Antibodies to Islet 37k Antigen, But Not to Glutamate Decarboxylase, Discriminate Rapid Progression to IDDM in Endocrine Autoimmunity

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Apart from islet cell antibodies (ICAs), antibodies to glutamate decarboxylase (GAD), insulin autoantibodies (IAAs), and a novel islet antigen (37k antigen) are potential markers for insulin-dependent diabetes mellitus (IDDM). GAD is also an antigen in stiff-man syndrome (SMS), and both SMS and IDDM are associated with ICAs and autoimmunity to other endocrine organs. We investigated possible links between antibody responses to islet antigens with autoimmunity to other endocrine organs and determined which specific antibodies can identify individuals who progress to IDDM. Antibodies to GAD were detected in ≥90% of both diabetic and nondiabetic patients with ICAs and other endocrine autoimmunity, in 59% of ICA-positive IDDM patients without endocrine autoimmunity, in all patients with SMS, but in only 1–3% of healthy (nondiabetic) and autoimmune disease control subjects. GAD antibody levels were increased in ICA-positive IDDM patients with polyendocrine autoimmunity compared with those without. In contrast, antibodies to 37k antigen were only detected in patients who developed acute-onset IDDM. IAAs were also associated with IDDM. Thus, certain factors enhance antibody responses to GAD in polyendocrine autoimmunity, but this does not necessarily lead to development of IDDM or SMS. Antibodies to 37k antigen are strongly associated with acute-onset IDDM and are useful serological markers for disease. *Diabetes* 43:1254–1259, 1994

Insulin-dependent diabetes mellitus (IDDM) is an autoimmune disease first found to be associated with islet cell antibodies (ICAs) (1). More recently, antibodies to glutamic acid decarboxylase (GAD) have been detected in the majority of IDDM patients at and before onset (2–7), and T-cell reactivity against GAD has been reported in the peripheral blood of IDDM patients (8). However, GAD autoantibodies are not restricted to IDDM but are also detected in patients with stiff-man syndrome (SMS), a rare neurological disorder affecting the GABA-ergic (γ-amino-*n*-butyric acid) nervous system (9,10). GAD autoimmunity in SMS is strongly associated with the presence of other endocrine/organ-specific autoimmune manifestations (10). Interestingly, we have also detected GAD antibodies in nondiabetic ICA$^+$ patients with autoimmunity to other endocrine organs (11). Common features of SMS and polyendocrine autoimmunity may point to possible links between autoimmune responses to GAD and to other endocrine organs. In both of these ICA$^+$ populations, which are predominantly female, IDDM develops in only ~30% of individuals, despite very high titers of ICAs (10,12).

IDDM is associated with autoimmunity to islet antigens other than GAD, including insulin (13), and a 64,000 M_r antigen that is distinct from GAD (14,15). Antibodies to the latter antigen (37k antigen) can be detected by measuring antibody binding to 37,000 and 40,000 M_r fragments generated by tryptic proteolysis of the 64,000 M_r antigen (14). Antibodies to the 37k antigen are more closely associated with progression to diabetes than are antibodies to GAD in identical twins of diabetic patients (16). In this study, we have investigated antibody responses to a variety of islet antigens in a cohort of patients with ICAs and other endocrine autoimmunity and have determined which of these antibodies identify individuals who progress to IDDM.

RESEARCH DESIGN AND METHODS

ICA$^+$ endocrine autoimmune patients. Endocrine autoimmune patients positive for ICA were selected from a large prospective study on the pathogenesis of IDDM (U.K. Polyendocrine Study) (12). The basis for recruitment was the detection of ICA on at least two occasions in the absence of IDDM and the presence of at least one other organ-specific antibody and/or organ-specific autoimmune disease. The project was initiated in 1985, and 186 individuals are currently enrolled. Participants have been followed prospectively for the development of IDDM. Two groups of patients were selected for this study. The first group included all patients in the U.K. Polyendocrine Study who progressed to IDDM and who had a serum sample available before, or <1 year after, disease diagnosis. Seventeen patients (11 women) met these criteria, and the earliest serum sample and a serum sample drawn close to diabetes onset were selected for antibody analysis. Of these 17 patients, 9 required insulin at diagnosis of diabetes, and 1 additional patient required insulin treatment within 4 months of diagnosis of non-insulin-dependent diabetes mellitus (NIDDM) (acute-onset diabetes). The mean age (± SE) at onset of IDDM for these patients was 51.0 ± 6.1 years (range 17–75). The remaining 7 patients (4 women) had NIDDM at initial presentation and did not require insulin for >1 year (range 15–48 months) before

