Better HbA1c during the first years after diagnosis of type 1 diabetes is associated with residual C peptide 10 years later

Annika Grönberg [ID],[1] Daniel Espes,[2,3] Per-Ola Carlsson[2,3]

[1]Department of Women's and Children's Health, Uppsala Universitet Institutionen for kvinnors och barns halsa, Uppsala, Sweden
[2]Department of Medical Cell Biology, Uppsala University, Uppsala, Sweden
[3]Department of Medical Sciences, Uppsala University, Uppsala, Sweden

Correspondence to
Dr Annika Grönberg;
annika.gronberg@kbh.uu.se

ABSTRACT

Objective To identify the factors associated with residual C peptide production at least 10 years after diagnosis in children and adolescents with type 1 diabetes.

Research design and methods 73 children and adolescents (<25 years), born in 1988–2005, diagnosed with type 1 diabetes were included during the 4-year study period (2013–2016). At least 10 years after diagnosis, we measured any remaining C peptide concentration using an ultrasensitive C peptide ELISA (≥1.17 pmol/L). The average hemoglobin A1c (HbA1c) was calculated during each of the 10 years after diagnosis and further grand average was calculated for the entire study period.

Results C peptide was detectable in 38% of participants. The C peptide concentration was 4.3±5.3 pmol/L. At onset of type 1 diabetes, participants were on average approximately 5 years of age, and their average HbA1c was 9.4% (79 mmol/mol). During the first 3 years after diagnosis, HbA1c was lower in the group with detectable C peptide at follow-up ≥10 years later. Moreover, detectable C peptide was more common among female participants. Body mass index SD scores had not increased since the 1-year follow-up, but were higher in patients with measurable C peptide. Nine participants (12%) had been diagnosed with celiac disease and two (3%) with hypothyreosis. Eighteen (25%) participants had retinopathy.

Conclusions Children and adolescents with detectable C peptide after more than 10 years of diabetes duration were predominantly female and had better HbA1c than others during the first 3 years after diagnosis.

INTRODUCTION

The development of ultrasensitive methods for C peptide analysis has led to findings that show glucose-responsive residual insulin production may occur for decades after diagnosis of type 1 diabetes.[1–3] Although most patients 10 years after onset of disease do not have levels of residual insulin secretion high enough to lower mean blood glucose concentration and provide less blood glucose fluctuations,[4] surviving immune-resistant beta cells are an attractive target for new protocols aiming to expand beta cell mass.

Significance of this study

What is already known about this subject?
► Some immunological factors and genotypes have been associated with long-term preserved residual beta cell function.

What are the new findings?
► Female adolescents and children with better hemoglobin A1c during the first 3 years after diagnosis were more likely to have preserved beta cell function more than 10 years later.

How might these results change the focus of research or clinical practice?
► The findings emphasize strategies to implement strict glycemic control during the first years after diagnosis of type 1 diabetes to preserve long-standing residual beta cell mass.

There is little knowledge on the factors that correlate to a long-term preserved residual beta cell mass in type 1 diabetes. However, we recently identified immunological factors, particularly higher production of interleukin-35 by immune cells, in patients with residual C peptide for more than 10 years.[5] Also the higher frequency of the HLADR3 genotype[1] and the level of ZnT8 autoantibodies[2] have been correlated with C peptide concentrations remaining for decades.

The present study took advantage of the unique longitudinal follow-up data of the Swedish Childhood Diabetes Registry SWEDIABKIDS and combined these with participants' records, aiming to identify other potential factors, aside from immunological factors, that are associated with long-standing preservation of beta cell function in type 1 diabetes.

RESEARCH DESIGN AND METHODS
Participants

Adolescents and young adults with at least 10 years' duration of type 1 diabetes were recruited from the Uppsala University

Children's Hospital (n=51) and the Department of Endocrinology and Diabetology, Uppsala University Hospital (n=22) in 2013–2016. All included patients were born in 1988–2005 and presented with type 1 diabetes in 1991–2007. All participants had been hospitalized at the Children's Hospital at onset of disease and were started on multiple insulin injection therapy (MIT), usually with human insulin (Actrapid and Insulatard). During the course of treatment, most of the participants had been switched to insulin aspart (NovoRapid) and insulin detemir (Levemir), or alternatively to insulin pump treatment. Follow-up was done at the Children's Hospital and according to the framework of the Swedish national program for pediatric diabetes.[6] At the age of 18, participants were transferred to the diabetes service of the Department of Endocrinology and Diabetology. The clinical diagnosis of type 1 diabetes did not need to be revised in any of the participants. Any presence of retinopathy was assessed every second year, with assessment becoming more frequent on occurrence of changes and with participants being 10 years of age.

