A1C Variability Predicts Incident Cardiovascular Events, Microalbuminuria, and Overt Diabetic Nephropathy in Patients With Type 1 Diabetes

Johan Wadén,[1,2] Carol Forsblom,[1,2] Lena M. Thorn,[1,2] Daniel Gordin,[1,2] Markku Saraheimo,[1,2] and Per-Henrik Groop,[1,2] on behalf of the Finnish Diabetic Nephropathy Study Group

OBJECTIVE—Recent data from the Diabetes Control and Complications Trial (DCCT) indicated that A1C variability is associated with the risk of diabetes microvascular complications. However, these results might have been influenced by the interventional study design. Therefore, we investigated the longitudinal associations between A1C variability and diabetes complications in patients with type 1 diabetes in the observational Finnish Diabetic Nephropathy (FinnDiane) Study.

RESEARCH DESIGN AND METHODS—A total of 2,107 patients in the FinnDiane Study had complete data on renal status and serial measurements of A1C from baseline to follow-up (median 5.7 years), and 1,845 patients had similar data on cardiovascular disease (CVD) events. Intrapersonal SD of serially measured A1C was considered a measure of variability.

RESULTS—During follow-up, 10.2% progressed to a higher albuminuria level or to end-stage renal disease, whereas 8.6% had a CVD event. The SD of serial A1C was 1.01 versus 0.75 ($P < 0.001$) for renal status and 0.87 versus 0.79 ($P = 0.023$) for CVD in progressors versus nonprogressors, respectively. In a Cox regression model, SD of serial A1C was independently associated with progression of renal disease (hazard ratio 1.92 [95% CI 1.49–2.47]) and of a CVD event (1.98 [1.39–2.82]) even when adjusting for mean A1C and traditional risk factors. Interestingly for CVD, mean serial A1C itself was not predictive even though SD of A1C was.

CONCLUSIONS—In patients with type 1 diabetes, A1C variability was not only predictive of incident microalbuminuria and progression of renal disease but also of incident CVD events.
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A substantial proportion of patients with type 1 diabetes develops diabetes complications, such as microvascular complications (nephropathy, retinopathy, and neuropathy) and cardiovascular disease (CVD). Among the risk factors that have been identified for the microvascular complications, glycemic control is perhaps one of the most important. There is compelling evidence that the level of long-term glycemia, clinically determined by the A1C test, influences the risk of microvascular disease in patients with type 1 diabetes. The landmark study DCCT (Diabetes Control and Complications Trial) showed that a mean reduction in A1C by 2% units reduced the incidence of nephropathy by 54% and retinopathy by 76% (1). However, regarding CVD, the results were ambiguous because of the low incidence of CVD in this young cohort. Nevertheless, in the extended follow-up, the EDIC (Epidemiology of Diabetes Interventions and Complications) study, the risk of coronary heart disease was reduced by 42% in patients originally allocated to the intensively treated group (2).

In addition to the level of long-term glycemia, it has been debated whether glycemic variability confers additional risk of diabetes complications. Patients with identical A1C values may still show large variation in their glycemic excursions. In theory, a highly variable glucose profile could confer a greater risk of diabetes complications through increased oxidative stress (3). The clinical evidence is so far scarce, which may be because of a lack of a standardized method to measure glycemic variability in a large number of patients. In the DCCT, variation in seven-point laboratory-measured glucose profiles did not predict diabetes complications (4). However, a recent analysis of the DCCT data indicated that variation in the long-term glycemia, defined as intrapersonal SDs of quarterly measured A1C, added to the predictive value of A1C level alone for both nephropathy and retinopathy (5). This is a potentially important clinical finding, but since the DCCT was an intervention focusing on the A1C, the results might not reflect the situation in a normal clinical setting. Therefore, we analyzed the effect of both the mean A1C and the variability of A1C on the prediction of diabetes complications in the observational, longitudinal Finnish Diabetic Nephropathy (FinnDiane) Study.

