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Lower HbA1c in patients with type 1 diabetes and celiac disease who reached celiac-specific antibody-negativity—A multicenter DPV analysis

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Abstract

Objectives: To study celiac-specific antibody status over 3 years in patients with type 1 diabetes and biopsy-proven celiac disease (T1D + CD). Furthermore, to determine clinical differences after diagnosis between patients reaching constant antibody-negativity (Ab-neg) and staying antibody-positive (Ab-pos).

Methods: A total of 608 pediatric T1D + CD patients from the multicenter DPV registry were studied longitudinally regarding their CD specific antibody-status. Differences between Ab-neg (n = 218) and Ab-pos (n = 158) patients 3 years after biopsy were assessed and compared with 26 833 T1D patients without CD by linear and logistic regression adjusted for age, gender, diabetes duration and migration background.

Results: Thirty-six percent of T1D + CD patients reached and sustained antibody-negativity 3 years after CD diagnosis. The median time until patients returned to Ab-

Abbreviations: Ab, antibody; Ab-neg, antibody-negative, antibody-negativity; Ab-pos, antibody-positive, antibody-positivity; Ab-status, antibody-status; anti-tTG, transglutaminase autoantibodies; BMI, body-mass-index; CD, celiac disease; CGM, continuous glucose monitoring; CI, confidence interval; DKA, diabetic ketoacidosis; DPV, Diabetes Patienten Verlaufsdokumentation; EMA, endomysial autoantibodies; ESPGHAN, European Society for Pediatric Gastroenterology, Hepatology, and Nutrition; FDR, false-discovery-rate; GFD, gluten-free diet; HbA1c, glycated hemoglobin A1c; HDL, high-density lipoprotein; ISPAD, International Society of Pediatric and Adolescent Diabetes; LDL, low-density lipoprotein; T1D + CD, type 1 diabetes and concomitant celiac disease; T1D, type 1 diabetes.

neg was 0.86 (0.51;1.16) years. Three years after diagnosis, HbA1c was lowest in Ab-neg and highest in Ab-pos patients compared to T1D-only patients (adjusted mean (95%CI): 7.72 (7.51-7.92) % vs 8.44 (8.20-8.68) % vs 8.19 (8.17-8.21) %, adjusted $P < 0.001$, respectively). Total cholesterol, LDL-cholesterol and frequency of dyslipidemia were significantly lower in Ab-neg compared to T1D-only patients (167 (161-173) mg/dl vs 179 (178-179) mg/dl, $P < .001$; 90 (84-96) mg/dl vs 99 (98-99) mg/dl, $P = .005$; 15.7 (10.5-22.9) % vs 25.9 (25.2-26.6) %, $P = .017$). In longitudinal analyses over 6 years after diagnosis, a constantly higher HbA1c ($P < .001$) and a lower height-SDS ($P = .044$) was observed in Ab-pos compared to Ab-neg patients.

Conclusion: Only one third of T1D + CD patients reached constant Ab-negativity after CD diagnosis. Achieving Ab-negativity after diagnosis seems to be associated with better metabolic control and growth, supposedly due to a higher adherence to therapy in general.

KEYWORDS

antibody negativity, antibody positivity, celiac disease, metabolic control, type 1 diabetes

1 | INTRODUCTION

Celiac disease (CD) is an immune-mediated systemic autoimmune disease in genetically susceptible persons (HLA DQ2 and/or HLA DQ8) triggered by gluten and related prolamins characterized by a wide variety of gluten dependent symptoms and an enteropathy with a specific histopathology of the small gut mucosa. Due to HLA relation CD is a frequent co-morbidity in type 1 diabetes (T1D) with a wide range of prevalence of up to 10% worldwide[1] compared to 1% in the general population. Based on registry data from the multicenter, standardized DPV (Diabetes Patienten Verlaufsdokumentation) database, biopsy-confirmed CD was documented in 3.2% of pediatric T1D-patients.[2]

CD may manifest with gastrointestinal symptoms due to malabsorption, and growth retardation,[3,4] however the majority of patients, including those with T1D, have no or non-specific symptoms. Therefore, the ISPAD Clinical Practice Consensus Guidelines recommend a regular screening for CD at T1D onset and every 1-2 years thereafter.[1] Irrespective of symptoms, T1D patients with CD show significant differences in height and weight percentiles compared to patients with T1D alone even years after diagnosis.[1,5]

Coexisting CD is associated with a 2-fold increased risk for diabetic retinopathy after 15 years[6] and with the development of diabetic nephropathy.[7] Lately, CD in T1D is also being related to the pathogenesis of cardiovascular diseases (CVD).[8] Possible explanations for this might be systemic inflammation[9] and low HDL-cholesterol levels in T1D patients with CD.[10] It has been reported that after initiation of a gluten-free-diet (GFD) HDL-cholesterol levels increased in patients with CD.[10,11] Most strongly, however, this effect occurred in a group of patients who were completely adherent to GFD.[11]

Other studies, however, found no association of T1D + CD with the development of CVD or diabetic nephropathy.[12,13] Children with both CD and T1D, who had been compliant with GFD, showed lower levels of albuminuria compared to matched patients with T1D alone.[13]

Strict GFD reduces symptoms of CD and is recommended to patients with CD in order to prevent CD-related complications.[14] In asymptomatic patients benefits from GFD are sometimes apparent in retrospective comparison,[15] but in many cases they are limited to more subtle long-term changes such as increase in body weight[16] and better bone mineralization.[17] Strict GFD limits food choice and has thereby a large impact on the individual's daily life. This might lead to a reduced quality of life,[18] especially in patients who are already faced with restrictions due to diabetes. In particular, for patients who do not experience direct benefits and improvement of their subjective well-being it is a large burden to adhere to a strict GFD.[19,20] According to literature about 20% to 50% of T1D patients with CD do not adhere to GFD, also depending on age and socio-cultural background.[17,21] The European Society for Pediatric Gastroenterology, Hepatology, and Nutrition (ESPGHAN) recommends regular monitoring of CD patients including testing for auto-antibodies against tissue-transglutaminase (TGA-Ab) which, on a GFD, should show a continuous decline and finally turn negative after 12 months, depending on the initial level.[14]

The aim of this study was, firstly, to examine the longitudinal development of celiac disease specific antibody-status (Ab-status) in patients with T1D after CD diagnosis. Secondly, after an observation period of 3 years, this study aimed to assess how many T1D patients with coexisting CD within the DPV registry reached continuous antibody negativity after CD diagnosis and whether these patients

clinically differed from T1D + CD patients remaining antibody-positive, or patients with T1D alone.

