Interleukin-6 and Cardiovascular and Kidney Outcomes in Patients With Type 2 Diabetes: New Insights From CANVAS

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Diabetes Care 2022;45:2644–2652 | https://doi.org/10.2337/dc22-0866

OBJECTIVE

The inflammatory cytokine interleukin-6 (IL-6) is associated with cardiovascular (CV) and kidney outcomes in various populations. However, data in patients with type 2 diabetes are limited. We assessed the association of IL-6 with CV and kidney outcomes in the Canagliflozin Cardiovascular Assessment Study (CANVAS) and determined the effect of canagliflozin on IL-6.

RESEARCH DESIGN AND METHODS

Patients with type 2 diabetes at high CV risk were randomly assigned to canagliflozin or placebo. Plasma IL-6 was measured at baseline and years 1, 3, and 6. The composite CV outcome was nonfatal myocardial infarction, nonfatal stroke, or CV death; the composite kidney outcome was sustained ≥40% estimated glomerular filtration rate decline, end-stage kidney disease, or kidney-related death. Multivariable-adjusted Cox proportional hazards regression was used to estimate the associations between IL-6 and the outcomes. The effect of canagliflozin on IL-6 over time was assessed with a repeated-measures mixed-effects model.

RESULTS

The geometric mean IL-6 at baseline, available in 3,503 (80.2%) participants, was 1.7 pg/mL. Each doubling of baseline IL-6 was associated with 14% (95% CI 4, 24) and 21% (95% CI 1, 45) increased risk of CV and kidney outcomes, respectively. Over 6 years, IL-6 increased by 5.8% (95% CI 3.4, 8.3) in the placebo group. Canagliflozin modestly attenuated the IL-6 increase (absolute percentage difference vs. placebo 4.4% [95% CI 1.3, 9.9; $P = 0.01$]). At year 1, each 25% lower level of IL-6 compared with baseline was associated with 7% (95% CI 1, 22) and 14% (95% CI 5, 22) lower risks for the CV and kidney outcome, respectively.

CONCLUSIONS

In patients with type 2 diabetes at high CV risk, baseline IL-6 and its 1-year change were associated with CV and kidney outcomes. The effect of IL-6–lowering therapy on CV, kidney, and safety outcomes remains to be tested.

PATHOPHYSIOLOGY/COMPLICATIONS

Systemic inflammation plays an important role in the development and progression of cardiovascular (CV) disease and chronic kidney disease (CKD), especially in patients with type 2 diabetes (1–8). The cytokine interleukin-6 (IL-6), which is commonly elevated in patients with type 2 diabetes or CV disease, regulates inflammatory

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Received 3 May 2022 and accepted 11 July 2022

Clinical trial reg. no. NCT01032629, clinicaltrials.gov

This article contains supplementary material online at https://doi.org/10.2337/figshare.20387259.

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responses by binding to the IL-6 receptor (9,10). The receptor can be found in its soluble form in the systemic circulation and also in a membrane-bound form on specific cells including leukocytes and kidney podocytes (10). Plasma levels of IL-6 are associated with adverse CV outcomes in patients at varying degrees of CV risk (11–14). Circulating IL-6 levels are also associated with kidney function decline in patients with CKD without type 2 diabetes (15). Only a few studies have included assessment of the associations between IL-6 and CKD in patients with type 2 diabetes. These studies were limited by their cross-sectional design (16), small sample size, short follow-up, and unclear end point definitions (17). Additional data in patients with type 2 diabetes on the association of IL-6 with CV and kidney outcomes would help in the development of a better understanding of the role of IL-6 in the pathophysiology of diabetes-related complications.

Sodium–glucose cotransporter 2 (SGLT2) inhibitors reduce the risk of CV disease and kidney failure in patients with type 2 diabetes at high CV risk and established CKD (18–20). Several studies have suggested that SGLT2 inhibitors also reduce markers of inflammation, indicating that they may have anti-inflammatory properties (21–23). However, the effect of SGLT2 inhibitors on IL-6 has not been investigated in a large cohort of patients with type 2 diabetes with global representation.