Procedure and biochemical analyses
Non-fasting blood samples were obtained during regular follow-up visits to respective diabetes services. Hemoglobin A1c (HbA1c), serum creatinine, blood glucose, glutamic acid decarboxylase antibodies (GADA; detection limit 5 IU/mL) and islet antigen-2 antibodies (IA-2A; detection limit 7.5 kU/L) were analyzed according to standards and routine at the Department of Clinical Chemistry at Uppsala University Hospital. The laboratory is certified by a Swedish government authority (Swedac). To analyze C peptide concentrations, plasma was separated from samples of EDTA blood and stored frozen at −80°C until analysis. Plasma C peptide concentrations were analyzed by Mercodia (Uppsala, Sweden) using an ultrasensitive ELISA (catalog no 10-1141-01; Mercodia). The assay was calibrated against the international reference reagent for C peptide, C peptide 84/510, which is the WHO standard and is categorized by the US Food and Drug Administration as class I in vitro diagnostic device. The detection limit of the assay is set to 1.17 pmol/L, with interassay and intra-assay coefficients of variation of 5.5% and 3.8% at 37 pmol/L.

Data collection
Data were collected from patient records and the SWEDIABKIDS, a national incidence and quality control register.[7]

From SWEDIABKIDS, baseline data for each patient at the time of diagnosis of type 1 diabetes were collected. These included age, gender and comorbid diseases diagnosed before the onset of diabetes, weight, height, HbA1c and blood pH at the time of admission, and weight, height, HbA1c, insulin dose (units/kg body weight/day) and use of insulin pump registered at follow-up visits. Body mass index (BMI) (kg/m^2) and BMI SD scores (BMI SDS) are generated automatically by the SWEDIABKIDS register.[8]

The presence of retinopathy and persistent microalbuminuria are also registered.

From October 2010, HbA1c has been analyzed according to the International Federation of Clinical Chemistry (IFCC) standard and expressed as mmol/mol. Prior to this, analyses were according to the Mono S standard and expressed in percent. Analyses performed with the Mono S standard were recalculated using the following expression: HbA1c (IFCC; mmol/mol)=10.45×HbA1c (Mono S; %) − 10.62. Mono S standard was also recalculated according to the Diabetes Control and Complications Trial (DCCT) standard. HbA1c was finally expressed following both DCCT and IFCC standards (http://www.diabetes.co.uk).

For the first 10 years of follow-up, the average HbA1c was calculated for each individual year. For each year, one to six (median 3) HbA1c measurements were usually available. HbA1c grand average was calculated as the average of the 10 individual 1-year averages. A further grand average was calculated for the entire study period when participants had been followed up for more than 10 years.

Statistics
Statistical analyses were performed using SPSS V.20.0.0. Data are presented as mean±SD. Student's t-test for independent samples and χ^2 test were used to analyze continuous and categorical data, respectively. Predictors of detectable C peptide secretion as a binary outcome (detectable or not) were analyzed using univariate and multivariate logistic regression analyses, expressed with OR and 95% CI. The multivariate models included variables that were statistically significant at the univariate level (gender and celiac disease). BMI SDS was also significant, but due to the high proportion of missing data this was not included in the final model.

RESULTS
At diagnosis of type 1 diabetes, participants were on average approximately 5 years old (table 1) and 55% were male. The average HbA1c at diagnosis was as high as 9.4%±3.8% (79±18 mmol/mol), and six (8%) participants presented with ketoacidosis (pH <7.3). Seven (10%) participants started treatment in an intensive care unit. Three participants had previously been diagnosed with celiac disease and one with hypothyreosis.

At 1-year follow-up, glycemic control improved, with HbA1c of 6.6%±2.9% (49±9 mmol/mol). All participants but one were on MIT with a daily dose of insulin of 0.7±0.2 (U/kg).

