

Effects of Duration and Age at Onset of Type 1 Diabetes on Preclinical Manifestations of Nephropathy

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Several studies have reported that the effect of type 1 diabetes disease duration on nephropathy may be greater during or after puberty than an equivalent number of years before puberty. The International Diabetic Nephropathy Study examined the effects of disease duration and age at onset on glomerular morphometry obtained from kidney biopsy in 243 young type 1 diabetic subjects with albumin excretion rates <100 µg/min: 184 with prepubertal onset, 35 with pubertal onset, and 24 with postpuberty onset. Outcomes included the volume fraction of the glomerulus occupied by the mesangium [Vv(Mes/glom)], glomerular basement membrane (GBM) width, and the surface density of peripheral glomerular capillary basement membrane per glomerulus Sv(PGBM/glom). Vv(Mes/glom) progressed slowly in the first 14 or 15 years after disease onset but more rapidly thereafter. GBM width increased, while Sv(PGBM/glom) decreased with increasing disease duration. No statistically significant differences in the effect of duration were observed among the three age-at-onset subgroups, even with multivariate adjustment for sex, center, baseline HbA_{1c} concentration, diastolic blood pressure, height, and BMI. Our results may explain the conflicting results of previous studies regarding the effects of pre- versus postpuberty disease duration. *Diabetes* 52:1818–1824, 2003

The microvascular complications of type 1 diabetes are known to increase with duration of the disease (1–4). The results of several studies, however, suggest that the effect of duration on nephropathy and retinopathy may be modified by the age at onset of diabetes and, in particular, that duration after puberty has a larger impact than an equivalent duration before puberty (2,4–19). The mechanisms proposed to explain this phenomenon include the adverse effects of sex steroids on vascular basement membrane and a dete-

rioration of glycemic control with greater independence from parents during adolescence and young adulthood (5,20,21). The inference that the prepubertal years have little or no effect on the development of diabetes complications could theoretically reduce the aggressiveness of treatment of young children with type 1 diabetes. Nonetheless, the evidence for a lesser effect of prepubertal duration is inconsistent; several studies have yielded conflicting results (2,12,22).

Previous studies of pre- versus postpubertal duration effects on diabetic nephropathy have generally used albuminuria as an end point, since albuminuria usually reflects well-established diabetic nephropathy (2,3,5,7,12). But in children and young adults, albuminuria and overt nephropathy are relatively uncommon. Moreover, several recent studies have reported that microalbuminuria is sometimes reversible (13,19,23). With the exception of one small study (22), the effect of age at onset of diabetes on earlier stages of nephropathy has not been previously reported because investigators have heretofore lacked renal morphometric measures to detect these earlier stages. Because albuminuria rarely develops before puberty and biopsies are usually performed only after the onset of albuminuria, little information is available on nephropathic lesions that develop before adolescence. In addition to these issues related to end points, previous studies have been limited by several methodologic problems, including small sample sizes; collinearity of age at onset with total disease duration; confounding by glycemic control, BMI, blood pressure, and sex; and inconsistent definitions of puberty.

In this article, we report findings from the International Diabetic Nephropathy Study (24), which overcomes most of these methodologic problems by obtaining kidney biopsies on type 1 diabetic subjects before the onset of microalbuminuria to assess the critical mesangial and basement membrane morphometric changes characteristic of diabetic nephropathy (24,25). Detailed information is available on glycemic control (glycosylated hemoglobin), BMI, blood pressure, and Tanner staging at study enrollment. In addition, we have used statistical methods that separate the effect of age of onset from that of total disease duration. These morphometric and methodologic advantages permit a rigorous test of the hypothesis that prepubertal duration has a smaller effect than postpubertal duration on the early development of diabetic nephropathy.

RESEARCH DESIGN AND METHODS

The International Diabetic Nephropathy Study is a prospective cohort study of 243 children and young adults with type 1 (insulin-dependent) diabetes.

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GBM, glomerular basement membrane; Sv(PGBM/glom), surface density of peripheral glomerular capillary basement membrane per glomerulus; Vv(Mes/glom), volume fraction of the glomerulus occupied by the mesangium.

