# Immunogenetic and Clinical Characterization of Slowly Progressive IDDM

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**OBJECTIVE** — To examine the clinical and immunogenetic heterogeneity of IDDM.

**RESEARCH DESIGN AND METHODS** — We divided 207 IDDM patients into groups based on the interval from clinical onset to initiation of insulin therapy: group A (<3 mo, acute clinical-onset group, $n$ = 134), group B (3–12 mo, intermediate group, $n$ = 31), and group C (≥13 mo, slowly progressive group, $n$ = 42). Immunogenetic and clinical markers were compared between group A and group C.

**RESULTS** — The mode age of onset was higher in group C (52 yr) than group A (10 yr). Group C had a higher prevalence of islet cell antibodies (42.9%, 18 of 42) than group A (25.4%, 34 of 134, $P$ = 0.05). Serum C-peptide immunoreactivity assayed by radioimmunoassay in response to a 100-g oral glucose tolerance test was significantly higher in group C than in group A. Group C patients were also more likely to have a family history of NIDDM (26.1%, 11 of 42) among their first-degree relatives than group A patients (11.2%, 15 of 134, $P$ = 0.039). The prevalences of family history of IDDM and endocrine autoimmune diseases were not different between groups C and A. The frequency of complications of endocrine autoimmune disease was not different between group A (6.7%, 9 of 134) and group C (2.3%, 1 of 42). Significant associations with two class I major histocompatibility complex antigens (HLA-A24 and -Bw54) and one class II antigen (HLA-DR4) were observed in group A. Group A patients were assocciated with three diabetogenic HLA-DQ haplotypes including DQA1*0301–DQB1*0401, DQA1*0301–DQB1*0302, and DQA1*0301–DQB1*0303. In contrast, group C lacked the association with class I antigens, although HLA-DR4 and HLA-DQA1*0301–DQB1*0401 were more common in this group than in control subjects.

**CONCLUSIONS** — These results indicate that the clinical subtype with slowly progressive course (slowly progressive IDDM) has distinct findings including late-age onset, high prevalence of islet cell antibodies, preserved β-cell function, and high family history of NIDDM. An additive effect of class I and class II major histocompatibility complex antigens is suggested as an explanation for the acute clinical manifestations and more severe β-cell destruction in group A patients.

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IDDM, INSULIN-DEPENDENT DIABETES MELLITUS; ICA, ISLET CELL ANTIBODIES; CPR, C-PEPTIDE IMMUNOREACTIVITY; RIA, RADIOIMMUNOASSAY; NIDDM, NON-INSULIN-DEPENDENT DIABETES MELLITUS; EAD, ENDOCRINE AUTOIMMUNE DISEASE; MHC, MAJOR HISTOCOMPATIBILITY COMPLEX; HLA, HUMAN LEUKOCYTE ANTIGEN; OGTT, ORAL GLUCOSE TOLERANCE TEST; TMA, THYROID MICROSOMAL ANTIBODY; BW, BODY WEIGHT; HPLC, HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY; JDF U, JUVENILE DIABETES FOUNDATION UNIT; CV, COEFFICIENT OF VARIATION; PCR, POLYMERASE CHAIN REACTION; RFLP, RESTRICTION FRAGMENT-LENGTH POLYMORPHISM; BMI, BODY-MASS INDEX; HP, HAPTOGLOBIN PHENOTYPE.

Several prospective studies demonstrated that the natural history of IDDM is characterized by a heterogeneous clinical course before and after the clinical onset. Some cases are preceded by an $ICA^+$ subclinical prediabetic period, and these patients later develop acute clinical β-cell failure with classical symptoms of IDDM (1–3). In these cases, the persistence of ICA is usually short. Other patients are characterized by persistently positive ICA and slowly progressive deterioration of β-cell function through a non–insulin-dependent stage to ultimately an insulin-dependent state (4,5). This form of diabetes is clinically classified as NIDDM during the period when insulin is not required. Furthermore, some patients with fluctuating or transient ICA positivity have been reported, and β-cell function improved or did not deteriorate after ICA converted to negative (5–8). The specific clinical course of IDDM may be related to the immunogenetic background, including HLA (5,9), sex (10), race (9,11,12), and other predisposing factor(s) (9). However, scant knowledge is available regarding the differences in immunogenetic backgrounds and other clinical findings between acute-onset IDDM and slowly progressive IDDM.

In this study, possible heterogeneity in immunogenetic and clinical aspects was examined by comparing the findings between acute clinical-onset and slowly progressive IDDM in a large Japanese diabetic population.