RESEARCH DESIGN AND METHODS

The FinnDiane Study is an ongoing, nationwide, prospective multicenter study seeking for clinical, genetic, biochemical, and environmental risk factors for diabetes complications, with emphasis on diabetic nephropathy. Participating study centers comprise diabetes and renal outpatient clinics at all five university central hospitals, all 16 central hospitals, the majority ($N = 27$) of all regional hospitals, and 31 major primary health care centers in Finland. Patients with type 1 diabetes (ICD-10 code E10) were recruited at routine outpatient visits. Based on medical records, the attending physician completed a standardized check-list regarding diabetes complications and medication. Patients answered a self-report questionnaire regarding lifestyle factors and socioeconomic status. Total leisure-time physical activity was assessed by a separate questionnaire as previously described (6), and low physical activity was defined as <10 MET*h/week. Physical activity data were available for 775 eligible patients, and we also performed a subanalysis including only patients ($N = 534$) with physical activity data and normal urinary albumin excretion rate (UAER) because of a possible confounding effect of diabetes complications as we have shown decreased physical activity in patients with diabetes complications (7). Serum lipids were determined by

From the [1]Folkhälsan Institute of Genetics, Folkhälsan Research Center, Helsinki, Finland; and the [2]Department of Medicine, Division of Nephrology, Helsinki University Central Hospital, Helsinki, Finland.
Corresponding author: Per-Henrik Groop, per-henrik.groop@helsinki.fi.
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TABLE 1
Baseline characteristics according to progression in renal status (defined as any increase in albuminuria level or progression to ESRD) and occurrence of a hard CVD event (coronary event, stroke, peripheral vascular event) during follow-up

	Progression in renal status		CVD event	
	Nonprogressors	Progressors	No event	Event
n	1,893	214	1,687	158
Sex (% male)	52.0	64.8*	52.0	62.4#
Age (years)	36.5 ± 11.9	36.1 ± 11.1	35.2 ± 11.4	47.9 ± 9.0#
Duration of diabetes (years)	21.8 ± 11.9	23.1 ± 11.1	20.8 ± 11.4	34.6 ± 8.6#
A1C (%)	8.4 ± 1.4	9.4 ± 1.7*	8.5 ± 1.5	8.7 ± 1.4#
Systolic blood pressure (mmHg)	132 ± 18	138 ± 21*	131 ± 17	151 ± 21#
Ever smoking (%)	44.1	55.2*	44.2	53.7#
Total cholesterol (mmol/l)	4.9 ± 0.9	5.2 ± 1.1*	4.9 ± 0.9	5.3 ± 1.0#
Diabetic nephropathy (%)	NA	NA	15.7	63.3#

Values are expressed as number, means ± SD, or percentage. *$P < 0.05$ vs. nonprogressors. #$P < 0.05$ vs. no event. NA, not applicable.

automated enzymatic methods using a Cobas Mira analyzer (Hoffman-LaRoche, Basel, Switzerland). As an estimate of insulin sensitivity, the formula for estimated glucose disposal rate (eGDR) was used as previously described (8). Blood pressure was measured in the sitting position twice, and the mean value was used.

The study was initiated in November 1997 and follow-up data have been collected since 2004 either by reexamination of the patients or review of the medical files. Written informed consent was obtained from each patient, and the study protocol was conducted in accordance with the Declaration of Helsinki. In the present analyses, type 1 diabetes was restricted to age at onset of diabetes below 35 years and insulin treatment initiated within 1 year of diagnosis. In January 2009, the FinnDiane database comprised 2,107 patients with type 1 diabetes, serial data on A1C, and known renal status both at baseline and at follow-up, and these were included in the analyses.

Serial A1C measurements. A1C was measured at the local study centers with standardized assays (normal range 4.0–6.0%). The nationwide validity of A1C measurements in Finland has previously been studied, showing a high correlation with the DCCT reference method (9). Laboratory records of A1C measurements from baseline to follow-up were obtained. During a median follow-up of 5.7 years, the median number of A1C measurements per patient was 13 (interquartile range 7–20), that is 2.3 measurements per patient and year. For each patient, intrapersonal mean and SDs of all recorded A1C measurements were calculated, and the SD was considered a measure of A1C variability. As a normalized measure of variability, the coefficient of variation for A1C was calculated as the ratio of intrapersonal SD and mean to correct for larger SD because of higher absolute values of A1C.