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TABLE 1
Baseline characteristics of overall cohort and three age-at-onset subgroups

| | Overall | Prepuberty | Puberty | Postpuberty |
|---------------------------|----------------------------|----------------------------|----------------------------|----------------------------|
| <i>n</i> | 243 | 184 | 35 | 24 |
| Age at onset (years) | 8.8 ± 4.8 (0.5, 31.5) | 6.8 ± 2.9 (0.5, 12.0) | 12.8 ± 1.0 (11.1, 14.3) | 18.5 ± 4.3 (14.0, 31.5) |
| Age at biopsy (years) | 16.8 ± 6.0 (9.5, 38.0) | 14.9 ± 4.0 (9.5, 29.1) | 18.6 ± 3.7 (13.0, 32.3) | 28.9 ± 6.3 (16.5, 38.0) |
| Diabetes duration (years) | 8.0 ± 4.2 (1.8, 20.3) | 8.1 ± 4.0 (2.0, 19.8) | 5.8 ± 3.7 (1.8, 19.1) | 10.4 ± 5.1 (2.1, 20.3) |
| HbA _{1c} (%) | 8.7 ± 1.5 (5.3, 13.9) | 8.7 ± 1.4 (5.7, 13.5) | 9.0 ± 1.8 (6.2, 13.9) | 8.1 ± 1.6 (5.3, 11.9) |
| Systolic BP (mmHg) | 113.5 ± 11.1 (85.5, 145.0) | 112.2 ± 10.6 (85.5, 140.0) | 113.2 ± 10.1 (92.0, 135.0) | 124.2 ± 11.0 (99.0, 145.0) |
| Diastolic BP (mmHg) | 62.8 ± 8.9 (44.5, 89.5) | 61.7 ± 7.9 (45.0, 82.0) | 61.4 ± 8.3 (44.5, 75.5) | 74.6 ± 8.7 (59.0, 89.5) |
| Vv(Mes/glom) | 0.22 ± .05 (0.13, 0.42) | 0.22 ± .04 (0.14, 0.41) | 0.21 ± 0.05 (0.13, 0.34) | 0.25 ± 0.06 (0.15, 0.42) |
| GBM (nm) | 428 ± 77 (266, 715) | 425 ± 76 (266, 692) | 427 ± 60 (314, 575) | 452 ± 102 (330, 715) |
| Sv(PGBM/glom) | 0.14 ± 0.02 (0.09, 0.20) | 0.14 ± 0.02 (0.09, 0.19) | 0.14 ± 0.02 (0.09, 0.20) | 0.13 ± 0.03 (0.09, 0.18) |

Data are means ± SD (min, max). BP, blood pressure.

Subjects between the ages of 9 and 30 years at three study sites (Montreal, Minneapolis, and Paris) with a duration of type 1 diabetes of 2–20 years, normal blood pressure, and albumin excretion rate <100 µg/min (all but eight had rates <20 µg/min) were recruited and underwent a renal biopsy. A detailed description of the study methods is given elsewhere (24). The primary goal was to relate changes in glomerular morphometry (as they reflect the early, prealbuminuric phase of diabetic nephropathy) to a variety of potential etiologic determinants, including age at onset, disease duration, degree of glycemic control, blood pressure, and lipid abnormalities. A study of the relationships among these factors and of biopsy at study entry is the subject of a recent publication (25). In this article, we investigate the relationships among age at onset, pre- and postpubertal duration of disease, and morphometric glomerular abnormalities at baseline. Tanner staging was assessed at the time of the first study visit, usually within 3 months of the baseline biopsy, in 176 (72%) of the 243 study subjects.

Morphometric measurements. The following three morphometric measures obtained from renal biopsy tissue were used as the principal outcomes in this investigation because of their demonstrated importance as precursor lesions for more overt clinical manifestations of diabetic nephropathy (26,27): the volume fraction of the glomerulus occupied by the mesangium [Vv(Mes/glom)], glomerular basement membrane (GBM) width, and the surface density of peripheral glomerular capillary basement membrane per glomerulus [Sv(PGBM/glom)]. Increases in relative mesangial volume reduce the glomerular surface available for filtration (18,19). The methods for obtaining the morphometric measurements are detailed in an earlier report (25). The sample size of the study was based on estimates of the expected change in Vv(Mes/glom) over the 5-year period between the baseline and 5-year biopsies (24).

Data analysis. To estimate the effect of duration on all morphometric measures, we stratified the study population into three subgroups based on biologic age at onset of diabetes: 1) the prepuberty group consisted of 184 subjects who were prepubertal at the time of enrollment and also, therefore, at the time of diabetes onset ($n = 18$) [males: both pubic hair and genital Tanner stages = 1, or (in the absence of Tanner staging) age <12 years; females: both pubic hair and breast Tanner stages = 1 or age <11 years] or reported diagnosis of diabetes before age 12 years in males or age 11 years in females ($n = 166$); 2) the puberty group consisted of 35 subjects who reported a diagnosis of diabetes at age 12–15 years in males or 11–14 years in females; and 3) the postpuberty group consisted of 24 subjects who were enrolled postpuberty (males: pubic hair and/or genital Tanner stage 5 or age >15 years; females: pubic hair and/or breast Tanner stage 5 or age >14 years) and whose diabetes was also diagnosed after age 15 years in males and after age 14 years in females.