In this post hoc analysis of the Canagliflozin Cardiovascular Assessment Study (CANVAS), we studied the association of baseline plasma IL-6 with adverse CV and kidney outcomes. We also assessed the effect of the SGLT2 inhibitor canagliflozin on plasma IL-6 and whether changes in plasma IL-6 were associated with CV and kidney outcomes.

RESEARCH DESIGN AND METHODS

Participants and Study Design

The CANVAS Program consisted of two multicenter, double-blinded, placebo-controlled, randomized trials (CANVAS and CANVAS-Renal [CANVAS-R]) carried out to assess the effects of canagliflozin on CV, kidney, and safety outcomes in patients with type 2 diabetes who had a history of CV disease or multiple CV risk markers, as previously described (18). Blood and urine samples for exploratory biomarker research were stored during CANVAS (but not CANVAS-R). In this study, we therefore only included data from CANVAS. CANVAS included 4,330 subjects, who were followed for a median of 6.1 years. Eligible participants had type 2 diabetes with HbA_{1c} $\geq$7.0% (58 mmol/mol) and $\leq$10.5% (91 mmol/mol), estimated glomerular filtration rate (eGFR) $>$30 mL/min/1.73 m^2, and either a history of CV disease (58.9% of participants) or multiple CV risk factors. Participants with type 1 diabetes or a history of a severe hypoglycemic episode within 6 months prior to enrollment were excluded. Participants randomly received 100 mg or 300 mg canagliflozin or matching placebo in a 1:1:1 ratio. We combined the 100 mg and 300 mg arms in our main analyses. Study participants, treating teams, trial staff, and members of the outcome adjudication committee were blinded to treatment allocation. The conduct of CANVAS followed the Declaration of Helsinki principles (ClinicalTrials.gov identifier NCT01032629) (18). The protocol was approved by the independent ethics committee of each participating site. Written informed consent was obtained from all participants. All volunteers were also offered the opportunity to take part in the exploratory biomarker initiative, and those who agreed signed a separate optional informed consent form.

IL-6 Measurements

Plasma samples for exploratory biomarker measurements were collected at baseline and 1, 3, and 6 years after randomization. Collected samples were stored at $-80^{\circ}C$ for future analyses. Plasma IL-6 was measured with a V-PLEX Human IL-6 immunoassay kit (Meso Scale Discovery, Rockville, MD). Electrochemiluminescence was detected with use of the MESO QuickPlex SQ 120 platform. Each plate included a negative control, a calibration curve, and spiked pooled samples with predefined IL-6 concentrations. Samples underwent a maximum of three freeze-thawing cycles prior to measurement. Consistent with a prior study (24), we confirmed in our laboratory that plasma IL-6 levels were unchanged after at least five freeze-thawing cycles. Of 8,358 samples, 381 (4.6%) were randomly selected for duplicate assessment. For verification of measurement quality and consistency across plates, the values of the pooled sample in the different plates were entered into a Levey-Jennings plot. With this plot, the 1_{3s} and the 2_{2s} rules of the Westgard rules were applied to detect the plates that were out of control range (25). The mean intra- and interassay coefficients of variation were 5.1% and 8.3%, respectively. Measurements were carried out between June 2020 and July 2021 at the Department of Clinical Pharmacy and Pharmacology, University Medical Center Groningen.

Outcomes

The CV outcome was the prespecified primary outcome of CANVAS, composed of nonfatal myocardial infarction, nonfatal stroke, or death due to CV disease. Other prespecified outcomes were a composite of CV death or hospitalization for heart failure (HHF), HHF alone, and a composite kidney outcome defined as a sustained $\geq$40% reduction from baseline in eGFR, end-stage kidney disease (defined as eGFR $<$15 mL/min/1.73 m^2 or the need for chronic dialysis or kidney transplantation), or death due to kidney failure. All outcomes were adjudicated by a masked independent event adjudication committee.

