Diabetic Autoimmune Markers in Children and Adolescents With Type 2 Diabetes

Eba H. Hathout, MD; Wendy Thomas; Mohamed El-Shahawy, MD, MHA; Fadi Nahab; and John W. Mace, MD

ABSTRACT. Background. There is an increase in the incidence of type 2 diabetes in children and adolescents. Absence of known diabetes autoimmune markers is sometimes required to confirm the diagnosis.

Objective. To identify clinical and autoimmune characteristics of type 2 diabetes in a pediatric population.

Method. We report an analysis of 48 children and adolescents with type 2 diabetes, compared with 39 randomly selected children with type 1 diabetes, diagnosed and followed at the Loma Linda University Pediatric Diabetes Center. Ethnic, familial, seasonal, and autoimmune marker characteristics are outlined. To determine the reliability of antibody testing in confirming the type of diabetes at diagnosis, we studied the incidence of positive islet cell antibodies (ICAs), glutamic acid decarboxylase antibodies (GADs), and insulin autoantibodies (IAAs) at diagnosis in both groups. ICAS12, GADs, and IAAs were measured by radioimmunoassay.

Results. The cohort with type 2 diabetes had a similar gender distribution as the group with type 1 diabetes but a significantly higher age at diagnosis. Ethnic background was significantly different between the 2 groups, predominantly Hispanic in type 2 and white in type 1. Body mass index was significantly higher in type 2 diabetes (mean = 31.24 kg/m²). Among the patients with type 2 diabetes, 33% presented in diabetic ketoacidosis, random blood glucose at diagnosis ranged from 11.4 to 22.25 mmol/L (228 – 445 mg/dL), fasting C-peptide levels were 0.89 to 2.7 mmol/L (2.7–8.2 ng/mL) and hemoglobin A1C was 10.8 ± 3.5% (normal: <6.6%). None of these parameters was significantly different from the type 1 diabetes group. Although the incidence of diabetes antibody markers was significantly lower in type 2 versus type 1 diabetes, 8.1% of patients with type 2 diabetes had positive ICAs, 30.3% had positive GADs, and 34.8% had positive IAAs without ever being treated with insulin. In the type 2 diabetes group, none of the Hispanic patients had ICAs. However, there was no significant correlation between any of the diabetes antibodies and obesity, presence of acanthosis nigricans, or family history of diabetes. The frequency of thyroid antibodies was not significantly different from the group with type 1 diabetes. Daily insulin requirements 1 year after diagnosis were significantly lower in type 2 diabetes, ranging from 0 to 1.2 U/kg with a mean of 0.33.


ABBREVIATIONS. ICA, islet cell antibody; GAD, glutamic acid decarboxylase antibody; IAA, insulin autoantibody; BMI, body mass index; DKA, diabetic ketoacidosis; TGA, thyroglobulin antibody; TPOA, thyroid peroxidase antibody; SD, standard deviation.
factors: Hispanic, Pacific Island or African, native or Asian-American ancestry, history of type 2 diabetes in a first-degree relative, obesity (body mass index [BMI]: >24 kg/m²), and presence of acanthosis nigricans. Evidence of endogenous insulin production by measuring basal and stimulated C-peptide levels in the non-ketotic state was used to confirm the diagnosis. Patients with suspected or genetically documented maturity-onset diabetes of the young or iatrogenic diabetes were given their respective diagnoses. The control group of 39 patients was selected at random (every 10th name in the database given a diagnosis of type 1 diabetes with age of onset of >3 years and current follow-up at Pediatric Diabetes Center of Loma Linda University Children’s Hospital). All patient histories were obtained by members of the Pediatric Diabetes Center at the initial clinic visit. Clinical information was obtained retrospectively by hospital and clinic record review.

Demographic characteristics examined in this study include age at diagnosis, gender, ethnic background, rank among siblings, household size at diagnosis, season of diagnosis, and family history of diabetes of either type. Past medical history also includes mode of delivery and parental ages at birth. Clinical characteristics at the time of diagnosis include symptoms of a viral illness, such as rhinorrhea or cough, BMI, presence of acanthosis nigricans, blood glucose, hemoglobin A1C and C-peptide levels, insulin treatment, and presence of diabetic ketoacidosis (DKA).