At long-term follow-up, with diabetes duration of 12.3±2.4 years and age of 16.9±3.3 years, HbA1c had increased to 8.1%±3.1% (65±11 mmol/mol). The BMI SDS had not increased since the 1-year follow-up. Nine participants (12%) had now been diagnosed with celiac disease and two (3%) with hypothyreosis. The majority (56%) were on MIT, usually with insulin aspart

Table 1 Characteristics of 73 children and adolescents with type 1 diabetes followed up for at least 10 years

		Range
At diagnosis		
Male	40 (55%)	
Age (years)	5.1±3.0	0.6 to 12.5
BMI SDS	−0.81±1.44	−4.17 to 2.57
Blood glucose (mmol/L)	25.1±8.7	7.9 to 58
Hemoglobin A1c (%)	9.4±3.8	6.0 to 13.6
Hemoglobin A1c (mmol/mol)	79±18	42 to 125
Ketoacidosis (pH <7.30)	6 (8%)	
Intensive care	7 (10%)	
Previously diagnosed celiac disease	3 (4%)	
Previously diagnosed hypothyreosis	1 (1%)	
At 1-year follow-up		
Age (years)	6.2±3.0	1.6 to 13.7
Duration of diabetes (years)	1.0±0.1	0.8 to 1.4
BMI SDS	0.60±1.1	−2.35 to 2.71
Hemoglobin A1c (%)	6.6±2.9	5.3 to 9.8
Hemoglobin (mmol/mol)	49±9	34 to 84
Celiac disease	4 (5%)	
Hypothyreosis	1 (1%)	
Insulin treatment		
Multiple injection therapy	72 (99%)	
Insulin pump	1 (1%)	
Daily insulin dose (U/kg)	0.7±0.2	0.2 to 1.1
At long-term follow-up		
Age (years)	16.9±3.3	10.6 to 24.0
Duration of type 1 diabetes (years)	12.3±2.4	10 to 20
BMI SDS	0.52±1.06	−2.40 to 2.57
Hemoglobin A1c (%)	8.1±3.1	6.1 to 11.2
Hemoglobin A1c (mmol/mol)	65±11	43 to 99
Celiac disease	9 (12%)	
Hypothyreosis	2 (3%)	
Insulin treatment		
Multiple injection therapy	41 (56%)	
Insulin pump	32 (44%)	
Daily insulin dose (U/kg)	0.9±0.3	0.5 to 1.7
Retinopathy	18 (25%)	
Creatinine (µmol/L)	61±12	37 to 96
Blood glucose (mmol/L)	10.6±5.4	2.8 to 21.9
C peptide (pmol/L)	1.17±5.31	1.17 to 22.7
GADA detectable	32 (44%)	
IA-2A detectable	23 (32%)	
GADA and IA-2A antibodies detectable	10 (14%)	

Values are given as mean±SD (range).
BMI SDS, body mass index SD score; GADA, glutamic acid decarboxylase antibodies; IA-2A, islet antigen-2 antibodies.

(NovoRapid) and insulin detemir (Levemir). The daily dose of insulin was 0.9±0.3 (U/kg). Eighteen (25%) participants had retinopathy, with 16 classified as having mild retinopathy and 2 as having moderate retinopathy.

Serum creatinine was within the reference range for young adults (men: 60–105 µmol/L; women: 45–90 µmol/L). Urine albumin secretion had not been consistently monitored during the follow-up. GADA was detectable in 32 (44%) participants and IA-2A in 23 (32%), and both GADA and IA-2A were detectable in 10 (14%).

At long-term follow-up, C peptide was detectable (≥1.17 pmol/L) in 28 (38%) participants. In these participants, the C peptide concentration was 4.3±5.3 pmol/L, ranging from 1.17 to 22.7 pmol/L (table 2). C peptide was detectable in a larger proportion of female participants compared with male participants, while age, BMI SDS, HbA1c, presence of ketoacidosis, or GADA and IA-2A at diagnosis were not related to whether C peptide was detectable ≥10 years later (table 2). The daily insulin needs at 1-year follow-up were similar in the groups of participants with or without detectable C peptide at long-term follow-up (0.7±0.2 U insulin/kg vs 0.7±0.2 U insulin/kg). During the course of treatment, HbA1c increased in both groups, but the participants with detectable C peptide at long-term follow-up had lower HbA1c during the first 3 years of treatment (table 2). The participants with detectable C peptide also had higher BMI SDS and celiac disease was more prevalent. There was a tendency of an increased prevalence of retinopathy in those without detectable C peptide (p=0.105). The two participants with retinopathy classified as moderate were found in the latter group.