Diabetes complications. Renal status was defined based on UAER in at least two of three consecutive timed (either 24 h or overnight) urine collections. Microalbuminuria was defined as UAER ≥30 and <300 mg/24 h or ≥20 and <200 μg/min, respectively. Macroalbuminuria was defined as UAER ≥300 mg/24 h or ≥200 μg/min, respectively. End-stage renal disease (ESRD) was defined as hemodialysis, peritoneal dialysis, or renal transplantation. Diabetic nephropathy was defined as macroalbuminuria and/or ESRD. Renal status was prospectively assessed by review of all recorded values of UAER and of medical records. Progression of renal disease was defined as a shift to a higher albuminuria level in any two (of three) consecutive urine collections or to ESRD. CVD was defined based on medical records both at baseline and at follow-up as any of the following hard events: myocardial infarction, coronary artery procedure (by-pass surgery or angioplasty), stroke (ischemic or hemorrhagic), limb amputation because of ischemia, or a peripheral artery procedure. Of the 2,107 patients with complete data on renal status and serial A1C measurements, 1,845 patients (87.6%) had follow-up data on CVD events. Patients with and without CVD data did not differ according to sex, age, duration of diabetes, CVD, or nephropathy status at baseline.

Statistical analyses. Continuous variables are expressed as means ± SD if normally distributed, otherwise as median (interquartile range). Categorical variables are reported as percentages. Differences between groups were analyzed by Student *t* test (two groups) or ANOVA (over two groups) for normally distributed continuous variables, otherwise the Mann-Whitney or Kruskal-Wallis was used, respectively. The χ^2 test was used for categorical variables. When *P* values were adjusted for age, ANCOVA was used for continuous variables and logistic regression for categorical variables. In cross-sectional data, logistic regression was used as multivariate analysis. Longitudinal data were analyzed with Kaplan-Meier survival curves with log-rank tests and Cox proportional hazard survival regression with results as hazard ratio (HR) and 95% CI. SPSS version 15.0 (SPSS, Chicago, IL) was used for statistical calculations, and survival cures were drawn by GraphPad Prism 5 (GraphPad Software, La Jolla, CA).

RESULTS

Patient characteristics. Of the eligible 2,107 patients, 53.2% of patients were men and the mean age was 36.4 ± 11.8 years, duration of diabetes 22.0 ± 11.9 years, BMI 25.0 ± 3.5 kg/m^2, and A1C 8.5 ± 1.5% at baseline. Altogether 1,381 patients had normal UAER, 316 microalbuminuria, 302 macroalbuminuria, and 108 ESRD. During a median follow-up of 5.7 years, 214 patients (10.2%) progressed to a higher level of albuminuria or to ESRD and 158 suffered a CVD event (of 1,845 with data on CVD; 8.6%). In Table 1, baseline characteristics according to progression in renal status and occurrence of a CVD event are given. For A1C there was a considerably larger difference for progression in renal status (1.0% unit) than for a CVD event (0.2% unit) (Table 1).

Serial A1C measurements. Intrapersonal mean of serially measured A1C was 8.5% and SD 0.78. There was a clear correlation between baseline single-measured A1C and the mean of serial A1C (Pearson $r = 0.72$, $P < 0.001$). According to baseline renal status, patients with normo-, micro-, or macroalbuminuria and ESRD had an SD of A1C during follow-up as follows: 0.75, 0.82, 0.89, and 0.79 (overall $P < 0.0001$), respectively. Patients that progressed regarding their renal status had a median of 15 A1C measurements, whereas nonprogressors had a median of 13 measurements ($P = 0.012$), but for CVD events there was no such corresponding difference ($P = 0.907$). In Table 2, mean and SD of serial A1C is given according to progression in renal and CVD status. The level of mean serial A1C was higher in patients that progressed to a higher albuminuria level (but not to ESRD), but no difference was seen in mean A1C between patients with and without a CVD event (Table 2). The variability (SD) of A1C, on the other hand, was greater in all subgroups of patients that progressed in renal status and in patients with a CVD event during follow-up (Table 2). In addition, the finding for coefficient of variation was similar to that of SD, indicating that the differences in SD were not solely because of differences in absolute A1C level between progressors and nonprogressors (Table 2).

