



The natural history of emerging diabetic retinopathy and microalbuminuria from prepuberty to early adulthood in Type 1 diabetes: A 19-year prospective clinical follow-up study

Journal:	<i>Diabetic Medicine</i>
Manuscript ID	DME-2021-00551.R1
Wiley - Manuscript type:	Research Article
Date Submitted by the Author:	13-Jul-2021
Complete List of Authors:	Tienhaara, Emmi; Oulun yliopisto Falck, Aura; Oulun yliopisto Pokka, Tytti; Oulun yliopisto Tossavainen, Päivi ; Oulun yliopisto
Keywords:	type 1 diabetes, children and adolescents, retinopathy, microalbuminuria, puberty

SCHOLARONE™
Manuscripts

Title:

The natural history of emerging diabetic retinopathy and microalbuminuria from prepuberty to early adulthood in Type 1 diabetes: A 19-year prospective clinical follow-up study

Running title:

Late complications of Type 1 diabetes in puberty and early adulthood

Emmi Tienhaara^{1,2}, MD, Aura A K Falck², MD, PhD, Tytti M-L Pokka, MSc¹, Päivi H Tossavainen¹, MD, PhD

Departments of Pediatrics¹ and Ophthalmology², PEDEGO Research Group and Medical Research Center, University of Oulu and Oulu University Hospital, PO Box 23, 90029 OYS, Oulu, Finland

<https://orcid.org/0000-0003-0388-9039>

Email addresses:

emmi.tienhaara@gmail.com; aura.falck@oulu.fi; tytti.pokka@oulu.fi;

paivi.tossavainen@oulu.fi

Corresponding author:

Päivi H. Tossavainen; Email address: paivi.tossavainen@oulu.fi

Word count: main text 3135 words, abstract 250 words, 22 references

Tables: 2; Figures: (2); Supplemental Figures: (3)

Conflicts of interest

The authors do not have any relevant conflicts of interests to disclose.

- What is already known?

High long-term HbA1c and possibly high HbA1c variability carry a risk for late complications in adolescents with Type 1 diabetes.

- What this study had found?

Pubertal development, long diabetes duration and elevated HbA1c predispose young people with Type 1 diabetes to microvascular complications during early adulthood. Although girls seem to develop first microvascular complications at earlier pubertal stages than boys, the prevalence of diabetic retinopathy ends up being equal for the sexes by early adulthood.

- What are the clinical implications of the study?

The findings that adolescents with long duration of diabetes and long term high HbA1c are prone to microvascular complications by early adulthood underpin the need to prevent pubertal deterioration of glycemic control.

Acknowledgements

The authors are grateful to the participants and their families for their contribution during the follow-up, and to Päivi Tapanainen, MD, PhD, for reviewing the manuscript.

Funding: The study was funded (P.T.) by The Alma and K.A. Snellman Foundation, Oulu, Finland, and The Foundation for Pediatric Research, Finland

Objective

To evaluate the impact of long-term glycemic control and glycemic variability on microvascular complications in adolescents and young adults with childhood-onset Type 1 diabetes.

Methods

Twenty-six participants took part in a prospective follow-up study. We used univariate generalised estimating equations (GEE) analysis with first-order autoregressive AR(1) covariance structure for repeated measurements to evaluate relationship between emerging diabetic retinopathy (DR) and each single explanatory variable, namely age at developmental stages from late prepuberty until early adulthood, duration of diabetes and long-term HbA1c. Thereafter, the simultaneous effect of these three explanatory variables to DR was analysed in a multivariate model.

Results

Twenty-five participants developed DR by early adulthood after a median diabetes duration of 16.2 years (range 6.3 – 24.0). No participants had DR during prepuberty. Each of the three variables was independently associated with emerging DR: age (OR 1.47, 95% CI to 1.25 to 1.74, $P<0.001$) stronger than diabetes duration (OR 1.42, 95% CI 1.23 to 1.63, $P<0.001$) and HbA1c (OR 1.02, 95% CI 1.001 to 1.05, $P=0.041$) in this population. In the multivariate analysis of these three explanatory variables only age was associated with DR (adjusted OR 1.52, 95% CI 1.10 to 2.10, $P=0.012$).

Conclusions

Emergence of DR during adolescence and early adulthood is not rare and increases with age in patients with deteriorating metabolic control during puberty and thereafter. This underpins the need to prevent deterioration of glycemic control from taking place during puberty - seen again in this follow-up study - in children with diabetes.

Key words: Type 1 diabetes, adolescents, diabetic retinopathy, microalbuminuria,
puberty, follow-up study.

For Peer Review

Introduction

Microvascular complications are very seldom detectable in prepubertal children with Type 1 diabetes. However, at least early markers of diabetic retinopathy (DR) may be detected in thorough examinations (1). Changes in the vascular morphology of the retina, such as venular tortuosity, may precede DR lesions during the early course of diabetes, and decreased peripheral nerve conduction or markers of autonomic nerve dysfunction without any obvious symptoms may develop soon after diabetes diagnosis in children (2-4). According to results from some studies the effect of prepubertal duration of diabetes on late complications of Type 1 diabetes would be minimal (5), while some other series have shown that the prepubertal years matter (6). Onset age of diabetes under 15 years has been found to be a more notable risk factor for microvascular complications than an onset after puberty or during adulthood (7). High long-term HbA1c and possibly high HbA1c variability carry a risk for late complications (8), and together with a high-normal urine albumin excretion rate, it may predispose to microalbuminuria (MA) and increase the risk for early cardiovascular morbidity and dysfunction (9;10). Puberty may enhance the development of structural changes in microvasculature in children with Type 1 diabetes, especially in those with prolonged poor glycemic control. In the long term, measured high glycemic control, e.g., with HbA1c, is harmful to the endothelial system, while there are some controversies regarding whether HbA1c variability over time is a predisposing factor to long-term complications (8;11).

In the present study, we prospectively followed a cohort of adolescents with Type 1 diabetes from the onset of diabetes, i.e. from prepubertal years until early adulthood. The aim was to evaluate the impact of prepubertal and pubertal long-term glycemic control and glycemic variability on DR and MA, during and after adolescence.

Study participants, design and methods

Twenty-six prepubertal children with Type 1 diabetes participated. The participants of the present population based series initially took part in a previously described cross-sectional population-based cohort study of 138 children and adolescents over nine years of age of whom 101 agreed to take part. The 26 prepubertal participants of the cross-sectional study were recruited to the present follow-up survey (3;12). The inclusion criteria were: 1) participation in the cross-sectional study, 2) age >8 years, 3) duration of diabetes >2 years, and 4) Tanner pubertal stage 1 (prepubertal) (13).

Evaluation for DR and MA was performed once at every pubertal stage and thereafter at the time of transition to adult care as well as once during early adulthood, i.e., 20 – 30 years of age. All participants were followed in the same pediatric outpatient diabetes unit until an individually scheduled transition to an adult clinic. In Finland, there is a national health insurance for the entire population, and the people with Type 1 diabetes are followed up in specialised diabetes clinics in central hospitals (children and adolescents), or in public health centres or central hospitals (adults), and insulin treatment is fully covered for subjects with diagnosis of Type 1 diabetes.

During adolescence, pubertal staging was assessed according to Tanner pubertal stages based on breast development in girls and testes developmental stage in boys (13). Since some participants missed either visit T 4 or T 5, we used the data from the available visit in the analysis; the data from T 5 were taken into the final analysis for those who attended both T 4 and T 5 study visits. One participant had pubertal follow-up data only from T 1 to T 3 and data from early adulthood because of moving away from the hospital catchment area for a couple of years. The data regarding early adulthood were gathered from hospital registries.

All participants and their parents or guardians gave informed consent before entering the pubertal follow-up study. The study was approved by the Ethics Committee of the Faculty of Medicine, University of Oulu, Finland and the Regional Ethics Committee of the Northern Ostrobothnia Hospital District, Oulu, Finland. The study was carried out

according to the Good Clinical Practice guidelines and the provisions of the Declaration of Helsinki.

One of the researchers (P.T.) arranged the study visits, assessed the participants' pubertal stage during outpatient visits, and scheduled the clinical examinations during the study. E.T. and P.T. gathered the data from the participants' medical records.