## RESEARCH DESIGN AND METHODS

— We studied 207 IDDM patients (103 males and 104 females; mean ± SE age at onset, 26 ± 1 yr [range 2–62 yr]; mean ± SE duration of diabetes, 100 ± 7 mo [range 0–564 mo]). Of 207 patients, 178 (86%) were recruited from a primary-care base. The remaining 29 patients were referred from other institutions, and 18 of these 29 patients were seen within 3 mo of onset. All subjects were of Japanese ethnic stock

and were residents of the Tokyo and Yokohama areas. The diagnosis of IDDM was based on the criteria of the National Diabetes Data Group (13), with the following additional requirements; daily urinary CPR excretion <6.6 nmol and an integrated serum CPR value during the 100-g OGTT (ΣCPR) <3.3 nM. These two C-peptide criteria for distinguishing between IDDM and NIDDM were established on the basis of previous reports (5,14,15). The 207 subjects were recruited from the IDDM patients attending the outpatient clinic for diabetes of Toranomon Hospital from December 1980 through July 1989. The 100-g OGTT and blood sampling for blood glucose, CPR, ICA, $HbA_{1c}$, and TMA were conducted from 1987 to 1989. The rate of recruitment of IDDM patients in this study was 90% (207 of 230). No heavy alcohol users were among the subjects. All 207 IDDM patients were ketosis prone, and the blood glucose levels were unstable. Of the 207 IDDM patients, 12 (5.8%) had other EADs (16): 11 (5.3%) had Graves' disease and 1 (0.5%) had Hashimoto's thyroiditis. All patients underwent a 100-g OGTT and had blood glucose and serum CPR levels determined after a 12-h overnight fast. All subjects were on a diet containing >250 g of carbohydrate/day before OGTT. Blood samples were also drawn for ICA and TMA. Patients were also measured for daily urinary CPR excretion.

A family history of diabetes in first-degree relatives and a family history of EADs were collected for each patient at interview and/or by questionnaire. The questionnaire on family history included the following items: *1*) Do you have any family members with diabetes? If so, what kind of treatment do or did they have? 2) Do you have any family members with disease(s) of the thyroid (i.e, swollen thyroid gland), adrenals, pituitary gland, or other hormonal glands? If so, please specify.

The subjects were divided into three groups based on the duration of diabetes before insulin therapy. In group A (acute-onset IDDM), the duration of diabetes before insulin therapy was <3 mo; in group B (intermediate group), the duration before insulin was 3–12 mo; and in group C (slowly progressive IDDM), the duration before insulin was ≥13 mo. This division of the subjects was based on our prospective observation of the progression of β-cell failure in $ICA^+$ NIDDM (5,10), which showed that all 17 patients with ICA and progressive β-cell failure required insulin ≥13 mo (range 13–45 mo, mean 27 mo) after onset. Data on the duration of illness before insulin therapy were collected from the medical records ($n$ = 195) or at interview (group A, $n$ = 6; group B, $n$ = 3; group C, $n$ = 3). The data on age at onset, fasting blood glucose before insulin therapy, daily dose of insulin, maximum BW, BW at onset, the presence of complicating EAD, and the absence of other disorders including infection, hepatic disease, renal disease, and pregnancy before the initiation of insulin were obtained from medical records. The medical records used an itemized fill-in format. Classification and subdivision of the subjects were conducted independently by three investigators. The three groups were compared for age at onset, sex, fasting blood glucose level before insulin therapy, dose of insulin, prevalence of ICA, TMA, past maximum BW, BW at onset, the prevalence of complicated EADs, family history of diabetes in first-degree relatives of the patient, family history of EAD in first-degree relatives, prevalence of HLA, and serum CPR response to 100-g OGTT.

### ICA, TMA, and serum and urinary CPR and $HbA_{1c}$

Detection of ICA was performed by the indirect immunofluorescence method as described previously (5,17). The quality of the assay was class A (cutoff point, 5 JDF U; sensitivity, 90%; specificity, 92% [18]). Our laboratory participated in the second through fifth international workshops on the standardization of ICA assays. Serum CPR was measured by a sensitive RIA as described elsewhere (19). The intra-assay CV of the CPR assay at CPR concentrations of 0.760 and 0.056 nM were, respectively, 3.4 and 9.5%. The interassay CV values at CPR concentrations of 0.760 and 0.056 nM were, respectively, 8.1 and 14.2%. With respect to 24-h urine collection for the CPR assay, only the samples with an increased creatinine level (males: 1600 μmol [18 mg]/kg BW; females: 884 μmol [10 mg]/kg BW) were subjected to measurement. $HbA_{1c}$ levels were measured by HPLC (normal range: 4.8–6.3%). The intra-assay and interassay CVs of $HbA_{1c}$ were 0.9 and 1.2%, respectively. TMA were tested by the Microsome Test (Fujizoki, Tokyo). TMA tests were considered positive when the titer was >1:240.

### HLA typing

**HLA-DQ typing.** Typing of HLA-DQA1 and HLA-DQB1 alleles was performed by the previously described PCR–RFLP method (20–23). The sequences of the primers used were as follows.