Statistical Analysis

Descriptive analyses with categorical variables are reported as percentages. Continuous variables with normal distributions or skewed distributions are reported as mean (SD) or median (interquartile range), respectively. Urinary albumin-to-creatinine ratio (UACR) and IL-6 were logarithmic transformed before analyses to alleviate their skewness. Comparison of baseline IL-6 levels across patient groups defined by markers of kidney function and medical history was performed with use of a two-sided Student *t* test or one-way ANOVA, followed by Student *t* test with Bonferroni correction, as applicable.

Multivariable Cox proportional hazards regression models were used to assess the hazard ratios (HRs) for clinical outcomes associated with each doubling in baseline IL-6 concentration. In an additional analysis, baseline IL-6 was stratified into quartiles with the first quartile as a common reference for the other three quartiles to determine the HRs for clinical outcomes in each quartile of the IL-6

distribution. To explore the association of baseline IL-6 with each clinical outcome, we made four models with stepwise adjustment for risk factors of CV and kidney disease progression. Model 1 included adjustment for demographic variables (age, sex, and race) and treatment allocation (canagliflozin or placebo). Model 2 additionally included adjustment for the following medical history variables and clinical and laboratory measurements: history of CV disease, current smoking, BMI, systolic and diastolic blood pressure (BP), HbA_{1c}, and LDL cholesterol. Baseline eGFR was added in model 3, followed by the addition of log-transformed baseline UACR in model 4. The associations between IL-6 as a continuous variable and clinical outcomes were graphically presented in restricted cubic splines with the fully adjusted model (model 4). The association between baseline IL-6 and clinical outcomes with stratification by baseline eGFR and UACR categories was also assessed with the fully adjusted model. The Cox proportional hazards assumption was confirmed by visual inspection of the c-log-log plot.

The between-group difference in percentage changes in IL-6 levels over time was calculated with a repeated-measures mixed-effects model. We performed an additional analysis to assess the effect of the two canagliflozin doses (100 mg/day and 300 mg/day) on IL-6 separately. Treatment arm, study visit, and a term of interaction between treatment and visit were included as factors. Baseline IL-6 was included as a covariate. An unstructured covariance matrix was used in the model. The same models were also used to assess the effect of canagliflozin on IL-6 over time by baseline UACR and eGFR subgroups. The change in IL-6 from baseline to year 1 for each treatment arm was plotted in a histogram and Kernel density plot.

For the assessment of the association between change in IL-6 from baseline to year 1 and subsequent clinical outcomes, multivariable-adjusted Cox proportional hazards regression models were performed with use of a landmark approach. Only patients who had an IL-6 measurement at baseline and year 1 were included. Participants who experienced the outcome of interest during the first year of follow-up were excluded from analysis (Supplementary Fig. 1). A small portion ($<$0.5%) of the participants had missing covariate values at year 1; these missing values were imputed as means or medians, depending on their distribution. The change in IL-6 at year 1 was analyzed as a categorical or continuous variable. For the categorical analysis, the first quartile was selected as reference for the other three quartiles. For the continuous analysis, we expressed the change in IL-6 per 25% lower level since this value was close to the upper threshold of the first quartile of the 1-year change from baseline (24%). Model 1 was adjusted for baseline IL-6, demographic variables (age, sex, and race), and treatment allocation. Model 2 also included history of CV disease, current smoking, systolic and diastolic BP, BMI, HbA_{1c}, and LDL cholesterol, as well as 1-year change in systolic BP, BMI, HbA_{1c}, and LDL cholesterol from baseline. eGFR and its 1-year change from baseline were added in model 3, with the further addition of UACR and its 1-year change in model 4. The association between the change in IL-6 as a continuous variable and HR for each clinical outcome was illustrated in restricted cubic spline plots with the fully adjusted model (model 4). Model 4 was also used for assessment of the associations between IL-6 change and the outcomes by treatment allocation.

