequency in diabetic patients. Among the diabetics in group 1, the frequencies of HLA-B14, -B40, and -Cw4 were decreased in comparison with control subjects. Among patients in group 2, the frequencies of HLA-B14, -Cw2, and -Cw4 were all decreased when compared with those of controls: None of these decreased frequencies in HLA antigens were statistically significant. There were no notable deviations in the frequency of any of the other HLA-A, -B, or -C antigens studied, when diabetic patients were compared to the control population.

TABLE 1
Frequency of HLA-B and -C antigens in Mexican-American control subjects and diabetic patients

	Controls N = 332	Group 1 N = 33			Group 2 N = 34		
Antigen	%	%	χ^2	P*	%	χ^2	P*
B7	8	8	0.02	NS	18	2.10	NS
B8	6	9	0.02	NS	18	4.25	NS
B14	11	6	0.43	NS	3	1.53	NS
B18	6	21	8.01	NS	6	0.10	NS
B40	21	9	1.91	NS	26	0.30	NS
CW2	8	12	0.15	NS	0	3.00	NS
CW4	31	15	2.80	NS	21	1.00	NS

* Corrected for number of antigens.

The frequency of HLA-DRw antigens in Mexican-American diabetic patients and the control population is shown in Table 2. The incidence of DRw4 was significantly increased among patients in group 1 as compared with normal control subjects ($X^2 = 9.49$; $P = 0.016$). A moderate increase in the frequency of DRw3 was also noted among patients in group 1; this difference was not statistically significant, however. No Mexican-American diabetic patient in group 1 demonstrated the presence of the HLA-DRw2 antigen. As shown in the table, the relative risk to develop diabetes in a Mexican-American who was positive for both DRw3 (RR = 2.013) and DRw4 (RR = 3.693) was increased, while the relative risk for diabetes was extremely small in Mexican-Americans positive for HLA-DRw2.

When the frequencies of HLA-A, -B, and -C antigens in black-American diabetic patients were compared to those in the appropriate control population, no notable differences were found. However, as shown in Table 3, there was a significant increase in HLA-DRw4 in group 1 black-American diabetic patients when they were compared with the suitable control population ($X^2 = 30.6$; $P = 0.0001$). There was also an increase in the frequency of DRw3 among patients in group 1, but this increase was not statistically significant. Again, HLA-DRw2 was not detected in any diabetic patient in group. 1. The relative risk of developing diabetes before age 19 was markedly increased in black-Americans positive for HLA-DRw4 (RR = 14.238) and extremely low for black-Americans positive for HLA-DRw2. In black-Americans there was also an increased risk of developing diabetes between the ages of 19 and 30 in individuals positive for HLA-DRw3 (RR = 3.560) or DRw4 (RR = 4.333).

Our data were analyzed for the presence of an association between all HLA-A, -B, -C and -DRw antigens studied. As shown in Table 4, there was an association between HLA-Bw35 and HLA-Cw4 in both Mexican-American and black-American diabetic subjects. However, this association is no different from that found in control populations of both ethnic groups. A significant association between HLA-B18 and HLA-DRw3 was also found in Mexican-American diabetic patients in group 1, which was notably different from any association between these antigens among either patients in group 2 or Mexican-American control subjects. Black-American patients in group 1 also showed an association between these two antigens, but the degree of association was not statistically significant. This association was not present in either black-American diabetic patients in group 2 or the appropriate control population. There was no association between HLA-B18 and HLA-DRw4 in either Mexican-American or black-American diabetic patients.

DISCUSSION

We have studied the frequencies of HLA-A, -B, -C, and -DRw antigens in Mexican-American and black-American diabetic patients and in control subjects of comparable ethnic background. We did not find an increased frequency of

TABLE 2
Frequency of HLA-DRw antigens in Mexican-American control subjects and diabetic patients

	Controls N = 189	Group 1 N = 30				Group 2 N = 30			
Antigen	%	%	χ^2	P*	RR†	%	χ^2	P*	RR†
DRw1	20	10	0.99	NS	0.518	7	2.10	NS	0.375
DRw2	15	0	3.88	NS	0.092	17	0.06	NS	1.209
DRw3	20	33	1.89	NS	2.013	23	0.03	NS	1.254
DRw4	38	70	9.49	0.016	3.693	33	0.08	NS	0.836
DRw5	15	3	2.08	NS	0.285	7	0.86	NS	0.492
DRw7	14	3	1.72	NS	0.312	13	0.00	NS	1.043

* Corrected for number of antigens.
† Relative risk.