2 | METHODS

Using the continuous diabetes data acquisition system for prospective surveillance (Diabetes Patienten Verlaufsdokumentation [DPV]), we analyzed data on pediatric patients with T1D from 368 participating centers in Germany, Austria, Luxemburg, and Switzerland.

Since 1995, routinely-documented patient data (demographics, therapy and other clinical information) are collected at every visit by participating centers and are transmitted pseudonymized for benchmarking and central analysis twice a year to Ulm University, Germany. Inconsistent data are reported back, and centers are asked for corrections. All plausible data are aggregated into a cumulative database, the DPV registry.[22] The DPV initiative has been approved by the ethics committee of the University of Ulm and the local review boards of each participating center approved the anonymized data collection.

2.1 | Study population

For the present study, pediatric patients with T1D were included and grouped by comorbid CD (Figure 1). "No-Celiac Disease" (no-CD) was defined as being CD specific antibody negative, having had no duodenal biopsy and no documented CD diagnosis. Autoantibodies considered in the database were endomysial autoantibodies (EMA), transglutaminase autoantibodies (anti-tTG) and gliadin autoantibodies, as previously described.[23] T1D patients with biopsy results corresponding to Marsh-classification ≥2 and positive CD specific auto-antibodies prior to biopsy were classified as patients with biopsy-proven celiac disease (CD).[10] Further inclusion criteria for patients with T1D + CD were: available date of biopsy, biopsy after T1D diagnosis, and clinical information available within 1 year prior to 3 years after biopsy. A time period of 1 year prior to 3 years after biopsy has been chosen, as a longer time interval would have led to a considerable reduction in observable patients (see also legend of Figure 2).

For each patient, multiple data entries per year were aggregated as median (quantitative parameters) and sums (count data).

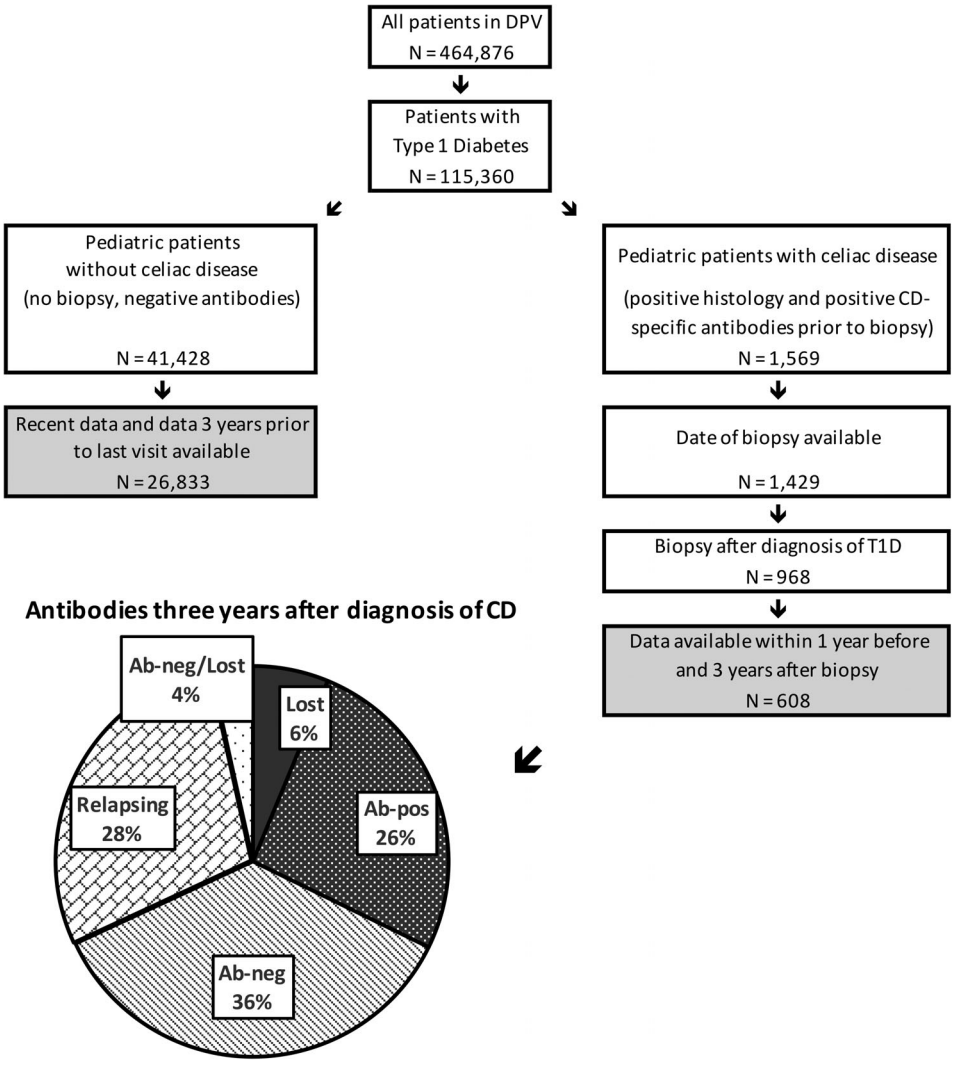

FIGURE 1 Selection of patients and proportion of patients (n = 608) grouped by CD specific antibody-status 3 years after CD diagnosis. Ab-neg = patients that had at least one antibody negative test result within the first 3 years after diagnosis and stayed continuously antibody negative (Ab-neg) thereafter, Relapsing = patients with at least one antibody negative test result within the first 3 years after diagnosis, but with antibody-positivity-relapse thereafter, Ab-neg/Lost = patients with at least one antibody negative test result within the first 3 years after diagnosis, but no further antibody test thereafter, Lost = patients without further information on antibody titers after diagnosis, Ab-pos = patients with persistent antibody positivity in all available tests (number depends on individual patient)