For DQA1 typing:

GH26 5′ GTGCTGCAGGTGTAAACTTGT ACCAG 3′ (20)

GH27 5′ CACGGATCCGGTAGCAGCGGTA GAGTTC 3′ (20)

For DQB1 (DQw1) typing:

P1bis 5′ TGTGCTACTTCACCAACGGG 3′ (22)

DQ202 5′ CAGGGCAGATCCCGCGGTACG CCACCTC 3′ (23)

For DQB1 (DQw 2, 3, and 4) typing:

P1bis 5′ TGTGCTACTTCACCAACGGG 3′ (22)

DQ204 5′ CACCTGCAGTGCGGAGCTCCA ACTGGTA 3′ (23)

Typing for these alleles was carried out in 146 randomly selected IDDM patients and 90 normal control subjects.

**HLA typing for A, B, C, and DR antigens.** HLA typing for the A, B, C, and DR antigens was performed in 190 randomly selected IDDM patients by a standard microcytotoxicity test (24). The 203 normal control subjects, who were all of Japanese ethnic stock and lived in the Tokyo and Yokohama area, were typed for HLA.

**Table 1—*Clinical characteristics of subjects divided by duration of diabetes before insulin treatment***

| | GROUP A | GROUP B | GROUP C |
|---|---|---|---|
| (*n*) | 134 | 31 | 42 |
| DURATION OF DIABETES (MO) | 99 ± 9 | 110 ± 21 | 96 ± 14 |
| SEX (M/F) | 67/67 | 12/19 | 24/18 |
| AGE AT ONSET (YR) | 21.6 ± 1.2* | 28.3 ± 3.0† | 38.0 ± 2.4‡ |
| | (2–60) | (5–62) | (6–62) |
| FASTING BLOOD GLUCOSE BEFORE INSULIN (MM) | 31.2 ± 3.2§ | 20.0 ± 2.5† | 13.3 ± 0.6‖ |
| | (12.4–71.7) | (15.7–28.1) | (10.4–20.8) |
| PEAK CPR (NM) | 0.17 ± 0.02 | 0.17 ± 0.03 | 0.26 ± 0.02¶ |
| 24-H URINE CPR (NMOL) | 4.3 ± 0.7 | 4.4 ± 0.9 | 5.4 ± 0.5¶ |
| DAILY DOSE OF INSULIN (U/KG BW) | 0.71 ± 0.08 | 0.69 ± 0.09 | 0.70 ± 0.09 |
| $HbA_{1c}$ (%) | 10.3 ± 0.2 | 10.2 ± 0.3 | 9.6 ± 0.2¶ |
| TMA (%) | 19.4 (26/134) | 9.7 (3/31) | 30.9 (13/42)# |
| OTHER EAD (%) | 6.7 (9/134) | 6.4 (2/31) | 2.3 (1/42)[a] |
| HISTORY OF DIABETES IN FIRST-DEGREE RELATIVES (%) | 18.7 (25/134) | 19.4 (6/31) | 38.0 (16/42)[a] |
| IDDM | 7.5 (10/134) | 6.4 (2/31) | 11.9 (5/42) |
| NIDDM | 11.2 (15/134) | 12.9 (4/31) | 26.1 (11/42)[b] |
| FAMILY HISTORY OF EAD (%) | 3.0 (4/134) | 3.2 (1/31) | 9.5 (4/42) |

Data are means ± SE.
*$P < 0.005$ vs. group C.
†$P < 0.05$ vs. group A.
‡$P < 0.01$ vs. group B.
§$P < 0.01$ vs. group C.
‖$P < 0.05$ vs. group B.
¶$P < 0.01$ vs. group A and group B.
#$P = 0.054$ (2-sided), 0.027 (1-sided) vs. group B.
[a]$P = 0.02$ vs. group A.
[b]$P = 0.04$ vs. group A.

### Statistical analysis

The Mann-Whitney *U* test was applied to compare values between the different groups and the Kolmogorov-Smirnov test was used to evaluate the differences of distributions. Fisher's exact test was used to compare the prevalence of ICA, TMA, family history, and HLA between different groups. The correction was accomplished by multiplying the value for *P* by the number of HLA antigens tested. The data were expressed as means ± SE.

## RESULTS

### Overall backgrounds of the IDDM subjects

The age at onset of diabetes ranged from 2 to 62 yr with a mode of 10 yr. Eighty-eight (43%) patients had diabetes onset at ≥30 yr of age. A difference in the male/female ratio was observed between the patients who developed IDDM at <30 yr of age (49/70) and those who developed IDDM at ≥30 yr of age (53/35, $P < 0.01$). There was no apparent seasonality in diabetes onset.

### Comparison of the clinical and immunogenetic characteristics

#### Subdivided subjects

No difference was noted in the duration of diabetes among the three groups. Single fasting blood glucose values before insulin treatment in group A were significantly higher than in group C (Table 1). In group C patients, insulin was started 45 ± 15 mo (13–300 mo) after the diagnosis. At the time of this investigation, all subjects were ketosis prone (>4 episodes of ketonuria and/or ketoacidosis; mean: 9 episodes), and their fasting blood glucose levels measured at the outpatient clinic were unstable. No difference was observed in the daily insulin dose between groups A and C (Table 1). $HbA_{1c}$ values in group C were significantly lower than those in groups A and B.