For all analyses, estimations were performed using multiple regression, with the duration of diabetes as the principal independent variable. Because age and disease duration are highly collinear (and nearly completely so, when examined within subgroups defined by age at disease onset), regression models did not include age terms. The resulting measure of effect is expressed as the mean difference in each morphometric measure for every 1-year increase in disease duration. Several regression models were assessed for goodness of fit, including a single straight line, a quadratic, two straight lines with different slopes, and combinations of a straight line and a quadratic. In all instances, residual analyses were performed to assess the goodness of fit of the models. Multivariate models were used to adjust the effect estimates for sex, center, and baseline HbA_{1c}, diastolic blood pressure, height, and BMI.

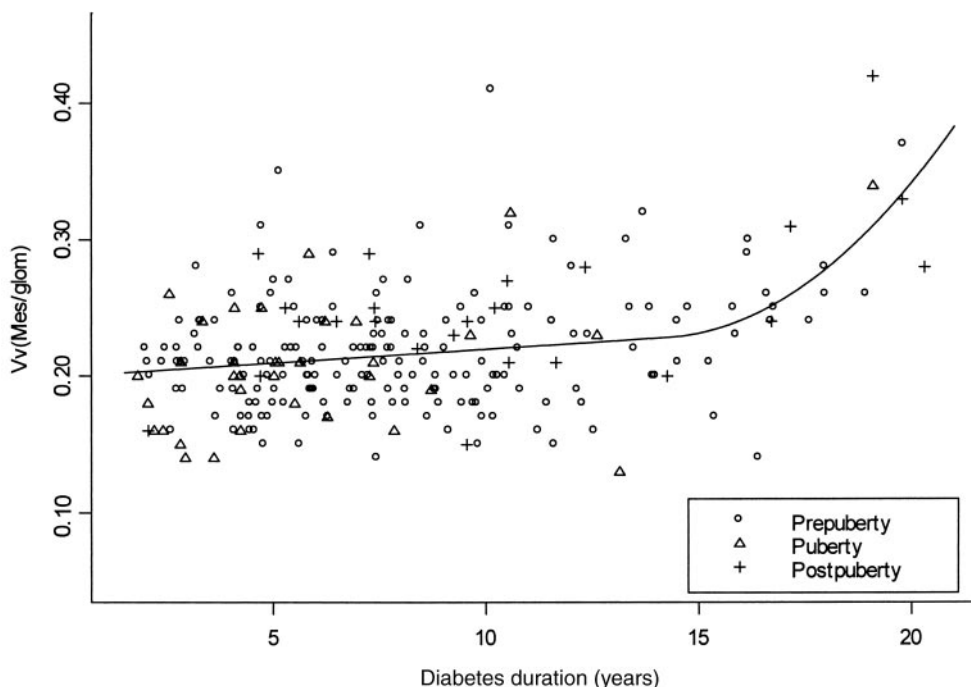


FIG. 1. Vv(Mes/glom) as a function of disease duration in three subgroups defined by age at onset of type 1 diabetes.

Because the estimates before and after adjustment were extremely similar, however, only the unadjusted estimates will be presented in the RESULTS.

RESULTS

Of the 243 subjects (118 males and 124 females) in the cohort, 184 were in the prepuberty group, 35 in the puberty group, and 24 in the postpuberty group. The baseline characteristics and morphometric outcomes are shown in Table 1 for the overall cohort and for each of the age-at-onset subgroups. The ages at onset and at biopsy increased across the three subgroups, as expected from their definition. The duration of diabetes at the time of biopsy was lower in the puberty group. Baseline HbA_{1c} values were similar in the three groups, while systolic and diastolic blood pressures were higher in the postpuberty group, reflecting their older age.

Vv(Mes/glom) increased with increasing duration of diabetes; a combination of a straight line and quadratic relationship provided a significantly ($P < 0.0001$) better overall fit of the data than a single straight line (Fig. 1). The inflection joint for the two model segments was at 14.2 years. This pattern was similar in the three age-at-onset subgroups, although few subjects in the puberty group had durations beyond the inflection point. These data suggest a nonlinear relationship between disease duration and Vv(Mes/glom) that is independent of age of disease onset but that begins to increase substantially only after 14 or 15 years duration. The values of the coefficients (and their 95% CIs) for both curve segments in the overall cohort and in each subgroup are shown in Table 2.