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IDDM, insulin-dependent diabetes mellitus; ICA, islet cell antibody; GAD, glutamic acid decarboxylase; SMS, stiff-man syndrome; NIDDM, non-insulin-dependent diabetes mellitus; JDF U, Juvenile Diabetes Foundation units; SLE, systemic lupus erythematosus; IFL, immunofluorescence; IAA, insulin autoantibody; TG-Ab, antibodies to thyroglobulin; TM-Ab, antibodies to thyroid microsomes; GPC-Ab, antibodies to gastric parietal cells; GAD-Ab, antibodies to GAD; 50k-Ab, antibodies to the 50,000 M_r fragment of islet GAD_{65}; 37k-Ab, antibodies to 37,000 M_r tryptic fragments of islet 37k antigen.

TABLE 1
Frequencies of antibodies to GAD, antibodies to the 50,000 M_r fragment of islet GAD_{65}, antibodies to 37,000 M_r tryptic fragments of islet 37k antigen, IAA, TG-Ab, TM-Ab, and GPC-Ab, in ICA^+ subjects with and without IDDM

Group	*n*	GAD-Ab	50k-Ab	37k-Ab	IAA	TG-Ab	TM-Ab	GPC-Ab
ICA^+ endocrine autoimmune								
Non-IDDM	20	18 (90)	19 (95)	0	1 (5)	6 (30)	15 (75)	14 (70)
Slow-onset IDDM	7	7 (100)	7 (100)	0	2 (29)	2 (29)	5 (71)	6 (86)
Acute-onset IDDM	10	10 (100)	10 (100)	9 (90)*	4 (40)†	4 (40)	8 (80)	10 (100)
ICA^+ nonendocrine autoimmune								
IDDM	22	13 (59)	13 (59)	18 (82)	NT	0	0	0
ICA^+ SMS								
Non-IDDM	3	3 (100)	3 (100)	0	NT	0	1 (33)	2 (67)
IDDM	3	3 (100)	3 (100)	2 (67)	NT	1 (33)	3 (100)	1 (33)

Data are *n* (%). GAD-Ab, antibodies to GAD; 50k-Ab, antibodies to the 50,000 M_r fragment of islet GAD_{65}; 37k-Ab, antibodies to 37,000 M_r tryptic fragments of islet 37k antigen; NT, not tested. *Significant difference in antibody frequency compared with endocrine autoimmune, non-IDDM ($P < 0.0001$) and slow-onset IDDM ($P < 0.005$) subjects; †significant difference in antibody frequency compared with endocrine autoimmune, non-IDDM subjects ($P < 0.05$).

progression to IDDM (slow-onset diabetes). In these patients, the mean age at diagnosis of IDDM was 54.3 ± 8.3 years (range 13–79). The second group consisted of 20 patients (15 women) selected at random from participants in the U.K. Polyendocrine Study who had ICAs >10 Juvenile Diabetes Foundation units (JDF U) and who had not developed diabetes on follow-up for at least 36 months (mean 67.7 months, range 36–102).

ICA^+ IDDM patients. A group of 22 patients (9 women) with acute-onset IDDM were included in the study. The mean age of the patients at serum sample was 16.4 ± 1.9 years (range 5–40). Serum samples were obtained within 12 months of diagnosis of disease. These patients were selected for having ICAs but no detectable antibodies to thyroglobulin, thyroid microsomes, or gastric parietal cells.

SMS patients. Six patients (four women) with SMS were included in the study. The mean age at serum sampling was 53.8 ± 2.5 years (range 47–63). Three SMS patients also had IDDM. All SMS patients had high titer ICAs (>80 JDF U).