#### BW, sex ratio, and age distribution

No significant differences were noted in maximum BW expressed as BMI or in the BMI at onset between groups A and C. No differences in BMI by sex within or among groups were observed (maximum BMI, males vs. females: group A, 24.0 ± 0.2 vs. 23.4 ± 0.2, NS; group B, 24.8 ± 0.6 vs. 25.9 ± 0.6, NS; group C, 24.3 ± 0.6 vs. 25.1 ± 0.6, NS. BMI at onset, males vs. females: group A, 20.5 ± 0.3 vs. 19.1 ± 0.3, NS; group B, 20.1 ± 0.6 vs. 19.6 ± 0.6, NS; group C, 21.3 ± 0.8 vs. 19.5 ± 0.7, NS). No differences were found in sex ratio between groups A, B, and C. The mean age at diabetes onset was significantly higher in group C than in group A. The modal age of onset in group A was 10 yr (Fig. 1*A*), and in group C it was 52 yr. The age distribution in group C was different from that in group A ($P < 0.01$ by the Kolmogorov-Smirnov test; Fig. 1*c*).

#### TMA and family history

The prevalence of TMA was 20.3% (42 of 207). The overall prevalence of family history of diabetes in first-degree relatives of the subjects was 18.7% in group A and 38.0% in group C ($P = 0.02$). The prevalence of family history of NIDDM in group C was significantly higher than in group A (Table 1). A family history of IDDM or other EADs was not significantly different between group A (4 Graves' disease) and group C (3 Graves' disease and 1 Hashimoto's thyroiditis).

To examine the possibility that older patients were more likely to have a

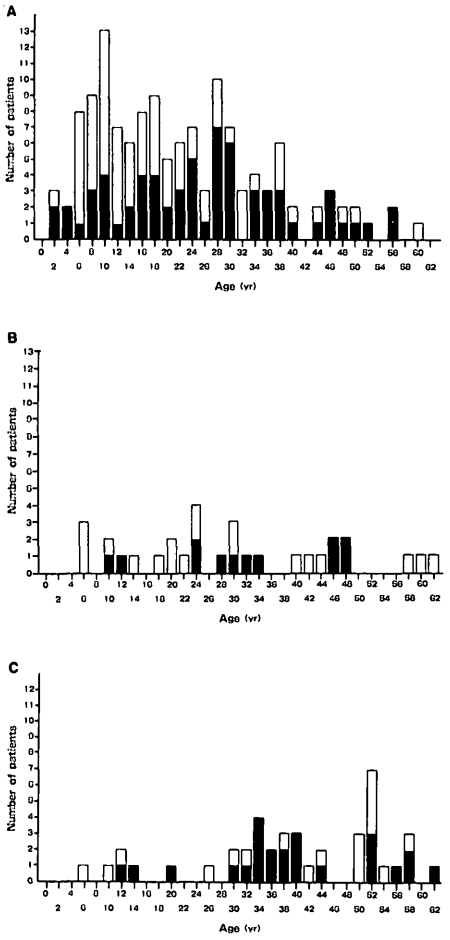


**Figure 1**—*Age distribution in groups A (A), B (B), and C (C). The period before insulin was <3 mo in group A, 3–12 mo in group B, and ≥13 mo in group C. (□), female; (■), male.*

Table 2—*Prevalence of ICA with duration of diabetes in groups A, B, and C*

| | Duration of diabetes (mo) | | | |
|---|---|---|---|---|
| | 0–12 | 13–59 | ≥60 | Overall prevalence |
| Group A (%) | 54 (20/37) | 32 (7/22) | 9 (7/75) | 25 (34/134) |
| Group B (%) | 50 (4/8) | 40 (2/5) | 6 (1/18) | 23 (7/31) |
| Group C (%) | | 52 (11/21) | 33 (7/21)* | 43 (18/42)† |

*$P$ = 0.023, group A vs. group C.
†$P$ = 0.05, group A vs. group C.

## ICA

The overall prevalence of ICA was 28.5% (59 of 207). The prevalences of ICA in group C was higher than that in group A (Table 2).

## Serum and 24-h urinary CPR

Serum CPR response represented by integrated value of serum CPR levels at six sampling points during OGTT. The 0, 30-, 60-, 90-, 120-, and 180-min and peak values were significantly higher in group C than in groups A and B (Tables 1 and 3). No significant difference was observed in CPR by sex in group A or group C. The 24-h urinary CPR value in group C was higher than in groups A or B (Table 1).

## HLA-A, -B, -C and -DR

HLA-A24 was significantly more common in group A than in normal control subjects (Table 4), and HLA-B52 was significantly less common. HLA-Bw54 was also increased in group A but was insignificant in group C compared with normal control subjects. HLA-Cw1 was also increased in group A, but insignificant in group C. HLA-DR2 was significantly lower in both groups A and C than in the normal control subjects (Table 5). HLA-DR4 was significantly higher in both groups A and C (Table 5). The relative risk of a heterozygous combination of HLA-DR4/DR9 was not increased in either group A or group C.

