



Published in final edited form as:

Diabetologia. 2021 October ; 64(10): 2247–2257. doi:10.1007/s00125-021-05514-3.

Characteristics of children diagnosed with type 1 diabetes before vs after 6 years of age in the TEDDY cohort study

Jeffrey P. Krischer¹, Xiang Liu¹, Åke Lernmark², William A. Hagopian³, Marian J. Rewers⁴, Jin-Xiong She⁵, Jorma Toppari^{6,7}, Anette G. Ziegler⁸, Beena Akolkar⁹ TEDDY Study Group

¹Health Informatics Institute, Morsani College of Medicine, University of South Florida, Tampa FL, USA

²Department of Clinical Sciences, Lund University/CRC, Skåne University Hospital SUS, Malmö, Sweden

³Pacific Northwest Diabetes Research Institute, Seattle WA, USA

⁴Barbara Davis Center for Childhood Diabetes, University of Colorado, Aurora CO, USA

⁵Center for Biotechnology and Genomic Medicine, Augusta University, Augusta GA, USA

⁶Department of Pediatrics, Turku University Hospital, Turku, Finland

⁷Institute of Biomedicine, Research Centre for Integrated Physiology and Pharmacology, and Population Research Centre, University of Turku, Turku, Finland

⁸Institute of Diabetes Research, Helmholtz Zentrum München, and Klinikum rechts der Isar, Technische Universität München, and Forschergruppe Diabetes e.V., Neuherberg, Germany

⁹National Institute of Diabetes & Digestive & Kidney Diseases, Bethesda MD, USA

Abstract

Aims/hypothesis—Prognostic factors and characteristics of children diagnosed with type 1 diabetes before 6 years of age were compared with those diagnosed at 6–13 years of age in the TEDDY study.

Terms of use and reuse: academic research for non-commercial purposes, see here for full terms. <http://www.springer.com/gb/open-access/authors-rights/aam-terms-v1>

Corresponding author: Jeffrey P. Krischer, jeffrey.krischer@epi.usf.edu.

A complete list of the TEDDY Study Group members can be found in the electronic supplementary material [ESM].

Contribution statement All authors attest to meeting ICMJE uniform requirements for authorship by making substantial contributions to conception and design of this paper; acquisition, analysis and interpretation of the data; drafting or revising the article for intellectual content; and giving final approval of the published version. JK designed the study, proposed the analysis, interpreted the findings, and wrote the manuscript. XL performed the analysis and contributed to writing the manuscript. ÅL, WH, MR., J-XS, JT, AZ, and BA designed the study and reviewed/edited the manuscript. JK and XL are the guarantors of this work and, as such, had full access to all the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. The authors declare that there are no relationships or activities that might bias, or be perceived to bias, their work.

Publisher's Disclaimer: This Author Accepted Manuscript is a PDF file of an unedited peer-reviewed manuscript that has been accepted for publication but has not been copyedited or corrected. The official version of record that is published in the journal is kept up to date and so may therefore differ from this version.

Duality of interest There are no conflicts of interest.

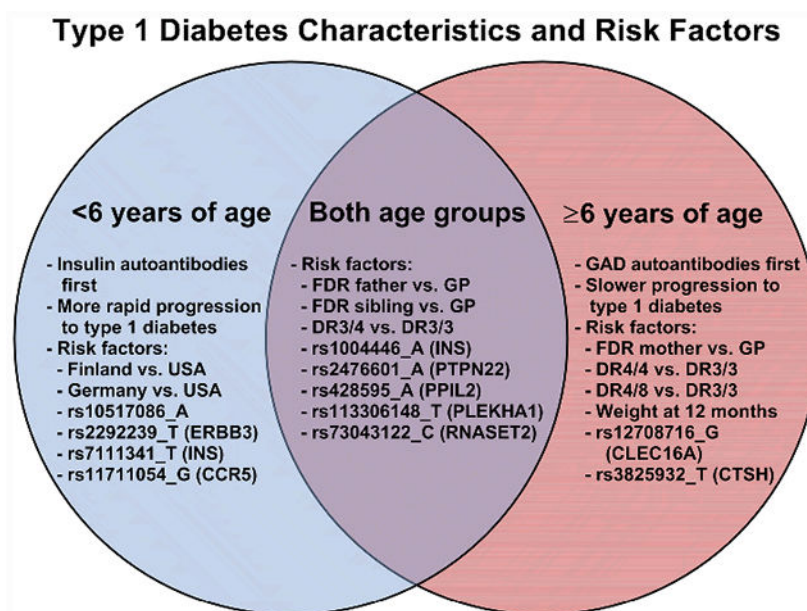
[ClinicalTrials.gov](https://clinicaltrials.gov) identifier: NCT00279318

Methods—Genetically high-risk children (n=8502) were followed from birth for a median of 9.9 years; 328 (3.9%) were diagnosed with type 1 diabetes. Cox proportional hazard model was used to assess the association of prognostic factors with the risk of type 1 diabetes in the two age groups.

Results—Children in the younger group tended to develop autoantibodies earlier than those in the older group did (mean age 1.5 vs 3.5 years), especially insulin autoantibodies (IAA), which developed earlier than GAD autoantibodies (GADA). Children in the younger group also progressed to diabetes more rapidly than the children in the older group did (mean age 1.9 vs 5.1 years). Children with autoantibodies first appearing against insulinoma antigen-2 (IA-2A) were found only in the older group. The significant diabetes risk associated with the country of origin in the younger group was no longer significant in the older group. Conversely, the diabetes risk associated with HLA genotypes was statistically significant also in the older group. Initial seroconversion before vs after 2 years of age was associated with decreased risk for diabetes diagnosis in children positive for multiple autoantibodies, but the diabetes risk did not decrease further with increasing age if initial seroconversion occurred after age 2. Diabetes risk associated with the minor alleles of rs1004446 (*INS*) was decreased in both the younger and older groups compared with other genotypes (HR 0.67). Diabetes risk was significantly increased (HR 2.07 and 1.72) with the minor alleles of rs2476601 (*PTPN22*), rs428595 (*PPIL2*) (HR 2.13 and 2.10), rs113306148 (*PLEKHA1*) (HR 2.24 and 3.21) and rs73043122 (*RNASET2*) (HR 2.31 and 2.54) (HR values represent the younger and older groups, respectively).

Conclusions/interpretations—Diabetes at an early age is likely to be preceded by IAA autoantibodies and is a more aggressive form of the disease. Among older children, once multiple autoantibodies have been observed there does not seem to be any association between progression to diabetes and the age of the child or family history.

Graphical Abstract



Research in context

What is already known about this subject?

- Diabetes-related insulin autoantibodies (IAA) appear first in very young children whereas GAD autoantibodies (GADA) usually only appear after the age of 5 years. After the age of 5 years, IAA is rarely the first autoantibody to appear

What is the key question?

- In addition to differences in the way that diabetes-related autoimmunity arises as children grow older, are there also different risk factors and rates of progression for diabetes that are associated with age?

What are the new findings?

