

Evidence for two distinct phenotypes of chronic kidney disease in individuals with type 1 diabetes mellitus

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Abstract

Aims/hypothesis In a retrospective, observational, cross-sectional, single-centre study, we assessed the prevalence and correlates of different CKD phenotypes (with and without albuminuria) in a large cohort of patients of white ethnicity with type 1 diabetes.

Methods From 2001 to 2009, 408 men and 369 women with type 1 diabetes (age 40.2 ± 11.7 years, diabetes duration 19.4 ± 12.2 years, HbA_{1c} $7.83 \pm 1.17\%$ [62.0 ± 12.9 mmol/mol]) were recruited consecutively. Albumin-to-creatinine ratio (ACR) and eGFR (Modification of Diet in Renal Disease) were obtained for all individuals, together with CKD stage. Diabetic retinopathy and peripheral polyneuropathy were detected in 41.5% and 8.1%, respectively, and cardiovascular disease (CVD) occurred in 8.5%. Adjudications of CKD phenotype were made by blinded investigators.

Results Normo- (ACR <3.4), micro- (ACR $3.4\text{--}34$) or macroalbuminuria (ACR ≥ 34 mg/mmol) were present in 91.6%, 6.4% and 1.9% of individuals, respectively. eGFR categories 1 (≥ 90 ml min⁻¹ [1.73 m]⁻²), 2 ($60\text{--}89$ ml min⁻¹ [1.73 m]⁻²) and 3 (<60 ml min⁻¹ [1.73 m]⁻²) were present in 57.3%, 39.0% and 3.7%, respectively. The majority of participants had no CKD (89.4%), while stages 1–2 and ≥ 3

CKD were detected in 6.8% and 3.7%, respectively. The albuminuric (Alb⁺) and non-albuminuric (Alb⁻) phenotypes were present in 12 (41.4%) and 17 (58.6%) individuals with stage ≥ 3 CKD, respectively. Individuals with an ACR <3.4 mg/mmol were subdivided into those with normal albuminuria (<1.1 mg/mmol; 77.2%) and mildly increased albuminuria ($1.1\text{--}3.4$ mg/mmol; 14.4%), and individuals with stage 2 CKD were subdivided into those with eGFR $75\text{--}89$ ml min⁻¹ [1.73 m]⁻² and $60\text{--}74$ ml min⁻¹ [1.73 m]⁻². ACR <3.4 mg/mmol (88.7%) and even <1.1 mg/mmol (70.4%) were common in individuals with eGFR $60\text{--}74$ ml min⁻¹ [1.73 m]⁻². The prevalence of ACR <1.1 mg/mmol was lower but still significant (34.5%) in those with stage ≥ 3 CKD. In logistic regression analysis, stages 1–2 and ≥ 3 CKD were independently associated with age, HbA_{1c} , γ -glutamyltransferase, fibrinogen, hypertension, but not with sex, BMI, smoking, HDL-cholesterol or triacylglycerol. Inclusion of advanced retinopathy removed HbA_{1c} from the model. The CKD Alb⁺ phenotype correlated with diabetes duration, HbA_{1c} , HDL-cholesterol, fibrinogen and hypertension, while the CKD Alb⁻ phenotype was associated with age and hypertension, but not with diabetes duration, HbA_{1c} and fibrinogen.

Conclusions/interpretation The Alb⁻ CKD phenotype is present in a significant proportion of individuals with type 1 diabetes supporting the hypothesis of two distinct pathways (Alb⁺ and Alb⁻) of progression towards advanced kidney disease in type 1 diabetes. These are probably distinct pathways as suggested by different sets of covariates associated with the two CKD phenotypes.

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Keywords Albuminuria · Chronic kidney disease · Diabetic retinopathy · Glomerular filtration rate · Type 1 diabetes mellitus

Abbreviations

ACR	Albumin-to-creatinine ratio
Alb ⁺	Albuminuric (CKD phenotype)
Alb [−]	Non-albuminuric (CKD phenotype)
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
CKD	Chronic kidney disease
CVD	Cardiovascular disease
ERFD	Early renal function decline
GGT	γ -glutamyltransferase
RAS	Renin–angiotensin system
UAE	Urinary albumin excretion

Introduction

Chronic kidney disease (CKD) is a major complication of diabetes and an independent risk factor for cardiovascular disease (CVD). Albuminuria and reduced GFR are hallmarks of renal damage in individuals with diabetes [1]. In the progression of CKD, albuminuria is commonly believed to precede GFR loss [2] with microalbuminuria being the earliest marker of renal damage [2, 3]. Thus, urinary albumin excretion (UAE) is routinely measured during screening, diagnosis and management of diabetic nephropathy [4].

Recently, this view has been challenged [5, 6] following identification of individuals with type 2 diabetes who have non-albuminuric renal function impairment [7–11]. In the UK Prospective Diabetes Study (UKPDS), over a median follow-up of 15 years, about half of the participants who developed renal impairment had no prior albuminuria [12]. Data from the National Health and Nutrition Examination Survey (NHANES) [13] suggest a trend in the change of CKD phenotypes. While the rate of any CKD in individuals with diabetes has not changed over time, the prevalence of an albumin-to-creatinine ratio (ACR) ≥ 3.4 mg/mmol decreased from 20.8% in 1988–1994 to 15.9% in 2009–2014. In contrast, the prevalence of eGFR < 60 ml min^{−1} [1.73 m]^{−2} increased from 9.2% to 14.1%.

Limited information is available concerning a similar heterogeneity in those with type 1 diabetes. GFR decline can occur with no increased albuminuria even in individuals with long-standing type 1 diabetes. A renal biopsy study of 105 normoalbuminuric individuals with type 1 diabetes of ≥ 10 years duration showed advanced diabetic glomerular lesions even in those with mild GFR reduction (to < 90 ml min^{−1} [1.73 m]^{−2}) [14]. Over 19 years of the Diabetes Control and Complications Trials/Epidemiology of Diabetes Interventions and Complication (DCCT/EDIC), 11.4% of participants with type 1 diabetes developed sustained reduction of eGFR to < 60 ml min^{−1} [1.73 m]^{−2}. Among these, 24% developed persistent reduction in eGFR while remaining normoalbuminuric [15]. Thus, although AER

was associated with eGFR decline in 76% of participants a sustained loss of eGFR could not be identified in 24% of cases.

Microalbuminuria was found to revert to normoalbuminuria in several studies of type 1 diabetes [16–18] suggesting that the appearance of microalbuminuria does not necessarily signal the start of the progression of diabetic nephropathy [18]. Rather, these findings support the concept of a non-albuminuric CKD phenotype where AER does not progress despite steadily declining GFR [19, 20]. Interestingly, albuminuria and loss of GFR seem to be associated with distinct clusters of specific risk factors, suggesting that the two phenotypes only partly share common pathogenic mechanisms [11, 20].