The associations between baseline IL-6 and the treatment effect of canagliflozin on the clinical outcomes were assessed with a Cox proportional hazards regression model with adjustment for randomized treatment, baseline IL-6 (as categorical or continuous variable), and the interaction term between treatment group and baseline IL-6.

STATA, version 17.1 (StataCorp, College Station, TX), was used for all data analyses, with $P < 0.05$ considered to indicate statistical significance. No correction for multiple testing was performed.

RESULTS

Study Population and IL-6 Levels

In total, 3,503 (80.9%) of the 4,330 CANVAS participants had available plasma IL-6 concentrations at baseline (Supplementary Fig. 1). Baseline characteristics were well balanced between treatment groups and were comparable with the overall CANVAS population (Supplementary Table 1). Mean age at baseline was 62.8 years, 33% were female, 59% had a history of CV disease, 13% had a history of heart failure (HF), and the majority of patients had UACR $<$30 mg/g (n = 2,525 [72%]) or eGFR $\geq$60 mL/min/1.73 m^2 (n = 3,007 [86%]). The geometric mean IL-6 concentration at baseline was 1.72 pg/mL (95% CI 1.68, 1.76). Higher IL-6 values were observed in participants with UACR $\geq$30 mg/g or eGFR $<$60 mL/min/1.73 m^2 compared with those with UACR $<$30 mg/g or eGFR $>$60 mL/min/1.73 m^2, respectively (both $P < 0.01$) (Supplementary Fig. 2). The highest IL-6 concentration was observed in those with both UACR $\geq$30 mg/g and eGFR $<$60 mL/min/1.73 m^2 (Supplementary Fig. 2). Participants with a history of HF or a history of CV disease had higher IL-6 values compared with those without a history of HF or CV disease, respectively (both $P < 0.01$) (Supplementary Fig. 2). There was no correlation between baseline IL-6 and other baseline participant characteristics, except for a moderate correlation between IL-6 and BMI (Pearson correlation coefficient = 0.21) (Supplementary Table 2).

Association Between Baseline IL-6 and CV and Kidney Outcomes

Participants were followed for a median duration of 6.1 years (interquartile range 5.9–6.4). During follow-up, the CV outcome, HHF or CV death, HHF outcome, and kidney outcome occurred in 548 (15.6%), 359 (10.2%), 128 (3.7%), and 136 (3.9%) participants, respectively. In a multivariable analysis with adjustment for treatment assignment and patient demographics, IL-6 levels were significantly associated with all outcomes (model 1) (Table 1). IL-6 remained independently associated with all outcomes after stepwise adjustment for medical history, vital signs, and laboratory values (model 2), eGFR (model 3), and UACR (model 4). In the fully adjusted model, each doubling of IL-6 at baseline was associated with a significant 14% (95% CI 4, 24; $P < 0.01$) increased risk for the CV outcome, 24% (95% CI 13, 37; $P < 0.01$) for combined CV death or HHF, 35% (95% CI 16, 57; $P < 0.01$) for the HHF outcome, and 21% (95% CI 1, 45; P = 0.04) for the kidney outcome (Fig. 1 and Table 1). The associations between IL-6 and individual components of the CV outcome are shown in Supplementary Table 3.