An additional analysis was performed where participants were dichotomously divided into those with HbA1c <7.0% (53 mmol/mol) and those with HbA1c ≥7.0% (53 mmol/mol) during the first years in relation to the presence or absence of detectable C peptide at long-term follow-up. During the first 2 years, but not the third year, HbA1c <7.0% (53 mmol/mol) predicted retained C peptide secretion at long-term follow-up (X^2 (1, n=73)=6.6, p=0.01 for the first year; X^2 (1, n=73)=7.5, p=0.006 for the second year) (table 3).

To analyze predictors of retained C peptide secretion, the presence of detectable C peptide was used as the dependent variable in a logistic regression analysis. A 10-unit change in the first-year, second-year or third-year average HbA1c was used as independent variable and was for all these years highly predictive of detectable C peptide, such that lower HbA1c was associated with detectable C peptide (table 4). Female gender, but not celiac disease, independently predicted detectable C peptide when added to the model. Data on BMI SDS at 10-year follow-up were available for only 50 participants. When a one-unit change in BMI SDS at 10-year follow-up was added to the model in this subgroup, it independently predicted detectable C peptide, such that higher BMI SDS was associated with detectable C peptide (OR 3.00, 95% CI 1.15 to 7.85, p=0.025). Adding age at diagnosis, ketoacidosis at presentation, BMI SDS at 1-year follow-up, or the presence of GADA and/or IA-2A at 10-year follow-up to the models did not improve prediction.

Table 2 Characteristics of 73 children and adolescents with type 1 diabetes and detectable C peptide with at least 10 years of follow-up

Ultrasensitive C peptide (pmol/L) N=73	C peptide detectable 28	C peptide not detectable 45	P value
At diagnosis			
Male/female	11/17 (39%/61%)	29/16 (64%/36%)	0.036*
Age (years)	5.2±3.1	5.0±2.9	0.753
BMI SDS	−0.44±1.4	−1.01±1.4	0.086
Hemoglobin A1c % (mmol/mol)	9.4±4.0 (79±20)	9.4±3.7 (79±17)	0.917
Ketoacidosis (pH <7.30)	1 (4%)	5 (11%)	0.244
Average hemoglobin A1c % (mmol/mol)			
First year	6.1±2.6 (43±6)	6.5±2.8 (49±8)	0.003**
Second year	6.6±2.7 (49±7)	7.0±2.8 (53±8)	0.029*
Third year	6.8±2.8 (51±8)	7.2±2.9 (56±9)	0.026*
Fourth year	7.1±2.8 (54±8)	7.5±3.0 (58±10)	0.065
Fifth year	7.5±2.9 (58±9)	7.5±3.1 (59±11)	0.505
Sixth year	7.5±2.8 (59±8)	7.7±3.1 (61±11)	0.575
Seventh year	7.8±2.7 (62±7)	7.7±2.9 (61±9)	0.560
Eighth year	8.0±2.7 (64±7)	7.7±2.9 (61±9)	0.247
Ninth year	7.9±2.9 (63±9)	7.8±2.9 (62±9)	0.519
Tenth year	7.9±3.0 (63±10)	8.1±2.9 (65±9)	0.401
Ten-year grand average	7.4±2.6 (57±6)	7.5±2.7 (58±7)	0.260
Grand average for the entire study period	7.5±2.6 (58±6)	7.6±2.7 (60±7)	0.218
At long-term follow-up			
Age (years)	16.4±3.2	17.2±3.3	0.331
Duration of type 1 diabetes (years)	11.8±2.0	12.7±2.6	0.098
BMI SDS (n=50)	0.90±0.75	0.28±1.17	0.043*
Hemoglobin A1c % (mmol/mol)	8.0±3.3 (64±13)	8.1±2.9 (65±9)	0.774
Treatment with insulin pump	11 (39%)	21 (47%)	0.537
Celiac disease	7 (25%)	2 (4%)	0.041*
Hypothyreosis	0 (0%)	2 (4%)	0.527
Retinopathy	4 (14%)	14 (31%)	0.105
Creatinine (µmol/L)	58±12	62±12	0.077
Blood glucose (mmol/L)	10±6	11±5	0.460
GADA detectable	15 (54%)	17 (38%)	0.186
IA-2A detectable	10 (36%)	13 (29%)	0.542
GADA and IA-2A detectable	5 (18%)	5 (11%)	0.415

Values are given as mean±SD.
Significance of the difference between subjects with and without detectable C peptide (>1.17 pmol/L): *p<0.05, **p<0.01.
BMI SDS, body mass index SD score; GADA, glutamic acid decarboxylase antibodies; IA-2A, islet antigen-2 antibodies.