HbA1c. All HbA1c (mmol/mol and %) results available from 1992 on were included in the analysis. HbA1c was analysed by high-pressure liquid chromatography up to May 2000, and thereafter, by an immunoassay-based method.

Diabetic retinopathy. A clinical ophthalmologist and A.F. from the present study group classified fundus photographs into five stages: grade 1) no DR; grade 2) mild background DR characterized by microaneurysms only; grade 3) intermediate background DR with intraretinal hemorrhages, hard exudates or cotton wool spots present in addition to microaneurysms; grade 4) advanced (preproliferative) background DR; and grade 5) proliferative DR (14).

Urine albumin excretion rate. The urine albumin excretion rate (AER) was measured at least once a year and at every study visit. The definition of persistent MA was two positive measurements out of at least three samples with an AER of 20-200 micrograms/ min in overnight collected urine or a urine albumin-to-creatinine ratio of 2.5-25 mg/mmol in boys/men and 3.5-25 mg/mmol in girls/women.

Statistical methods

Descriptive statistics are expressed as the mean and standard deviation (SD) or median and range. The *group mean HbA1c* data for various study time points were calculated.

1) Before follow-up; the mean of all available HbA1c measurements between the end of the remission phase (i.e. at least 12 months after diabetes onset) and before the first follow-up study visit. 2) At the beginning of the follow-up (Tanner stage 1), during puberty (Tanner stages 2-5) and transition; the mean of the HbA1c measurements during individual study visits. 3) Early adulthood; HbA1c at the nearest time-point

1 before or after the first available eye fundus photograph taken after transition to an adult
2 clinic.
3

4 The *long-term HbA1c parameters* were calculated to evaluate the effect of long-term
5 metabolic control to DR: Two long-term HbA1c parameters, namely, mean HbA1c
6 (HbA1c-total) and the variability of HbA1c (HbA1c-SD) based on all available HbA1c
7 measurements after the remission phase, were calculated for each participant. *HbA1c-*
8 *SD* was adjusted for the number of HbA1c (n) measurements by dividing HbA1c-SD by
9 the equation $\sqrt{n/(n-1)}$ (15). *HbA1c-total* included all available HbA1c values after the
10 remission phase until early adulthood. These two parameters were also calculated
11 separately for prepubertal phase i.e., before follow-up, pubertal follow-up from Tanner 1
12 to 5, transition, and early adulthood. The median number of HbA1c measurements for
13 an individual participant was 22 (range 14 - 47) during the entire duration after
14 remission phase, including prepubertal (median 8, range 3-11) measurements.
15 Additionally, Altman individual plots of HbA1c values from pubertal onset until transition
16 time were created (Supplemental Fig. S1).
17

18 *Univariate and multivariate generalised estimating equations (GEE)*. First we used
19 univariate generalised estimating equations (GEE) analysis with first-order
20 autoregressive AR(1) covariance structure for repeated measurement analysis to
21 evaluate relationship between DR and each single explanatory variable i.e. age at
22 developmental stages from late prepuberty until early adulthood, duration of diabetes
23 and HbA1c-total. Thereafter we used a multivariate model to examine the simultaneous
24 effect of these three explanatory variables on emerging DR. Since there was a
25 significant correlation between the HbA1c-total and HbA1c-SD ($r=0.56$), HbA1c-total
26 was chosen for the GEE analyses. The odds ratios in GEE analysis represent the effect
27 of every one unit change in explanatory variable to expected log odds of emerging DR.
28 We tested the differences between independent group means with a Student's t-test for
29 two groups, and with one-way ANOVA for more than two groups. A paired t-test was
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60

used to test the means of dependent variables. Pearson correlation coefficient (r) was used to find univariate correlation between the variables. The 95% CI for differences between the means are shown. Logistic regression analysis was carried out to estimate the independent associations of prepubertal and pubertal HbA1c parameters with emerging DR during early adulthood. Kaplan-Meier analysis was calculated to examine DR-free time in relation to long-term glycemic control: HbA1c-total above or equal vs. below the group mean of 74 mmol/mol (8.9%). A P value <0.05 was considered statistically significant. The statistical analyses were performed using IBM SPSS for Windows version 27 (IBM Corp, Armonk, NY).

Results

Twenty-six adolescents (15 boys and 11 girls) participated, and the study comprised 513 person-years of follow-up. The basic characteristics of the study participants at the various follow-up time points are shown in Table 1. The mean age at diabetes diagnosis was 5.1 years (± 2.7 , range 1.0 - 8.9), while the age at the end of the follow-up was 24.6 (± 3.1) years and the median duration of diabetes was 19.5 (± 3.3 , range 11.2 – 24.0) years. All participants were on multiple-dose insulin injections and none of them used continuous glucose monitoring by the time of transition to adult care. During early adulthood, 19 participants continued with injections and four started using an insulin pump. The information is missing in three cases.

Group mean HbA1c: The dynamics of the group mean HbA1c from prepuberty until early adulthood was different between female and male participants. Although HbA1c levels varied more in the female than in the male participants, there were no clear statistically significant differences between the sexes. The group mean HbA1c was highest during the transition phase at 82 (± 19) mmol/mol (9.7%), and lowest during the prepubertal years 63 (± 12) mmol/mol (7.9%). (Table 1 and Supplemental Fig. S2).

There was a no significant correlation between diabetes duration and any of the HbA1c parameters. There were no significant differences in HbA1c-SD between the sexes during prepuberty (female 1.01 ± 0.89 vs. male 1.29 ± 0.98 ; HbA1c %) or during the entire follow-up (1.54 ± 0.54 vs. 1.47 ± 0.50).

Retinopathy and the long-term HbA1c parameters: The earliest diagnostic lesions of mild DR were detected at Tanner stage 2 in a boy and in one girl at Tanner stage 3. The minimum age at the first detection of DR lesions was 12.2 years. The first DR lesions appeared after a median diabetes duration of 16.2 years (range 6.3 – 24.0). DR incidence and severity increased during pubertal maturation, and in early adulthood the majority of the participants had at least intermediate background DR, while one-fourth of participants (n=7; 27%) had DR grade 4 or 5 (Fig. 1A). In Figure 1 B the associations between HbA1c-total, diabetes duration and DR along pubertal maturation are shown. Figure 2 shows DR grades during early adulthood in relation to the duration of diabetes and individually calculated HbA1c-total. In the logistic regression analysis, HbA1c-total (P=0.021, OR 1.2) and mean pubertal HbA1c (P=0.033, OR 1.13) showed an independent association with advanced DR (grade 3 to 5), while mean prepubertal HbA1c, all of the HbA1c-SD parameters and diabetes duration did not associate with DR severity.

Each of the three variables was independently associated with emerging DR: age (OR 1.47, 95% CI to 1.25 to 1.74, P<0.001) stronger than diabetes duration (OR 1.42, 95% CI 1.23 to 1.63, P<0.001) and HbA1c (OR 1.02, 95% CI 1.001 to 1.05, P=0.041) in this population. In the multivariate analysis of these three explanatory variables only age was associated with DR (adjusted OR 1.52, 95% CI 1.10 to 2.10, P=0.012). (Table 2). The Kaplan-Meier analyses show that participants with HbA1c-total ≥ 74 mmol/mol (8.9%) tended to develop DR at an earlier age than those who had lower mean HbA1c (Fig. 3).

Microalbuminuria. The girls had significantly higher AER rates than the boys during Tanner 3 stage (ln alb 2.60 ± 1.08 vs. 1.60 ± 0.75 , $P=0.011$, 95% CI 0.26, 1.74) and late puberty (ln alb 2.71 ± 0.91 vs. 1.85 ± 0.50 , $P=0.009$, 95% CI 0.24, 1.48 for combined Tanner 4 and 5 stages) (Supplemental Fig. S3). The calculated long-term HbA1c parameters were not significantly associated with the appearance of MA by transition time (HbA1c-total in logistic regression analysis $P=0.069$, OR 3.6).