IAAs were measured by 113-25 binding capacity (normal: <5 μU/mL), GAD65 (GAA), and ICA512 were measured by radioimmunoprecipitation assay (normal: <1 U/mL, Endocrine Sciences Laboratories, Calabasas, CA). Thyroglobulin antibodies (TGA) and thyroid peroxidase antibodies (TPOAs) were measured by immunochemiluminometric assay (Quest Diagnostics at Nichols Institute, San Juan Capistrano, CA).

Data were analyzed in a cross-sectional manner. All data for this study were extracted from pediatric diabetes center charts, which include hospital admission information, onto a structured database form. Data were subsequently entered using Excel, Version 5.0 (Microsoft Co, Seattle, WA), verified by inspection, and checked for erroneous values.

Statistical Analysis
Continuous data were analyzed using Student’s 2-tailed t test. Analysis of categorical variables was accomplished using the χ² test. P values <.05 were assigned statistical significance. All P values are 2-tailed. Age, BMI, hemoglobin A1C, blood glucose, and C-peptide data are presented as mean values ± standard deviation (SD).

RESULTS
Age range at diagnosis was 9.7 to 17.9 years for the group with type 2 diabetes and 3.75 to 17 years for the group with type 1 diabetes (Table 1). Ethnic breakdown in type 1 diabetes was similar to that of the background population in distinction to the group with type 2 diabetes, which proportionately consisted of significantly more Hispanics (P = .001) and fewer whites (P = .011). No particular gender predominance was apparent in either group. There was no significant difference in the ranking of the participants in the family or household size at diagnosis between the 2 groups. Fall was the most common season for diagnosis of type 2 diabetes, compared with winter, which was the predominant season of presentation for the type 1 group (P = .01). There was a significantly higher frequency of type 2 diabetes in first-degree relatives of patients with type 2 diabetes (P = .001). Past medical history did not show a significant difference in the frequency of birth by cesarean section or parental ages at birth between the 2 groups.

BMI at diagnosis of type 2 diabetes ranged from 21.90 to 53.08 kg/m² (Table 2). This range was significantly higher than BMI at diagnosis of type 1 diabetes (range: 8.40–27.89; P = .0001). Acanthosis nigricans was exclusive to type 2 diabetes, occurring in 37% of patients in that group. The percentage of patients with type 2 diabetes initially treated with insulin was 68.6%. The rest were managed with a combination of diet and exercise, with or without metformin. Although blood glucose, hemoglobin A1C levels, and the percentage of patients presenting in DKA were all lower in type 2 (vs type 1) diabetes, their difference between the 2 groups was not statistically significant.

Among patients with type 2 diabetes, 30.3% had
positive GADs (>1.0 U/mL), 34.8% had positive IAAs (>5 μU/mL), and 8.1% had positive ICAs (>1.0 U/mL; Table 2). Positive ICAs were inversely correlated with Hispanic ancestry. Otherwise, there was no significant correlation among any of the 3 antibodies and race or presence of obesity, acanthosis, or a family history of diabetes of either type. ICA and GAD frequencies in type 2 patients were significantly lower than in type 1 diabetes patients (positive frequency range: 71%–76%; P < .01). However, the frequency of positive TGAs and TPOAs was not significantly different between the 2 groups. Subsequent clinical correlation showed that 3 of the type 2 diabetes patients with positive thyroid antibodies had concomitant hypothyroidism at the time of the study, whereas none of the type 1 group had evidence thereof.