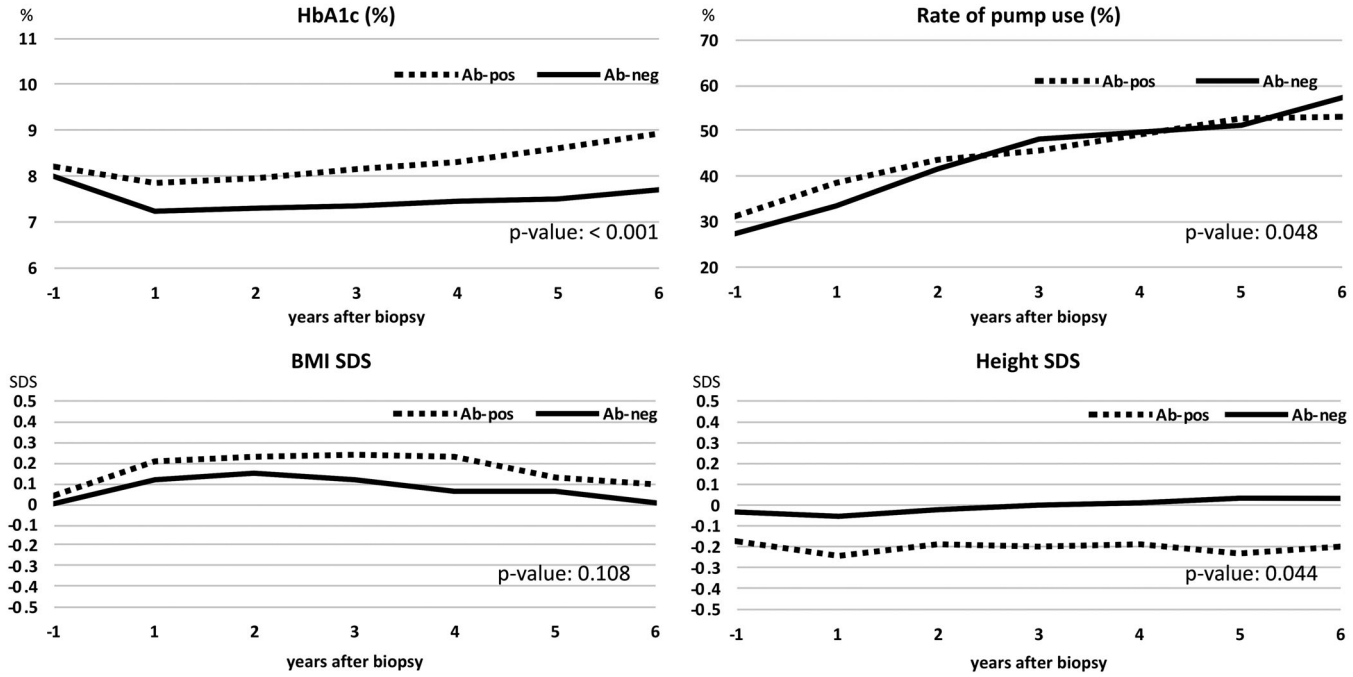

FIGURE 2 Data are adjusted means based on multivariable regression models with interaction term patient Ab-pos or Ab-neg and calendar year. Adjustments for age at biopsy, diabetes duration at biopsy, gender and migration background. *P* values for time-trend. Number of patients at various time-points from within 1 year prior biopsy to 6 years after biopsy, Ab-pos: n = 158, 156, 154, 158, 107, 73, 54; Ab-neg: n = 218; 218; 218; 218; 190; 141; 109

T1D + CD patients were stratified based on their celiac disease specific antibody status after CD diagnosis (antibodies considered, as available: EMA, anti-tTG and gliadin autoantibodies): (i) patients that had at least one antibody negative test result within the first 3 years after diagnosis and stayed continuously antibody negative (Ab-neg) thereafter, (ii) patients with persistent antibody positivity (Ab-pos) in all available tests (number depends on individual patient), (iii) patients with at least one antibody negative test result within the first 3 years after diagnosis, but with antibody-positivity-relapse thereafter (Relapsing), (iv) patients without further information on antibody titers after diagnosis (Lost), (v) patients with at least one antibody negative test result within the first 3 years after diagnosis, but no further antibody test thereafter (Ab-neg/Lost).

Only Ab-neg (i) and Ab-pos (ii) patients were considered in the later statistical analysis regarding diabetes outcomes and were compared with T1D patients with no-CD.

Finally, the study comprised 26 833 patients with T1D-only, with a median age of 16.9 (14.0; 18.0) years, 218 Ab-neg T1D + CD patients with a median age of 11.2 (8.1;14.3) years and 158 Ab-pos T1D + CD patients with a median age of 13.1 (9.5;16.0) years at the time of analysis (third year after biopsy for Ab-neg and Ab-pos patients and the most recent treatment year for patients with T1D alone).

2.2 | Variables of interest

As a marker of glycemic control, HbA1c was studied and mathematically standardized to the reference range of 4.0%-6.05% (IFCC 20.8-42.6 mmol/L) of the Diabetes Control and Complications Trial applying the multiple of the mean method, in order to correct for different laboratory methods.[24]

Severe hypoglycemic events and DKA were defined according current ISPAD guidelines and given as annual rates as described previously.[22] The annual rate of hospitalization (defined as at least one overnight stay) was determined too.

Anthropometric data (weight, height and body-mass-index [BMI]) are given as SD scores (SDS). Using recent national reference values,[25] SDS were computed applying the least mean squares method (LMS).[26] Microalbuminuria was defined according to the ISPAD guidelines, as described previously.[27] Further variables of interest were total daily insulin dose per body weight, number of blood sugar measurements per day, use of continuous glucose monitoring (CGM) (either real-time or intermittent "flash" CGM) and insulin therapy modality (percentage of insulin pump use).[28]

Elevated total cholesterol (>200 mg/dL), LDL-cholesterol (>130 mg/dL), triglycerides (>130 mg/dL) and/or low levels of HDL-cholesterol (<35 mg/dL) were reported as dyslipidemia. Migration background was defined as either patient or one parent not born in Germany, Austria, Switzerland or Luxemburg, respectively.

2.3 | Statistical analysis

Baseline characteristics of T1D patients with CD (Ab-neg and Ab-pos patients) and T1D patients without CD (most recent treatment year documented) were compared by using Wilcoxon test for continuous

parameters and χ^2 test for binomial parameters. To adjust for multiple comparisons, *P* values were corrected according to the Benjamini-Hochberg procedure controlling the false discovery rate (FDR).[29] Data are given as median (first; third quartile) or proportions.