## DQA1 and DQB1

The prevalence of DQA1*0301 was significantly increased in all three groups of patients (Table 6). DQA1*0103 was significantly decreased in all three groups. DQB1*0302, DQB1*0303, and DQB1*0401 were significantly increased in group

family history of diabetes, the incidence of a positive family history among the patients >40 yr of age was compared between groups A and C. The mean ± SE age of the patients >40 yr of age was 51.4 ± 1.4 yr in group A ($n$ = 45) and 53.5 ± 1.2 yr in group C ($n$ = 27, NS). The incidence of a positive family history of NIDDM in group A patients >40 yr of age was 9% (4 of 45), while it was 33% in group C (9 of 27, $P$ = 0.03 vs. group A). In addition, the incidence of a family history of IDDM in groups A and C patients >40 yr of age was 4% (2 of 45 and 1 of 27; NS).

Table 3—*Integrated serum C-peptide values during 100-g OGTT in terms of duration of diabetes in groups A, B, and C*

| | Duration of diabetes (mo) | | | |
|---|---|---|---|---|
| | 0–12 | 13–59 | ≥60 | Total |
| Group A (nM) | 1.1 ± 0.2 | 0.8 ± 0.2 | 0.9 ± 0.1 | 0.9 ± 0.1 (0–2.6) |
| $n$ | 37 | 22 | 75 | 134 |
| Group B (nM) | 0.9 ± 0.4 | 1.7 ± 0.8 | 0.7 ± 0.2* | 0.9 ± 0.2† (0–3.2) |
| $n$ | 8 | 5 | 18 | 31 |
| Group C (nM) | | 1.1 ± 0.3 | 1.6 ± 0.2‡ | 1.4 ± 0.1‡ (0–2.4) |
| $n$ | | 21 | 21 | 42 |

Data are means ± SE.
*$P$ < 0.01 group B vs. group C.
†$P$ < 0.05 group B vs. group C.
‡$P$ < 0.01 group A vs. group C.

Table 4—*HLA-A and -B antigen frequencies in groups A, B, and C and normal control subjects*

| Antigens | Group A ($n$ = 122) | Group B ($n$ = 31) | Group C ($n$ = 37) | Control ($n$ = 203) | Relative risk | | | $P$ value vs. control | | |
|---|---|---|---|---|---|---|---|---|---|---|
| HLA | $n$ (%) | $n$ (%) | $n$ (%) | $n$ (%) | Group A | Group B | Group C | Group A | Group B | Group C |
| A1 | 1 (1) | 0 (0) | 0 (0) | 1 (0) | | | | | | |
| A2 | 51 (42) | 15 (48) | 20 (54) | 74 (36) | 1.25 | 1.63 | 1.95 | NS | NS | NS |
| A3 | 1 (1) | 0 (0) | 0 (0) | 1 (0) | | | | | | |
| A11 | 17 (14) | 4 (13) | 6 (17) | 36 (18) | | | | | | |
| A24 | 86 (70) | 21 (68) | 21 (57) | 114 (56) | 1.87 | 1.64 | 1.02 | 0.01 | NS | NS |
| A26 | 21 (17) | 6 (19) | 12 (33) | 48 (24) | 0.67 | 0.78 | 1.61 | NS | NS | NS |
| A31 | 11 (9) | 1 (4) | 3 (8) | 25 (12) | | | | | | |
| A33 | 17 (14) | 6 (19) | 6 (17) | 28 (14) | | | | | | |
| B5 | 1 (1) | 0 (0) | 1 (3) | 0 (0) | | | | | | |
| B7 | 18 (15) | 6 (19) | 3 (8) | 10 (10) | | | | | | |
| B8 | 0 (0) | 0 (0) | 0 (0) | 1 (0) | | | | | | |
| B13 | 1 (1) | 0 (0) | 1 (3) | 3 (1) | | | | | | |
| B15 | 14 (11) | 2 (6) | 6 (15) | 33 (14) | | | | | | |
| B16 | 3 (2) | 0 (0) | 2 (6) | 1 (0) | | | | | | |
| B27 | 1 (1) | 0 (0) | 0 (0) | 1 (0) | | | | | | |
| B35 | 19 (15) | 1 (4) | 6 (17) | 22 (11) | | | | | | |
| B37 | 0 (0) | 0 (0) | 0 (0) | 0 (0) | | | | | | |
| B39 | 1 (1) | 0 (0) | 4 (11) | 16 (8) | | | | | | |
| B40 | 3 (2) | 0 (0) | 0 (0) | 0 (0) | | | | | | |
| Bw42 | 0 (0) | 0 (0) | 0 (0) | 0 (0) | | | | | | |
| B44 | 16 (13) | 5 (16) | 6 (17) | 27 (13) | | | | | | |
| Bw48 | 6 (5) | 0 (0) | 2 (6) | 7 (3) | | | | | | |
| B51 | 12 (10) | 0 (0) | 2 (6) | 26 (13) | | | | | | |
| Bw52 | 10 (8) | 1 (4) | 4 (11) | 45 (22) | 0.31 | 0.11 | 0.44 | 0.001* | 0.01 | NS |
| Bw54 | 50 (40) | 9 (29) | 9 (25) | 36 (18) | 3.13 | 1.90 | 1.72 | $0.001 \times 10^{-2}$† | NS | NS |
| Bw56 | 2 (2) | 0 (0) | 0 (0) | 2 (1) | | | | | | |
| Bw58 | 0 (0) | 0 (0) | 0 (0) | 1 (0) | | | | | | |
| Bw59 | 4 (3) | 1 (4) | 0 (0) | 3 (1) | | | | | | |
| Bw60 | 11 (9) | 7 (23) | 6 (17) | 29 (14) | | | | | | |
| Bw61 | 33 (27) | 1 (4) | 6 (17) | 50 (25) | | | | | | |