Here, we report the results of a retrospective analysis performed in a large cohort of individuals with type 1 diabetes designed to determine the prevalence of CKD phenotypes (i.e. with and without albuminuria), describe associations with other diabetes complications and identify any differences in the associated factors.

Methods

Participants Type 1 diabetes was defined as diagnosis at < 36 years with immediate insulin requirement and unbroken insulin need after the initial year since diagnosis [21]. All individuals with type 1 diabetes ($n = 843$) meeting those criteria who attended the diabetes unit of our institution from 1 January 2001 to 31 December 2009 for screening of complications were eligible for inclusion. Exclusion criteria included pregnancy, non-white ethnicity ($n = 32$, 4.0%), diabetes duration < 1 year ($n = 34$, 4.0%) and dialysis or renal transplantation. A total of 777 individuals were recruited. The Ethics Committee of the University of Pisa approved the study and consent procedures. Written informed consent was obtained from each participating individual.

All participants underwent a structured interview collecting information about onset of diabetes, its duration, smoking and pharmacological treatments [21]. Body weight, height and waist circumference were measured. BP was measured after 5 min rest in a sitting position, and the average of two measurements (taken 5 min apart) was calculated; hypertension was defined for systolic BP > 140 mmHg and/or diastolic BP > 80 mmHg and/or use of antihypertensive medication(s). Blood was drawn once, on entry, after overnight fasting for determination of creatinine, HbA_{1c}, lipids and other variables (see ESM Methods).

Measurements ACR was determined in at least three first-void urine samples obtained at about 1 month intervals in the year preceding recruitment. Urine samples ($n = 175$, 6.7%) with abnormal sediments (nitrites or ≥ 250 leucocytes/ml)

were discarded. Albumin and creatinine were assayed on the morning of collection.

HbA_{1c} was measured by HPLC using DCCT-aligned methods [22]. Triacylglycerol, total cholesterol and HDL-cholesterol were determined by enzymatic methods and LDL-cholesterol was calculated using the Friedewald formula [23]. Standard methods were employed for the measurement of the other variables (see ESM [Methods](#)).

Definition of renal outcomes Based on the geometric mean of three ACR values the following categories were defined: normoalbuminuria (<3.4 mg/mmol), microalbuminuria or moderately increased albuminuria (3.4–34 mg/mmol) and macroalbuminuria or severely increased albuminuria (≥34 mg/mmol). Normoalbuminuria was subdivided into normal albuminuria (<1.1 mg/mmol) and mildly increased albuminuria (1.1–3.4 mg/mmol) (see ESM [Methods](#)).

eGFR was calculated by the Modification of Diet in Renal Disease (MDRD) Study equation [24] and the following categories were identified: category 1, eGFR ≥90; category 2, eGFR 60–89; and category 3, eGFR <60 ml min⁻¹ [1.73 m]⁻². Category 2 was subdivided into eGFR 75–89 ml min⁻¹ [1.73 m]⁻² and 60–74 ml min⁻¹ [1.73 m]⁻².

CKD was defined as: no CKD (eGFR ≥60 ml min⁻¹ [1.73 m]⁻² and normoalbuminuria), stages 1–2 CKD (eGFR ≥60 ml min⁻¹ [1.73 m]⁻² and albuminuria) and stages ≥3 CKD (eGFR <60 ml min⁻¹ [1.73 m]⁻² irrespective of albuminuria), according to the National Kidney Foundation's Kidney Disease Outcomes Quality Initiative [25]. CKD stages 3–5 were pooled.

Participants were further classified as having one of the following CKD phenotypes: albuminuria alone (stage 1–2 CKD), reduced eGFR alone (stage ≥3 CKD without albuminuria, Alb⁻) or both albuminuria and reduced eGFR (stage ≥3 CKD with albuminuria, Alb⁺).

Assessment of diabetes complications Diabetic retinopathy was assessed by retinal photography [21] and classified according to the Global Diabetic Retinopathy Project Group criteria [26] as non-advanced or advanced, sight-threatening retinopathy. Diabetic neuropathy was assessed with a validated questionnaire [27] and routine screening tests including vibration perception threshold [21]. The presence of previous CVD was determined based on a medical history of all documented major acute events. A 12 lead resting ECG was recorded and coded according to the Minnesota Code [28]. Peripheral vascular disease was assessed by search for femoral and foot pulses and measurement of ankle/brachial ratio (see ESM [Methods](#)).

Statistics For this analysis, we first stratified the study population according to AER or eGFR and then assessed the prevalence and implication of CKD stage for each

combination of the two. Data are expressed as median (interquartile range [IQR]) and/or mean ± SD for continuous variables, and number of cases and percentage for categorical variables. Continuous variables were compared by the Student's *t* test or one-way ANOVA for normally distributed variables, and by the Wilcoxon Sum-of-Ranks (Mann–Whitney) *U* test or Kruskal–Wallis test for variables with skewed distribution. Pearson χ^2 or Fisher exact tests were applied to categories. For post hoc comparisons, Scheffé's test, Mann–Whitney *U* test and χ^2 test were used for normally distributed, not normally distributed and categorical variables, respectively. Full model binary logistic regression analyses (including all variables of interest) were applied to assess the independent association of several continuous and categorical variables with each CKD phenotype. Results of these analyses were expressed as OR with 95% CI. A *p* value ≤0.05 was considered to be statistically significant (see ESM [Methods](#)).

Results

The main demographic and clinical characteristics of the study cohort are shown in Table 1 along with current pharmacological treatment.

Clinical characteristics by UAE category Normo-, micro- and macroalbuminuria were found in 712 (91.6%), 50 (6.4%) and 15 (1.9%) participants, respectively. Albuminuric individuals were older and had longer diabetes duration, higher HbA_{1c}, BMI, systolic and diastolic BP, LDL-cholesterol and triacylglycerol, and lower eGFR (ESM Table 1). They also had higher γ -glutamyltransferase (GGT), fibrinogen and uric acid levels. There were more current smokers in the micro-, and fewer in the macro-, compared with the normoalbuminuric groups. Albuminuric individuals were more frequently hypertensive, on BP-lowering agents (and renin–angiotensin system [RAS] blockers) and antiplatelet agents, and had higher daily insulin requirements. Rates of advanced retinopathy, polyneuropathy and CVD increased across the albuminuria categories.

Compared with participants with normal albuminuria (*n* = 600, 77.2%), those with mildly increased albuminuria (*n* = 112, 14.4%) had higher HbA_{1c} and fibrinogen, higher rates of hypertension and advanced retinopathy, and marginally increased rates of CVD. Use of BP-lowering agents and RAS blockers was more common in the group with mildly increased albuminuria (ESM Table 2).