The results for GBM width are shown graphically in Fig. 2. GBM width increased with increasing disease duration; a single quadratic function with a constantly decreasing slope provided the best fit for these data ($P = 0.033$ vs. a straight line). Both the linear (duration) and quadratic (duration²) terms for disease duration were significant predictors of GBM width in the overall sample (Table 3). Results were similar in the three age-at-onset subgroups, although the CIs were wide in the puberty and postpuberty groups.

The results for Sv(PGBM/glom) are shown graphically in Fig. 3. As disease duration increased, Sv(PGBM/glom) decreased; a downward-curving quadratic provided the best fit ($P = 0.002$ vs. a straight line). The quantitative results are given in Table 3. Consistent with the results for Vv(Mes/glom) and GBM width, no difference was observed in the duration-Sv(PGBM/glom) relation according to age at onset of diabetes.

DISCUSSION

Previous studies have yielded inconsistent findings as to whether nephropathy in type 1 diabetes progresses more slowly before puberty than thereafter, but these studies have generally been limited to later stages of progression (i.e., after development of albuminuria) and have suffered from several important methodologic limitations. Our results suggest that it is total duration of disease, rather than duration before or after puberty, that affects the rate of progression of the GBM and mesangial lesions characteristic of diabetic nephropathy.

Two key features of our study have enabled us to clarify this issue and help to explain the conflicting evidence.

TABLE 2
Yearly effect of diabetes duration on Vv(Mes/glom) in overall cohort and three age-at-onset subgroups

| | Overall | Prepuberty | Puberty | Postpuberty |
|--|-----------------------------|-----------------------------|----------------------------|-----------------------------|
| <i>n</i> | 243 | 184 | 35 | 24 |
| Duration (if <14.2 years) | 0.0020 (0.0003 to 0.0038) | 0.0017 (0.00002 to 0.0034) | 0.0028 (-0.0024 to 0.0081) | 0.0014 (-0.0072 to 0.0099) |
| Duration (if >14.2 years) | -0.0878 (-0.2216 to 0.0460) | -0.1689 (-0.4302 to 0.0924) | Not estimable | -0.0315 (-0.1783 to 0.1154) |
| Duration ² (if >14.2 years) | 0.0031 (-0.0009 to 0.0072) | 0.0055 (-0.0022 to 0.0132) | 0.0004 (0.0002 to 0.0007) | 0.0014 (0.0033 to 0.0060) |

Data are estimated regression coefficient (95% CI).