Table 3 Prediction of detectable C peptide in 73 children and adolescents with type 1 diabetes

Dependent variable	Independent variable	Odds	OR	P value	95% CI
Detectable C peptide	First-year average HbA1c <7%	0.87	6.5	0.01	1.4 to 31.1
Detectable C peptide	Second-year average HbA1c <7%	1.11	4.1	0.006	1.5 to 11.6
Detectable C peptide	Third-year average HbA1c <7%	0.94	2.1	0.13	0.8 to 5.5

HbA1c, hemoglobin A1c.

Table 4 Prediction of detectable C peptide in 73 children and adolescents with type 1 diabetes

Dependent variable	Independent variable	OR	95% CI	P value
Detectable C peptide	First-year average HbA1c	0.35	0.17 to 0.74	0.005
Detectable C peptide	First-year average HbA1c	0.3	0.13 to 0.68	0.004
	Gender	3.85	1.26 to 11.82	0.018
	Celiac disease	1.45	0.87 to 2.44	0.157
Detectable C peptide	Second-year average HbA1c	0.48	0.24 to 0.95	0.034
Detectable C peptide	Third-year average HbA1c	0.48	0.25 to 0.94	0.032

HbA1c, hemoglobin A1c.

Retinopathy was predicted by a 10-unit change in the 10-year grand average HbA1c (OR 4.42, 95% CI 1.48 to 13.2, p=0.008). Adding first-year average HbA1c or presence of detectable C peptide did not improve prediction.

DISCUSSION

Using an ultrasensitive C peptide assay with a detection limit in the low picomolar range, we have confirmed that a substantial proportion of individuals with pediatric type 1 diabetes have preserved C peptide secretions, that is, beta cells, many years after the onset of diabetes.[1–3 5] The present results show that tight blood glucose control during the first years of treatment, rather than average long-term control, is associated with preserved C peptide production. Since C peptide secretion declines by each year after diagnosis,[9–12] preservation of some remaining secretion would depend on secretion remaining at presentation and the rate of decline. C peptide concentrations at diagnosis were not available in our study, and we can therefore not exclude that the better HbA1c during the first years and the long-term preservation of C peptide both reflected higher remaining residual insulin production at diagnosis. However, insulin dosing per kilogram during the first year after diagnosis did not differ between the groups of participants with detectable and not detectable C peptide at long-term follow-up. There may also be other yet unidentified factors, aside from intensive management, that may have contributed to a lower HbA1c during the first years after diagnosis, eventually leading to residual C peptide 10 years later.

Several factors such as low age at presentation, ketoacidosis at presentation, high HbA1c at presentation and BMI have been related to C peptide concentrations measured at diagnosis[9 12–16] and to the decline of C peptide secretion during the first years of treatment.[9 17 18] These factors were not related to the long-term preservation of C peptide secretion in the present study. It is noteworthy that age at diagnosis, which is an important predictor in many studies,[9 12 13 17] did not presently emerge as a predictor. However, the present sample represents young children in a narrow age range, which makes it difficult to detect age-related differences. The young age at diagnosis of our studied participants also distinguishes them from the populations investigated in other studies, which may at least partially explain the observed differences

in findings. In this aspect, the present study adds to the existing literature that factors associated with C peptide decline may differ pending on age group.

GADA and IA-2A have also been associated with a more rapid decline in C peptide secretion.[18] In the present study, autoantibody titers were not available at presentation or during the first years of treatment, only at long-term follow-up. Therefore, the role of persisting antibodies in the preservation of C peptide secretion cannot be inferred, but their presence or absence at long-term follow-up did not differ. However, autoantibodies may not be the most important predictors of decline in C peptide secretion.[9]