Table 1—Associations of baseline IL-6 with CV and kidney outcomes

Outcome	No. of events (%)	Model 1	*P*	Model 2	*P*	Model 3	*P*	Model 4	*P*
CV outcome									
Per doubling		1.21 (1.07, 1.36)	<0.01	1.16 (1.07, 1.26)	<0.01	1.16 (1.06, 1.26)	<0.01	1.14 (1.04, 1.24)	<0.01
Quartile 1	102 (11.6)	Reference		Reference		Reference		Reference	
Quartile 2	124 (14.2)	1.21 (0.93, 1.58)	0.15	1.16 (0.88, 1.51)	0.29	1.15 (0.88, 1.50)	0.30	1.12 (0.85, 1.46)	0.43
Quartile 3	151 (17.2)	1.53 (1.19, 1.98)	<0.01	1.36 (1.05, 1.76)	0.02	1.34 (1.04, 1.74)	0.03	1.30 (1.00, 1.68)	0.05
Quartile 4	171 (19.5)	1.75 (1.37, 2.25)	<0.01	1.51 (1.17, 1.95)	<0.01	1.48 (1.14, 1.91)	<0.01	1.38 (1.07, 1.79)	0.02
P for trend across quartiles			<0.01		<0.01		<0.01		<0.01
Composite of CV death or HHF									
Per doubling		1.35 (1.25, 1.46)	<0.01	1.28 (1.17, 1.40)	<0.01	1.28 (1.17, 1.41)	<0.01	1.24 (1.13, 1.37)	<0.01
Quartile 1	50 (5.8)	Reference		Reference		Reference		Reference	
Quartile 2	74 (8.6)	1.47 (1.03, 2.11)	0.04	1.35 (0.94, 1.93)	0.11	1.34 (0.94, 1.93)	0.11	1.23 (0.86, 1.77)	0.26
Quartile 3	103 (11.7)	2.02 (1.44, 2.83)	<0.01	1.63 (1.15, 2.30)	<0.01	1.60 (1.13, 2.27)	<0.01	1.47 (1.04, 2.09)	0.03
Quartile 4	132 (14.5)	2.61 (1.88, 3.63)	<0.01	2.01 (1.44, 2.82)	<0.01	1.93 (1.38, 2.71)	<0.01	1.71 (1.21, 2.40)	<0.01
P for trend across quartiles			<0.01		<0.01		<0.01		<0.01
HHF									
Per doubling		1.47 (1.31, 1.65)	<0.01	1.39 (1.21, 1.60)	<0.01	1.38 (1.19, 1.60)	<0.01	1.35 (1.16, 1.57)	<0.01
Quartile 1	15 (1.7)	Reference		Reference		Reference		Reference	
Quartile 2	20 (2.3)	1.20 (0.61, 2.39)	0.60	1.06 (0.53, 2.12)	0.87	1.06 (0.53, 2.12)	0.86	0.97 (0.48, 1.93)	0.92
Quartile 3	37 (4.2)	2.60 (1.43, 4.72)	<0.01	1.79 (0.97, 3.29)	0.06	1.75 (0.95, 3.22)	0.07	1.57 (0.85, 2.90)	0.15
Quartile 4	56 (6.4)	3.74 (2.11, 6.63)	<0.01	2.34 (1.30, 4.25)	<0.01	2.21 (1.22, 4.00)	<0.01	1.96 (1.08, 3.57)	0.03
P for trend across quartiles			<0.01		<0.01		<0.01		<0.01
Kidney outcome									
Per doubling		1.31 (1.13, 1.52)	<0.01	1.27 (1.08, 1.50)	<0.01	1.26 (1.07, 1.50)	<0.01	1.21 (1.01, 1.45)	0.04
Quartile 1	21 (2.4)	Reference		Reference		Reference		Reference	
Quartile 2	36 (4.1)	1.72 (0.99, 3.00)	0.05	1.69 (0.97, 2.94)	0.07	1.70 (0.97, 2.96)	0.06	1.31 (0.75, 2.30)	0.34
Quartile 3	39 (4.5)	2.14 (1.25, 3.66)	<0.01	1.93 (1.12, 3.34)	0.02	1.90 (1.10, 3.29)	0.02	1.53 (0.88, 2.66)	0.13
Quartile 4	40 (4.6)	2.21 (1.29, 3.80)	<0.01	1.94 (1.11, 3.37)	0.02	1.88 (1.09, 3.27)	0.03	1.47 (0.84, 2.56)	0.18
P for trend across quartiles			<0.01		0.02		0.03		0.17