Control groups. The first group included 111 ICA^- nondiabetic patients who had other autoimmune diseases. Of these, 30 patients had autoimmune thyroid disease, 10 had Addison's disease, 10 had pernicious anemia, 10 had autoimmune polyendocrine disease, 27 had rheumatoid arthritis, and 24 had systemic lupus erythematosus (SLE). The second control group comprised 80 healthy individuals with no known family history of IDDM or other autoimmune diseases.

ICAs. These were measured by indirect immunofluorescence (IFL) on 4-mm cryostat sections of blood group O human pancreas, as described previously (17). Positive samples were titrated to end point in doubling dilutions in 10 mmol/l phosphate-buffered saline (pH 7.2). Local standard sera calibrated to 2, 4, 8, 16, 32, and 80 JDF U were included in each assay. End-point titers of test samples were converted to JDF U by comparison with a standard curve of $\log_2$ JDF U versus $\log_2$ of end-point titer of the standard sera. The threshold of ICA detection was 5 JDF U.

Insulin autoantibodies (IAAs). IAAs were measured by radioimmunoassay, with displacement with unlabeled insulin to correct for nonspecific binding (18). The mean (± SD) corrected binding for 140 control sera was 0.149 ± 0.298%. Sera with binding >3 SD above the mean for these control sera were considered positive. Since the introduction of the Immunology and Diabetes Workshops for the standardization of ICA and IAA, our laboratories have repeatedly participated with a high degree of performance.

Antibodies to GAD. These were measured by determining the enzyme activity immunoprecipitated by antibodies in sera from a soluble extract of rat brain as described previously (3). Serial dilutions of selected sera from each group demonstrated an approximately linear relationship between antibody concentration and enzyme activity immunoprecipitated for most samples. However, dilution experiments with all SMS patients demonstrated a marked prozone effect at low dilutions of serum. For quantification of GAD antibody levels, sera from SMS patients were analyzed at a dilution of 1:250 and all other sera at 1:5. Enzyme activity immunoprecipitated was calculated relative to that of the same standard antibody-positive control serum used in previous analyzes of GAD antibody activities (14,16). Sera were regarded as positive if the relative antibody activity exceeded 2 SD of the activity in sera from a group of 30 healthy (nondiabetic) control individuals (mean ± SD; 6.2 ± 3.4% of positive control). The interassay coefficient of variation was 15.4%.

Antibodies to 50,000, 40,000, and 37,000 M_r fragments of islet 64,000 M_r antigens. These antibodies were measured by immunoprecipitation of [^{35}S]methionine-labeled polypeptides released from particulate fractions of neonatal rat islets by trypsin treatment as described previously (14). Serum samples were regarded as positive for a specific antibody activity if a band corresponding to the appropriate polypeptide could be detected on the autoradiogram. Antibody levels were quantified by densitometric scanning of bands on autoradiograms, expressing band density as a proportion of a standard antibody-positive control serum included in each antibody analysis (14).

Other autoantibodies. Antibodies to thyroglobulin (TG-Ab) and thyroid microsomes (TM-Ab) were detected by indirect hemagglutination using a commercial kit (Serodia, Japan). The threshold titer for antibody positivity was 1/20 for TG-Ab and 1/400 for TM-Ab. Antibodies to gastric parietal cells (GPC-Ab) were detected by IFL using 4-mm cryostat sections of human blood group O stomach (19).

Statistical analysis. The significance of differences between antibody frequencies in populations was determined by χ^2 analysis with Yates' correction or by Fisher's exact test as appropriate. The significance of differences between antibody levels in patient groups was determined by the Mann-Whitney *U* test. The degree of correlation between antibody levels and titers was calculated as the Spearman coefficient of rank correlation.