To address the possibility of bias by the number of A1C measurements, we compared SD of progression versus nonprogression in renal status in patients with different numbers of measurements; <10 measurements: 0.98 ver-

TABLE 2
Intrapersonal mean, SD, and coefficient of variation of serially measured A1C values are given compared with progression vs. nonprogression of renal status or a CVD event (coronary event, stroke, peripheral vascular event) during follow-up

	Nonprogressors			Progressors			*P*		
	Mean	SD	CV	Mean	SD	CV	Mean	SD	CV
Renal status (n = 1,893/214)	8.5	0.76	8.9	9.2	1.01	11.0	<0.001	<0.001	<0.001
Normo to Micro (n = 1,283/98)	8.3	0.74	8.9	9.2	0.94	10.1	<0.001	<0.001	0.016
Micro to Macro (n = 271/45)	8.8	0.77	8.7	9.6	1.08	11.2	0.001	<0.001	<0.001
Macro to ESRD (n = 231/71)	9.0	0.84	9.2	8.8	1.07	12.0	0.261	0.005	0.001
CVD event (n = 1,687/158)	8.5	0.79	9.1	8.6	0.87	10.0	0.423	0.023	0.027

Numbers of patients are given as nonprogressors/progressors. CV is given as percentage. *P* values are given for nonprogressors vs. progressors within a given group. CV, coefficient of variation; Normo, normal UAER; Micro, microalbuminuria; Macro, macroalbuminuria.

sus 0.68 (P = 0.002), 10–30 measurements: 1.00 versus 0.79 (P < 0.001), and >30 measurements: 1.11 versus 0.87 (P = 0.001). This indicates greater variation with increasing number of measurements but with conserved differences in SD between progressors and nonprogressors. There was no correlation between follow-up time and SD of serial A1C (Pearson r = 0.006, P = 0.789).

Kaplan-Meier survival curves with quartiles of SD of A1C revealed a more clear association between the entire distribution of variability for progression in renal status (Fig. 1*A*) than for CVD events (Fig. 1*B*), for which only the highest quartile of SD diverged. In Fig. 2*A* and *B*, survival curves are given for patients with mean and SD of serial A1C above and below the population medians. For progression in renal status (Fig. 2*A*), the highest incidence was observed when both mean and SD were above the medians and conversely the lowest incidence when both were below the medians. Interestingly, patients above median for mean and below for SD had similar incidence for progression in renal status as patients below median for mean and above for SD, suggesting a distinct and equally important effect of both absolute value and variability of A1C in the risk of progression in renal disease. For CVD events, however, a similar finding was not observed (Fig. 2*B*) as all corresponding lines were superimposed, probably because of the fact that mean serial A1C did not differ between patients with and without a CVD event and all lines contain a prerequisite regarding the mean serial A1C.

Clinical and lifestyle factors. To depict which baseline patient characteristics were associated with the variability