HLA-B8, -B15, or -Cw3 in Mexican-American diabetics, as has been demonstrated in comparable Caucasian patients. This finding suggests the possibility that there is an ethnic difference in the HLA antigens associated with the development of insulin-dependent diabetes mellitus, particularly in patients who have the onset of their disease before age 19.

We were unable to demonstrate any differences in the frequency of HLA-A, -B, or -C antigens in black-American diabetic patients and in controls. This may also represent an ethnic difference in the HLA antigens associated with the development of diabetes mellitus or may be the result of the relatively small number of black-American patients studied. HLA typing is currently being carried out in larger groups of both black-American and Mexican-American diabetic patients.

The relative frequencies of HLA-DRw antigens in Mexican-American diabetic patients have not been reported previously, and only a preliminary report of the association between HLA-DRw antigens and diabetes in black-American patients has appeared.[26] Our data demonstrate a significant increase in the frequency of DRw4 in Mexican-American juvenile-onset–diabetic patients. The frequency of HLA-DRw3 was also increased in this group of patients, but the difference from control subjects was not statistically significant. HLA-DRw2 was completely absent in this group of young diabetics. These findings are in agreement with those reported for Caucasians with juvenile-onset diabetes mellitus by Farid et al.[12] and by Solow et al.[21] Black-American diabetics with an early onset of their disease also showed a significant increase of HLA-DRw4 and, again, DRw2 was not detected in any juvenile-onset black-American diabetic patient. We were unable to demonstrate a significant increase in the frequency of HLA-DRw3 in young black-American diabetics, probably because of the small number of patients studied. Our findings in black-Americans with diabetes mellitus are similar to those recently reported in preliminary form.[26]

Our results confirm the known association between HLA-Bw35 and HLA-Cw4 in both Mexican-American and black-American populations. This same association was detected in diabetic patients of both ethnic groups. An unexpected association between HLA-B18 and HLA-DRw3 was found in Mexican-American juvenile-onset–diabetic patients that was significantly different from the association of these antigens in older diabetics or in Mexican-American controls. A similar association, which did not reach the level of statistical significance, was found in young diabetics of black-American origin. We did not observe an association between HLA-B18 and HLA-DRw4, as has been noted in Caucasian diabetics.

Our findings add considerable support to the previously postulated importance of HLA-DRw antigens in relationship to the development of juvenile-onset diabetes mellitus. In Mexican-American and black-American populations, as well as in Caucasians, the presence of HLA-DRw4 and/or HLA-DRw3 may confer a susceptibility to the development of diabetes mellitus while the presence of HLA-DRw2 may in some way reduce the susceptibility. In all ethnic populations studied thus far, the presence of HLA-DRw4 and DRw3 and the absence of DRw2 appear to be important markers for the development of insulin-dependent diabetes mellitus.

Of particular importance in further elucidating these rela-

TABLE 3
Frequency of HLA-DRw antigens in black-American control subjects and diabetic patients

	Controls N = 145	Group 1 N = 21				Group 2 N = 13			
Antigen	%	%	χ^2	P*	RR†	%	χ^2	P*	RR†
DRw1	15	14	0.01	NS	1.047	8	0.10	NS	0.664
DRw2	36	0	9.26	0.016	0.042	8	3.02	NS	0.216
DRw3	30	38	0.24	NS	1.450	62	4.02	NS	3.560
DRw4	7	52	30.60	0.0001	14.238	23	2.30	NS	4.333
DRw5	24	5	2.95	NS	0.230	23	0.07	NS	1.047
DRw7	11	19	0.50	NS	2.034	23	0.01	NS	0.949

* Corrected for number of antigens.
† Relative risk.

TABLE 4
Association between HLA-B, -C, and HLA-DRw antigens in normal control subjects and diabetic patients of black-American and Mexican-American origin*

		Mexican			Black		
		Controls N = 142	Group 1 N = 30	Group 2 N = 30	Controls N = 178	Group 1 N = 22	Group 2 N = 13
HLA-BW35	HLA-CW4	0.85	0.64	0.60	0.73	0.89	0.54
HLA-B18	HLA-DRw3	0.15	0.71†	0.10	0.08	0.13	0.001
HLA-B18	HLA-DRw4	−0.03	−0.22	0.26	−0.05	−0.22	0.00

* The data are expressed as correlation coefficients.
† $P < 0.01$ when compared with control and with patients in group 1.

tionships are family studies of the distribution of HLA antigens in diabetic and nondiabetic family members. These studies are currently being pursued in both Mexican-American and black-American patients.

ADDENDUM

After our paper was submitted for publication, Rodey et al. reported data comparable to ours with regard to HLA-DRw antigens in black-American diabetic patients.[27]

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