Relative risk was calculated as follows: (a) × (d)/(b) × (c), where a = patients positive for antigen, b = patients negative for antigen, c = control subjects positive for antigen, and d = control subjects negative for antigen.
*Corrected $P < 0.05$.
†Corrected $P < 0.01$.

A, whereas only DQB1*0401 was increased in group C. DQB1*0601 was significantly decreased in all three groups.

### DQA1-DQB1 haplotypes

In group A, three haplotypes (DQA1*0301–DQB1*0401, DQA1*0301–DQB1*0302, and DQA1*0301–DQB1*0303) showed an increased prevalence, whereas only DQA1*0301–DQB1*0401 was significantly increased in group C (Table 7). DQA1*0103–DQB1*0601 was decreased in all three groups.

**CONCLUSIONS**— Scant knowledge is available on the natural history of ICA positivity among patients with NIDDM despite the large number of such patients (~10% of all Japanese NIDDM patients; 25). Recently, we reported that some NIDDM patients with persistent ICA and/or insulin autoantibodies progressed gradually to insulin dependency over several years (5,10,26). This slowly progressive clinical course is one of the characteristics observed in some Japanese IDDM patients (11,25). A comparison of the immunogenetic and clinical findings in these slowly progressive patients with those with typical acute-onset IDDM may help identify some of the pathogenetic factors contributing to this clinical subtype of IDDM.

In this clinic-based study, 20% of Japanese IDDM patients had a history of slowly progressive β-cell failure for >13 mo before the initiation of insulin treatment. Slowly progressive IDDM (group C) was characterized by a late age of onset and a different age distribution

Table 5—*HLA-C and -DR antigen frequencies in groups A, B, and C and normal control subjects*

| | Group A ($n$ = 122) | Group B ($n$ = 31) | Group C ($n$ = 37) | Control ($n$ = 203) | Relative risk | | | $P$ value vs. control | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Antigens | $n$ (%) | $n$ (%) | $n$ (%) | $n$ (%) | Group A | Group B | Group C | Group A | Group B | Group C |
| Cw1 | 66 (54) | 14 (45) | 14 (39) | 70 (34) | 2.20 | 1.56 | 1.30 | 0.004* | NS | NS |
| Cw2 | 0 (0) | 0 (0) | 0 (0) | 0 (0) | | | | | | |
| Cw3 | 48 (39) | 11 (36) | 17 (47) | 102 (50) | 0.63 | 0.54 | 0.89 | NS | NS | NS |
| Cw4 | 9 (7) | 4 (12) | 0 (0) | 20 (10) | | | | | | |
| Cw5 | 0 (0) | 0 (0) | 0 (0) | 0 (0) | | | | | | |
| Cw7 | 23 (19) | 7 (23) | 10 (28) | 43 (21) | | | | | | |
| Cw9 | 0 (0) | 0 (0) | 1 (3) | 0 (0) | | | | | | |
| Cw11 | 0 (0) | 0 (0) | 0 (0) | 0 (0) | | | | | | |
| DR1 | 11 (9) | 4 (12) | 2 (6) | 26 (13) | | | | | | |
| DR2 | 13 (10) | 1 (4) | 4 (11) | 68 (33) | 0.23 | 0.07 | 0.25 | $0.003 \times 10^{-3}$† | 0.0003† | 0.006* |
| DR3 | 0 (0) | 0 (0) | 0 (0) | 0 (0) | | | | | | |
| DR4 | 88 (71) | 21 (68) | 23 (62) | 81 (40) | 3.68 | 3.16 | 2.47 | $0.002 \times 10^{-15}$† | 0.007 | 0.02 |
| DR7 | 1 (1) | 1 (4) | 1 (3) | 0 (0) | | | | | | |
| DR8 | 20 (16) | 5 (16) | 6 (17) | 40 (20) | | | | | | |
| DR9 | 47 (38) | 11 (36) | 11 (31) | 63 (31) | 1.36 | 1.22 | 0.98 | | | |
| DR10 | 0 (0) | 0 (0) | 0 (0) | 1 (0) | | | | | | |
| DR4/DR9 | 22 (18) | 6 (19) | 4 (11) | 15 (7) | 2.76 | 3.00 | 1.52 | 0.007 | 0.029 | NS |

*Corrected $P < 0.05$.
†Corrected $P < 0.01$.

than acute-onset IDDM as well as a higher prevalence of ICA, longer persistence of β-cell function, and a more common family history of NIDDM compared with acute-onset patients. An elaborate population-based study may be required to further characterize these cases.