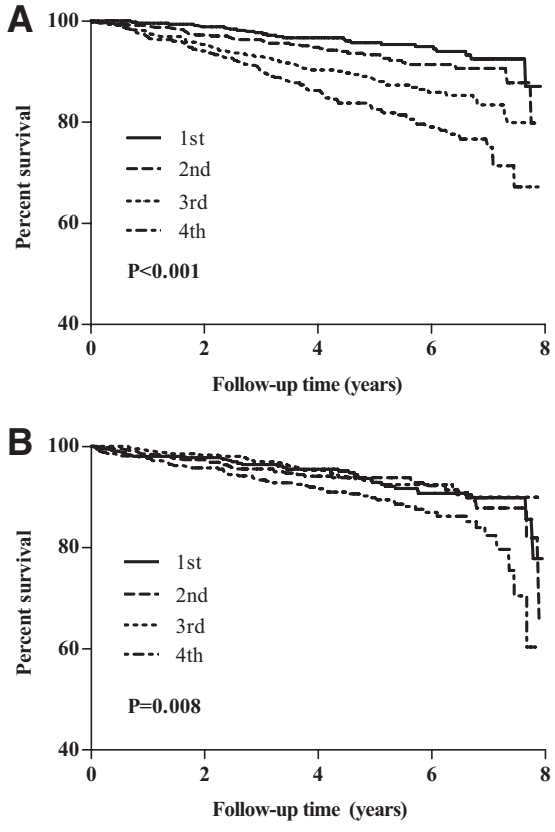

FIG. 1. *A*: **Kaplan-Meier survival curves for any progression in renal status (defined as any increase in albuminuria level or progression to ESRD) by quartiles of SD of serially measured A1C values.** *B*: **Kaplan-Meier survival curves for a CVD event (coronary event, stroke, peripheral vascular event) by quartiles of SD of serially measured A1C values.**

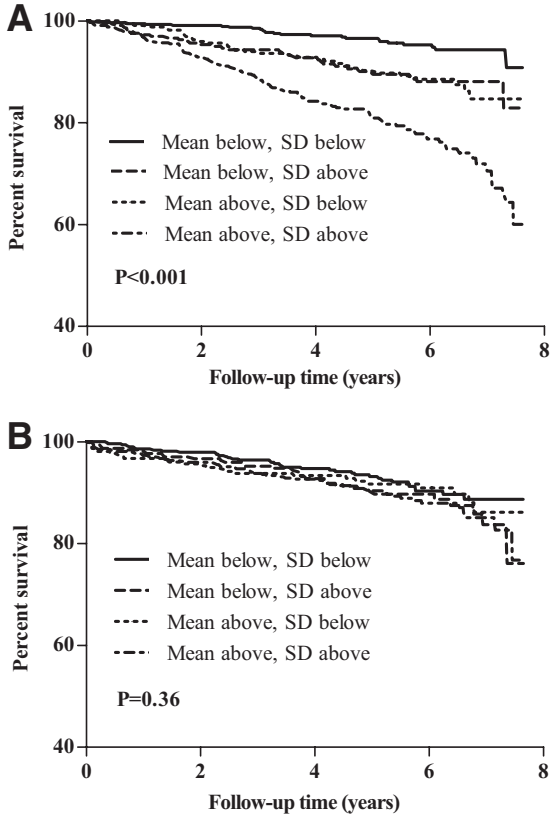

FIG. 2. *A*: **Kaplan-Meier survival curves for any progression in renal status (defined as any increase in albuminuria level or progression to ESRD) according to mean and SD of serial A1C above and below the population median.** *B*: **Kaplan-Meier survival curves for a CVD event (coronary event, stroke, peripheral vascular event) according to mean and SD of serial A1C above and below the population median.**

TABLE 3
Baseline patient characteristics according to quartiles of A1C variability defined as intrapersonal SD of serial A1C measurements during follow-up