- The association of country of origin on diabetes risk found in the younger group declined in the older age group, while the genotypic association significantly increased
- After age 2 years at seroconversion there was no longer a decreasing risk for diabetes in children positive for multiple autoantibodies associated with the age of initial seroconversion
- After multiple autoantibodies were observed, there was not a discernible difference in risk by family history of type 1 diabetes

How might this impact on clinical practice in the foreseeable future?

- This study adds to the growing body of evidence that type 1 diabetes is not a single, homogeneous disease. It differs in its presentation and, perhaps, aetiology. Much of the observed difference in the relationships between genes and exposures is explained by the age at appearance of autoantibodies, establishing that factors associated with diabetes risk need to be conditioned on age to be properly understood

Keywords

Autoimmunity; Type 1 diabetes

Introduction

The Environmental Determinants of Diabetes in the Young (TEDDY) study has enrolled and followed a cohort of 8676 infants at elevated genetic risk for autoimmune type 1 diabetes from three months of age (1–3). TEDDY is designed to follow children for 15 years. The characteristics of children progressing to type 1 diabetes during the first 6 years of age (the first third of the planned follow-up period) has been published (4), and there is now additional follow-up for the middle third of the planned follow-up period, i.e., through to 12 years of age. This paper describes the characteristics of children developing autoimmunity and type 1 diabetes during their second 6 years of life and seeks to identify differences in the pattern of islet autoantibody development and the changing relationship between previously identified risk factors for autoantibodies and type 1 diabetes endpoints. The aim is to explore whether the younger cohort developing type 1 diabetes differs from the older cohort, suggesting the emergence of a different form of type 1 diabetes in children as they get older.

The age at the first appearance of islet autoantibodies has been shown to be related to which autoantibody appears first, which has, in turn, been linked to specific genotypic subtypes and associated environmental exposures (4). It was also noted that the incidence of insulin autoantibodies (IAA) as the first-appearing autoantibody during the first 6 years of life, declined with age, almost disappearing, while the incidence of GAD autoantibodies (GADA), as the first-appearing autoantibody, increased and remained stable throughout follow-up (5). The changing incidence of the first-appearing autoantibody, as children age, signifies a possible difference in both the aetiology and the pathogenesis of type 1 diabetes. Therefore, this study explores differences in characteristics of children who progressed to diabetes among the younger <6 years of age cohort, presumably arising from those who

predominantly developed IAA first and compared them with the characteristics of children who progressed to diabetes among the 6–12 years of age cohort, presumably arising from those who predominantly developed GADA first, with particular emphasis on factors that were prognostic for disease initiation (aetiology) and progression (pathogenesis) specific to these age intervals.

Methods

Participants

TEDDY is a prospective cohort study funded by the National Institutes of Health with the primary goal of identifying environmental causes of type 1 diabetes. It includes six clinical research centres – three in the US (Colorado, Georgia/Florida and Washington State) and three in Europe (Finland, Germany and Sweden). Detailed study design and methods have been previously published (1–3). Written informed consents was obtained for all study participants from a parent or primary caretaker, separately, for genetic screening and participation in the prospective follow-up. The high-risk genotypes for participants screened from the general population were as follows: *DRB1*04-DQA1*03-DQB1*03:02/DRB1*03-DQA1*05-DQB1*02:01* (DR3/4), *DRB1*04-DQA1*03-DQB1*03:02/DRB1*04-DQA1*03-DQB1*03:02* (DR4/4), *DRB1*04-DQA1*03-DQB1*03:02/DRB1*08-DQA1*04-DQB1*04:02* (DR4/8) and *DRB1*03-DQA1*05-DQB1*02:01/DRB1*03-DQA1*05-DQB1*02:01* (DR3/3). Additional genotypes were included for first-degree relatives (FDRs) of an individual with type 1 diabetes: *DRB1*04-DQA1*03-DQB1*03:02/DRB1*04-DQA1*03-DQB1*02:02* (DR4/4b), *DRB1*04-DQA1*03-DQB1*03:02/DRB1*01-DQA1*01-DQB1*05:01* (DR4/1), *DRB1*04-DQA1*03-DQB1*03:02/DRB1*13-DQA1*01-DQB1*06:04* (DR4/13), *DRB1*04-DQA1*03-DQB1*03:02/DRB1*09-DQA1*03-DQB1*03:03* (DR4/9) and *DRB1*03-DQA1*05-DQB1*02:01/DRB1*09-DQA1*03-DQB1*03:03* (DR3/9). The HLA-DR-DQ genotype abbreviations shown in parentheses will be used throughout this paper. Genotyping was confirmed by reverse blot hybridisation at the central HLA Reference Laboratory at Roche Molecular Systems, Oakland, CA (3), along with the *INS*-23Hph1 (rs689), *CTLA4* T17A (rs231775) and *PTPN22* R620W (rs2476601) SNP primer pairs. The study was approved by local institutional review or ethics boards and is being monitored by an external evaluation committee formed by the National Institutes of Health.

SNP genotyping was performed by the Center for Public Health Genomics at the University of Virginia using the Illumina ImmunoChip, which is a custom array for genotyping SNPs selected from regions of the human genome firmly associated with autoimmune diseases (6). The final selection containing approximately 186,000 SNPs in 186 regions for 12 autoimmune diseases was decided by the ImmunoChip Consortium. TEDDY previously examined whether any of 41 non-HLA SNPs previously shown to be associated with type 1 diabetes conferred risk for islet autoimmunity (7).

Islet autoantibodies

Islet autoantibodies to insulin (IAA), GAD (GADA) or insulinoma antigen-2 (IA-2A) were measured in two laboratories by radiobinding assays. In the US, all sera were assayed at the Barbara Davis Center for Childhood Diabetes at the University of Colorado Denver; in Europe, all sera were assayed at the University of Bristol, UK. Both laboratories demonstrated high sensitivity and specificity as well as concordance (8). All positive islet autoantibody samples and 5% of the negative samples were re-tested in the other reference laboratory and deemed confirmed if concordant. Persistent islet autoimmunity was defined as confirmed positive IAA, GADA or IA-2A in at least two consecutive samples. Zinc transporter autoantibodies (ZnT8A) were measured in samples positive for one of the other autoantibodies.

Statistical methods

Characteristics of <6 year olds who progressed to diabetes diagnosis were compared with those of children between 6 and 12 years of age who progressed to diabetes diagnosis by non-parametric (Wilcoxon rank sum) tests for continuous variables and Pearson's χ^2 tests for categorical variables. Multiple Cox proportional hazard (PH) models were applied to examine factors related to the risk of diabetes previously published in the TEDDY study (5, 7, 9–12) with and without the adjustment of age at onset of multiple autoantibodies as a time-dependent covariate. The associations between the factors and the risk of type 1 diabetes during the first 6 years of age and in the range of 6–12 years of age were examined, as was the interaction between the factors and the dichotomised time variable (the two age intervals) treated as time-dependent covariates (13). The magnitudes of the associations were described by HRs with 95% CIs. Comparisons between the younger and older HRs were conducted by testing whether the ratio of the two HRs differed from 1 based on Wald tests. Adjustments for population stratification were made by using the top two principal components from the Immunochip SNP data as covariates in the Cox PH models (14). Hazard rates of progression to type 1 diabetes since the onset of multiple autoantibodies stratified by the age at initial seroconversion were calculated using the life-table approach. Data were analysed using the Statistical Analysis System software (version 9.4; SAS Institute, Cary, NC). Two-tailed p values less than 0.05 were considered to be statistically significant. No adjustment for type 1 error was made for multiple comparisons except in the context of the multiple Cox regression model.