Clinical characteristics by eGFR category eGFR categories 1, 2 and 3 included 445 (57.3%), 303 (39.0%) and 29 (3.7%) participants, respectively. Age, diabetes duration and BMI increased progressively across these categories. HbA_{1c}, systolic and diastolic BP, triacylglycerol, GGT, fibrinogen,

Table 1 Clinical characteristics of the type 1 diabetes cohort as a whole and after stratification by CKD stage

	All patients (<i>n</i> = 777)	No CKD (<i>n</i> = 695)	Stage 1–2 CKD (<i>n</i> = 53)	Stage ≥3 CKD (<i>n</i> = 29)	<i>p</i> value
Men/women, <i>n</i> (%)	408/369 (52.5/47.5)	359/336 (51.7/48.3)	31/22 (58.5/41.5)	18/11 (62.1/37.9)	0.369
Age, years	40.2 ± 11.7	39.4 ± 11.1	41.8 ± 12.3	56.2 ± 13.1*** †††	<0.0001
Age at diabetes diagnosis, years	20.8 ± 10.9	20.9 ± 10.8	18.1 ± 10.3	24.6 ± 12.8†	0.031
Duration of diabetes, years	19.4 ± 12.2	18.5 ± 12.0	23.7 ± 11.3**	31.7 ± 12.1*** †	<0.0001
BMI, kg/m ²	24.8 ± 3.6	24.6 ± 3.4	26.2 ± 4.8**	26.2 ± 3.7*	<0.001
Waist circumference, cm	90.8 ± 10.7	90.3 ± 10.3	95.7 ± 13.4**	94.1 ± 12.5	<0.0001
Smoking habit (non-smoker/ current smoker) (<i>n</i> = 762), <i>n</i> (%)	536/226 (70.4/29.6)	482/200 (70.7/29.3)	32/19 (62.7/37.3)	22/7 (75.9/24.1)	0.497
Fasting glucose, mmol/l	9.44 ± 4.56	9.22 ± 4.45	11.22 ± 4.79**	11.58 ± 5.52*	<0.0001
HbA _{1c} , % (mmol/mol)	7.83 ± 1.17 (62.0 ± 12.9)	7.76 ± 1.15 (61.4 ± 12.6)	8.46 ± 1.23*** (69.0 ± 13.4)	8.17 ± 1.26 (65.7 ± 13.8)	<0.0001
Systolic BP, mmHg	127 ± 18	125 ± 17	134 ± 19***	152 ± 18*** †††	<0.0001
Diastolic BP, mmHg	73 ± 9	73 ± 8	76 ± 12*	79 ± 8**	<0.0001
Total cholesterol, mmol/l	4.85 ± 0.88	4.81 ± 0.85	5.27 ± 1.14***	4.88 ± 1.01	<0.001
LDL-cholesterol, mmol/l	3.01 ± 0.76	2.99 ± 0.74	3.30 ± 0.95*	3.04 ± 0.82	0.014
HDL-cholesterol, mmol/l					
Men	1.45 (1.24–1.71)	1.45 (1.24–1.68)	1.61 (1.35–1.86)	1.37 (0.99–1.58) †	0.036
Women	1.74 (1.48–2.07)	1.73 (1.48–2.07)	1.75 (1.39–2.15)	1.79 (1.50–1.97)	0.981
Triacylglycerol, mmol/l					
Men	0.90 (0.71–1.24)	0.89 (0.70–1.19)	1.00 (0.82–1.58)	1.25 (0.86–2.01)	0.044
Women	0.78 (0.60–1.06)	0.77 (0.59–1.04)	0.98 (0.75–1.15)**	1.21 (0.70–1.32)	0.005
ALT, U/l	20 ± 11	20 ± 10	23 ± 16	21 ± 6	0.095
AST, U/l	22 ± 32	22 ± 33	25 ± 27	22 ± 13	0.812
GGT, U/l	21 ± 33	19 ± 25	32 ± 85*	36 ± 33*	<0.001
Uric acid, μmol/l	223.8 ± 67.5	217.4 ± 58.2	241.4 ± 72.3*	343.3 ± 126.9*** †††	<0.0001
Fibrinogen, μmol/l	9.89 ± 2.00	9.75 ± 1.91	10.93 ± 2.33***	11.41 ± 2.15***	<0.0001
Creatinine, μmol/l	73.4 ± 18.6	71.2 ± 12.7	72.2 ± 13.5	127.1 ± 46.2*** †††	<0.0001
ACR, mg/mmol	0.49 (0.26–1.02)	0.44 (0.24–0.81)	8.11 (5.21–22.74)***	1.96 (0.58–40.38)*** †	<0.0001
eGFR ^a , ml min ^{−1} [1.73 m] ^{−2}	95.1 ± 21.4	97.0 ± 19.8	95.7 ± 18.3	49.3 ± 9.5*** †††	<0.0001
Daily insulin dose, U/kg body weight	0.66 ± 0.20	0.66 ± 0.20	0.71 ± 0.15	0.71 ± 0.28	0.103
MDI/CSII, <i>n</i> (%)	694/83 (89.3/10.7)	617/78 (88.8/11.2)	48/5 (90.6/9.4)	29/0 (100/0)	0.151
Treatment, <i>n</i> (%)					
BP-lowering agents	151 (19.4)	99 (14.3)	31 (58.5)***	21 (72.4)***	<0.0001
RAS blockers	136 (17.5)	87 (12.5)	30 (56.6)***	19 (65.5)***	<0.0001
Lipid-lowering agents	100 (12.9)	79 (11.4)	8 (15.1)	13 (44.8)*** ††	<0.0001
Antiplatelet agents	50 (6.4)	33 (4.8)	7 (13.2)*	10 (34.5)*** †	<0.0001
Metformin	46 (5.9)	41 (5.9)	1 (1.9)	4 (13.8)	0.092
Hypertension, <i>n</i> (%)	273 (35.1)	207 (29.9)	39 (73.6)***	27 (93.1)*** †	<0.0001
Retinopathy: no/non-advanced/ advanced (<i>n</i> = 753), <i>n</i> (%)	441/194/118 (58.5/25.8/15.7)	423/175/77 (62.7/25.9/11.4)	15/12/24*** (29.4/23.5/47.1)	3/7/17*** (11.1/25.9/63.0)	<0.0001
Peripheral polyneuropathy: no/yes (<i>n</i> = 731), <i>n</i> (%)	671/60 (91.8/8.2)	619/39 (94.1/5.9)	39/9** (81.2/18.8)	13/12*** †† (52.0/48.0)	<0.0001
Cardiovascular events, <i>n</i> (%)	66 (8.5)	50 (7.2)	6 (11.3)	10 (34.5)*** †	<0.0001
Autoimmune thyropathy, <i>n</i> (%)	113 (14.5)	103 (14.8)	7 (13.2)	3 (10.3)	0.765

Quantitative variables are shown as mean ± SD or median (IQR)