	1st	2nd	3rd	4th	*P* value	*P* value adjusted for age
Patients (*n*)	529	541	512	525	—	—
Sex (% male)	52.8	55.0	51.4	53.7	0.69	0.94
Age (years)	38.1 ± 11.6	37.8 ± 11.7	36.5 ± 11.7	33.2 ± 11.4	<0.001	—
Age at onset of diabetes (years)	15.5 ± 8.5	15.0 ± 8.2	14.2 ± 8.5	13.2 ± 8.6	<0.001	0.13
Duration of diabetes (years)	22.6 ± 12.3	22.9 ± 11.6	22.4 ± 11.5	20.0 ± 11.8	<0.001	0.13
BMI (kg/m^2)	25.0 ± 3.2	25.0 ± 3.1	25.2 ± 3.7	24.9 ± 3.8	0.44	0.47
Waist-to-hip ratio, male	0.90 ± 0.07	0.91 ± 0.08	0.91 ± 0.07	0.90 ± 0.08	0.23	0.22
Waist-to-hip ratio, female	0.82 ± 0.06	0.80 ± 0.06	0.81 ± 0.06	0.82 ± 0.07	0.002	<0.001
A1C (%)	7.9 ± 1.2	8.3 ± 1.2	8.6 ± 1.3	9.2 ± 1.9	<0.001	<0.001
Systolic blood pressure (mmHg)	133 ± 17	133 ± 17	134 ± 19	132 ± 19	0.72	0.10
Diastolic blood pressure (mmHg)	79 ± 9	79 ± 10	80 ± 10	80 ± 10	0.73	0.54
Total cholesterol (mmol/l)	4.9 ± 0.9	4.9 ± 0.9	4.9 ± 0.9	5.0 ± 1.0	0.025	<0.001
HDL cholesterol (mmol/l)	1.30 ± 0.37	1.31 ± 0.37	1.32 ± 0.38	1.26 ± 0.36	0.043	0.19
LDL cholesterol (mmol/l)	3.06 ± 0.83	3.02 ± 0.81	3.06 ± 0.82	3.12 ± 0.93	0.25	0.005
Triglycerides (mmol/l)	0.97 (0.73–1.33)	1.02 (0.76–1.43)	1.01 (0.76–1.45)	1.13 (0.85–1.68)	<0.001	<0.001
Insulin dose (IU/kg)	0.69 ± 0.26	0.71 ± 0.23	0.72 ± 0.24	0.78 ± 0.28	<0.001	0.004
eGDR (mg · kg^{-1} · min^{-1})	6.6 (4.7–8.9)	6.5 (4.4–8.6)	6.3 (4.3–8.4)	6.0 (3.8–8.2)	<0.001	<0.001
Ever smoker (%)	37.1	44.1	47.3	52.8	<0.001	<0.001
Current smoker (%)	17.5	21.8	24.2	33.1	<0.001	<0.001
Blue-collar worker (%)	60.7	63.5	67.9	76.2	<0.001	<0.001
Sedentary (%), n = 775*	19.5	19.1	24.3	28.9	0.087	0.009
Sedentary (%), n = 534†	16.0	14.4	23.7	27.7	0.026	0.004

Values are expressed as means ± SD, percent, or median (interquartile range). *Patients with available data on physical activity. †Only patients with normal UAER included. Sedentary, leisure-time physical activity <10 MET*h/week.

of A1C during follow-up, we examined quartiles of SD of serial A1C. We found that a higher variability was associated with younger age, lower age at onset of diabetes, shorter duration of diabetes, lower insulin sensitivity, dyslipidemia, higher baseline A1C, both current and ever smoking, lower socioeconomic class, and lower leisure-time physical activity (Table 3). For the physical activity, we included in a separate analysis only patients with normal UAER, which did not change the results (Table 3). In patients with normal UAER, the associations for insulin dose ($P < 0.001$), current smoking ($P < 0.001$), and social class ($P < 0.001$) with quartiles of SD were similar as for all patients, suggesting that as for physical activity, the associations were not solely because of collinearity with diabetes complications. Finally, because there was a clear association between age and A1C variability, we also adjusted the *P* values for age (Table 3); however, the abovementioned associations did not change except for age at onset and duration of diabetes, which were no longer associated with A1C variability.

Multivariate models. Because of several potential confounding factors for the association between the variability of serial A1C and the progression of renal disease and CVD events, we used Cox regression as a multivariate analysis. As covariates, we used duration of diabetes, sex, systolic blood pressure, total cholesterol, ever smoking, intrapersonal mean of serial A1C measurements, and number (ln-transformed) of A1C measurements. For incident CVD events, we additionally adjusted for the presence of diabetic nephropathy and CVD events at baseline. As shown in Table 4, SD of serial A1C measurements predicted progression in renal status even independently of mean serial A1C. Moreover, SD was also an independent predictor of CVD events, whereas mean serial A1C was not.

DISCUSSION

In this study, we show that the variability of A1C predicts the development and progression of incipient and overt renal disease in patients with type 1 diabetes. These data support a recent analysis from the DCCT intervention where A1C variability predicted the development of diabetic nephropathy and retinopathy (5). We add to that study by showing the same finding for nephropathy in an observational cohort and further covering the entire spectrum of renal disease (from microalbuminuria to ESRD) in diabetes. However, the most interesting novel finding was that the A1C variability also predicted CVD events even though the mean A1C during follow-up was similar in patients with and without a CVD event. This association was independent of nephropathy status at baseline, indicating that the association was not entirely driven by renal disease, which is a strong CVD risk factor in patients with type 1 diabetes (10). We further show that a variable A1C profile is associated with a cluster of baseline factors such as lower insulin sensitivity, dyslipidemia, lower socioeconomic status, physical inactivity, and smoking.