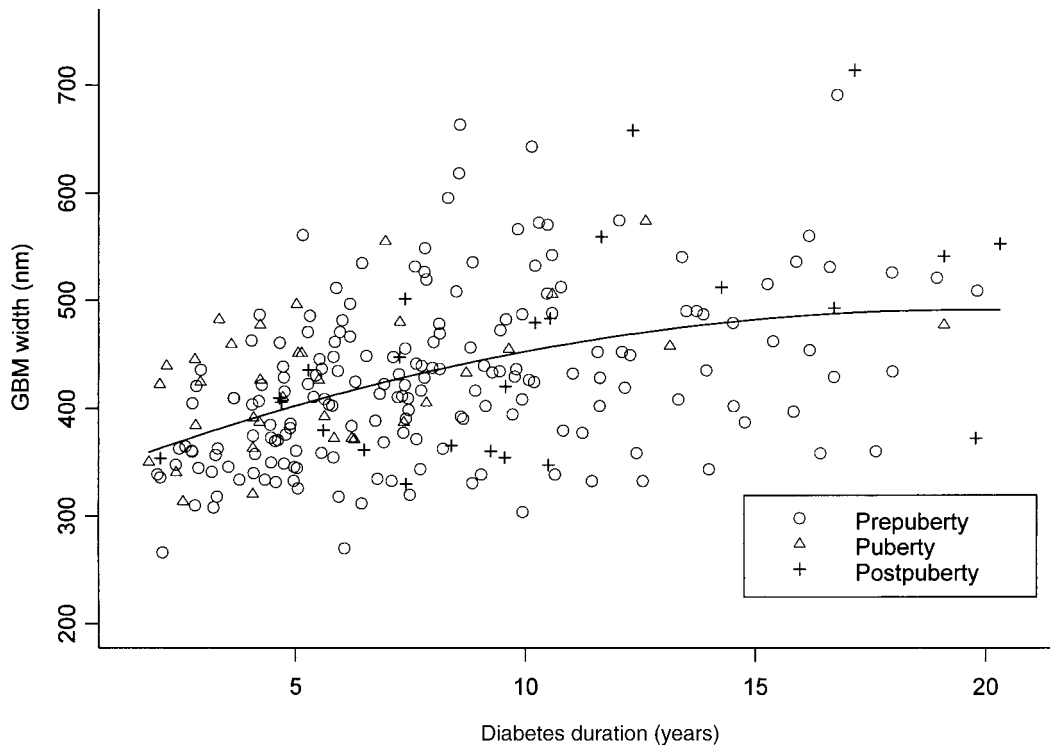


FIG. 2. GBM width as a function of disease duration in three subgroups defined by age at onset of type 1 diabetes.

First, our cohort consisted of young diabetic patients with albumin excretion rates $<100 \mu\text{g}/\text{min}$, and all but 8 of the 243 were normoalbuminuric (24), thus permitting us to study the effects of disease duration in young diabetic subjects before the clinical onset of nephropathy. Second, our morphometric analysis of renal biopsy specimens provides a quantitative assessment of glomerular histologic lesions that have previously been shown to be associated with clinical nephropathy (26,27). Third, we have controlled for such potentially confounding variables as sex, BMI, blood pressure, and glycemic control at the time of biopsy in order to isolate the effect of disease duration and its timing on the development of critical glomerular lesions. As a result, we have been able to show that the subclinical development of the principal glomerular lesion of diabetic nephropathy, i.e., the increase in $\text{Vv}(\text{Mes}/\text{glom})$, is nonlinear with respect to disease duration. $\text{Vv}(\text{Mes}/\text{glom})$ appears to progress slowly in the first 14 or 15 years of the disease, regardless of age (or pubertal stage) at onset. These results are consistent with those previously reported by Donaghue et al. (12) for microalbuminuria and Andersen et al. (2) for frank proteinuria. Similarly, Ellis et al. (22) did not find differences in $\text{Vv}(\text{Mes}/\text{glom})$ in diabetic patients who were prepubertal versus those who were pubertal or postpubertal at the time of biopsy, although none of the subjects in that study had a disease duration >12 years.

Our results help explain the conflicting findings of previous studies regarding the effects of pre- versus postpuberty disease duration. Because it is impossible to accumulate 14 or 15 years of disease duration before puberty, all of the disease duration beyond 14–15 years will be included in the postpubertal component. The postpubertal effect will thus appear larger, on average, than the prepubertal effect, i.e., the “protective effect” of

prepubertal duration is an artifact of the shape of the duration-nephropathy relation.

The apparent increased effect of disease duration after 14–15 years, even if real, might represent a period effect. In other words, disease duration earlier than 14–15 years before study enrollment may reflect an earlier calendar period during which type 1 diabetes was less aggressively treated; therefore, poorer glycemic control typical of disease management in the more distant past (which is not reflected in our baseline HbA_{1c} measurements) would be confounded with longer disease duration. Subjects with shorter disease duration have been treated for most or all of their duration in the recent past, thus creating a period effect by falsely suggesting that the early years of disease had a lesser influence on progression of nephropathy.

The apparent nonlinearity in the rate of development of mesangial expansion, a critical lesion in diabetic nephropathy (26), may have important pathophysiologic implications. For example, Fioretto et al. (28) found that successful pancreas transplantation did not result in the reversal of established diabetic nephropathy lesions during the first 5 years of normoglycemia, whereas substantial reversal was seen after 10 years (29). The reason for this delay in reversal of lesions is unknown but could represent the same pathophysiologic processes involved in the delayed expression of early diabetic nephropathy lesions. Thus, lesions may develop only after renal extracellular matrix molecules become heavily glycosylated, after which they may induce a reduction in their local degradation (30,31). Conversely, lesions may become reversible only after long-lived glycosylated extracellular matrix molecules are replaced by nonglycosylated molecules following re-establishment of normoglycemia.

