A Longitudinal Study of GAD65 and ICA512 Autoantibodies During the Progression to Type 1 Diabetes in Diabetes Prevention Trial–Type 1 (DPT-1) Participants

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OBJECTIVE—We examined changes in GAD65 and ICA-512 autoantibodies (GADA and IA-2A) during progression to type 1 diabetes (T1D).

RESEARCH DESIGN AND METHODS—Diabetes Prevention Trial–Type 1 (DPT-1) participants were assessed for changes in positivity and titers of GADA and IA-2A during the progression to T1D.

RESULTS—Among 99 progressors to T1D with GADA and IA-2A measurements at baseline and diagnosis (mean interval = 3.3 ± 1.5 years), GADA positivity changed little and GADA titers decreased ($P < 0.01$). In contrast, both IA-2A positivity and titers increased substantially ($P < 0.001$). Even among those positive at baseline, IA-2A titers increased from baseline to diagnosis ($n = 57$; $P < 0.001$), whereas GADA titers decreased ($n = 80$; $P < 0.01$). The same patterns of change were also evident among those positive for both autoantibodies ($n = 48$) at baseline.

CONCLUSIONS—IA-2A titers increase during the years before the diagnosis of T1D, even among those positive for IA-2A. In contrast, GADA titers tend to decline during those years.

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Pancreatic autoantibodies are commonly present years before the diagnosis of type 1 diabetes (T1D) (1–4), and they tend to occur according to a certain sequence (5,6). Yet there is little known about changes in both autoantibody positivity and overall titers as the onset of T1D approaches. In the Diabetes Prevention Trial–Type 1 (DPT-1) (7,8), serial measurements of autoantibodies were obtained before diagnosis. These measurements, together with the large number of participants diagnosed with T1D in DPT-1, provided unique data for studying how autoantibody positivity and titers change over time with progression to T1D.

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RESEARCH DESIGN AND METHODS

Subjects

DPT-1 parenteral and oral insulin trial participants (7,8) were all islet cell autoantibody (ICA)-positive relatives of T1D patients. Autoantibodies to GAD65 (GADA) and to ICA512 (IA-2A) were measured along with ICA at baseline. Individuals included in the analyses were selected according to whether they also had autoantibody measurements at the time of diagnosis.

Clinic procedures

Two-hour oral glucose tolerance tests were performed every 6 months for diagnostic surveillance. The majority of individuals were diagnosed with T1D by oral glucose tolerance test criteria (fasting glucose ≥126 mg/dL and/or 2-h glucose ≥200 mg/dL) at a routine study visit. The others were diagnosed clinically. There was no overall effect of the intervention in either trial (7,8).

Laboratory measures

DPT-1 autoantibody procedures have been described previously (9). ICA was determined by an immunofluorescence assay on frozen sections of blood type O human pancreas in the DPT-1 ICA Core Laboratory (Gainesville, FL, February 1994 to September 1997 and January 1999 to October 2003; New Orleans, LA, September 1997 to January 1999). Combined GADA and IA-2A radioassays were performed at the Barbara Davis Center. Positive testing for ICA, GADA, and IA-2A was defined as ≥10 JDF units, ≥0.33, and ≥0.50, respectively. Although quantitative measurements of GADA and IA-2A were based on indexes, for simplicity we have characterized those measurements as "titers."

Data analysis

Student t tests and χ^2 tests were used for comparisons. McNemar tests were used

for intraindividual comparisons of categorical variables. The SAS 9.1.3 version was used for the analyses. All *P* values are two-sided. A *P* value <0.05 was considered statistically significant.

RESULTS—There were 99 progressors (mean ± SD age: 11.5 ± 8.3 years; sex: 65% male) who had measurements of both GADA and IA-2A at baseline (3.3 ± 1.5 years before diagnosis) and at diagnosis. A substantial percentage was positive for GADA at baseline, with a slightly smaller percentage positive at diagnosis (80 of 99 [81%] and 76 of 99 [77%], respectively; $P = 0.32$). In contrast with GADA, IA-2A positivity increased markedly from baseline to diagnosis (57 of 99 [58%] and 80 of 99 [81%], respectively; $P < 0.001$).

Figure 1 shows that GADA titers (Fig. 1*A*) declined ($P < 0.01$), whereas IA-2A titers (Fig. 1*B*) increased ($P < 0.001$) from baseline to diagnosis. When the parenteral ($n = 51$) and oral ($n = 48$) trials were analyzed separately for titers, the same difference in directionality was evident (parenteral: $P = 0.14$ for GADA, $P < 0.001$ for IA-2A; oral: $P < 0.05$ for GADA, $P < 0.001$ for IA-2A).