Discussion

In the present population based study, we evaluated the impact of long-term glycemic control, diabetes duration and advancing pubertal development on microvascular complications in a prospective follow-up cohort of adolescents with Type 1 diabetes. In this small but very thoroughly examined patient cohort, with a mean diabetes duration of 19 years and a follow-up time of over 500 person years, most participants developed DR by early adulthood. In univariate GEE analysis, significant associations between emerging DR and age at study follow-up stages, diabetes duration and HbA1c-total were found. However, in multivariate analysis only advancing age along study follow-up stages was associated with DR when adjusted with diabetes duration and HbA1c-total. These findings reflect the complex nature of development of late complications of diabetes in young age.

The study participants were prospectively followed from prepuberty until early adulthood, and glycemic parameters were calculated. Follow-up evaluations were performed uniformly during the study for all the participants along pubertal maturation, exact data were also available from the patient records during early adulthood, and all participants stayed with the study, which are the main strengths of the present study. The main weakness of the present study is the relatively small number of subjects. This is related to the fact that the large original cross-sectional cohort of 138 young participants with Type 1 diabetes. The cohort was divided into five groups (12)

1 according to Tanner pubertal staging (13), and only the prepubertal children were asked
2 to take part in this follow-up study with the aim to evaluate diabetic microvascular
3 changes during adolescence in every pubertal stage in a clinical setting.
4

5 Secondly, some participants missed either the Tanner 4 or the Tanner 5 visit. Arranging
6 a prospective clinical follow-up study of adolescent subjects has its challenges as it may
7 be especially difficult for an adolescent to engage in a long-term follow-up study, and
8 that some lapses seem almost inevitable in pubertal participants. Beyond the present
9 study, there are not very many other studies on the long-term prognosis of Type 1
10 diabetes with pubertal staging (12;16).
11

12 There could be selection bias, e.g. children and adolescents with no major problems in
13 diabetes care could be more likely to take part, and those not willing to participate in a
14 study would likely have more rather than less problems related to diabetes. During the
15 data analyses of the original larger cross-sectional study, the mean HbA1c levels
16 between participants who entered and those who did not enter the study, were found
17 similar, however. The early adulthood clinical data were mainly gathered retrospectively
18 from patient records. The fact that health care of young adults with Type 1 diabetes is
19 centralised to special follow-up clinics was helpful for data collection. The patients under
20 19 years of age with childhood onset Type 1 diabetes (around 500 patients) living in the
21 catchment area of the Oulu University Hospital are followed in the same tertiary clinic.
22 This in part ensures that the present cohort right represents children and adolescents
23 with Type 1 diabetes. Our center in the Northern Ostrobothnia Hospital District is the
24 second largest pediatric diabetes center in Finland, and the incidence of new diagnoses
25 in children under 16 years is 55-65/ 100000/ year.
26

27 The follow-up period was long and the median number of individual HbA1c
28 measurements was relatively high and covered well the entire diabetes duration for
29 every participant in this study. We found no statistically significant associations between
30 long-term HbA1c-SD and DR in the present cohort, but as the patient number was
31

small, a connection cannot be ruled out. There are somewhat contrary findings in other studies regarding long-term variation in HbA1c in relation to microvascular complications in adults and adolescents with Type 1 diabetes (8;15).

The participants in the present study had rather high mean HbA1c levels, which underpins the need for strict metabolic control during prepubertal and pubertal years.

The study was initiated before the era of any or wide use of glucose sensors and insulin pumps, and none of the participants had sensor augmented pump before reaching early adulthood. Pubertal development, characterised by hormonal and psychological changes, makes Type 1 diabetes treatment demanding and challenging. Diabetes clinics and caretakers have a responsibility to provide an opportunity for good self-care; however, failure to accept a diabetes diagnosis, especially during puberty, can lead to treatment neglect, which then hampers reaching good metabolic control (17;18).

Generally, people with Type 1 diabetes diagnosis during early childhood, seem to be more prone to developing DR than those diagnosed in adulthood. In the FinnDiane Study, the risk for proliferative DR was highest in people with diabetes diagnosed during childhood (7). Additionally, the finding that one-quarter of the participants in the present follow-up cohort had developed proliferative DR by early adulthood is in line with earlier long-term studies (7). Drawing definitive quantitative conclusions about the long-term impact of childhood and pubertal glycemic control or HbA1c variability on advanced microvascular changes during early adulthood is not possible based on the present study.

Glycemic control often deteriorates during adolescence and improving insulin sensitivity again after the pubertal years is a phenomenon that is at least partly explained by both increased self-treatment compliance and increased tissue insulin sensitivity. The glycemic control in the present cohort deteriorated from prepuberty to late puberty, especially among the girls, and the HbA1c levels were highest during late puberty and transition. The finding of slightly higher glycemic control may be clinically important as

1 girls were found to develop DR lesions earlier than boys. Girls generally experience
2 earlier pubertal development, which may predispose them to earlier pubertal-induced
3 insulin resistance, deteriorating glycemic control and microvascular lesions. These
4 findings of increasing impairment of glycemic control during pubertal years in the
5 present study are in line with earlier surveys (19).
6
7
8
9
10

11 The finding that no long-term HbA1c parameters were associated significantly with the
12 appearance of MA during puberty in the study population underpins the theory that
13 other parameters beyond metabolic control may interfere during puberty. Additionally,
14 poor metabolic control during puberty in individuals with diabetes may reflect a deviant
15 growth hormone – IGF-1 axis (20;21). In our cohort, MA and HbA1c levels were higher
16 in female than in male participants during puberty, which may be related to such
17 deterioration of hormonal balance during puberty.
18
19
20
21
22
23
24
25
26

27 The high prevalence of any or proliferative DR in the present series of participants with
28 Type 1 diabetes onset before the era of glucose sensors and wide use of insulin pumps
29 is in line with earlier reports, with a DR prevalence of over 80% after a duration of
30 diabetes of over 20 years (22). The main goal in diabetes care is to maintain glycemic
31 levels near normal, and there are also encouraging data showing that the incidence of
32 DR may decline in the future, and there are numerous treatment options in addition to
33 good glycemic control, which improve the prognosis of people with DR and diabetic
34 kidney disease.
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52

53 References

- 54 (1) Donaghue KC, Fairchild JM, Chan A, Hing SJ, King J, Howard NJ, et al.
55 Diabetes microvascular complications in prepubertal children. J Pediatr
56 Endocrinol Metab 1997 Nov;10(6):579-85.
57
- 58 (2) Liew G, Benitez-Aguirre P, Craig ME, Jenkins AJ, Hodgson LAB, Kifley A, et al.
59 Progressive Retinal Vasodilation in Patients With Type 1 Diabetes: A
60

Longitudinal Study of Retinal Vascular Geometry. Invest Ophthalmol Vis Sci 2017 May 1;58(5):2503-9.

- (3) Riihimaa PH, Suominen K, Tolonen U, Jantti V, Knip M, Tapanainen P. Peripheral nerve function is increasingly impaired during puberty in adolescents with type 1 diabetes. Diabetes Care 2001 Jun;24(6):1087-92.
- (4) Tang M, Donaghue KC, Cho YH, Craig ME. Autonomic neuropathy in young people with type 1 diabetes: a systematic review. Pediatr Diabetes 2013 Jun;14(4):239-48.
- (5) Kostraba JN, Dorman JS, Orchard TJ, Becker DJ, Ohki Y, Ellis D, et al. Contribution of diabetes duration before puberty to development of microvascular complications in IDDM subjects. Diabetes Care 1989 Nov;12(10):686-93.
- (6) Donaghue KC, Fung AT, Hing S, Fairchild J, King J, Chan A, et al. The effect of prepubertal diabetes duration on diabetes. Microvascular complications in early and late adolescence. Diabetes Care 1997 Jan;20(1):77-80.
- (7) Hietala K, Harjutsalo V, Forsblom C, Summanen P, Groop PH. Age at onset and the risk of proliferative retinopathy in type 1 diabetes. Diabetes Care 2010 Jun;33(6):1315-9.
- (8) Virk SA, Donaghue KC, Cho YH, Benitez-Aguirre P, Hing S, Pryke A, et al. Association Between HbA1c Variability and Risk of Microvascular Complications in Adolescents With Type 1 Diabetes. J Clin Endocrinol Metab 2016 Sep;101(9):3257-63.
- (9) Lu L, Marcovecchio ML, Dalton RN, Dunger D. Cardiovascular autonomic dysfunction predicts increasing albumin excretion in type 1 diabetes. Pediatr Diabetes 2018 May;19(3):464-9.
- (10) Marcovecchio ML, Dalton RN, Daneman D, Deanfield J, Jones TW, Neil HAW, et al. A new strategy for vascular complications in young people with type 1 diabetes mellitus. Nat Rev Endocrinol 2019 Apr 17.
- (11) DCCT Research Group. The relationship of glycemic exposure (HbA1c) to the risk of development and progression of retinopathy in the diabetes control and complications trial. Diabetes 1995 Aug;44(8):968-83.
- (12) Riihimaa PH, Suominen K, Knip M, Tapanainen P, Tolonen U. Cardiovascular autonomic reactivity is decreased in adolescents with Type 1 diabetes. Diabet Med 2002 Nov;19(11):932-8.
- (13) Tanner JM. Normal growth and techniques of growth assessment. Clin Endocrinol Metab 1986 Aug;15(3):411-51.
- (14) Wilkinson CP, Ferris FL, III, Klein RE, Lee PP, Agardh CD, Davis M, et al. Proposed international clinical diabetic retinopathy and diabetic macular edema disease severity scales. Ophthalmology 2003 Sep;110(9):1677-82.
- (15) Kilpatrick ES, Rigby AS, Atkin SL. A1C variability and the risk of microvascular complications in type 1 diabetes: data from the Diabetes Control and Complications Trial. Diabetes Care 2008 Nov;31(11):2198-202.