^aBased on the MDRD equation**p* < 0.05; ***p* < 0.01; ****p* < 0.001 vs no CKD† *p* < 0.05; †† *p* < 0.01; ††† *p* < 0.001 vs stage 1–2 CKD

ALT, alanine aminotransferase; AST, aspartate aminotransferase; MDI/CSII, multiple daily insulin injections/continuous subcutaneous insulin infusion

uric acid, ACR, rates of treatment with BP-lowering (and RAS blockers), lipid-lowering and antiplatelet agents, and prevalence of hypertension, advanced retinopathy, polyneuropathy and CVD were all higher, and HDL-cholesterol lower in eGFR category 3 compared with categories 1 and 2. Categories 1 and 2 showed similar levels for all variables (ESM Table 3). Compared with individuals with eGFR 75–89 ($n = 232$, 29.9%), those with eGFR 60–74 $\text{ml min}^{-1} [1.73 \text{ m}]^{-2}$ ($n = 71$, 9.1%) had longer diabetes duration, higher rates of treatment with BP-lowering agents and higher prevalence of hypertension and advanced diabetic retinopathy (ESM Table 4).

Clinical characteristics by CKD stage Based on eGFR and ACR, 695 individuals (89.4%) had no CKD, with stages 1–2 and ≥ 3 CKD including 6.8% ($n = 53$) and 3.7% ($n = 29$) of the participants, respectively. Microalbuminuria was more prevalent in individuals with stage 1–2 CKD (46/53, 86.8%), while macroalbuminuria was more common in those with stage ≥ 3 CKD (8/29, 27.6%; $p = 0.0004$; Fig. 1). Sex distribution, smoking habits, and daily insulin dose were comparable across the CKD stages. Those with stage ≥ 3 CKD were older, had higher uric acid and, by definition, higher creatinine and lower eGFR. These variables did not differ between stage 1–2 CKD and no CKD (Table 1). BMI, waist, HbA_{1c}, diastolic BP, GGT, fibrinogen, treatment with BP-lowering agents and RAS blockers, and prevalence of hypertension were higher in those with stages 1–2 and ≥ 3 CKD compared with no CKD. Finally, diabetes duration, systolic BP, use of lipid-lowering and antiplatelet agents, and

prevalence of advanced retinopathy (Fig. 1), polyneuropathy and CVD progressively increased with CKD stage (Table 1). Lipid-lowering treatment most probably accounts for the higher LDL-cholesterol level observed in those with stage 1–2, but not in stage ≥ 3 CKD, compared with no CKD.

Albuminuric and non-albuminuric phenotypes in individuals with stage ≥ 3 CKD

Of 29 participants with stage ≥ 3 CKD, albuminuria ($\geq 3.4 \text{ mg/mm}$; Alb⁺) was detected in 12 (41.4%), with the remaining 17 (58.6%) having a non-albuminuric (Alb⁻) phenotype (Table 2). There were no significant differences between the phenotypes for most of the variables considered including age, diabetes duration and sex, with the exception of lower eGFR (45.2 ± 10.8 vs $52.2 \pm 7.4 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$, $p = 0.048$) and marginally higher HbA_{1c} (8.62 ± 1.33 vs $7.84 \pm 1.13\%$ [70.8 ± 14.6 vs $62.2 \pm 12.4 \text{ mmol/mol}$]; $p = 0.080$) in the Alb⁺ group (ESM Table 5). Interestingly, there were no differences between the phenotypes for the distribution of retinopathy (Fig. 1), rate of hypertension or CVD, or the use of BP-lowering agents or RAS blockers.

Alb⁺ and Alb⁻ phenotypes in individuals with eGFR <75 $\text{ml min}^{-1} [1.73 \text{ m}]^{-2}$

Of the 71 individuals with eGFR 60–74 $\text{ml min}^{-1} [1.73 \text{ m}]^{-2}$, eight (11.3%) and 63 (88.7%) had Alb⁺ and Alb⁻ phenotypes, respectively (Table 3). Within this sub-category, albuminuria was normal (ACR <1.1 mg/mm) in most of the participants (50/71, 70.4%). The percentage of individuals with normal albuminuria was lower, although still relevant, in patients in stage ≥ 3 CKD (10/29, 34.5%; Table 2).

Fig. 1 CKD stage and phenotype distribution in the study cohort. Microalbuminuria was more prevalent among individuals with stage 1–2 CKD (86.8%) and macroalbuminuria among individuals with stage ≥ 3 CKD (66.7%; $p = 0.0004$). Prevalence of advanced retinopathy increased with increasing CKD stage (no CKD < stage 1–2 CKD < stage ≥ 3 CKD; $p < 0.0001$) with no differences between non-albuminuric and albuminuric stage ≥ 3 CKD. Data on retinopathy were available for 753 individuals. Data are expressed as n (%)

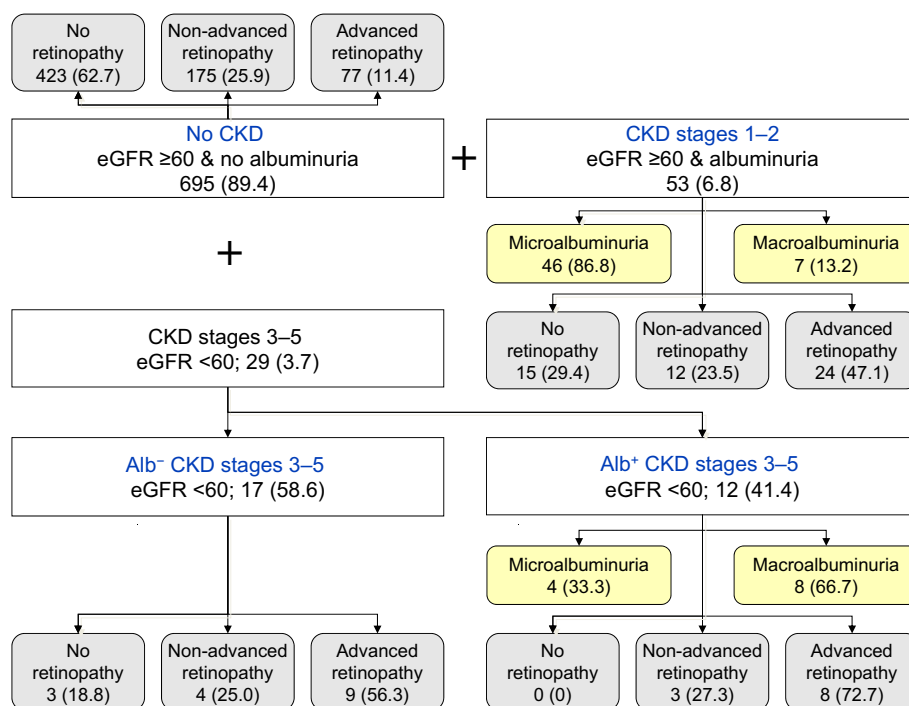


Table 2 Distribution of albuminuric and non-albuminuric phenotypes by eGFR category. Alb[−] and Alb⁺ CKD phenotypes are defined as for the binary logistic regression analysis described in Tables 4 and 5