To compare clinical parameters in T1D + CD patients (Ab-neg and Ab-pos patients) at time of biopsy (to a maximum of 1 year prior biopsy) and T1D-only patients, separate regression models for each parameter were created. In the group of T1D-only patients, data from the third year before the most recent treatment year documented in the DPV registry were used. Data are given as adjusted means with 95% confidence intervals (CI), estimated by linear regression for continuous parameters and logistic regression for binomial parameters and adjusted for age (categorized by <10, 10 to <15 and ≥15 years), gender, diabetes duration (categorized by <5 and ≥5 years) and migration background. Pump and CGM use were additionally adjusted for year of treatment, as use of pumps and CGM increased over years. Total daily insulin dose was additionally adjusted for pump use. Total-cholesterol, LDL-cholesterol, HDL-cholesterol and triglycerides were additionally adjusted for overweight (≥90th percentile) and HbA1c (≥7% and <7%). To adjust for multiple comparisons, all *P* values were corrected according to the Benjamini-Hochberg procedure controlling the false discovery rate (FDR).[29]

Ab-neg patients were compared to Ab-pos patients 3 years after CD diagnosis by using linear regression for continuous parameters and logistic regression for binomial distributed parameters. Additionally, both groups were compared to T1D patients without CD (most recent treatment year). All regression models were adjusted for age (categorized by <10, 10 to <15 and ≥ 15 years), gender, diabetes duration (categorized by <5 and ≥ 5 years) and migration background. Comparisons regarding insulin pump use were additionally adjusted for treatment year. Insulin dosage was additionally adjusted for pump use. Regression models for lipids were additionally adjusted for overweight (≥90th percentile) and HbA1c-level (≥7% and < 7%). The model for the occurrence of microalbuminuria was additionally adjusted for HbA1c-level (≥7% and < 7%). All regression models were adjusted for multiple comparisons (FDR).

Additionally, longitudinal differences between Ab-neg and Ab-pos patients were analyzed annually over a period of within 1 year before and 6 years after biopsy for some variables of interest. Linear (for continuous parameters) or logistic (for binomial distributed parameters) regression models were computed with interaction term patient Ab-pos or Ab-neg and calendar year. Adjustments were made for age at biopsy, diabetes duration at biopsy, gender and migration background. *P* values for time trends are given.

A two-sided *P* value <.05 was considered significant. SAS version 9.4 (SAS Institute, Cary, North California) was used for statistical analysis.

3 | RESULTS

3.1 | Antibody negativity—frequency and time period

Of all T1D patients with CD (n = 608), 68% had at least one negative antibody test within the first 3 years after CD diagnosis. Persistent antibody negativity was achieved by 36% of patients (n = 218), whereas 26% of patients (n = 158) stayed continuously antibody positive throughout the observation period. It took a median duration of 0.86 (0.51; 1.16) years until patients reached continuous Ab-negativity. Figure 1 depicts the frequency of patients with relapsing CD-Ab-status or missing information.

On average (mean ± SD), patients in the Ab-pos group had 4.6 ± 2.5 and patients in the Ab-neg group had 4.9 ± 2.1 CD specific antibody measurements during their individual observation period.

3.2 | Comparisons between Ab-pos and Ab-neg CD patients and no-CD patients

3.2.1 | Baseline demographics

Patients with CD who reached antibody negativity and stayed continuously Ab-neg thereafter were significantly younger at CD diagnosis compared to patients who stayed Ab-pos (Table 1). There was no significant difference regarding age at T1D-onset, diabetes duration, gender or migration background between the two groups. Compared to T1D patients without CD, T1D + CD patients were younger at T1D onset, at the time of analysis and had a shorter diabetes duration as well as a female preponderance, independent of their CD specific antibody status.

At time of CD diagnosis, both groups, patients, who later reached continuous Ab-negativity and those with persistent Ab-positivity, showed higher HbA1c values than T1D-only patients, after adjustment for age, gender, diabetes duration and migration background. Yet, HbA1c values did not differ between Ab-neg and Ab-pos patients at time of CD diagnosis. There were no differences in the documented numbers of daily blood glucose measurements between the three groups (Table 1).

Regarding anthropometric data, BMI, weight and height SDS did not differ between Ab-neg and Ab-pos patients at time of CD diagnosis, but both groups had significantly lower SDS values than T1D patients without CD.

Levels of total cholesterol, LDL-cholesterol and triglycerides levels did not differ between groups at time of CD diagnosis, but both Ab-neg and Ab-pos patients had lower levels of HDL-cholesterol compared to patients with T1D alone.

Frequencies regarding pump or CGM use, DKA and severe hypoglycemia were similar in all three groups, but both Ab-pos and Ab-neg patients were significantly hospitalized more often compared with T1D patients without CD.

3.2.2 | Three years after CD diagnosis

After adjustment for age, gender, diabetes duration and migration background (Table 2), there was no difference in any outcome parameter between Ab-pos and Ab-neg patients 3 years after CD diagnosis, except for glycemic control. HbA1c levels were lowest in Ab-neg and highest in Ab-pos patients. Moreover, when compared to T1D-only

TABLE 1 Baseline-characteristics: Ab-neg and Ab-pos T1D-patients with CD and T1D patients without CD