- 1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60
- (16) Zhu J, Volkeneing LK, Laffel LM. Distinct Patterns of Daily Glucose Variability by Pubertal Status in Youth With Type 1 Diabetes. *Diabetes Care* 2020 Jan;43(1):22-8.
- (17) Borus JS, Laffel L. Adherence challenges in the management of type 1 diabetes in adolescents: prevention and intervention. *Curr Opin Pediatr* 2010 Aug;22(4):405-11.
- (18) Court JM, Cameron FJ, Berg-Kelly K, Swift PG. Diabetes in adolescence. *Pediatr Diabetes* 2009 Sep;10 Suppl 12:185-94.
- (19) Rausch JR, Hood KK, Delamater A, Shroff PJ, Rohan JM, Reeves G, et al. Changes in treatment adherence and glycemic control during the transition to adolescence in type 1 diabetes. *Diabetes Care* 2012 Jun;35(6):1219-24.
- (20) Amin R, Schultz C, Ong K, Frystyk J, Dalton RN, Perry L, et al. Low IGF-I and elevated testosterone during puberty in subjects with type 1 diabetes developing microalbuminuria in comparison to normoalbuminuric control subjects: the Oxford Regional Prospective Study. *Diabetes Care* 2003 May;26(5):1456-61.
- (21) Dunger DB. Diabetes in puberty. *Arch Dis Child* 1992 May;67(5):569-70.
- (22) Klein R, Knudtson MD, Lee KE, Gangnon R, Klein BE. The Wisconsin Epidemiologic Study of Diabetic Retinopathy: XXII the twenty-five-year progression of retinopathy in persons with type 1 diabetes. *Ophthalmology* 2008 Nov;115(11):1859-68.

Table 1. The descriptive data of the study population.

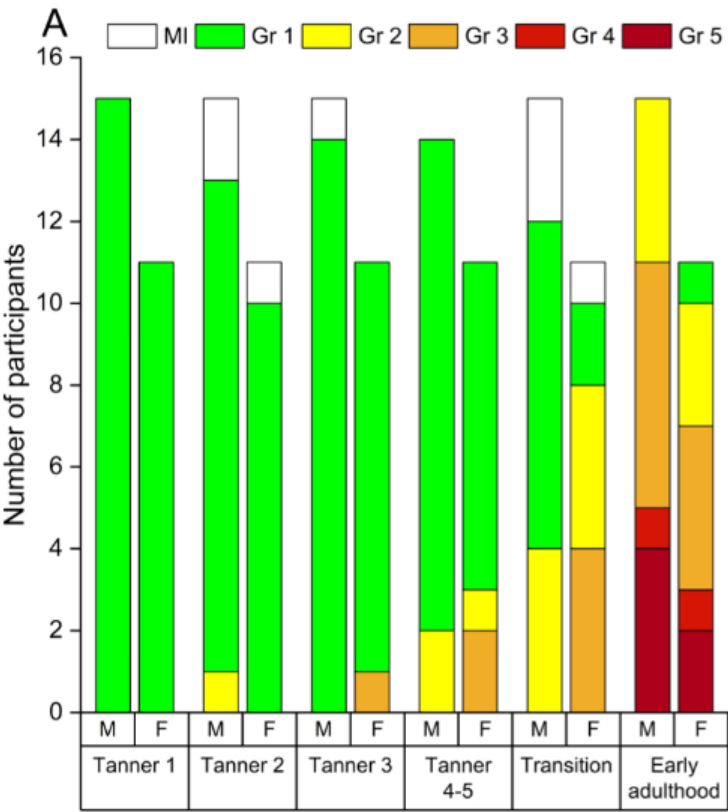
The study follow-up stages						
	Tanner 1	Tanner 2	Tanner 3	Tanner 4-5	Transition	Early adulthood
Age, yr						
Female	10.5 ± 0.7	12.0 ± 0.7	12.9 ± 0.6	15.7 ± 1.0	18.0 ± 0.8	23.0 ± 2.9
Male	11.7 ± 0.6	12.7 ± 0.8	13.8 ± 0.7	14.9 ± 0.7	17.0 ± 0.9	25.8 ± 2.7
Total	11.2 ± 0.9	12.4 ± 0.8	13.4 ± 0.8	15.3 ± 0.9	17.5 ± 1.0	24.6 ± 3.1
Diabetes duration, yr						
Female	5.8 ± 2.9	7.3 ± 2.8	8.1 ± 2.9	11.0 ± 3.2	13.3 ± 2.6	18.3 ± 4.1
Male	6.3 ± 2.7	7.5 ± 3.1	8.5 ± 2.9	9.7 ± 2.9	11.8 ± 2.8	20.4 ± 2.2
Total	6.1 ± 2.8	7.4 ± 2.9	8.3 ± 2.8	10.3 ± 3.0	12.5 ± 2.8	19.5 ± 3.3
HbA1c, mmol/mol (%)						
Female	63 ± 17	70 ± 15	80 ± 23	86 ± 26	84 ± 23	70 ± 14
	(7.9 ± 1.5)	(8.6 ± 1.4)	(9.5 ± 2.1)	(10.0 ± 2.4)	(9.5 ± 2.1)	(8.5 ± 2.6)
Male	59 ± 14	67 ± 15	74 ± 13	79 ± 18	80 ± 15	77 ± 17
	(7.6 ± 1.2)	(8.2 ± 1.4)	(8.9 ± 1.2)	(9.2 ± 1.6)	(9.4 ± 1.4)	(8.9 ± 1.7)

Basic characteristics of the participants at various pubertal stages (Tanner 1 to Tanner 5), at the time of transition and during early adulthood. N=26. Data are mean ± SD. The differences in the group mean HbA1c: P<0.001, Transition vs. Tanner stage 1 (95% CI 14, 29) and P=0.038 Transition vs. Early adulthood (95% CI 0, 15); a paired t-test.

Figure 1A and 1B. Development of diabetic retinopathy from Tanner 1 stage to early adulthood in a follow-up cohort of children with type 1 diabetes (1A). Associations between HbA1c-total, diabetes duration and diabetic retinopathy along pubertal maturation (1B).

Figure 2. Stages of diabetic retinopathy relative to overall duration of diabetes and mean HbA1c (mmol/mol) calculated from pubertal onset until early adulthood in a cohort of 26 young participants with type 1 diabetes.

Figure 3. Kaplan-Meier survival analysis for the appearance of first diabetic retinopathy lesions in type 1 diabetes, when the group of participants was divided into two groups according to HbA1c-total.



M; Male, F; Female. Tanner pubertal staging 1-5; Transition, transition phase to adult clinic follow up. MI; missing information, Gr1-5 Retinopathy grades (15).