RESULTS

Antibodies to brain GAD and 50,000 M_r fragments of islet GAD. Antibodies to GAD were measured by two independent assays: *1*) by measuring GAD enzyme activity immunoprecipitated from extracts of rat brain (GAD antibodies) and *2*) by immunoprecipitation of 50,000 M_r fragments of rat islet antigen, previously shown to be islet GAD (15) (50k antibodies). GAD and 50k antibodies were detected in 18 (90%) and 19 (95%), respectively, of 20 ICA^+ endocrine autoimmune patients without IDDM, in all 17 ICA^+ endocrine autoimmune patients who developed IDDM, and in all 6 patients with SMS (Table 1). The two ICA^+ endocrine autoimmune patients who were negative for GAD antibodies had low titer ICA (Table 2). When present, GAD antibodies persisted with time; no patients converted from antibody negative to positive, and the antibodies were detected 84 months before diabetes onset in one patient (patient 4 in Table 2) and persisted for at least 6 years in ICA^+ endocrine autoimmune patients who did not develop diabetes. GAD antibodies in the absence of antibodies to 37k antigen or IAAs were found in 23 ICA^+ polyendocrine disease patients; 17 of these are nondiabetic, 5 progressed to IDDM slowly after a period of NIDDM, and 1 developed acute-onset IDDM (Table 2). Of the 22 ICA^+ IDDM patients without endocrine autoimmunity, 13 (59%) had GAD antibodies and 13 (59%)

TABLE 2
Characteristics and antibody data for individual ICA$^+$ endocrine autoimmune patients

Diseases	Sex	Age (years)	Period to IDDM (months)	ICA (JDF U)	GAD-Ab	50k-Ab	37k-Ab	IAA	TG-Ab	TM-Ab	GPC-Ab
Acute-onset IDDM											
1. 1°M, Tx, IDDM	M	40	0	>80	417	++	++	−	>320	40^2	++
2. PA, Tx, IDDM	F	70	−1	49	391	++	+	−	0	0	++
3. PA, 1°M, IDDM	F	72	−2	>80	75	+	+	−	640	20^2	++
4. 1°M, PA, IDDM	M	58	0	>80	199	+	+	+	0	20^2	++
5. 1°M, Ad, IDDM	F	23	−29	30	149	++	+	+	40	20^2	++
6. IDDM	M	16	−15	52	70	+	+	+	0	0	+
7. Tx, IDDM	F	46	−49	35	416	++	−	−	0	20^2	++
8. H, IDDM	F	50	−6	69	41	+	+	+	320	320^2	+
9. PA, 1°M, IDDM	F	75	−2	20	340	++	+	−	0	160^2	++
10. Tx, IDDM	F	50	−33	49	150	++	+	−	0	320^2	+
Slow-onset IDDM											
11. V, TC, IDDM	F	57	−21	80	277	++	−	−	160	0	++
12. 1°M, PA, IDDM	M	63	5	>80	128	+	−	−	0	40^2	+
13. IDDM	F	40	3	23	32	+	−	−	0	20^2	+
14. IDDM	F	79	0	>10	31	++	−	+	0	80^2	+
15. GE, CM, IDDM	M	12	−18	>80	31	+	−	−	0	0	+
16. 1°M, IDDM	M	70	−9	10	29	+	−	+	160	20^2	−
17. H, IDDM	F	52	−40	>80	288	+	−	−	0	40^2	++
18. Tx	F	39		69	254	+	−	−	80	20^2	+
19. V	M	56		80	291	+	−	−	0	0	++
20. Ad	M	23		15	9	+	−	−	0	0	−
21. PA	M	53		>80	86	++	−	−	0	40^2	++
22. Ad, Tx, V, POF	F	48		>80	437	++	−	−	0	20^2	++
23. Tx	F	43		>80	226	+	−	−	0	80^2	++
24. V, H	F	49		>80	222	+	−	−	160	160^2	−
25. HPT, Ad	F	30		>80	188	+	−	−	0	0	+
26. None	F	53		>80	178	+	−	−	20	160^2	++
27. PA, Tx	F	35		>80	126	+	−	−	80	40^2	++
28. None	F	21		>80	149	+	−	−	0	0	++
29. POF	F	28		>80	248	+	−	−	0	160^2	+
30. None	F	35		>80	138	+	−	−	320	160^2	−
31. None	M	22		52	95	+	−	+	0	40^2	−
32. TC	F	22		>80	61	+	−	−	0	160^2	++
33. Tx	F	53		20	0	−	−	−	0	0	++
34. Tx	F	24		18	170	+	−	−	0	40^2	−
35. None	M	26		11	121	+	−	−	40	40^2	−
36. 1°M	F	80		18	375	+	−	−	0	40^2	+
37. None	F	62		>80	161	+	−	−	0	20^2	++