In the present longitudinal analysis, the baseline A1C level was a predictor of incident microalbuminuria and overt diabetic nephropathy. This is in line with the large body of evidence showing that lower A1C translates into reduced risk of diabetic nephropathy and retinopathy in patients with type 1 diabetes. Before the landmark study DCCT (1), this was also indicated by the Steno Study (11), the Oslo Study (12), and the Stockholm Diabetes Intervention Study (13).

Regarding glycemic control and CVD the picture is, however, less clear. In type 2 diabetes, the apparent lack of benefit of improved glycemia on CVD outcomes at least in advanced diabetes has recently been debated (14). In type

TABLE 4
Cox regression models for progression in renal status (defined as any increase in albuminuria level or progression to ESRD) and a CVD event (coronary event, stroke, peripheral vascular event) during a median follow-up of 5.7 years

	Progression in renal status		CVD event	
	HR (95% CI)	*P*	HR (95% CI)	*P*
Duration of diabetes (years)	1.01 (0.99–1.02)	0.356	1.08 (1.06–1.10)	<0.001
Male sex	1.74 (1.30–2.33)	<0.001	1.17 (0.81–1.70)	0.399
Systolic blood pressure (mmHg)	1.01 (1.00–1.02)	0.032	1.02 (1.01–1.03)	0.001
Total cholesterol (mmol/l)	1.19 (1.04–1.36)	0.014	1.11 (0.92–1.34)	0.267
Ever smoking	1.22 (0.92–1.63)	0.162	1.00 (0.70–1.43)	0.985
Diabetic nephropathy	NA	NA	1.78 (1.20–2.64)	0.004
CVD event	NA	NA	3.11 (2.10–4.59)	<0.001
Number of serial A1C measurements	1.09 (0.90–1.31)	0.397	0.88 (0.71–1.09)	0.245
Intrapersonal mean of serial A1C (%)	1.34 (1.20–1.51)	<0.001	1.01 (0.87–1.18)	0.892
Intrapersonal SD of serial A1C	1.92 (1.49–2.47)	<0.001	1.98 (1.39–2.82)	<0.001

Diabetic nephropathy: macroalbuminuria and/or ESRD. Serial A1C represents all available A1C measurements from baseline to follow-up, for which intrapersonal mean and SD were calculated. All other variables are from baseline. NA, not applicable.

1 diabetes, there is evidence in some (15,16) but not in all epidemiological studies (17–20), as well as meta-analyses (21,22), for an independent relationship between A1C and development of CVD or CVD mortality. In the extended DCCT follow-up (EDIC), there was a delayed benefit of intensive treatment on incident CVD (2). There is further a report that the change in two A1C values (baseline minus follow-up) predicts incident coronary artery disease in type 1 diabetes, even though the baseline A1C was not in itself predictive (23). In the present study, baseline A1C was slightly higher (0.2% unit) in patients that suffered a CVD event during follow-up, but the mean serial A1C during follow-up did not differ between patients with and without a CVD event during follow-up. Thus, the role of the level of A1C for CVD is somewhat unclear also in our study but certainly seems less important than is the case for diabetic nephropathy.

In addition to the actual level of A1C, it has been postulated that variation in the glycemic profile also could contribute to risk of diabetes complications because glucose peaks seem to cause detrimental in vitro cellular effects (24–27). Furthermore, glycemic excursions measured by continuous glucose measurement (CGMS) correlate with oxidative stress in patients with type 2 diabetes (3) but not type 1 diabetes (28). Oxidative stress has been considered fundamental in the pathogenesis of diabetes complications (29). Furthermore, experimentally induced oscillating blood glucose may also impair endothelial function, thus possibly linking glucose fluctuations also to CVD (30).