Our study has several limitations. First, despite the overall sample size of 243 subjects with renal biopsies, the

TABLE 3
Yearly effect of diabetes duration on GBM width and Sv(PGBM/glom) in overall cohort and three age-at-onset subgroups

| | Overall | Prepuberty | Puberty | Postpuberty |
|-------------------------------|------------------------------|-------------------------------|-----------------------------|-----------------------------|
| <i>n</i> | 243 | 184 | 35 | 24 |
| GBM width (nm/year) | | | | |
| Duration | 16.31 (7.96 to 24.67) | 21.30 (10.74 to 31.86) | 15.37 (-2.04 to 32.78) | 22.65 (-12.53 to 57.83) |
| Duration ² | -0.413 (-0.823 to -0.003) | -0.689 (-1.220 to -0.159) | -0.417 (-1.344 to 0.509) | -0.496 (-1.951 to 0.958) |
| Sv(PGBM/glom) (fraction/year) | | | | |
| Duration | 0.0023 (-0.0003 to 0.0049) | 0.0028 (-0.0004 to 0.0060) | 0.0013 (-0.0052 to 0.0077) | -0.0013 (-0.0110 to 0.0085) |
| Duration ² | -0.0002 (-0.0003 to -0.0001) | -0.0002 (-0.0004 to -0.00004) | -0.0002 (-0.0005 to 0.0001) | -0.0001 (-0.0005 to 0.0003) |

Data are estimated regression coefficient (95% CI).

number of subjects is rather small in our puberty ($n = 35$) and postpuberty ($n = 24$) groups. The sample size of the International Diabetic Nephropathy Study was based on estimated changes in Vv(Mes/glom) over the 5-year period between the baseline and follow-up biopsies (24), not a comparison of baseline morphometric measures in the three age-at-onset groups. As shown by the wide CIs, the small sizes of the puberty and postpuberty groups provided low statistical power for detecting effects of disease duration. A second limitation is that our puberty group has few subjects with a duration beyond 14–15 years, again providing low power for detecting differential effects of prolonged disease duration in that group. Finally, Tanner staging was missing in 28% of the subjects, which may have led to misclassification of pubertal status at disease onset in a few subjects. For these reasons, and particularly in regard to sample size, the current findings should be interpreted with caution.

Members of our group have previously demonstrated that increase in mesangial fractional volume [Vv(Mes/glom)] is associated with the development of proteinuria, hypertension, and renal insufficiency (26,27). Moreover, the International Diabetic Nephropathy Study Group has shown elsewhere that previous duration of type 1 diabetes is strongly associated with Vv(Mes/glom) and other diabetic glomerular lesions at the time of the baseline biopsy (25). Microalbuminuria has been reported in cohorts of children and adolescents with type 1 diabetes (15,16,19,32,33) and precedes the occurrence of frank proteinuria by ~5 years (2). Nonetheless, we have no way of ensuring that the relationships we have found for histologic lesions of diabetic nephropathy will persist with respect to development of microalbuminuria, proteinuria, and renal insufficiency. A second renal biopsy obtained after 5 years of follow-up of our cohort will enable us to examine the progression of glomerular morphometric measures between the first and second biopsy. In this way, we will be able to assess the extent of further increases in Vv(Mes/glom) and other glomerular lesions with additional postpubertal years of type 1 diabetes.

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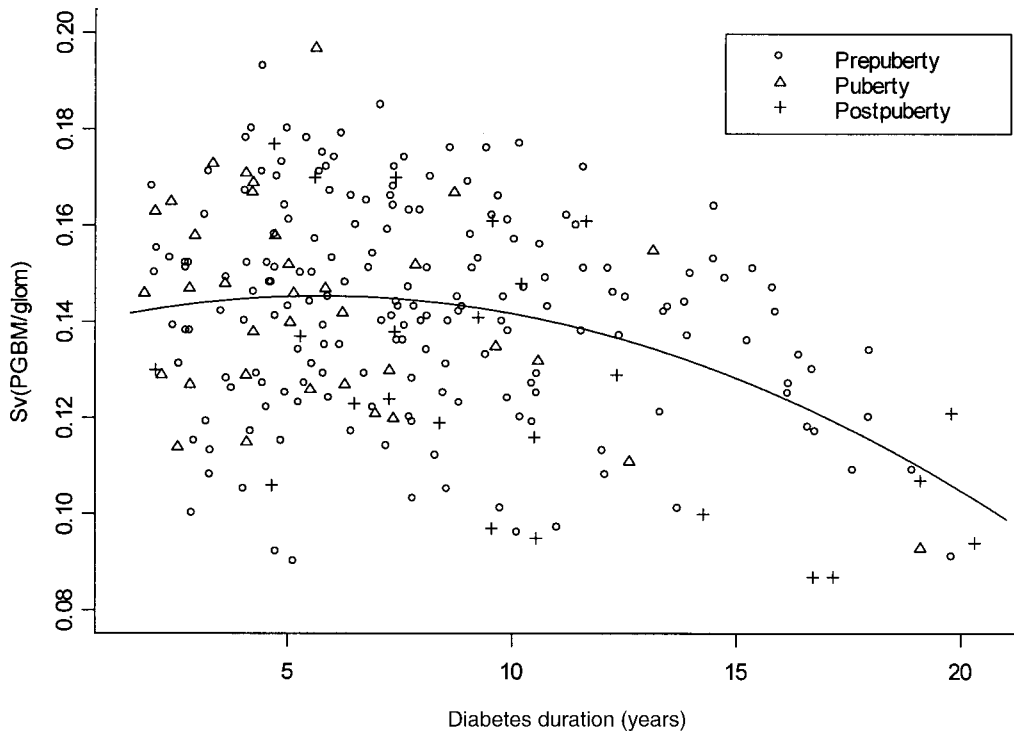