	T1D								
	With CD						*P* values		
N	Ab-neg 218		Ab-pos 158		Without CD 26 833		Ab-pos vs Ab-neg	Ab-neg vs no-CD	Ab-pos vs no-CD
Age at T1D onset (years)	5.1	(3.0;8.6)	6.2	(3.0;10.3)	8.4	(5.0;11.4)	.14	**<.001**	**<.001**
Age at biopsy (years)	8.8	(5.5;11.9)	10.6	(7.0;13.4)	—		**.005**	—	—
Age at analysis* (years)	11.2	(8.1;14.3)	13.1	(9.5;16.0)	16.9	(14.0;18.0)	**.005**	**<.001**	**<.001**
Diabetes duration at analysis* (years)	4.3	(2.8;6.8)	4.4	(3.3;7.0)	7.1	(4.7;10.2)	.25	**<.001**	**<.001**
Male (%)	45.0		33.5		53.4		.06	**.016**	**<.001**
Migration background (%)	17.4		17.7		17.9		.94	.86	.95
HbA1c (%)[a]	8.18	(7.98;8.39)	8.35	(8.11;8.59)	7.9	(7.89;7.93)	.62	**.029**	**.002**
HbA1c (mmol/mol)[a]	65.9	(63.7;68.2)	67.8	(65.1;70.4)	62.8	(62.7;63.2)			
Blood sugar measurements per day[a]	5.3	(5.1;5.7)	5.2	(4.9;5.4)	5.4	(5.4;5.4)	.62	.09	.95
Total daily insulin dose (IU/kg/d)[a]	0.80	(0.77;0.84)	0.84	(0.80;0.89)	0.84	(0.84;0.85)	.62	.09	.96
BMI-SDS[a]	−0.07	(−0.19;0.04)	−0.01	(−0.14;0.13)	0.26	(0.25;0.27)	.69	**<.001**	**<.001**
Weight-SDS[a]	−0.13	(−0.25;0.00)	−0.13	(−0.27;0.012)	0.26	(0.25;0.27)	.96	**<.001**	**<.001**
Height-SDS[a]	−0.13	(−0.27;0.01)	−0.25	(−0.41; −0.09)	0.14	(0.13;0.16)	.62	**<.001**	**<.001**
Total cholesterol (mg/dl)[a]	173.6	(168.1;179.2)	176.5	(169.3:183.8)	175.7	(175.2;176.3)	.74	.71	.90
HDL-cholesterol (mg/dl)[a]	55.2	(52.2;58.2)	55.0	(51.1;58.8)	61.9	(61.6:62.1)	.96	**<.001**	**.002**
LDL-cholesterol (mg/dl)[a]	93.9	(88.4;99.4)	102.1	(95.1;109.1)	94.3	(93.8;94.8)	.49	.94	.06
Triglycerides (mg/dl)[a]	117.4	(102.8;132.0)	130.8	(111.5;150.2)	116.2	(114.8;117.6)	.62	.94	.21
Dyslipidemia (%)[a]	26.2	(20.1;33.5)	30.8	(22.4;40.6)	22.7	(22.1;23.4)	.67	.50	.12
Pump use (%)[a]	24.4	(18.9;31.8)	23.9	(17.5:31.8)	28.8	(28.2;29.5)	.96	.43	.28
CGM use (%)[a]	4.0	(2.1;7.6)	1.2	(0.3;4.6)	2.6	(2.4;2.8)	.62	.44	.33
Severe hypoglycemia (%)[a]	10.2	(6.6;15.4)	11.4	(7.1;17.8)	7.9	(7.6;8.3)	.95	9.49	.20
DKA (%)[a]	1.9	(0.6;5.8)	2.3	(0.8;7.0)	2.3	(2.1;2.5)	.96	.91	.98
Hospitalization (%)[a]	6.7	(6.0;7.4)	6.7	(5.8;7.4)	3.8	(3.7;3.8)	.96	**<.001**	**<.001**

Note: Baseline Characteristics: data are median (first; third quartile) or proportion. Comparison between groups: Wilcoxon-test for continuous parameters, χ^2-test for binomial parameters. *CD: 3 years after biopsy, no-CD: most recent treatment year.

[a]Clinical parameters at biopsy (to 1 year prior biopsy max.) in T1D + CD patients and for the third year before most recent treatment year in no-CD patients. Data are adjusted means with 95% CI, estimated by linear regression for continuous parameters and logistic regression for binomial parameters. For each parameter, a separate model adjusted for age, gender, diabetes duration and migration background was created. Pump and CGM use were additionally adjusted for year of treatment. Total daily insulin dose was additionally adjusted for pump use. Total-cholesterol, LDL-cholesterol, HDL-cholesterol and triglycerides were additionally adjusted for overweight (≥90th percentile) and HbA1c (≥7% and <7%). To adjust for multiple comparisons, *P* values were corrected according to the Benjamini-Hochberg procedure controlling the false discovery rate (FDR).

patients (Table 2), HbA1c was observed to be even lower in Ab-neg patients.

There was no statistically significant difference in height-SDS, daily blood sugar measurements, rates of dyslipidemia, severe hypoglycemia or hospitalization in Ab-neg compared to Ab-pos patients. Compared to T1D without CD, weight and height SDS were significantly lower in Ab-pos and Ab-neg CD patients (Table 2). Further, Ab-neg patients had significantly less dyslipidemia and a more favorable lipid profile with specifically lower LDL-cholesterol levels in comparison to T1D-only patients.

Frequency of hospitalization was lower in T1D + CD patients, as compared to T1D-only patients, but the difference was only significant for Ab-neg patients. There were no significant differences in the frequencies of CGM and insulin pump use or microalbuminuria between the three groups.

TABLE 2 Linear/logistic regression: comparison between Ab-pos, Ab-neg and no-CD* 3 years after biopsy

	T1D								
	With CD						*P* values		
	Ab-neg		Ab-pos		Without CD		Ab-pos vs Ab-neg	Ab-neg vs no-CD	Ab-pos vs no-CD
HbA1c (%)	7.72	(7.51;7.92)	8.44	(8.20;8.68)	8.19	(8.17;8.21)	**<.001**	**<.001**	.13
HbA1c (mmol/mol)	60.9	(58.6; 63.1)	68.7	(66.1; 71.4)	66.0	(65.8; 66.2)			
Blood sugar measurements per day	5.1	(4.8;5.4)	4.8	(4.5;5.1)	4.9	(4.9;4.9)	.51	.22	.51
Total daily insulin dose (IU/kg/d)	0.88	(0.84;0.91)	0.94	(0.90;0.99)	0.92	(0.91;0.92)	.21	.06	.40
BMI-SDS	0.1	(−0.01;0.22)	0.2	(0.07;0.34)	0.37	(0.36;0.38)	.52	**<.001**	.09
Weight-SDS	0.06	(−0.06;0.19)	0.06	(−0.09;0.21)	0.35	(0.34;0.36)	.99	**<.001**	**.001**
Height-SDS	−0.08	(−0.22;0.06)	−0.25	(−0.42; −0.09)	0.10	(0.08;0.11)	.51	**.032**	**.001**
Total-cholesterol (mg/dl)	167.0	(161.4;172.6)	170.4	(163.0;177.7)	178.8	(178.3;179.4)	.61	**<.001**	.09
HDL-cholesterol (mg/dl)	60.0	(57.2;62.8)	57.9	(54.3;61.5)	62.8	(61.2;62.4)	.54	.19	.09
LDL-cholesterol (mg/dl)	89.9	(84.2;95.5)	93.7	(86.5;100.9)	98.9	(98.3;99.4)	.58	**.005**	.32
Triglycerides (mg/dl)	117.6	(103.9;131.2)	124.2	(106.1;142.2)	121.4	(120.1;122.8)	.69	.62	.77
Dyslipidemia (%)	15.7	(10.5;22.9)	27.8	(19.4;38.1)	25.9	(25.2;26.6)	.21	**.017**	.74
Microalbuminuria (%)	37.1	(30.8;43.3)	36.4	(29.0;43.7)	31.4	(30.8;32.0)	.96	.13	.34
Pump use (%)	43.4	(36.4;50.7)	37.6	(29.8;46.1)	42.1	(41.5;42.8)	.51	.79	.40
CGM use (%)	5.8	(3.7;8.9)	5.0	(3.0;8.5)	7.1	(6.7;7.5)	.80	.42	.34
Severe hypoglycemia (%)	8.3	(5.2;12.8)	11.5	(7.3;17.5)	8.1	(7.8;8.4)	.52	.93	.27
DKA (%)	3.3	(1.6;6.8)	4.3	(2.0;8.7)	2.9	(2.7;3.2)	.72	.81	.42
Hospitalization (%)	20.6	(16.0;26.2)	27.7	(21.53;35.0)	29.0	(28.4;29.5)	.51	**.013**	.76