We also studied changes in autoantibody titers in those positive at baseline. Among the 80 progressors positive for GADA at baseline with measurements at diagnosis, titers fell significantly (medians: 0.33 to 0.20; $P < 0.01$). Conversely, among the 57 progressors positive for IA-2A at baseline, titers increased from baseline to diagnosis (0.71 to 0.88; $P < 0.001$). Among those who were positive for both GADA and IA-2A at baseline ($n = 48$), GADA titers also decreased (0.30 to 0.14; $P < 0.001$) and IA-2A titers increased (0.73 to 0.88; $P < 0.001$).

Because DPT-1 participants were ICA positive, we assessed the representativeness of those positive for autoantibodies by examining individuals screened for trial eligibility ($n = 92,505$). Of the 3,560 GADA positive, 1,353 (38%) were also positive for ICA. Of the 1,484 IA-2A positive, 895 (60%) were also ICA positive. Thus, GADA positivity and IA-2A positivity were frequently associated with ICA positivity at screening.

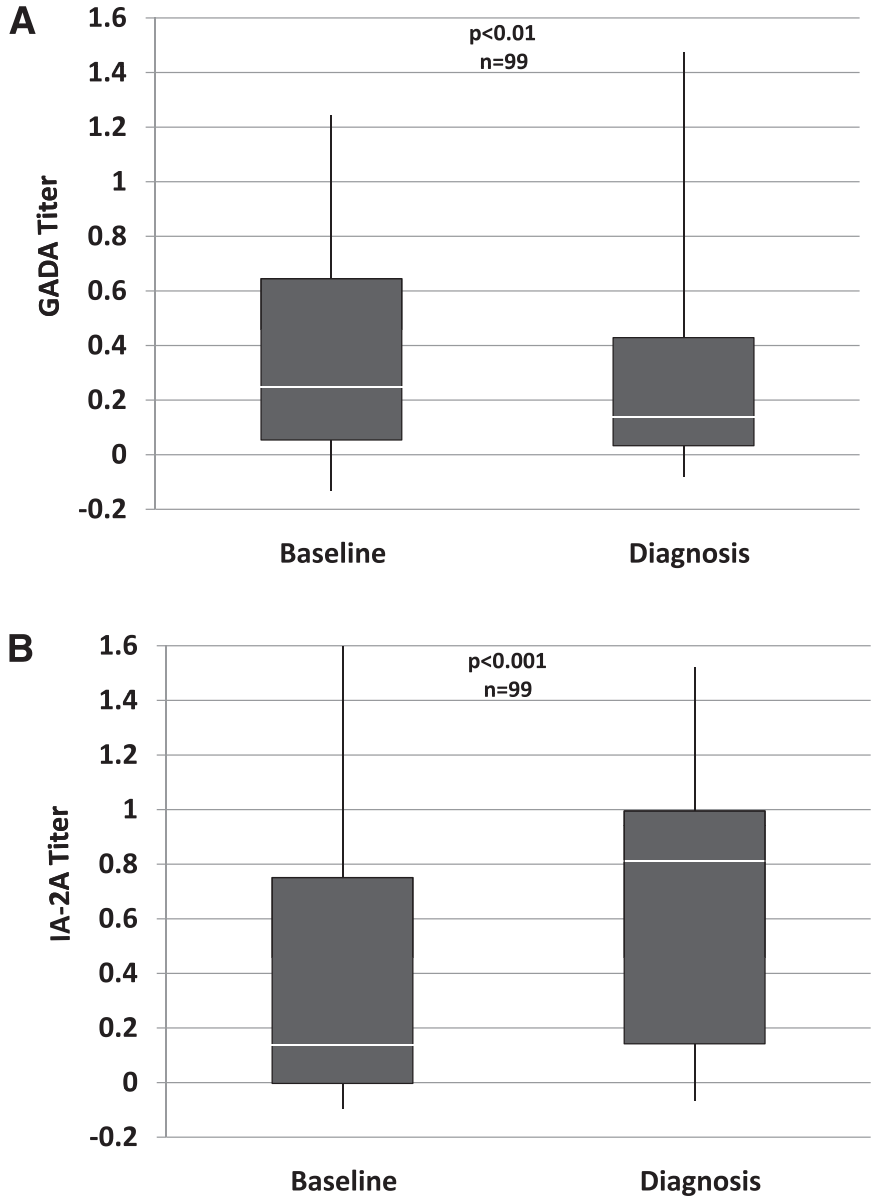

Figure 1—*Shown are titers of GADA* (A) *and IA-2A* (B) *at baseline (mean ± SD: 3.3 ± 1.5 years before diagnosis) and at diagnosis in the same individuals. Whereas there tends to be a decrease in the GADA titer, the IA-2A titer increases. White line, median; vertical line, range; bottom of box, 25th percentile; top of box, 75th percentile.*

CONCLUSIONS—This analysis indicates that GADA and IA-2A positivity are both common over 3 years before the diagnosis of T1D. Whereas GADA positivity shows little overall change with progression to T1D, there is a decrease in GADA titers. In contrast, IA-2A positivity and IA-2A titers increase appreciably. Among progressors already positive for autoantibodies, GADA titers decrease and IA-2A titers increase.