Female gender was an independent factor for preserving beta cell mass and function for at least 10 years in the present study. Although we lack data on C peptide levels of the included children at diagnosis, a registry of Swedish juvenile type 1 diabetes (Better Diabetes Diagnosis) has previously reported higher C peptide concentrations in female individuals at diagnosis, providing a potential explanation for this finding.[19] Practically, however, all patients with type 1 diabetes have measurable C peptide secretion at diagnosis, and the vast majority have retained some after 1 or 2 years later.[12] It is therefore evident that almost all patients initially have the prerequisite for C peptide secretion in the long term. In the short term, good glycemic control helps to preserve C peptide secretion, although a progressive decline appears inevitable.[20 21] The present results extend this observation to the long-term preservation of C peptide secretion following initial good glycemic control and show the first 3 years' glycemic control to be an independent predictor. However, a limitation of the present study was that there was low variability in glycemic control was low, with few participants having poorly controlled diabetes. It is notable that HbA1c beyond the first years of treatment did not influence the presence of detectable C peptide at long-term follow-up.

Interestingly, while BMI SDS at diagnosis did not predict residual C peptide concentrations at long-term follow-up, BMI SDS at follow-up was for unknown reasons an independent predictor of this favorable outcome. Insulin is an anabolic hormone, but the recorded low beta cell function is unlikely to have been contributed by its systemic effects. A possibility, however, may be local

trophic effects of residual insulin in the pancreas, since atrophy of the exocrine pancreas relates to loss of beta cell function, with exocrine pancreatic insufficiency and potential malnutrition as a consequence.[22]

Celiac disease was also more prevalent in patients with detectable C peptide concentrations at long-term follow-up. Previous studies have indicated that gluten-free diet may impact on the immune system, and at least after a 1-year follow-up, stimulated C peptide levels were higher in children with type 1 diabetes treated with a gluten-free diet when compared with matched children with type 1 diabetes on a regular diet.[23] Our study suggests that gluten-free diet may also have beneficial effects in a longer time frame, although an intervention study in individuals with type 1 diabetes without concomitant celiac disease would be needed to corroborate this matter.

At long-term follow-up, detectable C peptide concentrations, if any, were in the lower range, with a mean of $4.3 \, \text{pmol/L}$ (range $1.7–22.7 \, \text{pmol/L}$). While of potential importance as target for beta cell expansion therapies, HbA1c in participants with detectable C peptide concentrations was similar to those without detectable C peptide, indicating the recorded very low residual beta cell function had no impact on glycemic control. This is consistent with our previous study on older subjects with type 1 diabetes.[5] In the present study we also did not observe an association between the low remaining C peptide concentrations and protection from retinopathy, while retinopathy as expected was predicted by a 10-unit change in the 10-year grand average HbA1c. C peptide levels $>10 \, \text{pmol/L}$ have previously been associated with protection from microvascular and macrovascular complications,[17] while lower concentrations similar to those in the present study were not clearly associated.[24]

In our study, non-fasting C peptide measurements were performed at the long-term follow-up and C peptide was detectable in 38% of the participants. A previous study, using a C peptide assay with similar sensitivity to ours, has shown that even more patients (up to 19% of patients with undetectable non-fasting C peptide) may be identified using a mixed meal tolerance test.[25] Whether inclusion of such patients would have also impacted on the findings we obtained in the present study is unknown.

CONCLUSION

We conclude that better glycemic control during the first 3 years after diagnosis is associated with preserved beta cell mass and function.

ORCID iD
Annika Grönberg http://orcid.org/0000-0002-8502-8568

REFERENCES

1 Keenan HA, Sun JK, Levine J, et al. Residual insulin production and pancreatic ß-cell turnover after 50 years of diabetes: Joslin Medalist study. *Diabetes* 2010;59:2846–53.

2 Wang L, Lovejoy NF, Faustman DL. Persistence of prolonged C-peptide production in type 1 diabetes as measured with an ultrasensitive C-peptide assay. *Diabetes Care* 2012;35:465–70.

3 Oram RA, Jones AG, Besser REJ, et al. The majority of patients with long-duration type 1 diabetes are insulin microsecretors and have functioning beta cells. *Diabetologia* 2014;57:187–91.

4 Madsbad S, Alberti KG, Binder C, et al. Role of residual insulin secretion in protecting against ketoacidosis in insulin-dependent diabetes. *Br Med J* 1979;2:1257–9.

5 Espes D, Singh K, Sandler S, et al. Increased interleukin-35 levels in patients with type 1 diabetes with remaining C-peptide. *Diabetes Care* 2017;40:1090–5.