For epidemiological studies, the CGMS is often impractical and other measures of glycemic variability have therefore been used. In type 2 diabetes, there are some reports that variability of fasting plasma glucose predicts retinopathy (31) as well as cardiovascular and total mortality (32). In these studies, however, the patients had widely differing antihyperglycemic treatment modalities ranging from diet to insulin injections, which might have affected the variability. In type 1 diabetes, the variability of repeated measurements of blood glucose values has been investigated in the publicly available DCCT dataset by comparing the quarterly one-day measured pre- and postprandial (breakfast, lunch, and supper) and bedtime glucose values (4). The study by Kilpatrick et al. found no evidence of an effect of variation in blood glucose on the risk of diabetic nephropathy or retinopathy. Recently Kilpatrick et al. (5), however, reported that the variability of A1C in the DCCT independently predicted the incidence of diabetic nephropathy and retinopathy even after correction for the absolute level of A1C. Because the DCCT was an intervention study, the investigators excluded A1C measurements from the first 6 months of the study to decrease the risk of artificial variation in A1C because of a new insulin treatment regimen. The risk of bias by the intervention itself, however, is still possible, even though the main finding was seen in the conventionally treated nonintervened patient group, which supports an effect independent of the intervention. Our present study was performed as a longitudinal, observational study without intervention, and the results regarding the development of nephropathy are very similar to the DCCT data (5).

It is likely that the A1C variability reflects other processes compared with the short-term glycemic variability observed by CGMS monitoring and multiple blood glucose measurements. This may explain the difference in the predictive value of the variability in blood glucose (4) and the A1C (5) for nephropathy and retinopathy in the DCCT cohort. The relationship between short-term and long-term glycemic variability needs to be further clarified. In our study, the finding that A1C variability, but not mean serial A1C, was associated with CVD events might indicate that patients more severely affected by diabetes and its complications have a more variable A1C profile than patients with less complicated disease. Nevertheless, the association was still evident when controlling for the presence of diabetic nephropathy and CVD events at baseline (Table 4). For renal complications, those patients with a normal UAER at baseline but who progressed to microalbuminuria had higher A1C variability than those who remained with normal UAER, suggesting that A1C variability might not just be secondary to the presence of more severe diabetes complications.

The mechanisms behind the association between A1C variability and the risk of diabetes complications are at this stage merely speculative and cannot be determined in this observational study. However, several baseline factors indicative of a disadvantageous lifestyle and a low socioeconomic class were associated with a more variable A1C, which might indicate a suboptimal management of diabetes. A1C variability was also associated with insulin resistance, which in itself has been implicated in the pathogenesis of diabetes complications (33). It is also possible that the intrapersonal variation in A1C is secondary to variation in insulin sensitivity, which in turn may be

because of infections or other intermittent factors that decrease insulin sensitivity. In fact, we recently showed that serum lipopolysaccharide activity, a marker of gram-negative bacterial infections, is associated with progression of diabetic nephropathy (34). It is further possible that fluctuating long-term glycemic control causes homeostatic imbalance, for example, through intracellular sorbitol accumulation. It was suggested that the "normoglycemic re-entry phenomenon" (35) might tie A1C variability and risk of retinopathy (5). This phenomenon refers to an initial, but not sustained, worsening of retinopathy when lowering A1C in patients already affected by retinopathy. Similarly, in the kidneys, variable homeostatic circumstances may be detrimental. A further potential mechanism is induction of growth factors, for instance IGF-1 and vascular endothelial growth factor, by a changing glycemic environment (36,37), which may contribute to development of diabetes complications. Regarding CVD, A1C variability may cause vascular stress by a changing glycemic environment. An important remark at this stage, however, is that the available data on A1C variability and risk of diabetes complications cannot confirm a direct causal relationship because the association might be because of collinearity with other causal factors.

The strengths of this study are the large number of well-characterized patients and the prospective study design. There are, however, also limitations to this study. The serial A1C values were collected from available laboratory records as a part of the patient's routine clinical follow-up. Thus, there were no prespecified intervals between A1C measurements, and the number of measurements per individual patient varied, which was, however, statistically adjusted for. Another potential limitation is the use of A1C measured at the local study centers, not in a central laboratory. However, this should not affect the intrapersonal variability of A1C because the measurements were performed at the same center.

In conclusion, we show in an observational setting that a larger variability of A1C predicts not only incident microalbuminuria and progression of established renal disease but also CVD events in patients with type 1 diabetes.

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