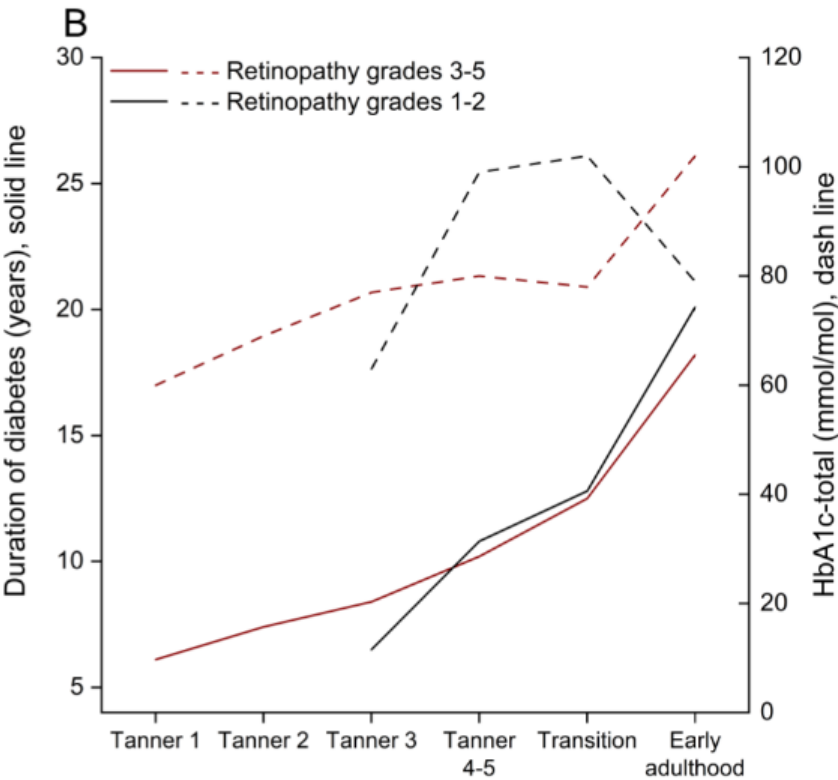
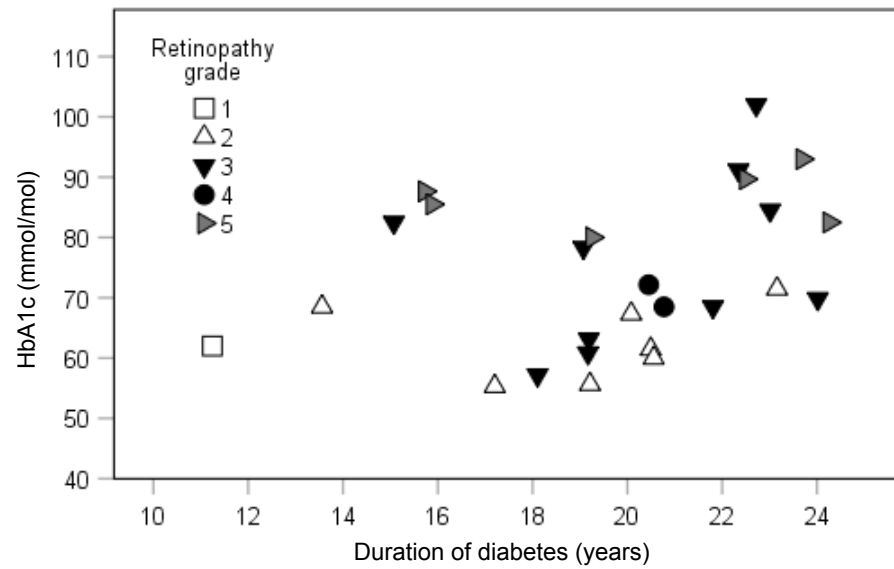
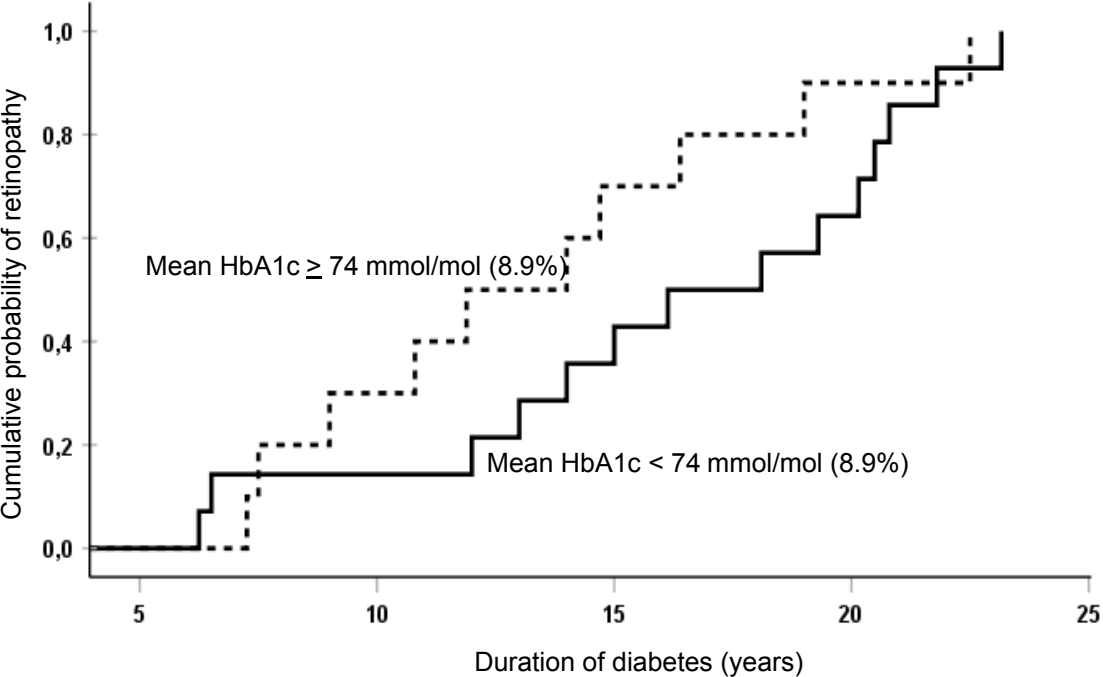


Figure 2



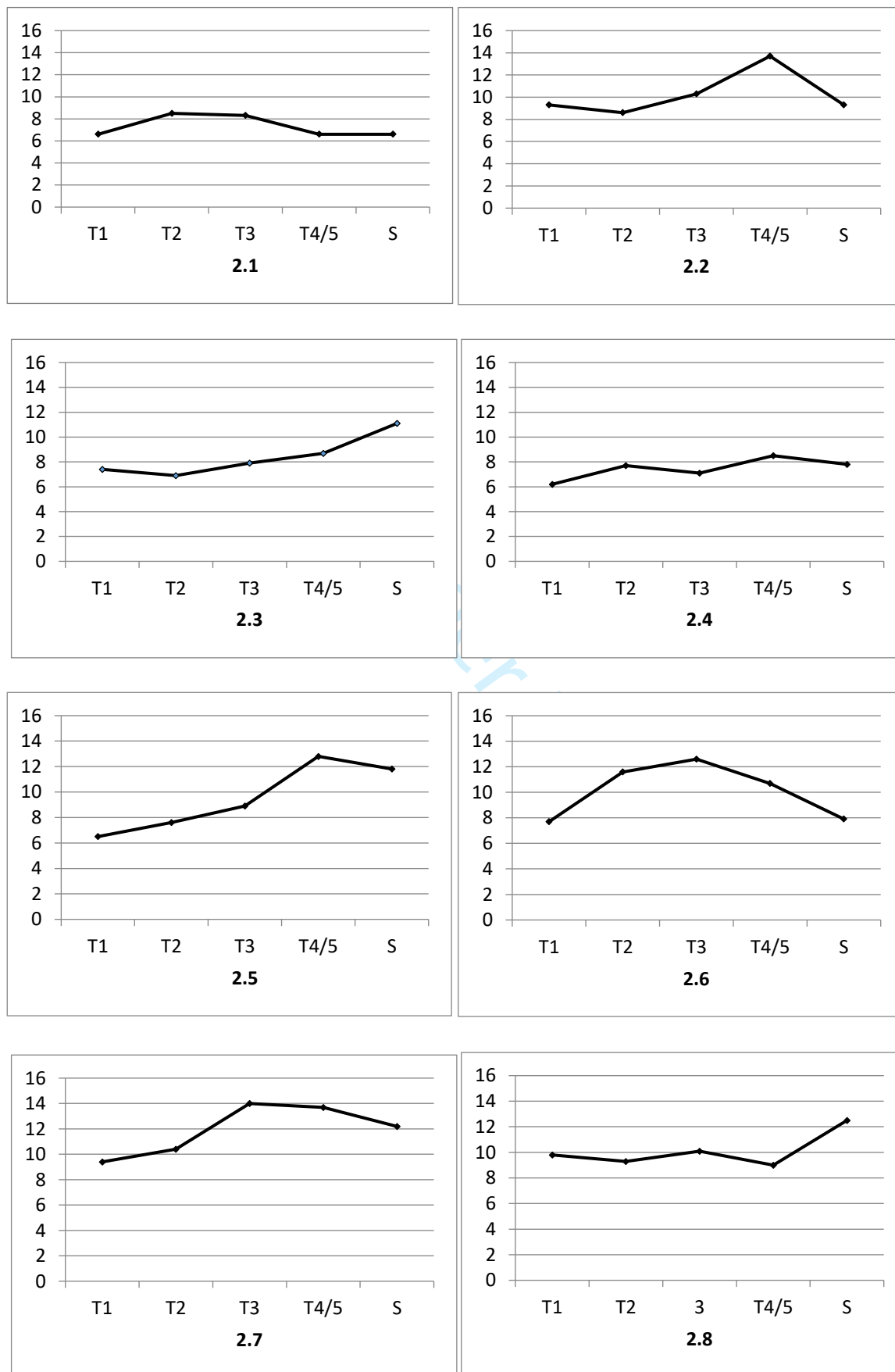
Numbers 1-5, diabetic retinopathy grades (15).