Note: Data are adjusted means with 95% CI, estimated by linear regression for continuous parameters and logistic regression for binomial parameters. For each parameter, a separate model adjusted for age, gender, diabetes duration and migration background was created. Pump and CGM use were additionally adjusted for year of treatment. Total daily insulin dose was additionally adjusted for pump use. Total-cholesterol, LDL-cholesterol, HDL-cholesterol and triglycerides were additionally adjusted for overweight (≥90th percentile) and HbA1c (≥7% and <7%). Microalbuminuria was additionally adjusted for HbA1c (≥7% and <7%). To adjust for multiple comparisons, *P* values were corrected according to the Benjamini-Hochberg procedure controlling the false discovery rate (FDR). *The most recent treatment year was considered in patients with no-CD.

3.3 | Longitudinal trends for Ab-neg and Ab-pos CD patients

Adjusted means of various characteristics of Ab-pos and Ab-neg patients were plotted from 1 year before biopsy to 6 years after biopsy (Figure 2). A significant difference over time was present for HbA1c. While HbA1c-values remained relatively stable at about 7% to 8% in Ab-neg patients, there was a constant increase of HbA1c in Ab-pos patients during the years after CD diagnosis. While Ab-neg patients showed a constantly higher height-SDS with a slight increase over time, Ab-pos patient did not show such a trend and remained constantly smaller than Ab-neg patients. BMI-SDS did not differ significantly between groups during observation period, however, Ab-pos patients showed a trend toward higher BMI-SDS values compared to Ab-neg ones (Figure 2).

Pump use over time differed significantly between the two groups, with higher pump use in Ab-pos patients around diagnosis, comparable pump use two and a half years after CD diagnosis and lower use 6 years after diagnosis.

Total daily insulin dosage increased steadily over time, with no group differences. Also, occurrence of severe hypoglycemia over time did not differ. In both groups, there was a reduction of hypoglycemic events after the fourth year of observation. Time trends for LDL- and HDL-cholesterol values in the Ab-neg compared to the Ab-pos group showed no statistical significance (Figure S1 in Data S1).

4 | DISCUSSION

Only 36% of the CD patients reached and sustained continuous antibody negativity throughout their individual observation period. Yet, other studies reported higher percentages of Ab-negativity after GFD initiation.[30,31] A study assessing GFD adherence in 35 children with T1D and CD, both clinically by a dietitian and serologically, found that 69% of patients were adherent to GFD. Furthermore, dietetic assessment and antibody titers showed complete concordance.[30] On the other hand, a 2 years follow-up study focusing on anti-tTG levels reported normalization of antibodies in only 22.2% after 2 years.[17]

The comparably low rate of antibody negativity after diagnosis of CD in the DPV register might be attributable to the multicenter observational design vs a single center studies emphasizing on GFD adherence.

According to the ESPGHAN 2012 guidelines, the timespan until antibody titers drop below the threshold for normal varies and depends on the initial level, generally a normalization should be achieved within 12 months after initiation of GFD.[14] Consequently, when celiac autoantibodies persist despite GFD for more than 2 years, insufficient compliance with the diet is generally assumed.[32]

In our study, HbA1c, as marker for glycemic control, was significantly higher in Ab-pos patients compared to the Ab-neg group, also after adjustment for age. In addition, there was a trend of more blood sugar measurements per day and a lower rate of acute complications such as DKA and severe hypoglycemia in the Ab-neg group compared to the Ab-pos, though this did not reach statistical significance. HbA1c values did not differ between Ab-pos and T1D-only patients. Hence, it is plausible, that it is not the Ab-pos patients having a worse glycemic control, but the GFD-adherent Ab-neg patients, who due to a particularly stricter adherence to therapy in general, also have a better metabolic control.

Compared to T1D-only patients, our data support the findings of others, that CD in T1D is not associated with a higher risk for DKA and severe hypoglycemia.[33] The trend for higher acute complications rates in the Ab-pos group might reflect a worse overall adherence with therapy. Supporting this argumentation, a recent study on quality of life and compliance with GFD found that adolescents with T1D and CD, who were non-adherent to diet had lower well-being scores and worse glycemic control.[30] Additionally, worse metabolic control in Ab-pos patients might also be caused by active celiac disease with chronic inflammation.[8,9] This might also be supported by the observation, that at the time of CD diagnosis and before the initiation of GFD, both groups, patients with later persistence of Ab-positivity and patients, who later showed continuous Ab-negativity, had higher Hba1c values than T1D-only patients, even after adjustment for age, gender, diabetes duration and migration background.

In addition to a constantly worse glycemic control, significantly lower height-SDS were observed in Ab-pos compared to Ab-neg patients over a time period of 1 year prior to 6 years after CD diagnosis. Growth retardation is a known complication in T1D with comorbid CD.[3,34] At CD diagnosis both groups, Ab-pos and Ab-neg patients, showed significantly lower values in weight, height and BMI-SDS in comparison to patients with T1D-only.

Remarkably, 3 years after CD diagnosis, both Ab-neg and Ab-pos patients still differed significantly with regard to body height from T1D-only patients. However, as we do not have antibody measurements before diabetes manifestation, we do not know how long undetected CD might have been present in these patients. A delay in the diagnosis of CD could also have caused a lasting growth retardation.