Treatment initiation and modification decisions for patients with type 2 diabetes in this study were made by 2 pediatric endocrinologists. One year after diagnosis, 50% of patients with type 2 diabetes were not on insulin therapy and were controlled with diet and exercise alone or in combination with metformin. Retrospective analysis showed that 47% of these patients had a first-degree relative with type 2 diabetes, 36% had acanthosis nigricans, 89% had BMI >24 kg/m², and 27% had DKA at diagnosis. In addition, 18.75% of these patients had positive GADs (Table 3), 37.5% had positive IAAs, and 0.0% had positive ICAs. The majority of patients with type 2 diabetes not requiring insulin 1 year after diagnosis remained off insulin up to the time of the present review, conducted 3 to 5 years after their diagnosis. The remaining patients were either diagnosed within 2 years of this review or lost to follow-up.

Addition of, or transition to, insulin therapy was based on lack of glycemic improvement (hemoglobin A₁C remains or rises to >8%) despite maximal attempts at diet and exercise and a trial of metformin. In patients treated with insulin 1 year after diagnosis, weight-adjusted daily insulin requirements were significantly lower than in patients with type 1 diabetes (P = .025), ranging from 0 to 1.2 U/kg with a mean of 0.33. All type 2 patients with positive ICA titers were on insulin therapy 1 year after diagnosis.

**DISCUSSION**

Type 2 diabetes in children and adolescents underwent a >10-fold increase over the past decade. This increase has been primarily attributed to a problem of childhood obesity, which has reached epidemic proportions. Genetic, ethnic, and environmental risk factors have also been considered. This, together with a potentially long asymptomatic period for type 2 diabetes, raise serious concerns for long-term microvascular and macrovascular complications.

Demographic characteristics of adolescents with type 2 diabetes in our study are similar to those reported previously by others in terms of predominance of Hispanic (Mexican American) ancestry, and association with obesity, acanthosis nigricans, and family history. However, given that the ethnic breakdown in our control group was similar to that of the background population, our study did not show the significantly higher incidence of type 2 diabetes in black adolescents described by others. Of interest is the 1 patient with type 2 diabetes with a family history of type 1 diabetes in a first-degree relative. This patient is a black female, diagnosed at 13 years old, with BMI of 41.6 kg/m², significant acanthosis, normal C-peptide, and negative diabetes and thyroid antibodies. Verification of the presumed type 1 diabetes in that relative was not possible.

The increase in BMI in type 2 versus type 1 diabetes might have been exaggerated by the fact that all our patients with type 2 diabetes were pubertal at the time of the study. The trend shown in this study for type 2 diabetes to be diagnosed in the fall might be the consequence of the summer vacation, a period of relative excess eating and decreased activity for many adolescents. In contrast, it might have been a coincidental finding brought to medical attention by requirements of a school physical examination.

The diagnosis of type 2 diabetes relies heavily on clinical criteria, such as Hispanic or black ancestry, positive family history, obesity, evidence of insulin resistance, and absent or milder-than-expected plasma glucose fluctuations, ketosis, or hypoglycemic reactions to insulin. Measurable fasting or stimulated C-peptide levels, reflecting endogenous pancreatic insulin secretion, are not exclusively diagnostic of type 2 diabetes, because they can theoretically be present in the honeymoon phase of type 1 diabetes. However, absence of β-cell autoimmune markers (ICA, GAD, and IAA without previous insulin therapy) has been a diagnostic prerequisite for type 2 diabetes in most pediatric cases, particularly pertaining to trials of noninsulin therapeutic modalities.

Evidence of β-cell autoimmunity in adults with type 2 diabetes, however, has been well documented in the literature. GAD positivity has been reported in 3.8% to 16.1% of patients with type 2 diabetes. ICA prevalence in type 2 diabetes was reported as 1.6% in 1 study and 4.8% in another. Moreover, some reports suggested that ICA positivity in type 2 diabetes predicted future insulin requirement and that GAD positivity correlated with a younger age of diagnosis, lower C-peptide at diagnosis, and increased probability of insulin requirement. No differences in BMI or serum C-peptide levels could be found between GAD-positive and GAD-negative type 2 patients at diagnosis. Of interest is a study showing that fasting C-peptide levels in ICA-positive patients with type 2 diabetes were similar to patients with type 1 diabetes, 3 years after diagnosis. In the present study, the higher incidence of β-cell