Figure 3

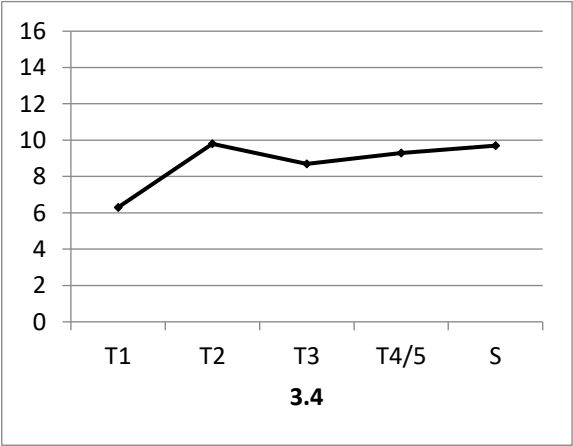
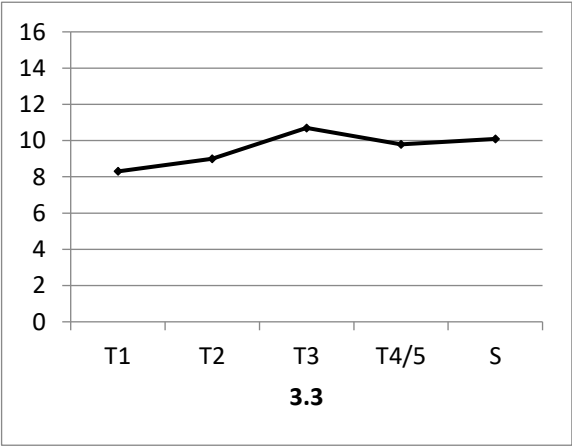
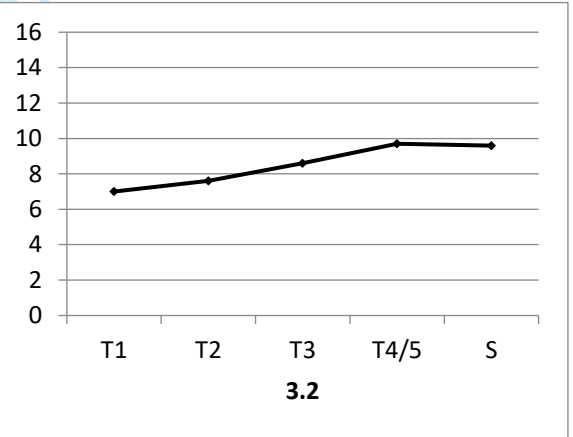
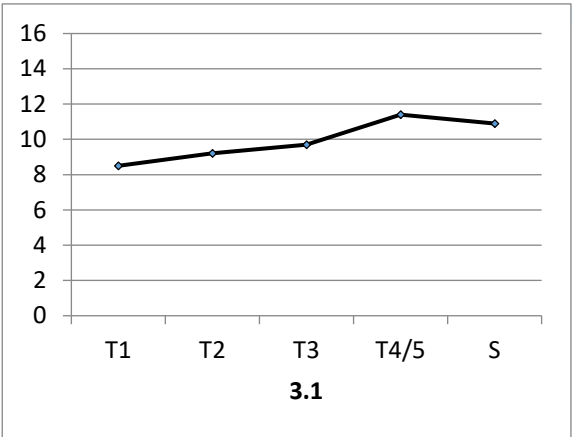
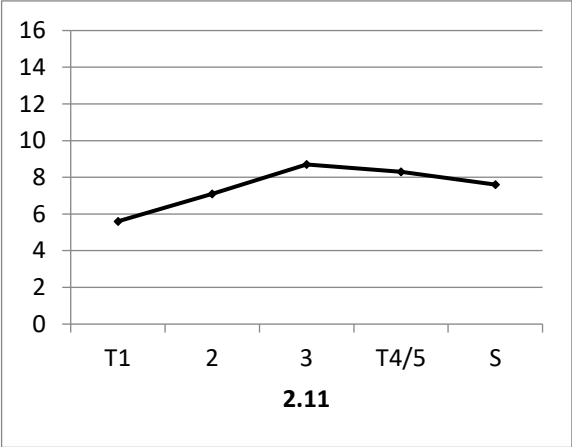
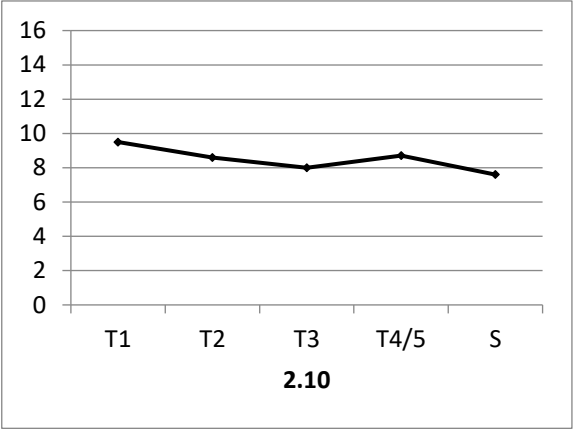
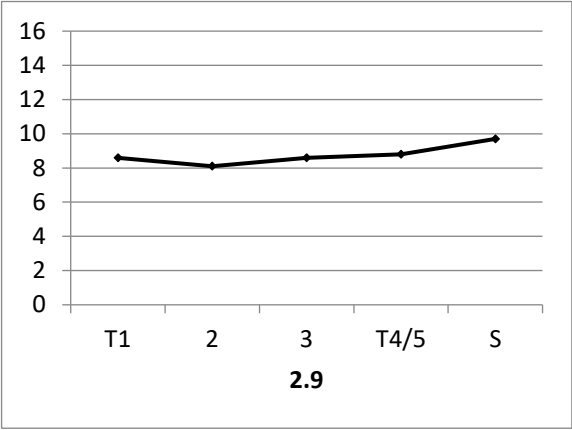