Previous studies showed impaired lipid profiles in smaller cohorts of T1D-children with untreated CD and reported an increase in cardio-protective HDL-cholesterol and decrease of triglycerides after initiation of GFD,[10,11,31] while LDL-cholesterol levels did not decrease after 1 year of GFD.[31] Interestingly, a significant increase of total cholesterol was reported, but only in those with good adherence to GFD. The authors hypothesized that this increase was attributable to an improvement of intestinal fat absorption.[31]

In our study, comparable to previous studies,[10,11] HDL-cholesterol values at time of CD diagnosis were lower in T1D patients with CD than in those without.

Longitudinal time trends for LDL- and HDL-cholesterol values after CD diagnosis were slightly more favorable in the Ab-neg compared to the Ab-pos group, although without statistical significance after adjustment for confounders.

However, 3 years after diagnosis, total cholesterol, LDL-cholesterol and the frequency of dyslipidemia were significantly lower in Ab-neg compared to T1D-only patients. The slightly better lipid profile in Ab-neg patients might be also partly related to the lower HbA1c values in this group of patients.

Whether antibody status is a valid marker for compliance with GFD remains controversial even in non-diabetic CD patients, but it is the best available non-invasive marker yet.[14] Spontaneous normalization of low to moderate elevated antibody titers (less than 10 time upper limit of normal) has been described in patients with T1D despite gluten consumption, but not in those with villous atrophy and therefore proven CD.[35] GFD is clearly beneficial in symptomatic patients with CD and reduces later complications.[15] On the other hand, gluten-free food might often have a high glycemic index and thereby lead to greater postprandial glucose excursions in patients with T1D.[19] Furthermore, GFD might lead to an inadequate intake of nutrients.[19] Detailed dietary counseling is needed to prevent negative effects on glycemic control and bodyweight.[16]

For future studies, it would be very interesting to assess the effect of GFD on glycemic variability and postprandial glucose excursions. However, as not enough CGM data downloads of patients included in this study were available for a more profound evaluation, we used HbA1c as a marker for glycemic control.

Due to the multicenter design, a further limitation is that antibody testing was performed by decentralized laboratories with different tests, which makes a direct comparison of antibody titers difficult. Moreover, many centers did not submit absolute antibody titers and provided solely the information whether the patients were antibody positive or negative. Concerning actual dietary habits of patients,

there is no information available in the registry. We do not know how strictly GFD was recommended by the various treatment centers and how much supportive diet counseling was provided to patients and families. Nonetheless, all centers, participating in the DPV initiative comply with internationally valid treatment guidelines.[1]

We focused on a 3-year observation period, since the number of observed patients decreased considerably after that time period. From 1 year prior to biopsy to 6 years after biopsy the number of observed patients in the Ab-pos group decreased from 158 to 54 patients and from 218 to 109 patients in the Ab-neg group. Then again, the resulting observation period of 3 years may have been too short to detect significant effects on, for example, microalbuminuria.

In conclusion, to the best of our knowledge, this is the first study to examine the longitudinal course of CD-antibodies after CD diagnosis and its relation to clinical outcome 3 years after diagnosis of CD in a large cohort of T1D patients with CD seroconversion to Ab-negativity as proxy for good dietary adherence.

Only 36% of CD patients were able to sustain antibody negativity 3 years after CD diagnosis. As opposed to persistent antibody positivity, achieving continuous antibody negativity seems to be associated with better growth and metabolic control. This may be related to a stricter adherence to both insulin therapy and GFD.

As the restrictions of GFD can be an additional burden for patients with T1D, we need to identify those patients who struggle early and provide additional support. Further prospective studies on the validity of CD specific antibodies as a marker for GFD adherence and the benefit of antibody negativity in CD with T1D are needed to derive clinical consequences from our findings.

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AUTHOR CONTRIBUTIONS

K.N. and N.P. designed the study. K.N., E.B., N.P. did or supported data analyses, including the statistical analyses. K.N. and N.P. interpreted the results. K.N. wrote the manuscript and prepared the figures and tables. E.B., S.L., J.R., S.K., A.P., A.N., E.F.R., C.D., J.W., P.S. and N.P. critically reviewed the manuscript.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

ETHICAL APPROVAL

Analysis of anonymized routine data within the DPV initiative was approved by the Ethics Committee of the Medical Faculty of the University of Ulm, Ulm, Germany and the institutional review boards at the participating centers.

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REFERENCES

1. Mahmud FH, Elbarbary NS, Fröhlich-Reiterer E, et al. ISPAD clinical practice consensus guidelines 2018 compendium other complications and associated conditions in children and adolescents with type 1 diabetes. *Pediatr Diabetes*. 2018;19:275-286.
2. Craig ME, Prinz N, Boyle CT, et al. Prevalence of celiac disease in 52,721 youth with type 1diabetes: international comparison across three continents. *Diabetes Care*. 2017;40(8):1034-1040.
3. Hansen D, Brock-Jacobsen B, Lund E, et al. Clinical benefit of a gluten-free diet in type 1 diabetic children with screening-detected celiac disease: a population-based screening study with 2 years' follow-up. *Diabetes Care*. 2006;29(11):2452-2456.
4. Diniz-Santos DR, Brandão F, Adan L, Moreira A, Vicente EJ, Silva LR. Bone mineralization in young patients with type 1 diabetes mellitus and screening-identified evidence of celiac disease. *Dig Dis Sci*. 2008;53(5):1240-1245.
5. Fröhlich-Reiterer EE, Rosenbauer J, Bechtold-Dalla Pozza S, Hofer SE, Schober E, Holl RW. Predictors of increasing BMI during the course of diabetes in children and adolescents with type 1 diabetes: data from the German/Austrian DPV multicentre survey. *Arch Dis Child*. 2014;99(8):738-743.
6. Mollazadegan K, Kugelberg M, Montgomery SM, Sanders DS, Ludvigsson J, Ludvigsson JF. A population-based study of the risk of diabetic retinopathy in patients with type 1 diabetes and celiac disease. *Diabetes Care*. 2013;36(2):316-321.
7. Rohrer TR, Wolf J, Liptay S, et al. Microvascular complications in childhood-onset type 1 diabetes and celiac disease: a Multicenter longitudinal analysis of 56,514 patients from the German-Austrian DPV database. *Diabetes Care*. 2015;38(5):801-807.
8. Korkmaz H, Sozen M, Kebapcilar L. Increased arterial stiffness and its relationship with inflammation, insulin, and insulin resistance in celiac disease. *Eur J Gastroenterol Hepatol*. 2015;27(10):1193-1199.
9. Björck S, Lindehammer SR, Fex M, Agardh D. Serum cytokine pattern in young children with screening detected coeliac disease. *Clin Exp Immunol*. 2015;179(2):230-235.
10. Warncke K, Liptay S, Fröhlich-Reiterer E, et al. Vascular risk factors in children, adolescents, and young adults with type 1 diabetes complicated by celiac disease: results from the DPV initiative. *Pediatr Diabetes*. 2016;17(3):191-198.