<table>
<thead>
<tr>
<th>TABLE 3. Diabetes Autoimmune Markers in Noninsulin-Requiring Patients With Type 2 Diabetes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Percentage With Positive Titer</td>
</tr>
<tr>
<td>GAD</td>
</tr>
<tr>
<td>IAA</td>
</tr>
<tr>
<td>ICA</td>
</tr>
</tbody>
</table>

http://www.pediatrics.org/cgi/content/full/107/6/e102
antibodies in adolescents with type 2 diabetes (Table 2) supports the previously reported correlation of positive diabetes antibodies with younger age at di-
gnosis.8 Our data also suggest that ICA antibody
positivity at diagnosis could be a predictor of future
insulin need.17,18

The possibility that positive diabetes antibodies in
adolescents with noninsulin-requiring diabetes may
represent a form of early-onset latent autoimmune
diabetes similar to that described in adults (latent
autoimmune diabetes in adults)21,22 needs to be con-
sidered. This form of diabetes, however, was not
reported to be specifically associated with obesity or
Hispanic ancestry. Long-term data on insulin re-
quirements can shed more light on this area. There is
also the possibility that some type 2 patients, par-
ticularly those requiring insulin, are actually obese pa-
tients with type 1 diabetes. Studying genetic deter-
minants specific to either type of diabetes, eg, human
leukocyte antigen-DQ/DR subtypes, might be
needed in the future for typing of diabetes in such
patients.23 The fact that autoimmunity should not
exclude the possibility of type 2 diabetes in adoles-
cents is supported by the presence of positive thyroid
(TPOA and TGA) antibodies and hypothyroidism in
type 2 patients, which was detected in 3 patients in
this series. Because the focus of this study was the
typing (and not the diagnosis) of diabetes in chil-
dren, antibody levels in healthy controls were not
included.

Data regarding insulin treatment in type 2 adoles-
cents, particularly at the time of diagnosis, should be
interpreted with caution. Many patients had been
misdiagnosed as having type 1 diabetes based on
either age or DKA at presentation. In addition, lack of
a Food and Drug Administration-approved step-
wise approach to the treatment of type 2 diabetes in
children often facilitated the use of insulin as initial
therapy. Moreover, social and cultural factors inter-
ferring with compliance with noninsulin regimens,
particularly in the adolescent age group, frequently
lead to use of insulin in noninsulin-requiring pa-
tients.

Another entity not addressed in this study consists of
patients who are both insulin-deficient and insu-
lin-resistant.24 This is exemplified by obese type 1
adolescent patients, with onset of diabetes in early
childhood, evidence of compliance in a supervised
setting, and daily insulin requirements exceeding 2
U/kg. Whether those patients, who have both types of
diabetes, could be categorized and treated as such
merits additional exploration.

The above data reflect the broad spectrum of pan-
creatic and peripheral abnormalities that could lead
to adolescent diabetes. Our study and many studies
in the adult literature suggest that the presence of
diabetes autoimmune markers should not be used to
exclude the diagnosis of type 2 diabetes in children
and adolescents. Larger prospective pediatric dia-
betes studies are needed to confirm the validity of this
conclusion. Acceptance of this concept in scientific
and research settings could open the door to trials of
a broader range of therapeutic modalities for a group

of children and adolescents who were previously
labeled insulin-dependent for life.