FIG. 3. Sv(PGBM/glom) as a function of disease duration in three subgroups defined by age at onset of type 1 diabetes.

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APPENDIX

Other members of the International Diabetic Nephropathy Study Group:

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REFERENCES

- West KM, Erdreich IJ, Tober JA: A detailed study of risk factors for retinopathy and nephropathy in diabetes. *Diabetes* 29:501-508, 1980
- Andersen AR, Christiansen JS, Andersen JK, Kreiner A, Deckert T: Diabetic nephropathy in type 1 (insulin-dependent) diabetes: an epidemiological study. *Diabetologia* 25:496-501, 1983
- Klein R, Klein BEK, Moss SE, Davis MD, DeMets DL: The Wisconsin Epidemiologic Study of Diabetic Retinopathy. II. Prevalence and risk of diabetic retinopathy when age at diagnosis is less than 30 years. *Arch Ophthalmol* 102:520-526, 1984
- Krolewski AS, Warram JH, Christlieb AR, Busick EJ, Kahn CR: The changing natural history of nephropathy in type I diabetes. *Am J Med* 78:785-794, 1985
- Kostraba JN, Dorman JS, Orchard TJ, Ohki Y, Ellis D, Doft BH, Lobes LA, LaPorte RE, Drash AL: Contribution of diabetes duration before puberty to development of microvascular complications in IDDM subjects. *Diabetes Care* 12:686-693, 1989
- Klein BEK, Moss SE, Klein R: Is menarche associated with diabetic retinopathy? *Diabetes Care* 13:1034-1038, 1990
- Lawson ML, Sochetti EB, Chait PG, Balfe JW, Daneman D: Effect of puberty on markers of glomerular hypertrophy and hypertension in IDDM. *Diabetes* 45:51-55, 1996
- Barkai L, Vamosi I, Lukacs K: Enhanced progression of urinary albumin excretion in IDDM during puberty. *Diabetes Care* 21:1019-1023, 1998
- Bruno G, Pagano G, the Piedmont Study Group for Diabetes Epidemiology: Low prevalence of microalbuminuria in young Italian insulin-dependent diabetic patients with short duration of disease: a population-based study. *Diabet Med* 13:889-893, 1996
- Cook JJ, Daneman D: Microalbuminuria in adolescents with insulin-dependent diabetes. *Am J Dis Child* 144:234-237, 1990
- Couper JJ, Clarke CF, Byrne GC, Jones TW, Donaghue KC, Nairn J, Boyce D, Russell M, Stephens M, Raymond J, Bates DJ, McCaul K: Progression of borderline increases in albuminuria in adolescents with insulin-dependent diabetes mellitus. *Diabet Med* 14:766-771, 1997
- Donaghue KC, Fung ATW, Hing S, Fairchild J, King J, Chan A, Howard NJ, Silink M: The effect of prepubertal diabetes duration on diabetes: microvascular complications in early and late adolescence. *Diabetes Care* 20:77-80, 1997
- Gorman D, Sochetti E, Daneman D: The natural history of microalbuminuria in adolescents with type 1 diabetes. *J Pediatr* 134:333-337, 1999
- Janner M, Knill SE, Diem P, Zuppinger KA, Mullis PE: Persistent microalbuminuria in adolescents with type 1 (insulin-dependent) diabetes is associated to early rather than late puberty. *Eur J Pediatr* 153:408-410, 1994
- Levy-Marchal C, Sahler C, Cahane M, Czernichow P: Risk factors for microalbuminuria in children and adolescents with type 1 diabetes. *J Pediatr Endocrinol Metab* 13:613-620, 2000
- Mortensen HB, Marinelli K, Norgaard K, Main K, Kastrup KW, Ibsen KK, Villumsen H: A nation-wide cross-sectional study of urinary albumin excretion rate, arterial blood pressure and blood glucose control in Danish children with type 1 diabetes mellitus. *Diabet Med* 7:887-897, 1990
- Rudberg S, Dahlquist G: Determinants of progression of microalbuminuria in adolescents with IDDM. *Diabetes Care* 19:369-371, 1996
- Salardi S, Cacciari E, Pascucci MG, Giambiasi E, Tacconi M, Tazzari R, Cicognani A, Boriani F, Puglioli R, Mantovani W, Donati S: Microalbuminuria in diabetic children and adolescents: relationship with puberty and growth hormone. *Acta Paediatr Scand* 79:437-443, 1990