	eGFR ^a (ml min ^{−1} [1.73 m] ^{−2})				Total
	≥90	75–89	60–74	<60	
<i>n</i>	445	232	71	29	777
Normal albuminuria (<1.1 mg/mmol)	353 (79.4)	187 (80.6)	50 (70.4)	10 (34.5)	600 (77.2)
Mildly increased albuminuria (1.1–3.4 mg/mmol)	61 (13.7)	31 (13.4)	13 (18.3)	7 (24.1)	112 (14.4)
Moderately increased albuminuria (3.4–34 mg/mmol)	25 (5.6)	14 (6.0)	7 (9.9)	4 (13.8)	50 (6.4)
Severely increased albuminuria (≥34 mg/mmol)	6 (1.3)	–	1 (1.4)	8 (27.6)	15 (1.9)

Data are expressed as number and percentage

Green, no CKD (*n* = 695); yellow, stages 1–2 CKD (*n* = 53); orange, non-albuminuric stages ≥3 CKD (*n* = 17); red, albuminuric stages ≥3 CKD (*n* = 12)

^a Based on the MDRD equation

The 21 individuals in the eGFR 60–74 ml min^{−1} [1.73 m]^{−2} sub-category with mildly-to-severely increased albuminuria were older, had longer diabetes duration, were heavier and had larger waist circumference, and higher HbA_{1c} (8.50 ± 1.28 vs $7.60 \pm 0.91\%$ [69.4 ± 13.9 vs 59.6 ± 10.1 mmol/mol]; *p* = 0.016) and fibrinogen levels compared with the 50 individuals with normal albuminuria.

When stage ≥3 CKD and the eGFR 60–74 ml min^{−1} [1.73 m]^{−2} sub-category were pooled (*n* = 100; ESM Table 6), the Alb⁺ group (*n* = 40) had higher BMI, larger waist circumference, higher HbA_{1c}, fibrinogen, triacylglycerol,

systolic BP and lower HDL-cholesterol (all *p* < 0.05) than the Alb[−] group (*n* = 60). There were no differences in age, diabetes duration, smoking habits and rate of CVD. The Alb⁺ group included more men (57.5 vs 33.9%, *p* = 0.024), had greater prevalence of hypertension (81.1 vs 44.8%, *p* = 0.001), and BP-lowering agents (60.0 vs 28.5%, *p* = 0.003) or RAS blockers (55.0 vs 23.7%, *p* = 0.003) and had higher rates of advanced retinopathy (62.5 vs 27.6%, *p* = 0.003).

Factors associated with different CKD phenotypes The independent association of continuous and categorical

Table 3 Distribution of albuminuric and non-albuminuric phenotypes by eGFR category. CKD phenotypes are defined as for the binary logistic regression analysis described in Table 5

	eGFR ^a (ml min ^{−1} [1.73 m] ^{−2})				Total
	≥90	75–89	60–74	<60	
<i>n</i>	445	232	71	29	777
Normal albuminuria (<1.1 mg/mmol)	353 (79.4)	187 (80.6)	50 (70.4)	10 (34.5)	600 (77.2)
Mildly increased albuminuria (1.1–3.4 mg/mmol)	61 (13.7)	31 (13.4)	13 (18.3)	7 (24.1)	112 (14.4)
Moderately increased albuminuria (3.4–34 mg/mmol)	25 (5.6)	14 (6.0)	7 (9.9)	4 (13.8)	50 (6.4)
Severely increased albuminuria (≥34 mg/mmol)	6 (1.3)	–	1 (1.4)	8 (27.6)	15 (1.9)

Data are expressed as number and percentage

Green, no CKD (*n* = 540); yellow, mildly-to-severely increased albuminuria and eGFR ≥75 ml min^{−1} [1.73 m]^{−2} (*n* = 137); orange, normal albuminuria and eGFR <75 ml min^{−1} [1.73 m]^{−2} (*n* = 60); red, mildly-to-severely increased albuminuria and eGFR <75 ml min^{−1} [1.73 m]^{−2} (*n* = 40)

^a Based on the MDRD equation

variables with the CKD phenotypes was assessed using binary logistic regression analysis (Tables 4, 5).

Stages 1–2 and ≥ 3 CKD were associated with systolic BP and/or hypertension, HbA_{1c}, GGT and fibrinogen. Age was directly related to stage ≥ 3 CKD and inversely related to stage 1–2 CKD. Diabetes duration and total cholesterol were only related to stage 1–2 CKD (model 1). The inclusion of retinopathy (model 2) removed diabetes duration and HbA_{1c} as independent covariates of stages 1–2 and ≥ 3 CKD, respectively (Table 4).

Notably, stage ≥ 3 CKD Alb⁺ was independently associated with diabetes duration and HbA_{1c}, systolic BP, fibrinogen and, in an inverse manner, HDL-cholesterol. In contrast, CKD Alb[−] was related to age, hypertension, systolic BP and GGT, but not to diabetes duration and HbA_{1c}. There was no association with use of antihypertensive agents (model 1). Inclusion of retinopathy (model 2) removed diabetes duration as a covariate for stage ≥ 3 CKD Alb⁺, with no effect on the covariates for stage ≥ 3 CKD Alb[−] (Table 5).

Overall, the sensitivity analysis performed in individuals with eGFR <75 ml min^{−1} [1.73 m]^{−2} confirmed the results (Table 5).

Discussion

Our results support the emerging view that in type 1 diabetes CKD is more heterogeneous than commonly believed. Even though the Alb⁺ phenotype remains predominant (79.3%), in our cohort more than half of the participants with stage ≥ 3 CKD (58.6%) displayed the Alb[−] phenotype. It may also be worth pointing out that the overall prevalence of the Alb[−] CKD phenotype (2%) is much lower than it has been reported in type 2 diabetes (10%) [11]; this probably reflects a greater prevalence of hypertension, CVD and obesity in the latter group [11]. These findings are of relevance since most studies investigating the prevalence of non-albuminuric CKD have been performed in type 2 diabetes, where as many as 36% [8] to 62% [10] of individuals with renal insufficiency do

Table 4 Independent correlates of stage 1–2 CKD and stage ≥ 3 CKD vs no CKD (the reference group)