Results

From September 2004 until February 2010, TEDDY enrolled 8676 children at birth, of whom 174 were excluded because of HLA ineligibility or indeterminate autoantibody status, leaving 8502 in the analysis. Children were followed quarterly for progression to diagnosis of diabetes. Follow-up of children with one or more islet autoantibodies continued on this schedule, whereas children who were autoantibody negative were followed semi-annually after 4 years of age. The median (IQR) age at last follow-up was 9.9 (8.1–11.5) years and the age range was 7–12 years.

As of 30 November 2018, 328 children (3.9%) had developed type 1 diabetes; 168 (2.0%) before 6 years of age and 160 (1.9%) at or after 6 years of age (Table 1). The incidence of type 1 diabetes remained fairly stable (Fig. 1), but the cumulative incidence differed by enrolment site ($p=0.0009$, Fig. 2). However, the excess risk associated with enrolment from Germany may be the consequence of the high proportion of FDRs enrolled (36.1%) compared with all the other TEDDY sites (9.2%). By 10 years of age, children from Germany and Finland had a comparable cumulative diabetes risk, as did children from Sweden and the US, but the risk in Germany and Finland remained higher than in the other two countries. The difference in geographic distribution of new type 1 diabetes patients reflects a drop in the proportion from Finland and Germany and an increase in the US while the proportion remained unchanged in Sweden ($p=0.001$).

The HLA distribution of diabetes patients was also different with an increase among those who were DR4/4 in the older age group and a corresponding decrease among those who were DR3/3 or had FDR-specific HLA genotypes ($p=0.005$). Those who were diagnosed with diabetes at 6 years of age or older developed a persistent confirmed autoantibody at an older mean age than those who became autoantibody positive before the age of 6 (3.5 vs 1.5 years, $p<0.001$). The mean duration of time between the first-appearing autoantibody and the diabetes diagnosis was also much longer (5.4 years vs 1.9 years, $p<0.001$) in older than in younger children. Similar patterns were observed when considering the appearance of multiple persistent confirmed autoantibodies (3.9 years and 1.8 years) and time until progression to diabetes (4.9 years vs 1.6 years) ($p<0.001$ for both).

The pattern of first-appearing autoantibody was also significantly different between those who were diagnosed with diabetes before 6 years of age and those who were diagnosed older ($p=0.001$). As might be expected, the percentage of those with GADA as the first-appearing autoantibody was higher in the older group (26.9% vs 18.5%) and the percentage of those presenting with IAA first was much higher in the younger group (44.6% vs 28.1%). Interestingly, no children in the younger group presented with IA-2A as the first-appearing autoantibody, but 10 (6.3%) among the older children did. In six of these individuals, ZnT8A autoantibodies were also present when IA-2A autoantibodies were detected.

No autoantibodies were detected in 36 (11.0%) of the children diagnosed with diabetes. The median (IQR) interval between the last autoantibody testing and the diabetes diagnosis in the 22 children from the older age group was 8.6 years (7.0–8.9) suggesting that autoantibody positivity at diagnosis, or long before it, was unknown because of lack of testing and poor protocol compliance. Among the 14 who developed type 1 diabetes in the younger age group without detected autoantibodies, the median (IQR) interval was 2.3 years (0.85–2.86). There were six children under 6 years of age whose last autoantibody test was negative within 2 years preceding their diabetes diagnosis. Whole genome sequencing was available for three, one of whom had a variant in the *HNFI1A* gene (rs762703502) associated with maturity-onset diabetes of the young, type 3.

Autoantibodies at the time of diabetes diagnosis showed that a higher per cent were IAA negative prior to diagnosis among the older group (28.8%) compared with the younger

group (13.1%), $p < 0.001$. A lower per cent were ZnT8A negative in the older group (34.4%) compared with in the younger group (61.9%) ($p < 0.001$).

The incidence of diabetic ketoacidosis (DKA) at diagnosis was marginally lower ($p = 0.046$) in the older age group compared with in the younger group, but the proportion who were symptomatic or not at diagnosis was not different.

The HRs from a multivariate PH model of risk factors published by the TEDDY study also revealed some differences in their association with diabetes comparing the two age groups (Table 2). The risk of type 1 diabetes in families with a mother who had type 1 diabetes significantly increased compared with families without an affected relative, in the older age group (HR 2.64, 95% CI 1.36, 5.12, $p = 0.004$) whereas it was not a significant risk factor in the younger age group (HR 1.51, 95% CI 0.75, 3.05, $p = 0.249$). However, this difference in HRs was not statistically significant, reflecting the wide confidence intervals of the individual HRs. Also, the risk of diabetes associated with DR4/4 vs DR3/3 was significant in the older (HR 4.16, 95% CI 1.99, 8.69, $p < 0.001$), but not in the younger (HR 1.26, 95% CI 0.65, 2.44, $p = 0.496$) age group. This increase in HRs was statistically significant at $p = 0.018$, suggesting that the DR4/4 genotype has a larger role in development of type 1 diabetes in older than in younger individuals. Conversely, in the older age group, the type 1 diabetes risk associated with children from Finland and Germany significantly declined (HR 3.20, 95% CI 1.88, 5.45, $p < 0.001$ to HR 1.26, 95% CI 0.73, 2.18, $p = 0.416$ and HR 2.19, 95% CI 1.27, 3.78, $p = 0.005$ to HR 0.56, 95% CI 0.25, 1.27, $p = 0.167$, respectively) ($p = 0.005$ and $p = 0.007$, respectively) compared with children from the US. Other risk factors were, or were not, statistically significant in both age groups. However, the HRs comparing the two groups were not significantly different.

Once multiple autoantibodies were observed, the rate of progression to type 1 diabetes decreased as the age at initial seroconversion increased ($p = 0.0003$, Fig. 3a). Children under 2 years of age at initial seroconversion progressed much more rapidly to type 1 diabetes once multiple autoantibodies were detected (hazard rate of 0.17) through 6 years of follow-up. If diabetes did not develop during this interval of time, the rate of progression from multiple autoantibodies to type 1 diabetes (hazard rate 0.127) was approximately the same as the rate of progression to type 1 diabetes in the group who initially developed autoantibodies at an older age (hazard rate range from 0.082 to 0.110) irrespective of the age of seroconversion. Additionally, the rate of progression from multiple autoantibodies to type 1 diabetes was statistically associated with a family history of type 1 diabetes in the younger age group, but not in the older age group ($p = 0.25$, Fig. 3b).