A striking male predominance was observed among IDDM patients >30 yr of age. Some IDDM patients in group C had a long duration of diabetes before insulin therapy. Male predominance was previously reported in $ICA^+$ NIDDM (10), and these findings suggest that maleness has some influence on slowly progressive β-cell failure through hormonal or immunological processes. Male predominance of IDDM has also been documented in Caucasians (27) and experimental animals (28), but the finding is not consistent.

The mechanisms underlying the different clinical courses of groups A and C remain speculative. Group C patients had a higher average age of onset than those in group A. Different clinical features based on the age at onset were reported (29). Adult-onset IDDM is characterized by a longer period before diagnosis and better preservation of residual β-cell function (29). The different β-cell mass in adults and children may possibly explain the different latencies before insulin treatment.

Genetic factors related to HLA may explain the differences in the severity of β-cell damage between groups A and C. Our haplotype analysis of DQA1–DQB1 provided some evidence of genetic differences between acute-onset IDDM and slowly progressive IDDM: group A patients featured three diabetogenic haplotypes (DQA1*0301–DQB1*0401, DQA1*0301–DQB1*0302, and DQA1*0301–DQB1*0303), whereas only DQA1*0301–DQB1*0401 was increased in group C (Tables 6 and 7). Thus, the difference in β-cell damage between groups A and C may perhaps be explained by the relative dose of DQ-α and -β heterodimers encoded by the DQA1 and DQB1 genes regulating the rate of β-cell destruction as well as susceptibility to IDDM (30). Group A patients also had a significant increase of HLA-A24, HLA-Bw54, HLA-Cw1, and HLA-DR4, as well as a decrease of HLA-Bw52 and HLA-DR2, which are characteristic findings in Japanese IDDM patients (17). Note that group C lacked any class I (HLA-A24, HLA-Bw54, or HLA-Cw1) association, whereas group A had such an association. A study on residual β-cell function in IDDM has demonstrated that a high proportion of IDDM patients with no β-cell function had a highest relative risk of 20 for HLA-A24 (31). In contrast, IDDM patients with a long duration, who had preserved β-cell function, did not have a significant association with HLA-A24 (31). The class I MHC is possibly associated with β-cell destruction in an additive manner with class II antigens in experimental animals (32).

One of the characteristics of group C patients was a high frequency of NIDDM in first-degree relatives, espe-

**Table 6—*Frequencies of HLA-DQA1 and -DQB1 alleles in groups A, B, and C and normal control subjects***

| | Group A ($n$ = 88) | Group B ($n$ = 26) | Group C ($n$ = 32) | Control ($n$ = 90) | Relative risk | | | $P$ value | | |
|---|---|---|---|---|---|---|---|---|---|---|
| HLA alleles | $n$ (%) | $n$ (%) | $n$ (%) | $n$ (%) | Group A | Group B | Group C | Group A | Group B | Group C |
| DQA1*0101/*0102 | 22 (25) | 16 (62) | 11 (34) | 46 (51) | | | | | | |
| DQA1*0103 | 2 (2) | 0 (0) | 6 (19) | 36 (42) | 0.07 | 0.03 | 0.34 | $1 \times 10^{-9}$ | $4 \times 10^{-12}$ | 0.02* |
| DQA1*0201 | 0 (0) | 0 (0) | 0 (0) | 2 (2) | | | | | | |
| DQA1*0301 | 84 (95) | 24 (92) | 30 (94) | 56 (62) | 12.75 | 7.29 | 9.11 | $2.8 \times 10^{-9}$ | 0.028 | 0.0049 |
| DQA1*0401 | 2 (2) | 2 (8) | 1 (3) | 2 (2) | | | | | | |
| DQA1*0501 | 4 (5) | 0 (0) | 1 (3) | 14 (16) | 0.26 | 0.10 | 0.18 | NS | 0.02* | NS |
| DQA1*0601 | 4 (5) | 0 (0) | 0 (0) | 4 (4) | | | | | | |
| DQB1*0501/*0502 | 2 (2) | 1 (4) | 2 (3) | 8 (9) | | | | | | |
| DQB1*0503 | 2 (2) | 0 (0) | 1 (3) | 10 (11) | | | | | | |
| DQB1*0601 | 2 (2) | 0 (0) | 2 (6) | 40 (44) | 0.03 | 0.02 | 0.08 | $4 \times 10^{-11}$ | $5 \times 10^{-5}$ | $6 \times 10^{-4}$ |
| DQB1*0602 | 4 (5) | 0 (0) | 1 (3) | 10 (11) | | | | | | |
| DQB1*0604 | 12 (14) | 11 (42) | 12 (38) | 15 (17) | | | | | | |
| DQB1*0201 | 0 (0) | 0 (0) | 0 (0) | 0 (0) | | | | | | |
| DQB1*0301 | 6 (7) | 0 (0) | 4 (13) | 17 (19) | | | | | | |
| DQB1*0302 | 28 (32) | 4 (15) | 6 (19) | 12 (14) | 3.03 | 1.18 | 1.50 | 0.028 | NS | NS |
| DQB1*0303 | 38 (43) | 11 (42) | 7 (22) | 20 (23) | 2.66 | 2.57 | 0.98 | 0.024 | NS | NS |
| DQB1*0401 | 41 (48) | 9 (35) | 17 (53) | 20 (23) | 3.05 | 3.50 | 3.97 | 0.006 | 0.020 | 0.015 |
| DQB1*0402 | 6 (7) | 4 (15) | 1 (3) | 6 (7) | | | | | | |