Tx, thyrotoxicosis; 1°M, primary myxedema; H, Hashimoto's disease; G, Graves' disease; TC, thyroid cyst; PA, pernicious anemia; Ad, Addison's disease; V, vitiligo; POF, premature ovarian failure; GE, gut enteropathy; CM, cardiomyopathy; HPT, hypoparathyroidism. Antibody data presented for diabetic patients are for samples collected closest to disease onset. Age given is at serum collection. The period from serum collection to development of IDDM is indicated; positive values indicate a postdiabetic sample. Quantitative antibody data are given for ICA (JDF U), GAD antibodies (% positive control) and antibodies to thyroglobulin and thyroid microsomes (reciprocal titers). Other antibodies are indicated as negative (−), positive (+), or strongly positive (++).

had 50k antibodies. Five of nine GAD antibody-negative IDDM patients had high titer ICAs (>80 JDF U), and GAD antibodies were not correlated with ICA titers in this group.

A quantitative analysis of GAD enzyme activity immunoprecipitated by antibodies in sera of ICA$^+$ patients is shown in Fig. 1. No significant difference was observed between GAD antibody levels in diabetic and nondiabetic ICA$^+$ endocrine autoimmune patients. However, GAD antibody levels in ICA$^+$ IDDM patients without endocrine autoimmunity were significantly lower than those in both diabetic and nondiabetic ICA$^+$ endocrine autoimmune groups ($P \leq 0.002$). SMS patients all had high GAD antibody levels (Fig. 1).

GAD antibodies were rarely detected in the ICA$^-$ groups. None of the 51 patients with rheumatoid arthritis or SLE were positive for GAD antibodies, and only 2 patients with endocrine autoimmune disease were positive, 1 with Graves' disease and 1 with Addison's disease and hypothyroidism (Table 3). The latter patient was also positive for 50k antibodies. Two of 80 (2.5%) normal control individuals were positive for antibodies to brain GAD and 50,000 M_r tryptic fragments of the islet antigen.

Antibodies to islet 37k antigen. In contrast to antibodies to GAD, antibodies to 37k antigen were only found in individuals who developed IDDM (Tables 1 and 2). Overall, 9 of 17 (53%) ICA$^+$ endocrine autoimmune patients who progressed to IDDM were positive for antibodies to 37k antigen. Nine of 10 patients (90%) with acute-onset IDDM had these antibodies, whereas all patients with slow-onset IDDM were negative ($P < 0.005$). Two patients with acute-onset IDDM converted from antibody negative to positive on follow-up, in one patient (patient 4) between 84 and 26 months before diabetes onset and in a second (patient 9) between 7 and 2 months before diagnosis. Of the six patients with SMS, two had antibodies to 37k antigen and both of them had IDDM. Antibodies to 37k antigen were detected in 18 of 22 (82%) ICA$^+$ IDDM patients without endocrine autoimmunity. ICA titers were significantly correlated with levels of antibodies to 37k antigen in these patients ($r = 0.71$, $P < 0.001$).

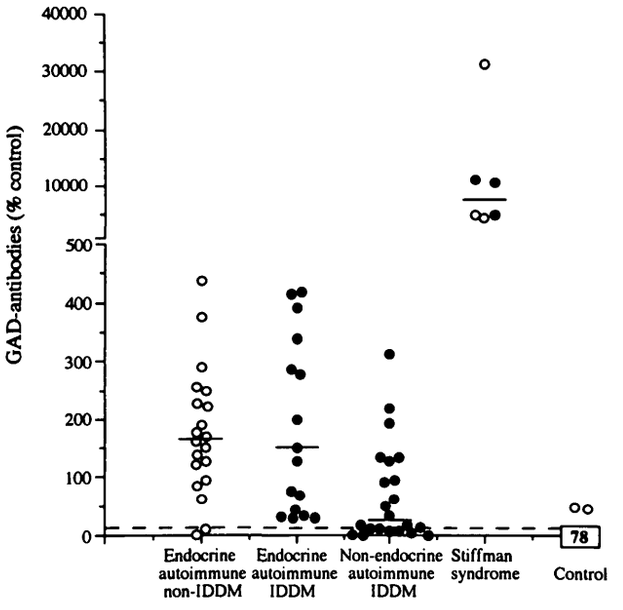

FIG. 1. Levels of antibodies to brain GAD in population groups with and without IDDM. Antibody levels are expressed as a proportion of those in a positive control serum. Note change of scale above 500% of control levels. ○, Nondiabetic subjects; ———, median values for each group; - - - -, cutoff for antibody positivity. The box indicates 78 control individuals negative for antibodies.