Age at the onset of multiple autoantibodies, as a continuous variable, was also included in the analysis to explore whether the age groupings introduced artificial associations when comparing risk factors for early type 1 diabetes onset compared with later type 1 diabetes onset. Age at the onset of multiple autoantibodies was a highly significant factor in each age group (HR 0.64, 95% CI 0.57, 0.72, $p < 0.001$ and HR 0.76, 95% CI 0.70, 0.83, $p < 0.001$) showing decreasing risk with increasing age in both. After adjusting for the age at the development of multiple autoantibodies, the only remaining type 1 diabetes risk factors were family history of type 1 diabetes (father with type 1 diabetes vs father without type

1 diabetes in the younger age group and mother or sibling with type 1 diabetes vs those without this type 1 diabetes family history in the older age group), and rs1004446_a (INS) (HR 0.63, $p=0.007$), rs2476601_A (PTPN22) and rs10517086_A (HR 1.49, $p=0.020$) in the younger age group and rs3825932_T (CTSH) (HR 0.60, $p=0.005$) and rs73043122_C (RNASET2) (HR 2.58, $p=0.010$) in the older age group.

Discussion

The proportion of children who were diagnosed with type 1 diabetes differed by geography between the younger and older age groups, despite the commonality of the high-risk genotypes across the TEDDY sites. Life-table analysis revealed that, over time, diabetes risk among the younger children residing in Finland was greatest, but declined proportionally as the children got older. Diabetes risk in the other sites remained relatively constant; the USA together with Sweden and Finland together with Germany were comparable, but the two country pairs were different. Older children more often presented with GADA as the first-appearing autoantibody and had a different HLA genotype (significantly less HLA DR3/3) than those who developed type 1 diabetes at an earlier age who predominantly presented with IAA as the first-appearing autoantibody. The number of children presenting with IAA as the first-appearing autoantibody declined with increasing age, whereas the incidence of GADA as the first-appearing autoantibody remained relatively constant over the age range. These results confirm earlier findings [e.g., the German BABYDIAB study (15), and the Finnish DIPP study (16,17)] even in this HLA-defined high-risk population.

A new finding was that diabetes risk after the appearance of multiple autoantibodies did not differ between type 1 diabetes-affected and unaffected families, nor did the relationship of the affected family member to the TEDDY child. Initial seroconversion before 2 years of age was accompanied by higher diabetes risk after the appearance of multiple autoantibodies, but in children whose initial seroconversion to autoantibody positive was after 2 years of age, the risk did not decrease with increasing age of initial seroconversion. To complete the picture, the risk of initial seroconversion and progression to multiple autoantibodies decreased with increasing age, but once multiple autoantibodies were detected, the risk of progression did not decline with increasing age at seroconversion. Coupled with the changing picture of autoantibody presentation, it appears that autoimmunity at an early age is a more aggressive form of the disease.

These results underline the importance of taking into account the age at development of multiple autoantibodies when evaluating risk factors for progression to diabetes diagnosis. The HLA genotype, most SNPs, and family history were not significantly associated with early vs late diabetes diagnosis when taking into account the age at seroconversion to multiple autoantibodies. The implication is that they are risk factors for islet autoimmunity, but not necessarily for progression to diabetes once multiple autoantibodies have been observed.

Type 1 diabetes risk factors were relatively consistent across the two age groups. Some reached statistical significance in one age group but not in the other, but the difference in HRs between the age groups was not statistically significant. This could be an artifact

caused by the age groupings. Nonetheless, it showcases the caution that should be exercised when trying to generalise findings beyond the population actually studied. The other important consideration is that the strength (i.e., the magnitude of the estimate of the HR along with its statistical significance) is derived from a multivariate analysis, which, by definition, adjusts for all the variables considered in the model. Hence, the results reflect the additive or independent contribution of each variable considered after the contribution of all the other variables have been considered. Hence, findings that are nonsignificant, or only marginally significant, might be completely different if all the variables in the model were independent (uncorrelated).

This study is not without limitations. The characteristics of children who develop type 1 diabetes in the TEDDY study might not be generalisable to other HLA-defined populations. Despite this study's size of 8502 children, there are still relatively few type 1 diabetes cases. Yet, the before and after age 6 groups consist of nearly exactly the same number of children. This suggests that differences between the two age-defined cohorts were not due to an imbalance in the numbers.

The predominance of IAA first as the single presenting autoantibody in the very young age group compared with the predominance of GADA-first as the single presenting autoantibody in the older age groups is entirely consistent with the observed age-related association of exposures and both HLA and non-HLA genotypes. However, caution should be exercised in generalising the results presented here beyond the age range in which they have been discovered and the selected high-risk HLA subgroups that make up the TEDDY population. Yet, the TEDDY children represent, depending on country, about 40–50% of children expected to develop diabetes before 18 years of age (3). The age at screening for the presence of islet autoantibodies in both the general population or among FDRs and its associated heterogeneity with respect to diabetes risk factors (18,19) should be taken into account when individuals are randomised in secondary prevention studies of type 1 diabetes.

Finally, the report of study findings in a large epidemiological study, like TEDDY, involves many statistical comparisons, increasing the chance of spurious findings. In our opinion, using multivariate analyses to incorporate adjustments is a practical approach to this issue, whereas adjusting the significance threshold for multiple comparisons is not. There are arguments to be made for and against these approaches (20–22). We have chosen to not make adjustments for multiple comparisons and advise readers to consider these findings in the context of the published literature on similar populations.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgements

The authors thank Sarah Austin-Gonzalez with the Health Informatics Institute at the University of South Florida for assistance with preparing the figures.

A special acknowledgement to the TEDDY families for their continued participation in this wonderful study.

Funding

Funded by U01 DK63829, U01 DK63861, U01 DK63821, U01 DK63865, U01 DK63863, U01 DK63836, U01 DK63790, UC4 DK63829, UC4 DK63861, UC4 DK63821, UC4 DK63865, UC4 DK63863, UC4 DK63836, UC4 DK95300, UC4 DK100238, UC4 DK106955, UC4 DK112243, UC4 DK117483, U01 DK124166 and Contract No. HHSN267200700014C from the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), National Institute of Allergy and Infectious Diseases (NIAID), Eunice Kennedy Shriver National Institute of Child Health and Human Development (NICHD), National Institute of Environmental Health Sciences (NIEHS), JDRF, and Centers for Disease Control and Prevention (CDC). This work supported in part by the NIH/NCATS Clinical and Translational Science Awards to the University of Florida (UL1 TR000064) and the University of Colorado (UL1 TR002535).

Data availability

The datasets generated and analysed during the current study will be made available in the NIDDK Central Repository at <https://www.niddkrepository.org/studies/teddy>.