*Uncorrected $P$ value.

cially in their parents. A misclassification of the NIDDM patients into group C is unlikely, because these group C patients had a high (42.9%) prevalence of ICA despite their long duration of diabetes and a low CPR. Recently, we found that up to 23% of the parents of IDDM probands had NIDDM, and some of these NIDDM parents had ICA and progressed slowly to IDDM (33). Thus, some of the parents with NIDDM documented in the present survey probably had IDDM in the pre–insulin-dependence stage. An association between IDDM probands and NIDDM parents

**Table 7—*DQA1-DQB1 haplotype frequencies in groups A, B, and C and normal control subjects***

| | Group A ($n$ = 88) | Group B ($n$ = 26) | Group C ($n$ = 32) | Control ($n$ = 90) | Relative risk | | | $P$ value | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Haplotype | $n$ (%) | $n$ (%) | $n$ (%) | $n$ (%) | Group A | Group B | Group C | Group A | Group B | Group C |
| DQA1*0103–DQB1*0601 | 2 (2) | 1 (4) | 1 (2) | 26 (29) | 0.06 | 0.09 | 0.08 | $0.007 \times 10^{-3}$ | 0.009 | 0.002 |
| DQA1*0102–DQB1*0602 | 4 (5) | 0 (0) | 1 (3) | 5 (6) | | | | | | |
| DQA1*0103–DQB1*0603 | 0 (0) | 0 (0) | 0 (0) | 1 (1) | | | | | | |
| DQA1*0102–DQB1*0604 | 12 (14) | 10 (38) | 7 (22) | 9 (10) | | | | | | |
| DQA1*0201–DQB1*0201 | 0 (0) | 0 (0) | 0 (0) | 0 (0) | | | | | | |
| DQA1*0501–DQB1*0201 | 0 (0) | 0 (0) | 0 (0) | 0 (0) | | | | | | |
| DQA1*0301–DQB1*0301 | 2 (2) | 0 (0) | 2 (3) | 1 (1) | | | | | | |
| DQA1*0501–DQB1*0301 | 2 (2) | 0 (0) | 1 (3) | 9 (10) | | | | | | |
| DQA1*0601–DQB1*0301 | 0 (0) | 0 (0) | 0 (0) | 4 (4) | | | | | | |
| DQA1*0301–DQB1*0302 | 27 (31) | 3 (12) | 6 (19) | 7 (8) | 5.2 | 1.5 | 2.7 | 0.0018 | NS | NS |
| DQA1*0301–DQB1*0303 | 39 (44) | 11 (42) | 7 (22) | 19 (21) | 3.0 | 2.7 | 1.0 | 0.016 | NS | NS |
| DQA1*0301–DQB1*0401 | 43 (49) | 10 (38) | 15 (47) | 21 (23) | 3.1 | 2.1 | 2.9 | 0.0064* | NS | 0.025* |

*Uncorrected $P$ value.

was reported as evidence of the heterogeneity of IDDM by Chern et al. (34). A high incidence of a family history of diabetes in $ICA^+$ late-onset diabetic patients was also reported by Groop et al. (35). The possibility cannot be ruled out that other genetic factor(s) contribute to the high prevalence of a family history of NIDDM in group C. We surveyed Hp, a putative genetic marker of NIDDM (36), and found that Hp 2–1 was significantly increased in the NIDDM parents of IDDM probands (37). This result suggests that a subgroup of NIDDM gene(s) may contribute to the development of IDDM as an additive risk factor (38).

In a Caucasian population, the familial occurrence of other EADs with IDDM has been reported (39). However, no significant difference was noted in the frequency of such diseases, a family history of such diseases, or the presence of TMA in our previous study (11). A weak genetic predisposition to autoimmunity in the Japanese population may explain this result.

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