Data are HR (95% CI) unless otherwise indicated. CV outcome is defined as a composite of nonfatal myocardial infarction, nonfatal stroke, or CV death. Kidney outcome is a composite of sustained ≥40% reduction from baseline eGFR, end-stage kidney disease, or kidney-related death. Models include adjustment for the following covariates: model 1, age, sex, race, and randomized treatment; model 2, covariates of model 1 + history of CV disease, current smoking, systolic and diastolic BP, HbA_{1c}, BMI, and LDL cholesterol; model 3, covariates of model 2 + eGFR; and model 4, covariates of model 3 + log-transformed UACR. Quartile 1, median baseline IL-6 0.9 pg/mL (range 0.0–1.2); quartile 2, median baseline IL-6 1.4 pg/mL (range 1.2–1.6); quartile 3, median baseline IL-6 1.9 pg/mL (range 1.6–2.3); and quartile 4, median baseline IL-6 3.2 pg/mL (range 2.3–29.1).

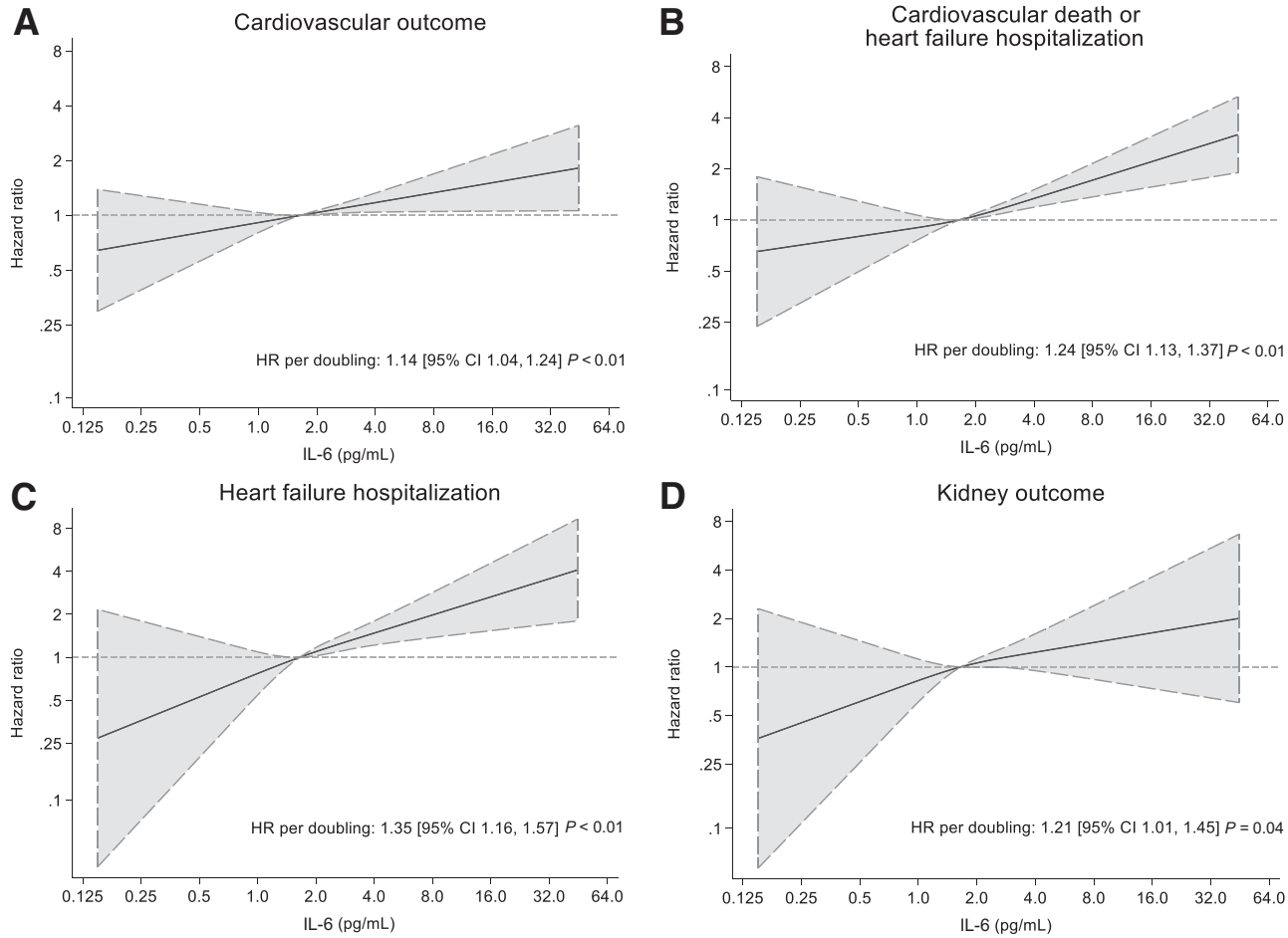

Figure 1—The associations between baseline plasma IL-6 and the risk for CV and kidney outcomes in the fully adjusted model. CV outcomes were as follows: a composite of nonfatal myocardial infarction, nonfatal stroke, and CV death (*A*); composite of CV death or HHF (*B*); or HHF (*C*). *D*: Kidney outcome was a composite of sustained ≥40% reduction from baseline eGFR, end-stage kidney disease, or kidney-related death. The restricted cubic splines were adjusted for the following covariates: age, sex, race, randomized treatment, history of CV disease, current smoking, systolic and diastolic BP, HbA_{1c}, BMI, LDL cholesterol, eGFR, and log-transformed UACR. The median IL-6 value, 1.6 pg/mL, was used as reference value in the model.

The associations of baseline IL-6 with the CV and kidney outcomes were generally consistent across eGFR and UACR subgroups. The exception was the CV outcome, for which there was some evidence that the association may be stronger for patients with baseline eGFR <60 mL/min/1.73 m^2 ($P_{interaction}$ = 0.02) (Supplementary Fig. 3). Evidence for this interaction was also present when baseline eGFR was categorized into three subgroups (<60, 60 to <90, and ≥90 mL/min/1.73 m^2; $P_{interaction}$ = 0.02), with the lowest eGFR subgroup showing the strongest association between baseline IL-6 and CV outcome (Fig. 2 and Supplementary Table 4).