Abbreviations

FDR	First-degree relative
GADA	GAD autoantibodies
IAA	Insulin autoantibodies
IA-2A	Insulinoma antigen-2
PH	Proportional hazard
TEDDY	The Environmental Determinants of Diabetes in the Young

References

1. TEDDY Study Group. The Environmental Determinants of Diabetes in the Young (TEDDY) study: study design. *Pediatr Diabetes*2007;8:286–298 [PubMed: 17850472]
2. TEDDY Study Group. The Environmental Determinants of Diabetes in the Young (TEDDY) study. *Ann N Y Acad Sci*2008;1150:1–13
3. Hagopian WA, Erlich H, Lernmark Å, et al. TEDDY Study Group. The Environmental Determinants of Diabetes in the Young (TEDDY): genetic criteria and international diabetes risk screening of 421 000 infants. *Pediatr Diabetes*2011;12:733–743 [PubMed: 21564455]
4. Krischer JP, Lynch KF, Schatz DA, et al. TEDDY Study Group. The 6 year incidence of diabetes-associated autoantibodies in genetically at-risk children: the TEDDY study. *Diabetologia*2015;58:980–987 [PubMed: 25660258]
5. Krischer JP, Lynch KF, Lernmark Å, et al. TEDDY Study Group. Genetic and Environmental Interactions Modify the Risk of Diabetes-Related Autoimmunity by 6 Years of Age: The TEDDY Study. *Diabetes Care*2017;40:1194–1202 [PubMed: 28646072]
6. Parkes M, Cortes A, van Heel DA, Brown MA. Genetic insights into common pathways and complex relationships among immune-mediated diseases. *Nat Rev Genet*2013;14:661–673 [PubMed: 23917628]
7. Törn C, Hadley D, Lee HS, et al. TEDDY Study Group. Role of type 1 diabetes associated SNPs on risk of autoantibody positivity in the TEDDY study. *Diabetes*2015;64:1818–1829 [PubMed: 25422107]
8. Bonifacio E, Yu L, Williams AK, et al. Harmonization of glutamic acid decarboxylase and islet antigen-2 autoantibody assays for national institute of diabetes and digestive and kidney diseases consortia. *J Clin Endocrinol Metab*2010; 95:3360–3367 [PubMed: 20444913]

9. Lee HS, Burkhardt BR, McLeod W, et al. the TEDDY Study Group: Biomarker Discovery Study Design for Type 1 Diabetes in The Environmental Determinants of Diabetes in the Young (TEDDY) Study. *Diabetes Metab Res Rev*30(5):424–34, 2014. [PubMed: 24339168]
10. Uusitalo U, Lee HS, Andren Aronsson C, et al. TEDDY Study Group. Early Infant Diet and Islet Autoimmunity in the TEDDY Study. *Diabetes Care*2018;41:522–530 [PubMed: 29343517]
11. Elding Larsson H, Vehik K, Haller MJ, et al. TEDDY Study Group. Growth and risk for islet autoimmunity and progression to type 1 diabetes in early childhood: The Environmental Determinants of Diabetes in the Young Study. *Diabetes*2016;65:1988–95 [PubMed: 26993064]
12. Sharma A, Liu X, Hadley D, et al. TEDDY Study Group. Identification of non-HLA genes associated with development of islet autoimmunity and type 1 diabetes in the prospective TEDDY cohort. *J Autoimmun*2018;89:90–100 [PubMed: 29310926]
13. Cantor AB. Extending SAS survival analysis techniques for medical research, 2nd ed. SAS Institute, 2003
14. Concannon P, Chen WM, Julier C, et al. Genome-wide scan for linkage to type 1 diabetes in 2,496 multiplex families from the Type 1 diabetes Genetics Consortium. *Diabetes*2009;58:1018–1022 [PubMed: 19136655]
15. Hummel M, Bonifacio E, Schmid S, et al. Brief communication: early appearance of islet autoantibodies predicts childhood type 1 diabetes in offspring of diabetic parents. *Ann Intern Med*2004;140:882–886. [PubMed: 15172902]
16. Bauer W, Veijola R, Lempainen J, et al. Age at Seroconversion, HLA Genotype and Specificity of Autoantibodies in Progression of Islet Autoimmunity in Childhood. *J Clin Endocrinol Metab*2019;104:4521–4530 [PubMed: 31120497]
17. Ilonen J, Hammais A, Laine AP, et al. Patterns of β -cell autoantibody appearance and genetic associations during the first years of life. *Diabetes*2013;62:3636–40 [PubMed: 23835325]
18. Frederiksen BN, Kroehl M, Barón A, et al. Assessing age-related etiologic heterogeneity in the onset of islet autoimmunity. *Biomed Res Int*2015;2015:708289 [PubMed: 25883970]
19. Xu P, Krischer JP; Type 1 Diabetes TrialNet Study Group. Prognostic Classification Factors Associated With Development of Multiple Autoantibodies, Dysglycemia, and Type 1 Diabetes-A Recursive Partitioning Analysis. *Diabetes Care*2016;39:1036–44 [PubMed: 27208341]
20. Thompson JR. Invited commentary: Re: “Multiple comparisons and related issues in the interpretation of epidemiologic data.” *Am J Epidemiol* 1998;147:801–806 [PubMed: 9583708]
21. Savitz DA and Olshan AF. Multiple Comparisons and Related Issues in the Interpretation of Epidemiologic Data. *Am J Epidemiol* 1995;142:904–908 [PubMed: 7572970]
22. Savitz DA and Olshan AF. Describing data requires no adjustment for multiple comparisons: a reply from Savitz and Olshan. *Am J Epidemiol* 1998;147:813–814 [PubMed: 9583710]