There have been no reports of changes in GADA and IA-2A positivity and titer with the approaching onset of T1D, although several prospective studies are currently examining incident autoantibody positivity in children at higher risk for T1D (4,6). Data from a prior report (5) are consistent with our finding that GADA positivity tended to be higher at baseline than IA-2A positivity.

The data indicate that more information about changes with progression to T1D can be obtained by examining autoantibody titers rather than just positivity. A significant decline in GADA titers was observed, but not in the frequency of positivity. Moreover, even within the positive range, IA-2A titers increased. The latter finding is consistent with prior observations that within the positive range, autoantibody titer can be predictive of T1D (9,10).

Because the DPT-1 participants were ICA positive, we examined the representativeness of the data for those GADA positive and those IA-2A positive at baseline. We found that when individuals are IA-2A positive, ICA positivity is frequently a concomitant. ICA positivity is less common among those GADA positive, but it is still substantial. Thus, the findings are likely to be representative of many who progress to T1D.

The decline in GADA among those positive could have been exaggerated by a regression toward the mean. However, this would not affect the overall trend. Moreover, a regression toward the mean would have actually dampened rather than have exaggerated the increase in IA-2A among those positive.

Even though autoantibodies are known predictors of T1D (9–11), and are commonly present at the time of diagnosis (1–4), their relevance to the pathogenesis of T1D is still unclear. The differing patterns of change between GADA and IA-2A in the years before diagnosis could be related to pathogenetic processes that are occurring during the progression to T1D.

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J.M.S. analyzed data and wrote the manuscript. J.S.S. conducted the study and reviewed the manuscript. J.P.P. conducted the study, reviewed the manuscript, and assisted in writing the manuscript. J.P.K. conducted the study and reviewed the manuscript. D.C. programmed for the study and reviewed the manuscript. L.Y. reviewed the manuscript. D.A.S., T.O., and G.E. conducted the study and reviewed the manuscript.

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References

1. Gorsuch AN, Spencer KM, Lister J, et al. Evidence for a long prediabetic period in type I (insulin-dependent) diabetes mellitus. Lancet 1981;2:1363–1365
2. Palmer JP, Asplin CM, Clemons P, et al. Insulin antibodies in insulin-dependent diabetics before insulin treatment. Science 1983;222:1337–1339
3. Baekkeskov S, Landin M, Kristensen JK, et al. Antibodies to a 64,000 Mr human islet cell antigen precede the clinical onset of insulin-dependent diabetes. J Clin Invest 1987;79:926–934
4. Barker JM, Barriga KJ, Yu L, et al.; Diabetes Autoimmunity Study in the Young. Prediction of autoantibody positivity and progression to type 1 diabetes: Diabetes Autoimmunity Study in the Young (DAISY). J Clin Endocrinol Metab 2004;89:3896–3902
5. Yu L, Rewers M, Gianani R, et al. Antiislet autoantibodies usually develop sequentially rather than simultaneously. J Clin Endocrinol Metab 1996;81:4264–4267
6. Ziegler A-G, Hummel M, Schenker M, Bonifacio E. Autoantibody appearance and risk for development of childhood diabetes in offspring of parents with type 1 diabetes: the 2-year analysis of the German BABYDIAB Study. Diabetes 1999;48:460–468
7. Diabetes Prevention Trial–Type 1 Diabetes Study Group. Effects of insulin in relatives of patients with type 1 diabetes mellitus. N Engl J Med 2002;346:1685–1691
8. Skyler JS, Krischer JP, Wolfsdorf J, et al. Effects of oral insulin in relatives of patients with type 1 diabetes: the Diabetes Prevention Trial–Type 1. Diabetes Care 2005;28:1068–1076
9. Orban T, Sosenko JM, Cuthbertson D, et al.; Diabetes Prevention Trial–Type 1 Study Group. Pancreatic islet autoantibodies as predictors of type 1 diabetes in the Diabetes Prevention Trial–Type 1. Diabetes Care 2009;32:2269–2274
10. Achenbach P, Warncke K, Reiter J, et al. Stratification of type 1 diabetes risk on the basis of islet autoantibody characteristics. Diabetes 2004;53:384–392
11. Kulmala P, Savola K, Petersen JS, et al.; The Childhood Diabetes in Finland Study Group. Prediction of insulin-dependent diabetes mellitus in siblings of children with diabetes. A population-based study. J Clin Invest 1998;101:327–336