6 Sjöblad S. Barn-och ungdomsdiabetes. In: *Barn-och ungdomsdiabetes*. 2nd ed. Lund, Sweden: Studentlitteratur AB, 2008.

7 Hanberger L, Samuelsson U, Lindblad B, et al. A1C in children and adolescents with diabetes in relation to certain clinical parameters: the Swedish childhood diabetes registry SWEDIABKIDS. *Diabetes Care* 2008;31:927–9.

8 Karlberg J, Luo ZC, Albertsson-Wikland K. Body mass index reference values (mean and SD) for Swedish children. *Acta Paediatr* 2001;90:1427–34.

9 Ludvigsson J, Carlsson A, Deli A, et al. Decline of C-peptide during the first year after diagnosis of type 1 diabetes in children and adolescents. *Diabetes Res Clin Pract* 2013;100:203–9.

10 Palmer JP. C-Peptide in the natural history of type 1 diabetes. *Diabetes Metab Res Rev* 2009;25:325–8.

11 Lachin JM, McGee P, Palmer JP, et al. Impact of C-peptide preservation on metabolic and clinical outcomes in the diabetes control and complications trial. *Diabetes* 2014;63:739–48.

12 Greenbaum CJ, Beam CA, Boulware D, et al. Fall in C-peptide during first 2 years from diagnosis: evidence of at least two distinct phases from composite type 1 diabetes TrialNet data. *Diabetes* 2012;61:2066–73.

13 Barker A, Lauria A, Schloot N, et al. Age-Dependent decline of ß-cell function in type 1 diabetes after diagnosis: a multi-centre longitudinal study. *Diabetes Obes Metab* 2014;16:262–7.

14 Komulainen J, Lounamaa R, Knip M, et al. Ketoacidosis at the diagnosis of type 1 (insulin dependent) diabetes mellitus is related to poor residual beta cell function. childhood diabetes in Finland Study Group. *Arch Dis Child* 1996;75:410–5.

15 Szypowska A, Groele L, Wysocka-Mincewicz M, et al. Factors associated with preservation of C-peptide levels at the diagnosis of type 1 diabetes. *J Diabetes Complications* 2018;32:570–4.

16 Lauria A, Barker A, Schloot N, et al. Bmi is an important driver of ß-cell loss in type 1 diabetes upon diagnosis in 10 to 18-year-old children. *Eur J Endocrinol* 2015;172:107–13.

17 Kuhtreiber WM, Washer SLL, Hsu E, *et al*. Low levels of C-peptide have clinical significance for established type 1 diabetes. *Diabet Med* 2015;32:1346–53.

18 Mortensen HB, Swift PGF, Holl RW, *et al*. Multinational study in children and adolescents with newly diagnosed type 1 diabetes: association of age, ketoacidosis, HLA status, and autoantibodies on residual beta-cell function and glycemic control 12 months after diagnosis. *Pediatr Diabetes* 2010;11:218–26.

19 Samuelsson U, Lindblad B, Carlsson A, *et al*. Residual beta cell function at diagnosis of type 1 diabetes in children and adolescents varies with gender and season. *Diabetes Metab Res Rev* 2013;29:85–9.

20 Hao W, Gitelman S, DiMeglio LA, *et al*. Fall in C-peptide during first 4 years from diagnosis of type 1 diabetes: variable relation to age, HbA1c, and insulin dose. *Diabetes Care* 2016;39:1664–70.

21 Nordwall M, Ludvigsson J. Clinical manifestations and beta cell function in Swedish diabetic children have remained unchanged during the last 25 years. *Diabetes Metab Res Rev* 2008;24:472–9.

22 Campbell-Thompson M, Rodriguez-Calvo T, Battaglia M. Abnormalities of the exocrine pancreas in type 1 diabetes. *Curr Diab Rep* 2015;15:79.

23 Svensson J, Sildorf SM, Pipper CB, *et al*. Potential beneficial effects of a gluten-free diet in newly diagnosed children with type 1 diabetes: a pilot study. *Springerplus* 2016;5:994.

24 Williams KV, Becker DJ, Orchard TJ, *et al*. Persistent C-peptide levels and microvascular complications in childhood onset type 1 diabetes of long duration. *J Diabetes Complications* 2019;33:657–61.

25 Davis AK, DuBose SN, Haller MJ, *et al*. Prevalence of detectable C-peptide according to age at diagnosis and duration of type 1 diabetes. *Diabetes Care* 2015;38:476–81.