Antibodies to 37k antigen were not detected in any of the 111 ICA^- patients with autoimmune disease (Table 3) or in the 80 healthy control individuals.

Antibodies to insulin. IAAs were measured only in ICA^+ endocrine autoimmune patients (Tables 1 and 2); in patients who developed IDDM, IAAs were measured in samples collected before initiation of insulin treatment. IAAs were detected in 4 of 10 (40%) patients who developed acute-onset IDDM, in 2 of 7 (29%) slow-onset IDDM patients, and in 1 of 20 (5%) nondiabetic individuals. The frequency of IAAs in acute-onset IDDM patients was significantly higher than that in ICA^+ endocrine autoimmune patients who have not developed IDDM ($P < 0.05$).

Thyrogastric antibodies. ICA^+ endocrine autoimmune patients and SMS patients had high frequencies of TG-Ab, TM-Ab, and GPC-Ab (Tables 1 and 2). The frequency of thyrogastric antibodies in ICA^- patients was similar to that expected for each group of diseases tested (Table 3).

DISCUSSION

A major goal of research into IDDM is to define genetic and/or immunological markers that precisely identify individuals predisposed to the disease (20). GAD is one of a number of novel islet cell antigens known to be associated with IDDM. However, GAD autoimmunity also develops in the absence of diabetes. Thus, GAD is an autoantigen in autoimmune SMS, but only one-third of SMS patients develop IDDM. We have also detected GAD antibodies in ~20% of nondiabetic identical twins who are long-term discordant for diabetes with their diabetic co-twin (16) and who are therefore at low risk for diabetes (21). Furthermore, GAD has been shown to be a target for a β-cell–selective ICA that is not predictive of diabetes development, in contrast with whole-islet ICA, which is associated with disease (11,22).

To further characterize the associations of GAD autoimmunity with diabetes and other autoimmune diseases, we have analyzed GAD antibodies in ICA^+ and ICA^- patients with endocrine autoimmunity. GAD antibodies, including antibodies to 50,000 M_r fragments of islet GAD_{65} that correlate well with the former (3), were detected in almost all ICA^+ endocrine autoimmune patients, whether or not they developed diabetes. GAD antibody levels were detected at similarly high levels in both diabetic and nondiabetic groups. The elevated frequency of GAD antibodies in this cohort of endocrine autoimmune patients, together with the strong association of GAD autoimmunity in SMS with the presence of ICA and other organ-specific autoimmunities (10), suggested possible links between endocrine autoimmunity and immune responses to GAD. However, GAD antibodies appeared in <2% of patients with autoimmune disease who were negative for ICA. Interestingly, when detected, the antibodies appeared only in endocrine autoimmune patients and not in patients with non-organ-specific disease. GAD antibodies thus appear predominantly in the ICA^+ endocrine autoimmune population. It has been estimated that 2.4% of patients with autoimmune disease possess ICA (23) (and therefore most likely GAD antibodies), a proportion similar to the frequency of GAD antibodies in our healthy control subjects (2.5%). Thus, development of GAD antibodies is not strongly associated with endocrine autoimmunity per se, other than autoimmunity to pancreatic islet cells or GAD-rich cells in the brain.

Despite this lack of association, the levels of GAD antibodies in IDDM patients with endocrine autoimmunity were significantly higher than in those who lacked organ-specific antibodies. These results suggest that, although the appearance of antibodies to GAD may not be dependent on autoimmune responses to other organs, the intensity of the antibody response to the enzyme, when it does develop, is enhanced in patients with coexistent endocrine autoimmunity. This concept is supported by a previous study, in which high levels of GAD antibodies were predominantly detected in diabetic patients with other organ-specific antibodies (24). Our series concentrated mainly on adult patients with type II autoimmune polyglandular syndrome (Schmidt syndrome extended [25]), but similar high-titer GAD antibodies have