11. Salardi S, Maltoni G, Zucchini S, et al. Celiac disease negatively influences lipid profiles in youngest children with type 1 diabetes: effect of the gluten-free diet. *Diabetes Care*. 2016;39:dc160717.
12. Picarelli A, Di Tola M, Sabbatella L, et al. Type 1 diabetes mellitus and celiac disease: endothelial dysfunction. *Acta Diabetol*. 2013;50(4):497-503.
13. Gopee E, van den Oever EL, Cameron F, Thomas MC. Coeliac disease, gluten-free diet and the development and progression of albuminuria in children with type 1 diabetes. *Pediatr Diabetes*. 2013;14(6):455-458.
14. Husby S, Koletzko S, Korponay-Szabó IR, et al. European Society for Pediatric Gastroenterology, Hepatology, and nutrition guidelines for the diagnosis of coeliac disease. *J Pediatr Gastroenterol Nutr*. 2012;54(1):136-160.
15. Kurppa K, Paavola A, Collin P, et al. Benefits of a gluten-free diet for asymptomatic patients with serologic markers of celiac disease. *Gastroenterology*. 2014;147(3):610-617.e1.
16. Scaramuzza AE, Mantegazza C, Bosetti A, Zuccotti GV. Type 1 diabetes and celiac disease: the effects of gluten free diet on metabolic control. *World J Diabetes*. 2013;4(4):130-134.
17. Simmons JH, Klingensmith GJ, McFann K, et al. Celiac autoimmunity in children with type 1 diabetes: a two-year follow-up. *J Pediatr*. 2011;158(2):276-281.e1.
18. Ukkola A, Mäki M, Kurppa K, et al. Diet improves perception of health and well-being in symptomatic, but not asymptomatic, patients with celiac disease. *Clin Gastroenterol Hepatol*. 2011;9(2):118-123.e1.
19. Pham-Short A, Donaghue KC, Ambler G, Garnett S, Craig ME. Greater postprandial glucose excursions and inadequate nutrient intake in youth with type 1 diabetes and celiac disease. *Sci Rep*. 2017;7(October 2016):45286.
20. Whitaker JKH, West J, Holmes GKT, Logan RFA. Patient perceptions of the burden of coeliac disease and its treatment in the UK. *Aliment Pharmacol Ther*. 2009;29(10):1131-1136.
21. Saadah OI, Zacharin M, O'Callaghan A, Oliver MR, Catto-Smith AG. Effect of gluten-free diet and adherence on growth and diabetic control in diabetics with coeliac disease. *Arch Dis Child*. 2004;89(9):871-876.
22. Hofer SE, Schwandt A, Holl RW. Standardized documentation in pediatric diabetology: experience from Austria and Germany. *J Diabetes Sci Technol*. 2016;10(5):1042-1049.
23. Binder E, Rohrer T, Denzer C, et al. Screening for coeliac disease in 1624 mainly asymptomatic children with type 1 diabetes: is genotyping for coeliac-specific human leucocyte antigen the right approach? *Arch Dis Child*. 2019;104(4):354-359.
24. Rosenbauer J, Dost A, Karges B, et al. Improved metabolic control in children and adolescents with type 1 diabetes: a trend analysis using prospective multicenter data from Germany and Austria. *Diabetes Care*. 2012;35(1):80-86.
25. Schaffrath Rosario A, Kurth BM, Stolzenberg H, Ellert U, Neuhauser H. Body mass index percentiles for children and adolescents in Germany based on a nationally representative sample (KiGGS 2003-2006). *Eur J Clin Nutr*. 2010;64(4):341-349.
26. Cole TJ, Green PJ. Smoothing reference centile curves: the LMS method and penalized likelihood. *Stat Med*. 1992;11:1305-1319.
27. Dost A, Bechtold-Dalla Pozza S, Bollow E, et al. for the Initiative DPV blood pressure regulation determined by ambulatory blood pressure profiles in children and adolescents with type 1 diabetes mellitus: impact on diabetic complications. *Pediatr Diabetes*. 2017;18(8):874-882.
28. DeSalvo DJ, Miller KM, Hermann JM, et al. Continuous glucose monitoring (CGM) and Glycemic control among youth with type 1 diabetes (T1D): international comparison from the T1D exchange and DPV initiative. *Pediatr Diabetes*. 2018;19(7):1271-1275.
29. Benjamini Y, Hochberg Y. Controlling the false discovery rate : a practical and powerful approach to multiple testing. *J R Stat Soc Ser B*. 1995;57(1):289-300.
30. Pham-Short A, Donaghue KC, Ambler G, Garnett S, Craig ME. Quality of life in type 1 diabetes and celiac disease: role of the gluten-free diet. *J Pediatr*. 2016;179:131-138.e1.
31. Salardi S, Maltoni G, Zucchini S, et al. Whole lipid profile and not only HDL cholesterol is impaired in children with coexisting type 1 diabetes and untreated celiac disease. *Acta Diabetol*. 2017;54(10):889-894.
32. Koletzko S, Auricchio R, Dolinsek J, et al. No need for routine endoscopy in children with celiac disease on a gluten-free diet. *J Pediatr Gastroenterol Nutr*. 2017;00(00):1.
33. Kurien M, Mollazadegan K, Sanders DS, Ludvigsson JF. A nationwide population-based study on the risk of coma, ketoacidosis and hypoglycemia in patients with celiac disease and type 1 diabetes. *Acta Diabetol*. 2015;52(6):1167-1174.
34. Fröhlich-Reiterer EE, Kaspers S, Hofer S, et al. Anthropometry, metabolic control, and follow-up in children and adolescents with type 1 diabetes mellitus and biopsy-proven celiac disease. *J Pediatr*. 2011;158:589-593.e2.
35. Castellaneta S, Piccinno E, Oliva M, et al. High rate of spontaneous normalization of celiac serology in a cohort of 446 children with type 1 diabetes: a prospective study. *Diabetes Care*. 2015;38(5):760-766.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of this article.

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