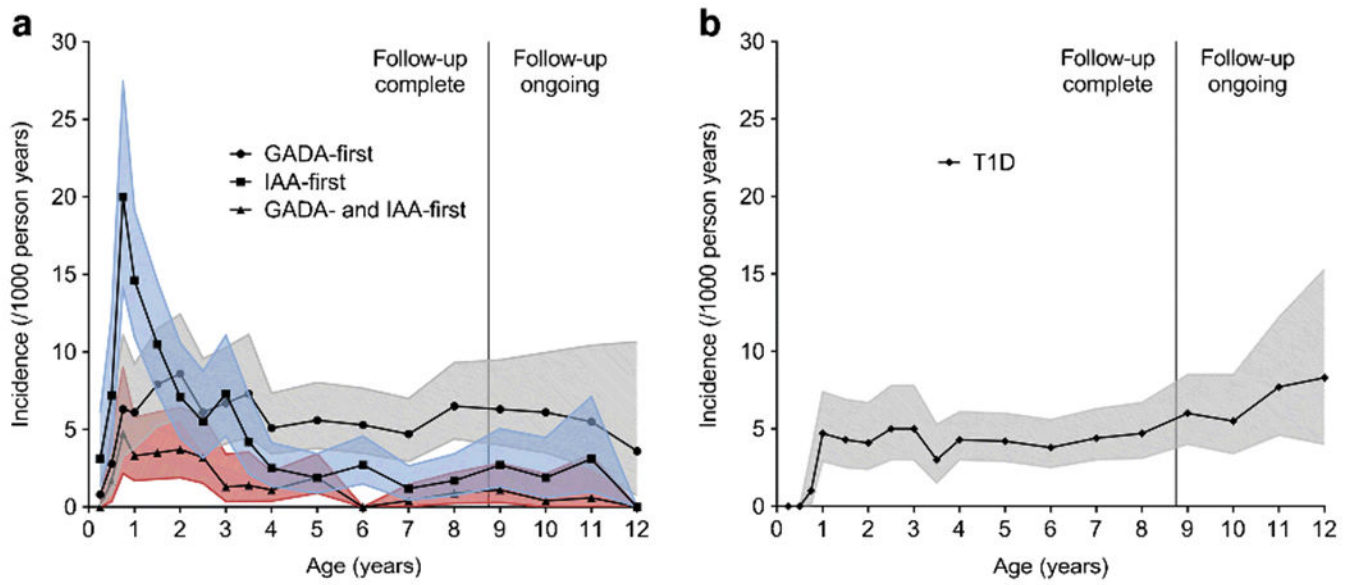
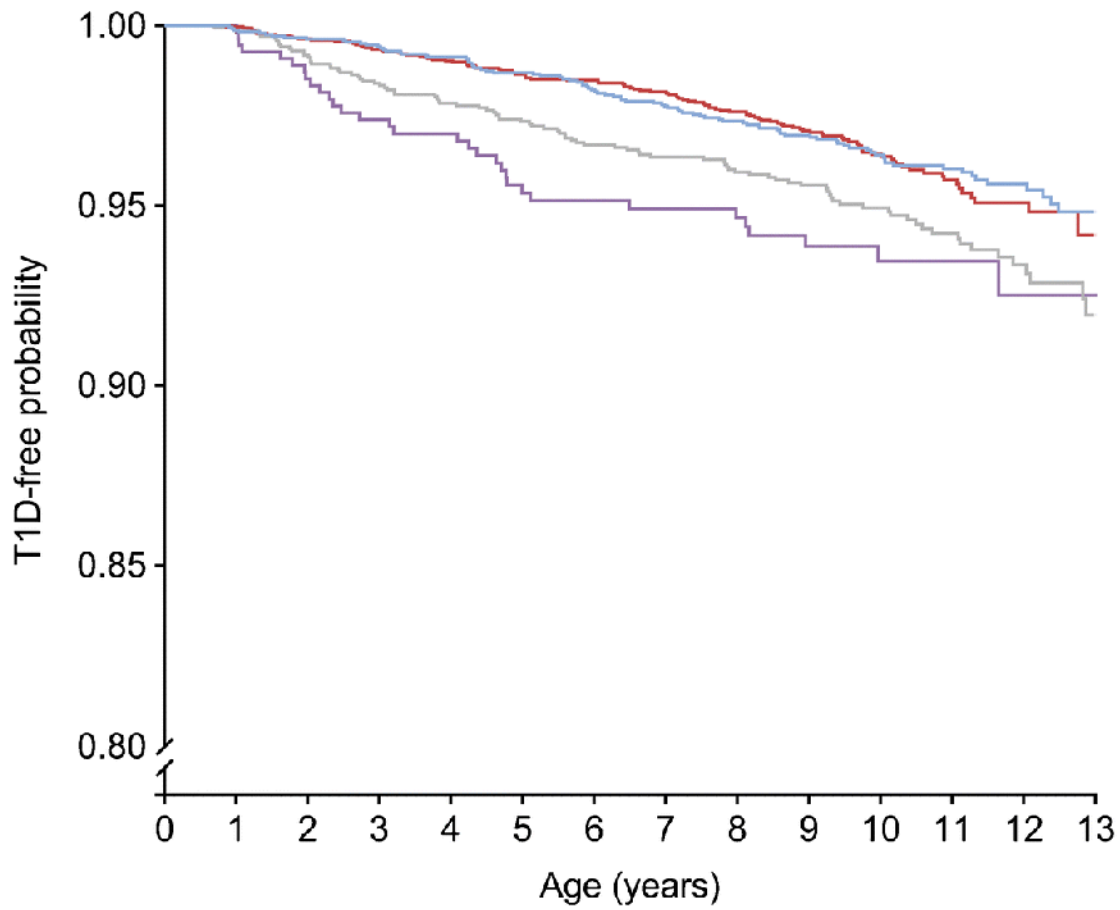


Fig. 1. Type 1 diabetes incidence by age-specific incidence of islet cell autoantibodies (a) and type 1 diabetes (b). T1D, type 1 diabetes



Number at risk:					
1803	1618	1467	1210	404	Finland —
574	505	433	318	79	Germany —
2498	2366	2222	1837	620	Sweden —
3627	3095	2813	2157	416	USA —

Fig. 2. Kaplan–Meier curve of type 1 diabetes (across all ages) by country of origin ($p=0.0009$ from logrank test). T1D, type 1 diabetes

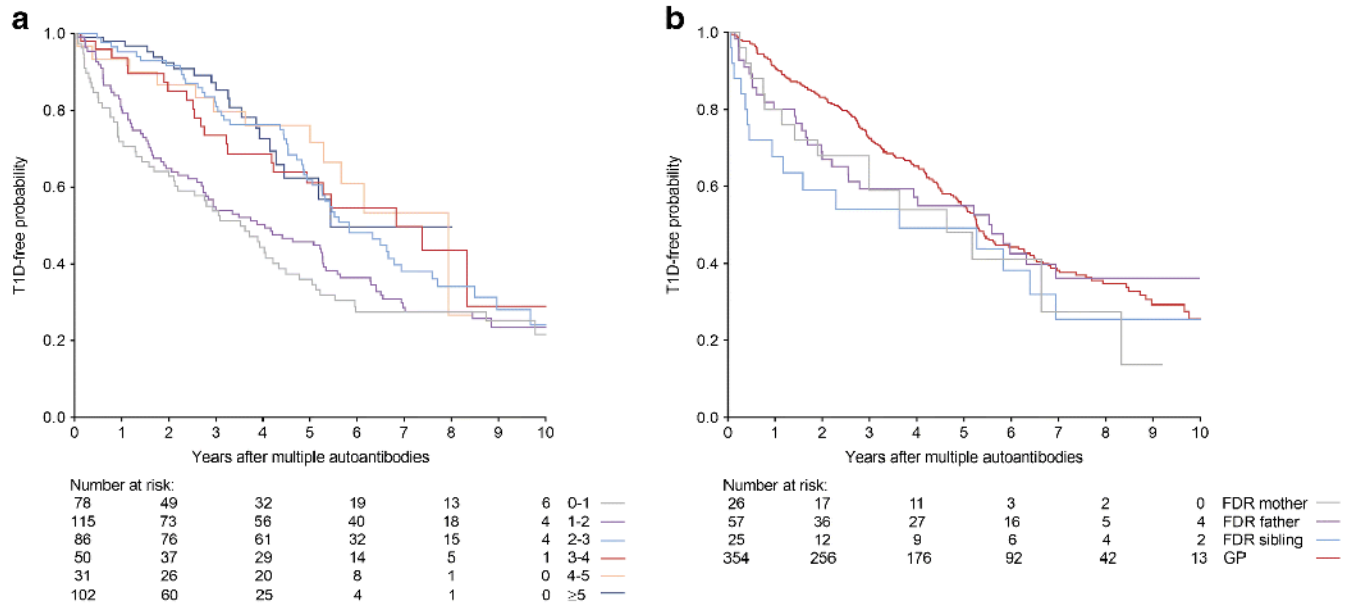


Fig. 3. Type 1 diabetes risk since developing multiple autoantibodies by **(a)** age of initial seroconversion and **(b)** family history of type 1 diabetes. T1D, type 1 diabetes