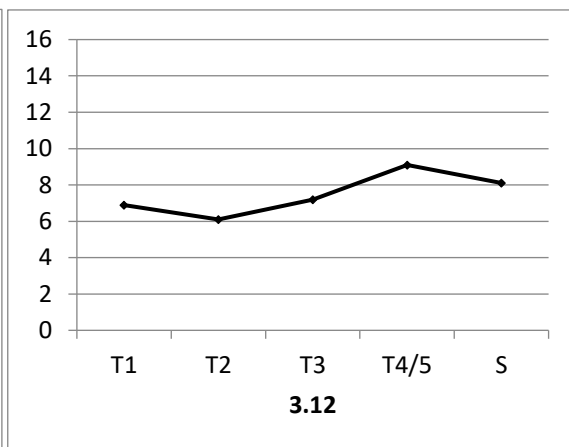
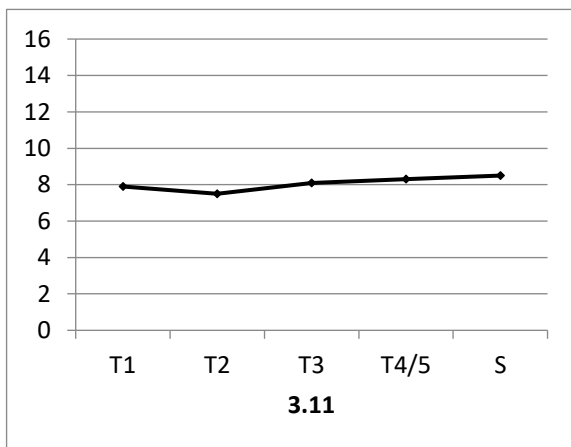
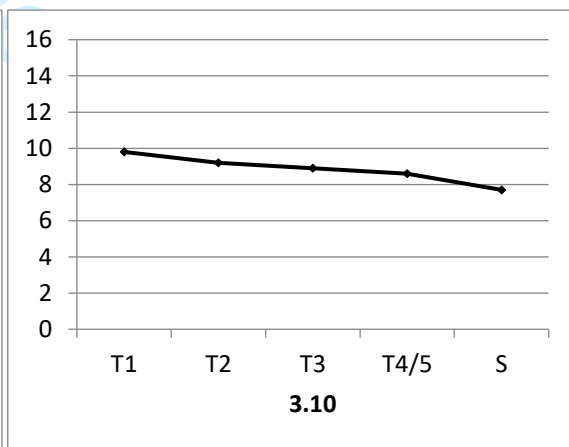
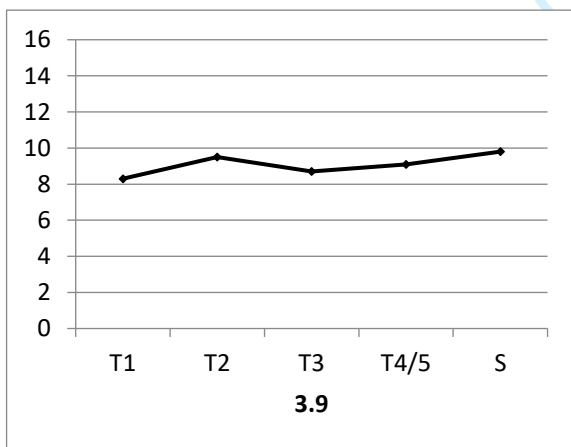
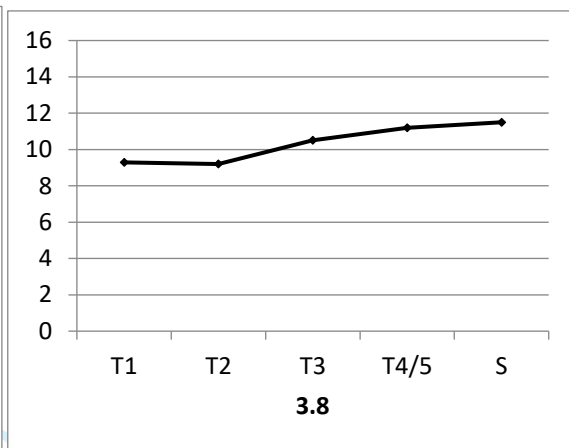
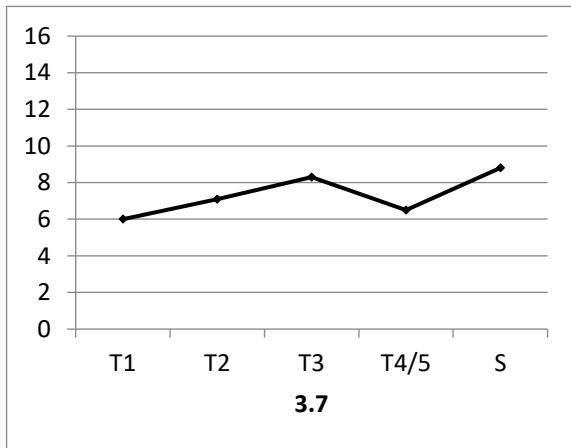
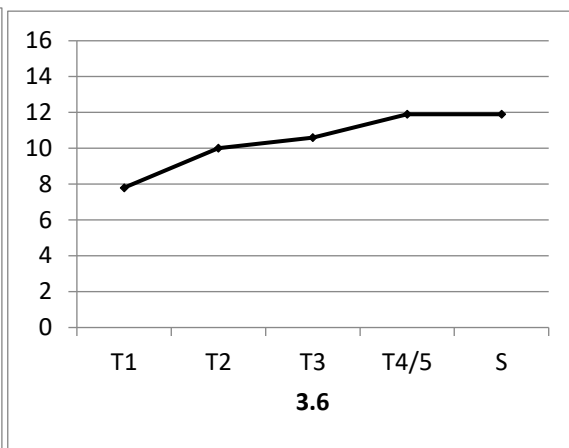
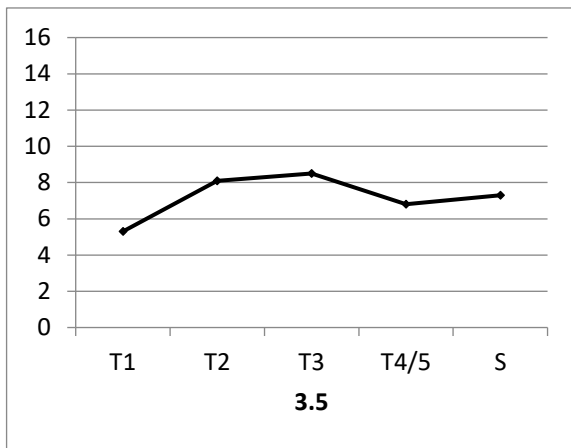
Supplemental Figure S1.

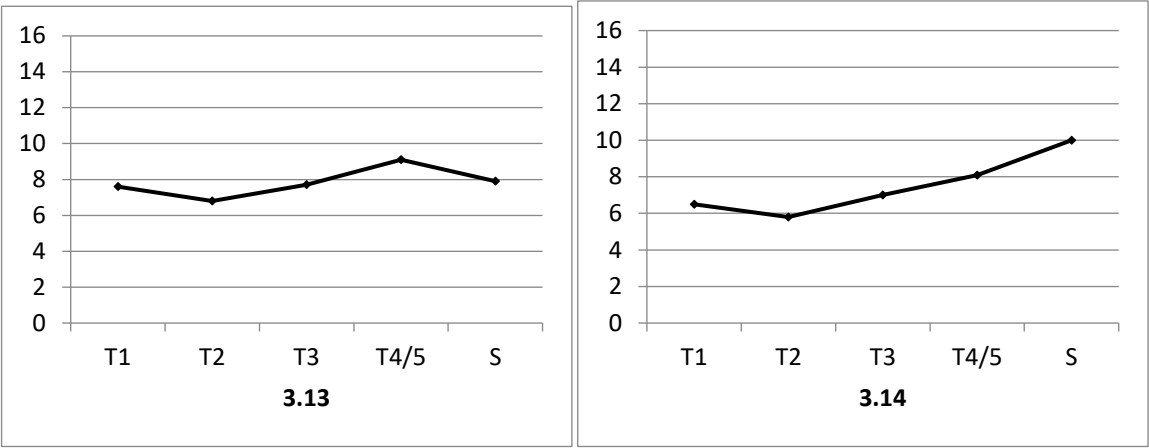
The mean of the HbA1c levels, presented as Altman individual plots of each participant at the developmental stages.