	Stage 1–2 CKD (<i>n</i> = 53)			Stage ≥ 3 CKD (<i>n</i> = 29)		
	OR	95% CI	<i>p</i> value	OR	95% CI	<i>p</i> value
Model 1^a						
Age, 1 year	0.950	0.912, 0.989	0.012	1.070	1.025, 1.117	0.002
Duration of diabetes, 1 year	1.048	1.012, 1.084	0.008	1.046	0.996, 1.099	0.072
HbA _{1c} , 1 mmol/mol	1.041	1.015, 1.067	0.002	1.044	1.005, 1.086	0.028
Total cholesterol, 1 mmol/l	1.406	1.007, 1.962	0.045	1.242	0.661, 2.334	0.501
GGT, 1 U/l	1.006	1.000, 1.012	0.035	1.017	1.006, 1.028	0.003
Fibrinogen, 1 μ mol/l	1.220	1.025, 1.451	0.025	1.440	1.110, 1.867	0.006
Hypertension	5.582	2.335, 13.341	0.0001	9.756	1.473, 64.629	0.018
Systolic BP, 1 mmHg	0.999	0.974, 1.025	0.956	1.027	1.001, 1.053	0.045
Model 2^b						
Age, 1 year	0.944	0.903, 0.986	0.009	1.071	1.008, 1.137	0.025
Duration of diabetes, 1 year	1.005	0.963, 1.050	0.810	1.010	0.951, 1.072	0.758
HbA _{1c} , 1 mmol/mol	1.036	1.009, 1.063	0.009	1.025	0.979, 1.073	0.293
Total cholesterol, 1 mmol/l	1.487	1.050, 2.106	0.025	1.334	0.669, 2.662	0.413
GGT, 1 U/l	1.006	1.000, 1.012	0.035	1.019	1.007, 1.031	0.003
Fibrinogen, 1 μ mol/l	1.204	0.999, 1.451	0.051	1.468	1.104, 1.951	0.008
Hypertension	5.102	2.037, 12.776	0.001	6.697	1.058, 42.399	0.043
Systolic BP, 1 mmHg	0.998	0.970, 1.027	0.895	1.025	0.998, 1.053	0.066
Retinopathy			0.0001			0.003
No	1			1		
Non-advanced	1.470	0.584, 3.698	0.413	1.747	0.547, 13.857	0.220
Advanced	10.242	4.216, 24.879	0.0001	9.877	2.200, 44.350	0.003

^a Other variables included in model 1 but never selected as significant independent covariates: sex, BMI, smoking, diastolic BP, HDL-cholesterol, triacylglycerol, uric acid, RAS blockers

^b Other variables included in model 2 but never selected as significant independent covariates: sex, BMI, smoking, diastolic BP, HDL-cholesterol, triacylglycerol, uric acid, RAS blockers

Table 5 Independent correlates of stage ≥ 3 CKD Alb⁺ and Alb⁻ vs no CKD and of individuals with eGFR <75 ml min⁻¹ [1.73 m]⁻² Alb⁺ and Alb⁻ vs no CKD (eGFR ≥ 75 ml min⁻¹ [1.73 m]⁻² without albuminuria)

	Stage ≥ 3 CKD Alb ⁻ (<i>n</i> = 17)				Stage ≥ 3 CKD Alb ⁺ (<i>n</i> = 12)				eGFR <75 ml min ⁻¹ [1.73 m] ⁻² Alb ⁻ (<i>n</i> = 60)				eGFR <75 ml min ⁻¹ [1.73 m] ⁻² Alb ⁺ (<i>n</i> = 40)			
	OR	95% CI	<i>p</i> value		OR	95% CI	<i>p</i> value		OR	95% CI	<i>p</i> value		OR	95% CI	<i>p</i> value	
Model 1^a																
Age, 1 year	1.089	1.033, 1.148	0.002	1.047	0.949, 1.154	0.360	1.044	1.015, 1.074	1.006	0.956, 1.059	0.003	1.004	1.006	0.956, 1.059	0.807	
Sex, women	1.558	0.452, 5.366	0.482	0.316	0.010, 1.309	0.092	2.357	1.207, 4.605	0.829	0.306, 2.245	0.012	2.357	0.829	0.306, 2.245	0.712	
Duration of diabetes, 1 year	1.024	0.967, 1.083	0.421	1.107	1.025, 1.197	0.010	1.016	0.934, 1.068	1.064	1.014, 1.116	0.534	1.016	1.064	1.014, 1.116	0.011	
HbA _{1c} , 1 mmol/mol	1.020	0.969, 1.074	0.440	1.104	1.037, 1.176	0.002	0.977	0.949, 1.005	1.046	1.008, 1.085	0.104	0.977	1.046	1.008, 1.085	0.017	
HDL-cholesterol, 0.1 mmol/l	0.926	0.781, 1.097	0.373	0.742	0.589, 0.935	0.012	1.076	0.986, 1.174	0.977	0.849, 1.125	0.100	1.076	0.977	0.849, 1.125	0.740	
Triacylglycerol, 0.1 mmol/l	1.009	0.966, 1.078	0.684	0.967	0.809, 1.156	0.714	1.066	1.007, 1.129	1.064	1.006, 1.125	0.029	1.066	1.064	1.006, 1.125	0.031	
GGT, 1 U/l	1.015	1.002, 1.028	0.020	1.016	0.998, 1.027	0.063	1.005	0.994, 1.017	1.005	0.993, 1.018	0.366	1.005	1.005	0.993, 1.018	0.414	
Fibrinogen, 1 μ mol/l	1.176	0.851, 1.626	0.325	2.155	1.431, 3.244	0.002	1.036	0.871, 1.232	1.486	1.117, 1.876	0.692	1.036	1.486	1.117, 1.876	0.001	
Hypertension	10.484	0.843, 130.392	0.068	2.170	0.401, 46.412	0.620	2.656	1.332, 5.296	4.564	1.314, 15.848	0.006	2.656	4.564	1.314, 15.848	0.017	
Systolic BP, 1 mmHg	1.025	0.995, 1.056	0.099	1.067	1.024, 1.113	0.002	0.998	0.976, 1.020	1.007	0.979, 1.037	0.829	0.998	1.007	0.979, 1.037	0.622	
Model 2^b																
Age, 1 year	1.086	1.028, 1.148	0.003	1.056	0.951, 1.173	0.305	1.037	1.009, 1.066	1.004	0.950, 1.061	0.010	1.037	1.004	0.950, 1.061	0.901	
Sex, women	1.642	0.423, 6.369	0.474	0.192	0.016, 2.309	0.193	2.571	1.284, 5.148	1.070	0.372, 3.075	0.008	2.571	1.070	0.372, 3.075	0.901	
Duration of diabetes, 1 year	0.989	0.922, 1.062	0.767	1.079	0.947, 1.229	0.254	1.006	0.972, 1.042	1.024	0.966, 1.085	0.719	1.006	1.024	0.966, 1.085	0.430	
HbA _{1c} , 1 mmol/mol	0.996	0.934, 1.062	0.899	1.098	1.005, 1.200	0.039	0.975	0.948, 1.004	1.021	0.978, 1.065	0.092	0.975	1.021	0.978, 1.065	0.341	
HDL-cholesterol, 0.1 mmol/l	0.961	0.783, 1.179	0.702	0.727	0.576, 0.919	0.008	1.123	1.015, 1.244	0.970	0.823, 1.144	0.025	1.123	0.970	0.823, 1.144	0.721	
Triacylglycerol, 0.1 mmol/l	1.014	0.969, 1.061	0.542	0.981	0.835, 1.154	0.820	1.084	1.016, 1.156	1.058	0.996, 1.125	0.015	1.084	1.058	0.996, 1.125	0.068	
GGT, 1 U/l	1.016	1.002, 1.031	0.023	1.020	1.004, 1.036	0.012	1.004	0.992, 1.017	1.006	0.992, 1.021	0.532	1.004	1.006	0.992, 1.021	0.419	
Fibrinogen, 1 μ mol/l	1.251	0.874, 1.790	0.222	2.237	1.272, 3.932	0.005	1.085	0.904, 1.304	1.489	1.153, 1.923	0.381	1.085	1.489	1.153, 1.923	0.002	
Hypertension	8.744	1.330, 117.632	0.028	3.380	0.408, 105.818	0.488	2.470	1.168, 5.222	3.601	1.766, 12.959	0.018	2.470	3.601	1.766, 12.959	0.003	
Systolic BP, 1 mmHg	1.022	0.985, 1.059	0.250	1.063	1.024, 1.103	0.002	0.997	0.974, 1.020	1.008	0.977, 1.041	0.779	0.997	1.008	0.977, 1.041	0.606	
Retinopathy			0.022			0.010					0.001				0.0001	
No	1			1			1		1				1			
Non-advanced	1.729	0.276, 10.821	0.558	8.935	0.901, 77.483	0.059	2.812	1.387, 5.699	3.060	0.809, 11.578	0.004	2.812	3.060	0.809, 11.578	0.100	
Advanced	6.683	1.251, 35.694	0.023	21.119	3.773, 222.051	0.004	4.879	2.095, 11.365	14.420	3.309, 42.863	0.0001	4.879	14.420	3.309, 42.863	0.0001	