Table 1

Characteristics of TEDDY children

Characteristic	No type 1 diabetes (n=8174)	Type 1 diabetes before 6 years of age (n=168)	Type 1 diabetes 6 years of age (n=160)	p value ^a
Country				
USA	3515 (43.0)	47 (28.0)	65 (40.6)	0.001
Finland	1711 (20.9)	54 (32.1)	38 (23.8)	
Germany	542 (6.6)	25 (14.9)	7 (4.4)	
Sweden	2406 (29.4)	42 (25.0)	50 (31.3)	
Family history				
FDR: Mother	315 (3.9)	10 (6.0)	13 (8.1)	0.106
FDR: Father	408 (5.0)	30 (17.9)	14 (8.8)	
FDR: Sibling	123 (1.5)	11 (6.5)	12 (7.5)	
General population	7328 (89.7)	117 (69.6)	121 (75.6)	
Sex				
Male	4142 (50.7)	85 (50.6)	84 (52.5)	0.730
Female	4032 (49.3)	83 (49.4)	76 (47.5)	
HLA genotype				
DR3/4	3140 (38.4)	96 (57.1)	82 (51.3)	0.005
DR4/4	1601 (19.6)	20 (11.9)	40 (25.0)	
DR4/8	1423 (17.4)	21 (12.5)	24 (15.0)	
DR3/3	1754 (21.5)	18 (10.7)	10 (6.3)	
FDR-specific ^b	256 (3.1)	13 (7.7)	4 (2.5)	
Probiotics introduction age				
28 days	7615 (93.2)	153 (91.1)	151 (94.4)	0.251
< 28 days	559 (6.8)	15 (8.9)	9 (5.6)	
Weight z score at 12 months				
n	7460	161	149	
Mean (SD)	-0.1 (1.0)	-0.0 (1.1)	0.0 (1.0)	
Median (IQR)	-0.1 (-0.8-0.6)	0.0 (-0.8-0.7)	-0.0 (-0.6-0.7)	0.582

Characteristic	No type 1 diabetes (n=8174)	Type 1 diabetes before 6 years of age (n=168)	Type 1 diabetes 6 years of age (n=160)	p value ^a
rs1004446_A (JNS)				
No	2946 (39.5)	84 (52.5)	73 (49.0)	0.538
Yes	4521 (60.5)	76 (47.5)	76 (51.0)	
rs2476601_A (PTPN22)				
No	5979 (80.1)	102 (63.8)	102 (68.5)	0.383
Yes	1488 (19.9)	58 (36.3)	47 (31.5)	
rs10517086_A				
No	3839 (51.4)	62 (38.8)	73 (49.0)	0.070
Yes	3628 (48.6)	98 (61.3)	76 (51.0)	
rs2292239_T (ERBB3)				
No	3438 (46.0)	52 (32.5)	67 (45.0)	0.024
Yes	4028 (54.0)	108 (67.5)	82 (55.0)	
rs3184504_T (SH2B3)				
No	2314 (31.0)	41 (25.6)	43 (28.9)	0.523
Yes	5153 (69.0)	119 (74.4)	106 (71.1)	
rs12708716_G (CLEC16A)				
No	3278 (44.0)	78 (48.8)	82 (55.0)	0.269
Yes	4170 (56.0)	82 (51.3)	67 (45.0)	
rs3825932_T (CTSH)				
No	3079 (41.2)	80 (50.0)	75 (50.3)	0.953
Yes	4387 (58.8)	80 (50.0)	74 (49.7)	
rs7111341_T (JNS)				
No	4031 (54.0)	104 (65.0)	91 (61.1)	0.475
Yes	3436 (46.0)	56 (35.0)	58 (38.9)	
rs11711054_G (CCR5)				
No	3694 (49.5)	65 (40.6)	69 (46.3)	0.314
Yes	3765 (50.5)	95 (59.4)	80 (53.7)	
rs428595_A (PPIL2)				
No	7134 (96.4)	145 (91.8)	133 (92.4)	0.850

Characteristic	No type 1 diabetes (n=8174)	Type 1 diabetes before 6 years of age (n=168)	Type 1 diabetes 6 years of age (n=160)	p value ^a
Yes	266 (3.6)	13 (8.2)	11 (7.6)	
rs113306148_T (PLEKHA1)				
No	7277 (97.5)	149 (93.1)	140 (94.0)	0.766
Yes	190 (2.5)	11 (6.9)	9 (6.0)	
rs73043122_C (RNASET2)				
No	7263 (97.3)	151 (95.0)	139 (93.3)	0.530
Yes	202 (2.7)	8 (5.0)	10 (6.7)	
Age at type 1 diabetes diagnosis (years)				
n		168	160	
Mean (SD)		3.3 (1.5)	8.9 (1.8)	
Median (IQR)		3.1 (2.0–4.6)	8.7 (7.5–10.1)	<0.001
Age at persistent confirmed autoantibody (years)				
n	499	154	138	
Mean (SD)	5.1 (3.1)	1.5 (0.8)	3.5 (2.4)	
Median (IQR)	4.6 (2.3–7.7)	1.2 (0.8–1.8)	2.6 (1.9–4.5)	<0.001
Duration of time between first-appearing autoantibody and diagnosis (years)				
n		154	138	
Mean (SD)		1.9 (1.4)	5.4 (2.5)	
Median (IQR)		1.6 (0.6–2.9)	5.4 (3.9–6.8)	<0.001
Age at multiple persistent confirmed autoantibodies (years)				
n	208	133	121	
Mean (SD)	5.7 (3.0)	1.8 (0.9)	3.9 (2.1)	
Median (IQR)	5.5 (3.1–8.4)	1.5 (1.1–2.2)	3.3 (2.3–5.1)	<0.001
Duration of time between multiple appearing autoantibodies and diagnosis (years)				
n		133	121	
Mean (SD)		1.6 (1.3)	4.9 (2.3)	
Median (IQR)		1.3 (0.6–2.7)	5.1 (3.3–6.3)	<0.001
Autoantibody status at seroconversion				