TABLE 3
Frequencies of antibodies in autoimmune disease patients negative for ICA

Disease	*n*	GAD-Ab	50k-Ab	37k-Ab	TG-Ab	TM-Ab	GPC-Ab
Autoimmune thyroid	30	1 (3)	0	0	13 (43)	24 (80)	3 (10)
Addison's disease	10	0	0	0	3 (30)	5 (50)	3 (30)
Pernicious anemia	10	0	0	0	0	5 (50)	7 (70)
Polyendocrine disease	10	1 (10)	1 (10)	0	7 (70)	9 (90)	6 (60)
Rheumatoid arthritis	27	0	0	0	4 (15)	8 (30)	6 (22)
SLE	24	0	0	0	4 (17)	7 (29)	3 (13)

Data are *n* (%).

also been described in type I autoimmune polyglandular syndrome, which primarily affects children (26).

Although possessing high levels of autoantibodies to GAD, only 30% of the ICA^+ endocrine autoimmune patients progress to IDDM (12). In another study (27), we had the opportunity to examine the pancreases of three such patients who died without developing IDDM. These pancreases displayed none of the immunohistological changes that are normally associated with autoimmune destruction of pancreatic β-cells in IDDM, suggesting that the strong antibody responses to GAD in these individuals occur independently of lymphocytic infiltration of islets, a potent indicator of β-cell destruction.

This study identifies a unique nondiabetic population, initially defined as possessing endocrine autoimmunity with ICA, with a high frequency (≥90%) and high levels of GAD antibodies, but which, in most cases, lacks other serological markers of IDDM. Is it possible that GAD antibodies, when they develop in the absence of other diabetes-associated immune markers, play a neutral, if not protective, role in determining susceptibility to IDDM? To try to answer this question, we investigated whether antibodies to insulin and 37k antigen could distinguish those GAD antibody-positive individuals who progressed to IDDM. Both markers were indeed associated with disease. IAAs were detected in 6 of 17 (35%) patients who progressed to diabetes but in only 1 of 20 (5%) nondiabetic patients. Antibodies to 37k antigen were only found in individuals who became diabetic, including two of three SMS patients with diabetes. More strikingly, antibodies to 37k antigen were detected in 90% of patients in our polyendocrine cohort who developed acute-onset IDDM, whereas all those in whom IDDM was preceded by a prolonged period of NIDDM were negative. Thus, antibodies to 37k antigen identify heterogeneity in the rate of progression to IDDM and are apparently markers of acute-onset diabetes in this population. In support of this, antibodies to 37k antigen were shown to be associated with rapid progression to IDDM in ICA^+ relatives of patients with IDDM (28).

Studies in patients with polyendocrinopathy have dissected two antibody specificities closely linked to IDDM development: whole islet ICAs and antibodies to 37k antigen. As previously shown in identical twins (16), levels of antibodies to 37k antigen were significantly correlated with ICA titers in the group of IDDM patients who lacked other organ-specific antibodies (and who tended to possess whole islet ICAs), suggesting that these antibodies might arise through common mechanisms. GAD antibodies, although disease-associated, appear in a wider population, only a subset of whom develop IDDM. Harrison et al. (29) have demonstrated an inverse correlation between antibody and T-cell responses to GAD in ICA^+ relatives of diabetic patients and have suggested that autoimmunity to GAD might deviate toward either humoral or cellular responses. Because IDDM is likely to be a T-cell–mediated disease, T-cell responses to GAD may be better indicators of progression to diabetes than are levels of antibodies to the antigen. Whether GAD antibodies in the absence of other diabetes-associated antibodies are markers of slow progression to diabetes remains to be substantiated.

The discovery of serological markers with high disease specificity offers the potential to identify individuals at risk for IDDM with increased accuracy. Among several now available, antibodies to 37k antigen seem to be closely associated with rapid progression to disease. Further characterization of the 37k antigen, including molecular cloning and expression of recombinant protein, should facilitate development of simpler screening assays for the detection of these antibodies. Large-scale screening studies in the general population are required to establish the true predictive value of different antibody specificities. With the availability of accurate markers for disease development, application of immune intervention protocols can be considered as a means of preventing IDDM.

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