1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60



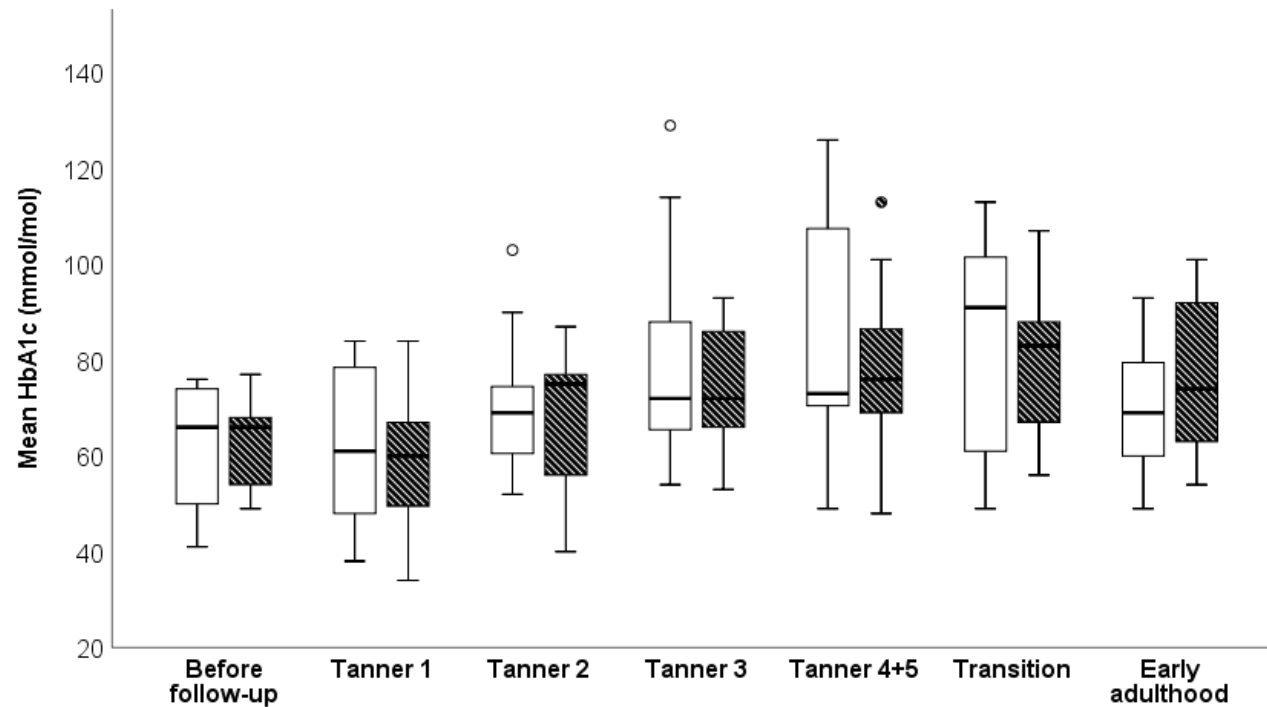




Figures 2.1-2.11. The mean HbA1c (% , vertical axis) levels in girls. **Figures 3.1-3.14.** The mean HbA1c (% , vertical axis) levels in boys. T1=Tanner 1, T2=Tanner 2, T3=Tanner 3, T4/5= Tanner 4/5 pubertal stage; S=Transition

Supplemental Figure S2.

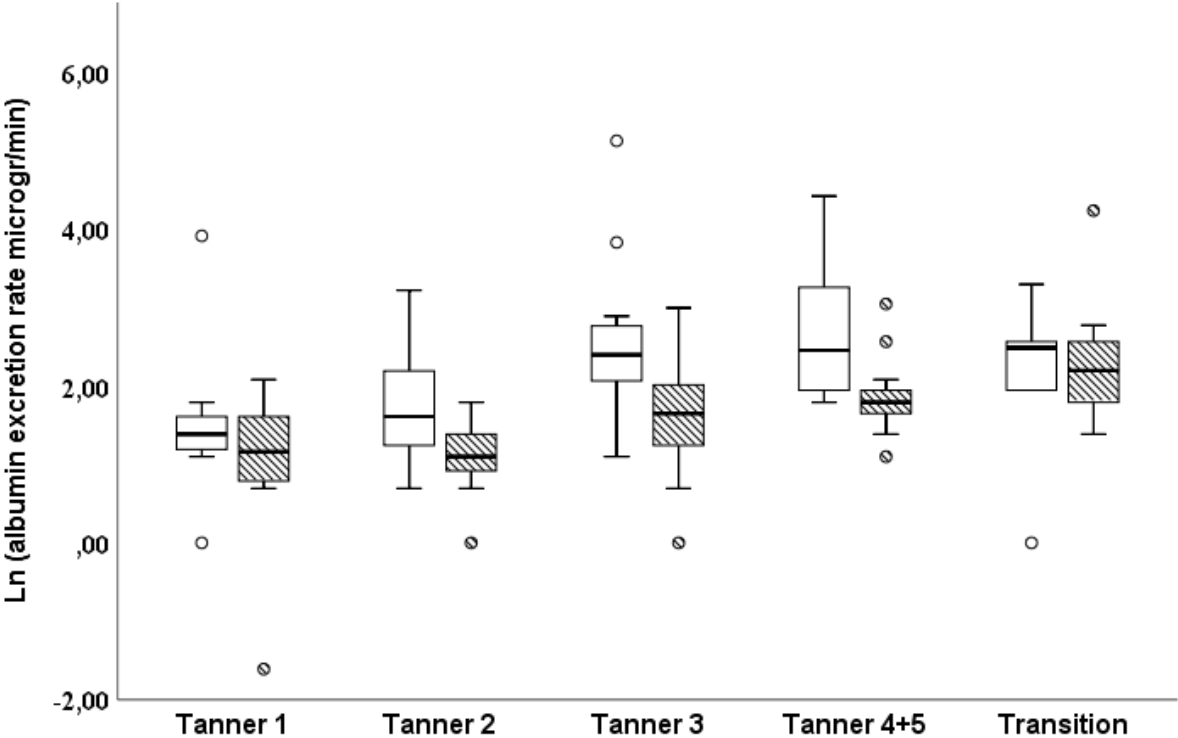
Box plots of HbA1c (mmol/mol) in various stages during the follow-up study.



Tanner pubertal stages (Tanner 4+5 means combined Tanner 4 and 5 stages).

White boxes represent girls and striped boxes represent boys. The small dots are outliers.

Supplemental Figure S3.
Box plot graph of Ln(urine albumin excretion rate) during the follow-up study.



The white boxes represent girls and striped boxes represent boys.
The small dots outside the boxes are outliers.
Tanner pubertal stages (T 4+5 means combined Tanner 4 and 5 stages).

Table 2. Associations between retinopathy (grades 3-5) and age at follow-up stages, duration of diabetes and HbA1c-total in univariate analysis [crude odds ratio (OR) with 95% confidence interval (95% CI)] and multivariate analysis (adjusted OR with 95% CI).

	Univariate analysis			Multivariate analysis		
	Crude OR	95% CI	P value ¹	Adjusted OR ²	95% CI	P value ¹
All participants						
Age at study follow-up stages (yr)	1.47	1.25 to 1.74	<0.001	1.52	1.10 to 2.10	0.012
Diabetes duration (yr)	1.42	1.23 to 1.63	0.001	1.04	0.87 to 1.26	0.659
HbA1c-total (mmol/mol)	1.02	1.001 to 1.05	0.041	1.04	0.99 to 1.09	0.104
Male						
Age at study follow-up stages (yr)	1.61	1.27 to 2.06	<0.001	1.54	0.81 to 2.94	0.191
Diabetes duration (yr)	1.68	1.32 to 2.16	<0.001	1.55	1.00 to 2.42	0.053
HbA1c-total (mmol/mol)	1.05	1.002 to 1.10	0.042	1.14	1.03 to 1.26	0.014
Female						
Age at study follow-up stages (yr)	1.54	1.25 to 1.89	<0.001	1.83	1.16 to 2.88	0.010
Diabetes duration (yr)	1.33	1.14 to 1.54	<0.001	0.88	0.66 to 1.19	0.432
HbA1c-total (mmol/mol)	1.002	0.98 to 1.03	0.983	1.02	0.97 to 1.07	0.411

Generalized estimating equations (GEE) analysis with first-order autoregressive AR(1) covariance structure for repeated measurements.

² Adjusted by other factors in model.