19. Schultz CJ, Konopelska-Bahu T, Dalton RN, Carroll TA, Stratton I, Gale EA, Neil A, Dunger DB: Microalbuminuria prevalence varies with age, sex, and puberty in children with type 1 diabetes followed from diagnosis in a longitudinal study: Oxford regional prospective study group. *Diabetes Care* 22:495–502, 1999
20. Williamson JR, Rowold E, Chang K, Marvel J, Tomlinson M, Sherman WR, Ackermann KE, Berger RA, Kilo C: Sex steroid dependency of diabetes-induced changes in polyol metabolism, vascular permeability, and collagen cross-linking. *Diabetes* 35:20–27, 1986
21. Cummings EA, Sochett EB, Dekker MG, Lawson ML, Daneman D: Contribution of growth hormone and IGF-1 to early diabetic nephropathy in type 1 diabetes. *Diabetes* 47:1341–1346, 1998
22. Ellis EN, Warady BA, Wood EG, Hassanein R, Richardson WP, Lane PH, Howard C, Kemp SF, Aceto T, Garibaldi L, Wiegmann TB, Savin VJ: Renal structural-functional relationships in early diabetes mellitus. *Pediatr Nephrol* 11:584–591, 1997
23. Schultz CJ, Neil HA, Dalton RN, Dunger DB: Risk of nephropathy can be detected before the onset of microalbuminuria during the early years after diagnosis of type 1 diabetes. *Diabetes Care* 23:1811–1815, 2000
24. Mauer M, Drummond K, for the International Diabetic Nephropathy Study Group: The early natural history of nephropathy in type 1 diabetes. I. Study design and baseline characteristics of the study participants. *Diabetes* 51:1572–1579, 2002
25. Drummond K, Mauer M, for the International Diabetic Nephropathy Study Group: The early natural history of nephropathy in type 1 diabetes. II. Early renal structural changes in type 1 diabetes. *Diabetes* 51:1580–1587, 2002
26. Mauer SM, Steffes MW, Ellis EN, Sutherland DER, Brown DM, Goetz FC: Structural-functional relationships in diabetic nephropathy. *J Clin Invest* 74:1143–1155, 1984
27. Ellis EN, Steffes MW, Goetz FC, Sutherland DER, Mauer SM: Glomerular filtration surface in type I diabetes mellitus. *Kidney Int* 29:889–894, 1986
28. Fioretto P, Mauer SM, Bilous RW, Goetz FC, Sutherland DER, Steffes MW: Effects of pancreas transplantation on glomerular structure in insulin-dependent diabetic patients with their own kidneys. *Lancet* 342:1193–1196, 1993
29. Fioretto P, Steffes MW, Sutherland DER, Goetz FC, Mauer M: Reversal of lesions of diabetic nephropathy after pancreas transplantation. *N Engl J Med* 339:69–75, 1998
30. McLennan SV, Fisher EJ, Yue DK, Turtle JR: High glucose concentration causes a decrease in mesangium degradation: a factor in the pathogenesis of diabetic nephropathy. *Diabetes* 43:1041–1045, 1994
31. Anderson SS, Kim Y, Tsilibary EC: Effects of matrix glycation on mesangial cell adhesion, spreading and proliferation. *Kidney Int* 46:1359–1367, 1994
32. Joner G, Brinchmann-Hansen O, Torres CG, Hansen KF: A nation-wide cross-sectional study of retinopathy and microalbuminuria in young Norwegian type 1 (insulin-dependent) diabetic patients. *Diabetologia* 35:1049–1054, 1992
33. Mathiesen ER, Saurbrey N, Hommel E, Parving HH: Prevalence of microalbuminuria in children with type 1 diabetes mellitus. *Diabetologia* 29:640–643, 1986