^a Other variables included in model 1 but never selected as significant independent covariates: BMI, smoking habits, diastolic BP, total cholesterol, uric acid, RAS blockers^b Other variables included in model 2 but never selected as significant independent covariates: BMI, smoking habits, diastolic BP, total cholesterol, uric acid, RAS blockers

not present with albuminuria. On the contrary, the prevalence of different CKD phenotypes (i.e. with and without albuminuria) and clinical and biochemical factors associated with increasing albuminuria vs decreasing GFR as separate entities have been poorly described in type 1 diabetes.

The appreciation of normoalbuminuric CKD is at variance with some of the classic studies. For example, The Pittsburgh Epidemiology of Diabetes Complications Study showed that eGFR reduction $<60 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$ occurred rarely in type 1 diabetes patients with no prior/concurrent albuminuria [29]. A different picture was described by the DCCT/EDIC where a sizeable percentage (24%) of individuals developing persistent eGFR reduction $<60 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$ had an AER $<30 \text{ mg/24 h}$ in the preceding 19 years [15]. Similarly, a cross-sectional survey of the UK National Diabetes Audit reported that albuminuria was absent in 54.4% of individuals with type 1 diabetes and stage ≥ 3 CKD [30]. In our cohort the overall prevalence of normoalbuminuric CKD was 2.2%, similar to that reported in the larger population of the FinnDiane Study ($n = 3809$, 2.0%) [31]. The two studies, however, differ in that in the FinnDiane Study non-albuminuric CKD accounted for 6.1% of all patients with any CKD phenotype and 15.5% of all individuals with stage ≥ 3 CKD, while these figures are 20.7% and 58.6%, respectively, in our cohort. These differences are not easy to reconcile but our population is representative of a specific geographical area and reflects clinical management in a single centre as opposed to a population collected at national level and from different centres, which could have introduced more variability. Our results remained substantially unaffected by including individuals with eGFR $60\text{--}74 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$ and those with mildly increased albuminuria (i.e. those with an ACR of 1.1–3.4 mg/mmol). Even with this broader categorisation, the Alb[−] phenotype (eGFR $<75 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$) accounted for a similar proportion (60%) of all participants. In summary, at least 50% of individuals with type 1 diabetes with an eGFR $<60 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$ are normoalbuminuric, and some may well progress to a more advanced stage of renal failure without manifesting albuminuria as indicated by normal UAE in 34.5% of individuals with stage ≥ 3 CKD.

Despite the cross-sectional nature of our study, we speculate that, in a sizeable proportion of the type 1 diabetes population, the decline in GFR can progress without albuminuria or even anticipate its appearance. These results are in line with those of Zerbini et al. [32] who demonstrated, over a 9.5 year follow-up in 146 young individuals with type 1 diabetes, a faster decline in GFR prior to the appearance of microalbuminuria. These observations have prompted the identification of a novel phenotype, the early renal function decline (ERFD), in the development of CKD in type 1 diabetes [33, 34]. Both the First and the Second Joslin Study of Natural History of Microalbuminuria in Type 1 Diabetes reported ERFD (defined as loss in GFR that exceeds 3.3% per year) to evolve in up to 9–10% of normoalbuminuric individuals with type 1 diabetes [33, 35]. Moreover, in the series

by Perkins et al. [36] about half of the individuals progressing to advanced CKD experienced only persistent microalbuminuria with some even reverting to normoalbuminuria [36]. These observations are in agreement with our data showing a high proportion of individuals (70.4%) with normal albuminuria (ACR $<1.1 \text{ mg/mmol}$) who already had a reduction, although mild, in eGFR ($60\text{--}74 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$). The two phenotypes may also differentiate in terms of outcomes, as ERFD appears to be more frequent in Alb⁺ than in Alb[−] patients [33, 35], and the rate of GFR loss tends to be faster in patients with micro- than in those with normoalbuminuria [36, 37].

In summary, our observations and the available evidence support the hypothesis that UAE and loss of GFR may not necessarily represent stages of a common pathway; rather they may be independent markers of progression of the disease [38]. This is further supported by the identification of distinct sets of risk factors for GFR loss vs progression of UAE.

In our cohort, age was an independent predictor in the Alb[−] but not in the Alb⁺ groups even though there was no difference in age between the two groups within stage ≥ 3 CKD and CKD stages with eGFR $<75 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$. The Alb[−] subgroup, however, was younger within the $60\text{--}74 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$ eGFR sub-category. In line with these results, Alb[−] CKD has been reported in relatively young patients with type 1 diabetes in several studies [14, 15]. The Alb[−] CKD phenotype within those with eGFR $<75 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$ was also independently related to female sex, similar to the findings of the FinnDiane Study [31].