Characteristic	No type 1 diabetes (n=8174)	Type 1 diabetes before 6 years of age (n=168)	Type 1 diabetes 6 years of age (n=160)	p value ^a
None	7675 (93.9)	14 (8.3)	22 (13.8)	0.001
GADA only	272 (3.3)	31 (18.5)	43 (26.9)	
IA2A only	11 (0.1)	0 (0.0)	10 (6.3)	
IAA only	174 (2.1)	75 (44.6)	45 (28.1)	
GADA, IA2A	5 (0.1)	1 (0.6)	3 (1.9)	
GADA, IAA	29 (0.4)	36 (21.4)	29 (18.1)	
IA2A, IAA	4 (0.0)	1 (0.6)	2 (1.3)	
GADA, IA2A, IAA	4 (0.0)	10 (6.0)	6 (3.8)	
Persistent confirmed GADA before or at type 1 diabetes diagnosis				
No		50 (29.8)	45 (28.1)	0.744
Yes		118 (70.2)	115 (71.9)	
Persistent confirmed IAA before or at type 1 diabetes diagnosis				
No		22 (13.1)	46 (28.8)	<0.001
Yes		146 (86.9)	114 (71.3)	
Persistent confirmed IA2A before or at type 1 diabetes diagnosis				
No		64 (38.1)	46 (28.8)	0.073
Yes		104 (61.9)	114 (71.3)	
Persistent confirmed ZnT8A before or at type 1 diabetes diagnosis				
No		104 (61.9)	55 (34.4)	<0.001
Yes		64 (38.1)	105 (65.6)	
DKA at type 1 diabetes diagnosis				
No		151 (89.9)	153 (95.6)	0.046
Yes		17 (10.1)	7 (4.4)	
Symptomatic at type 1 diabetes diagnosis				
No		68 (40.5)	76 (47.5)	0.200
Yes		100 (59.5)	84 (52.5)	

Data are presented as number (percentage) unless otherwise indicated

For each SNP, the minor allele is indicated and data are divided by children with (yes) and without this allele (no)

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

p value of comparing each characteristic between individuals with type 1 diabetes before 6 years of age and individuals with type 1 diabetes 6 years of age. Wilcoxon's rank tests were performed for continuous variables and χ^2 tests were performed for categorical variables

q FDR-specific HLA-DR-DQ genotypes are DR4/4b, DR4/1, DR4/13, DR4/9, and DR3/9

Table 2
HR, adjusted for covariates from the multiple PH analysis, of TEDDY published risk factors for type 1 diabetes

Factor	Comparison	Type 1 diabetes before 6 years of age			Type 1 diabetes 6 years of age		
		HR (95% CI)	p value	HR (95% CI)	p value	p value ^a	
Sex	Male vs female	1.06 (0.77, 1.46)	0.715	1.12 (0.80, 1.57)	0.507	0.819	
Family history of type 1 Diabetes	FDR mother vs GP	1.51 (0.75, 3.05)	0.249	2.64 (1.36, 5.12)	0.004	0.256	
	FDR father vs GP	3.24 (1.99, 5.26)	<0.001	2.41 (1.31, 4.41)	0.005	0.452	
	FDR sibling vs GP	4.55 (2.40, 8.62)	<0.001	5.13 (2.69, 9.80)	<0.001	0.795	
HLA genotype	DR4/4 vs DR3/3	1.26 (0.65, 2.44)	0.496	4.16 (1.99, 8.69)	<0.001	0.018	
	DR3/4 vs DR3/3	2.79 (1.63, 4.77)	<0.001	4.40 (2.19, 8.82)	<0.001	0.311	
	DR4/8 vs DR3/3	1.50 (0.77, 2.93)	0.237	3.36 (1.52, 7.45)	0.003	0.127	
	FDR-specific ^b vs DR3/3	1.60 (0.69, 3.70)	0.269	1.29 (0.38, 4.44)	0.683	0.777	
Country	Finland vs USA	3.20 (1.88, 5.45)	<0.001	1.26 (0.73, 2.18)	0.416	0.005	
	Germany vs USA	2.19 (1.27, 3.78)	0.005	0.56 (0.25, 1.27)	0.167	0.007	
	Sweden vs USA	1.20 (0.76, 1.89)	0.436	0.72 (0.47, 1.09)	0.123	0.098	
Probiotics introduction age	<28 days vs 28 days	0.86 (0.49, 1.51)	0.606	0.94 (0.46, 1.94)	0.871	0.852	
Weight at 12 months	z-score	1.10 (0.94, 1.30)	0.223	1.21 (1.03, 1.44)	0.024	0.426	
Child conditions before first clinical visit (age 3 months)	Upper resp. (Yes vs No)	1.43 (1.00, 2.04)	0.053	1.00 (0.67, 1.50)	0.999	0.198	
	Rash (Yes vs No)	0.66 (0.42, 1.04)	0.076	0.93 (0.62, 1.41)	0.747	0.275	
rs1004446_A (INS)	Yes vs No	0.67 (0.48, 0.92)	0.014	0.67 (0.48, 0.95)	0.022	0.964	
rs2476601_A (PTPN22)	Yes vs No	2.04 (1.46, 2.84)	<0.001	1.72 (1.20, 2.47)	0.003	0.503	
rs10517086_A	Yes vs No	1.48 (1.07, 2.05)	0.019	1.04 (0.75, 1.45)	0.808	0.141	
rs2292239_T (ERBB3)	Yes vs No	1.67 (1.19, 2.34)	0.003	1.06 (0.76, 1.48)	0.740	0.063	
rs3184504_T (SH2B3)	Yes vs No	1.23 (0.85, 1.77)	0.275	1.07 (0.74, 1.55)	0.731	0.599	

Factor	Comparison	Type 1 diabetes before 6 years of age		Type 1 diabetes 6 years of age	
		HR (95% CI)	p value	HR (95% CI)	p value
rs12708716_G (CLEC16A)	Yes vs No	0.82 (0.60, 1.13)	0.222	0.64 (0.46, 0.89)	0.009
rs3825932_T (CTSH)	Yes vs No	0.76 (0.55, 1.04)	0.087	0.66 (0.47, 0.93)	0.017
rs7111341_T (INS)	Yes vs No	0.68 (0.48, 0.96)	0.026	0.77 (0.54, 1.09)	0.142
rs11711054_G (CCR5)	Yes vs No	1.46 (1.06, 2.02)	0.021	1.15 (0.83, 1.61)	0.404
rs428595_A (PPIL2)	Yes vs No	2.13 (1.20, 3.80)	0.010	2.10 (1.13, 3.92)	0.019
rs113306148_T (PLEKHA1)	Yes vs No	2.34 (1.25, 4.38)	0.008	2.21 (1.07, 4.57)	0.032
rs73043122_C (RNASET2)	Yes vs No	2.31 (1.12, 4.76)	0.023	2.54 (1.27, 5.06)	0.008

7433 individuals with 296 type 1 diabetes events (155 in the first 6 years and 141 after 6 years of age) were included in the analysis because values of SNPs and/or weight at 12 months of age were missing. For each SNP, the minor allele is indicated and children with this allele (yes) were compared with children without it (no)

^a p value for comparing the HR in the first 6 years vs the HR after 6 years of age

^b FDR-specific HLA-DR-DQ genotypes are DR4/4b, DR4/1, DR4/13, DR4/9, and DR3/9

Upper resp., upper respiratory tract infection