REFERENCES
1. Rosenbloom AL. Emerging epidemic of type 2 diabetes in Mexican-
2. Young RS, Rosenbloom AL. Type 2 (non-insulin dependent) diabetes in
3. Glaser NS, Jones KL. Non-insulin dependent diabetes mellitus in Mex-
4. Clemons RD, Gottschalk ME, Jones KL. Basal and postprandial C-
peptide values in a racially mixed population of adolescents with type 2
diabetes mellitus. Diabetes. 1999;48(suppl 1):345
5. Niskanen LK, Tuomi T, Karjalainen J, et al. GAD antibodies in NIDDM:
dependent diabetes mellitus patients with autoantibodies to glutamic
with the initial diagnosis of NIDDM with positivity for antibodies to
glutamic acid decarboxylase. Exp Clin Endocrinol Diabetes. 1997;105:
327–330
131–139
(GAD65) antibodies and insulin auto-antibodies in Japanese patients
with non-insulin-dependent diabetes mellitus. Endocr J. 1997;44:43–51
11. Thai AK, Ng WY, Loke KY, et al. Anti-GAD antibodies in Chinese
patients with youth and adult onset IDDM and NIDDM. Diabetesologia.
1997;40:1425–1430
12. The Expert Committee on the Diagnosis and Classification of Diabetes
Mellitus. Committee report. Diabetes Care. 2000;23(suppl 1)
13. Pinhas-Hamiel O, Zeitzler P. A weighty problem: diagnosis and treat-
ment of type 2 diabetes in adolescents. Diabetes Spectrum. 1997;10:
219–224
14. Fagot-Campagna A. Type 2 diabetes among North American children
and adolescents: an epidemiologic review and a public health perspec-
15. Young TK. Childhood obesity in a population at high risk for type 2
and without family history of type 2 diabetes. Diabetes Care. 1999;22:
1325–1329
17. DiMario U, Irvine WJ, Borsey DQ, et al. Immune abnormalities in
diabetic patients not requiring insulin at diagnosis. Diabetesologia. 1983;
25:392–395
decarboxylase antibodies present at diagnosis of diabetes predict the
need for insulin treatment: a cohort study in young adults whose disease
was initially labeled as type 2 or unclassifiable diabetes. Diabetes Care.
isoform of glutamic acid decarboxylase detected in non-insulin-
relation to islet cell antibodies during the first 3 year after clinical
diagnosis of diabetes in type 2 diabetic patients. Diabetes Care. 1993;16:
902–910
autoimmune diabetes in white familial NIDDM pedigrees. Diabetes Care.
1997;20:1248–1251
autoimmune diabetes in adults (LADA) is associated with low level
glutamate decarboxylase (GAD65) and IA-2 autoantibodies: diabetes
type 1 diabetic patients lacking high risk HLA alleles in a Caucasian
population: are these type 1b diabetes cases? Diabetologia. 1997;40:
911–915
24. Withcfel SF, Arslanian S. Ovarian responses to hCG stimulation: insulin
resistance/hyperinsulinemia vs insulin deficiency. Clin Endocrinol (Oxf).
1999;51:127–130

Downloaded from http://pediatrics.aappublications.org/ by guest on October 22, 2017
Diabetic Autoimmune Markers in Children and Adolescents With Type 2 Diabetes
Eba H. Hathout, Wendy Thomas, Mohamed El-Shahawy, Fadi Nahab and John W. Mace

*Pediatrics* 2001;107:e102
DOI: 10.1542/peds.107.6.e102

**Updated Information & Services** Including high resolution figures, can be found at:
http://pediatrics.aappublications.org/content/107/6/e102

**References** This article cites 23 articles, 7 of which you can access for free at:
http://pediatrics.aappublications.org/content/107/6/e102.full#ref-list-1

**Subspecialty Collections** This article, along with others on similar topics, appears in the following collection(s):
- **Endocrinology**
  http://classic.pediatrics.aappublications.org/cgi/collection/endocrinology_sub
- **Diabetes Mellitus**
  http://classic.pediatrics.aappublications.org/cgi/collection/diabetes_mellitus_sub
- **Metabolic Disorders**
  http://classic.pediatrics.aappublications.org/cgi/collection/metabolic_disorders_sub

**Permissions & Licensing** Information about reproducing this article in parts (figures, tables) or in its entirety can be found online at:
https://shop.aap.org/licensing-permissions/

**Reprints** Information about ordering reprints can be found online:
http://classic.pediatrics.aappublications.org/content/reprints
Diabetic Autoimmune Markers in Children and Adolescents With Type 2 Diabetes
Eba H. Hathout, Wendy Thomas, Mohamed El-Shahawy, Fadi Nahab and John W. Mace

*Pediatrics* 2001;107;e102
DOI: 10.1542/peds.107.6.e102

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://pediatrics.aappublications.org/content/107/6/e102