In our survey, the Alb⁺ CKD phenotype was strongly associated with diabetes features. With individuals without CKD as a reference group, HbA_{1c} was independently associated with stages 1–2 and ≥ 3 CKD and with stage ≥ 3 CKD Alb⁺, but not with stage ≥ 3 CKD Alb[−]. Consistently, diabetes duration was related to stage 1–2 CKD and to stage ≥ 3 CKD Alb⁺, but not with stage ≥ 3 CKD Alb[−]. Sensitivity analysis confirmed the association of diabetes duration and HbA_{1c} with eGFR $<75 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$ Alb⁺, but not with eGFR $<75 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$ Alb[−]. These findings are in line with reports from the FinnDiane Study [31] and the Second Joslin Study [35]. In the latter, the risk of renal functional decline in normoalbuminuric individuals was independent of HbA_{1c} quartile, but increased from the lowest to the highest quartile in the microalbuminuric group.

When included in the model, retinopathy, a condition strongly associated with chronic hyperglycaemia, entered in the regression by displacing diabetes duration in stage 1–2 CKD, and HbA_{1c} in stage ≥ 3 CKD (Table 4). Furthermore, retinopathy displaced diabetes duration, not HbA_{1c}, in stage ≥ 3 CKD Alb⁺ and both diabetes duration and HbA_{1c} in eGFR $<75 \text{ ml min}^{-1} [1.73 \text{ m}]^{-2}$ Alb⁺, thus performing as an independent covariate for almost all CKD Alb⁺ phenotypes. Retinopathy was also independently related to

stage ≥ 3 CKD Alb⁻ and to eGFR < 75 ml min⁻¹ [1.73 m]⁻² Alb⁻. History of retinal laser treatment was also associated with both albuminuric and non-albuminuric CKD in the FinnDiane Study [31].

In our study, all CKD phenotypes were associated with hypertension and/or systolic BP, while no association was found with antihypertensive treatment or specific antihypertensive agents, such as RAS blockers. By indication, use of RAS blockers was more common in patients with stages 1–2 and ≥ 3 CKD than in individuals with no CKD, with no difference between stage ≥ 3 CKD Alb⁺ and Alb⁻ phenotypes. In particular, use of RAS blockers was more frequent among patients with eGFR < 75 ml min⁻¹ [1.73 m]⁻² Alb⁺ than in those with eGFR < 75 ml min⁻¹ [1.73 m]⁻² Alb⁻. Overall, these observations suggest that RAS blockage has no effect on non-albuminuric renal insufficiency. In the FinnDiane Study, non-albuminuric CKD was associated with the number of antihypertensive drugs, but not BP levels, antihypertensive treatment or specific renoprotecting antihypertensive agents [31]. In the Second Joslin Study [35], the risk of decline in renal function increased with systolic BP in both individuals with normo- as well as those with microalbuminuria and, at variance to both the FinnDiane Study [31] and our study, also increased with the number of prescribed nephroprotecting treatments. Increased renal vascular resistance in patients with hypertension can be related to systemic haemodynamics and the presence of subclinical atherosclerosis contributing to the decline in GFR in non-albuminuric CKD [39]. Furthermore, in a study of type 1 diabetes, renal vascular resistance, as reflected by raised systemic vascular stiffness and intraglomerular pressure, was lower in healthy nondiabetic individuals than in normoalbuminuric and microalbuminuric individuals with type 1 diabetes [40].

While uric acid has been proposed as a marker for decline in GFR in people with diabetes more than for progression of albuminuria [19, 37, 41], we could not find an independent association with any CKD phenotype. On the contrary plasma fibrinogen levels were independently related to the CKD Alb⁺ phenotypes (CKD stage 1–2, as well as both stage ≥ 3 Alb⁺ and individuals with eGFR < 75 ml min⁻¹ [1.73 m]⁻² Alb⁺), but not to Alb⁻ phenotypes. Fibrinogen is a marker of inflammatory vascular changes and endothelial dysfunction [42, 43] that, in the DCCT/EDIC, was related to albuminuria [44]. Finally, in our cohort, GGT was an independent covariate of stage 1–2 CKD and of stage ≥ 3 CKD Alb⁻ and Alb⁺ phenotypes, but not in individuals with eGFR < 75 ml min⁻¹ [1.73 m]⁻². GGT, a marker of cell oxidative stress, has been suggested as a predictor for low-grade albuminuria [45]. In a recent population-based study, the prevalence of albuminuria increased from the lowest to the highest GGT quartile, while eGFR correlated inversely across all GGT quartiles [45]. Moreover, it predicts microvascular

and/or renal complications in advanced CKD irrespective of UAE [46, 47].

Some limitations of our study must be considered. First, this is a single-centre study limited to individuals of white ethnicity. As in many other studies [31, 35] we have no direct measurements of GFR and its estimation was based on a single creatinine measurement. Hence, some individuals may have had temporary reductions in eGFR rather than persistent CKD. This could be especially the case in participants with eGFR 60–74 ml min⁻¹ [1.73 m]⁻². However, expanding the CKD definition to include this subset of individuals confirmed and strengthened the association with diabetic retinopathy and other covariates, supporting the overall correctness of our phenotype definition. The single assessment of eGFR does not allow us to describe changes over the time, which may be of relevance with respect to spontaneous or drug-mediated remission of microalbuminuria [16–18, 48], although the almost universal use of RAS blockers in type 1 diabetes with increased UAE tends to offset the latter. Furthermore, the cross-sectional nature of the study does not allow exploration of the predictive value of different CKD phenotypes for end-stage renal disease, cardiovascular events and all-cause mortality. Interestingly, in the FinnDiane Study [28], Alb⁻ CKD had the same risk of cardiovascular morbidity and all-cause mortality as patients with the Alb⁺ stage 1–2 CKD, but did not increase the risk of renal outcomes. Finally, our results are based on the MDRD formula for calculation of eGFR. However, the conclusions of our analysis did not change if the CKD-EPI equation was used (data not shown).

In conclusion, we found that non-albuminuric CKD phenotypes are present in a significant proportion of the type 1 diabetes population, in line with previous reports [35]. This challenges the paradigm of renal functional decline following the appearance of albuminuria. Rather, our findings support the hypothesis of two distinct pathways (Alb⁻ and Alb⁺) both potentially progressing to advanced kidney disease [38]. This hypothesis is corroborated by the appreciation that distinct covariates relate to the two CKD phenotypes.

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Data availability The datasets generated and/or analysed during the current study are available from the corresponding author on reasonable request.

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Author contribution GP, RM and SDP designed the study, drafted the manuscript and approved its final version. ER, MG, GD and DL contributed to the study design, and analysis and interpretation of data, and revised the article's intellectual content and approved the final version. ER, MG, LG, VSB, CB and AD acquired data, revised the article's intellectual content and approved the final version. GP and SDP are responsible for the integrity of this work as a whole.

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