There was no evidence that the effect of canagliflozin compared with placebo in reducing the relative risk of all assessed outcomes varied by baseline IL-6 levels (Supplementary Table 5).

Effect of Canagliflozin on IL-6 Over Time

IL-6 concentrations in the placebo arm increased by 3.3% (95% CI 0.5, 6.1), 6.0% (95% CI 1.8, 10.5), and 14.5% (95% CI 7.3, 22.3) compared with baseline at years 1, 3, and 6, respectively. Canagliflozin treatment significantly attenuated this increase compared with placebo (Fig. 3). Over the duration of the study, the least squares mean difference between the pooled canagliflozin and placebo groups was 4.4% (95% CI 1.3, 9.9; *P* = 0.01). This between-group difference was observed as early as year 1 and reached 9.1% (95% CI 1.8, 15.9) at year 6. When analyzed separately, each canagliflozin dose significantly attenuated IL-6 levels over time compared with placebo (least squares mean difference 4.2% [95% CI 1.1, 7.5] and 5.1% [95% CI 1.9, 8.2] for canagliflozin 100 mg and 300 mg, respectively; both $P < 0.01$), with no significant difference between the two canagliflozin doses (*P* = 0.67) (Supplementary Table 6 and Supplementary Fig. 4). There was no evidence that the effect of canagliflozin compared with placebo was different in patients with eGFR above or below 60 mL/min/1.73 m^2 and UACR above or below 30 mg/g ($P_{interaction}$ = 0.15 and 0.08 for UACR and eGFR categories, respectively) (Supplementary Fig. 5).

Associations Between First-Year Changes in IL-6 and Subsequent Outcomes

Of the participants with baseline IL-6 measurement, 80.7% (*n* = 2,826) had plasma IL-6 also measured at year 1 (Supplementary Fig. 1). Overall, one-quarter of the participants had a reduction in IL-6

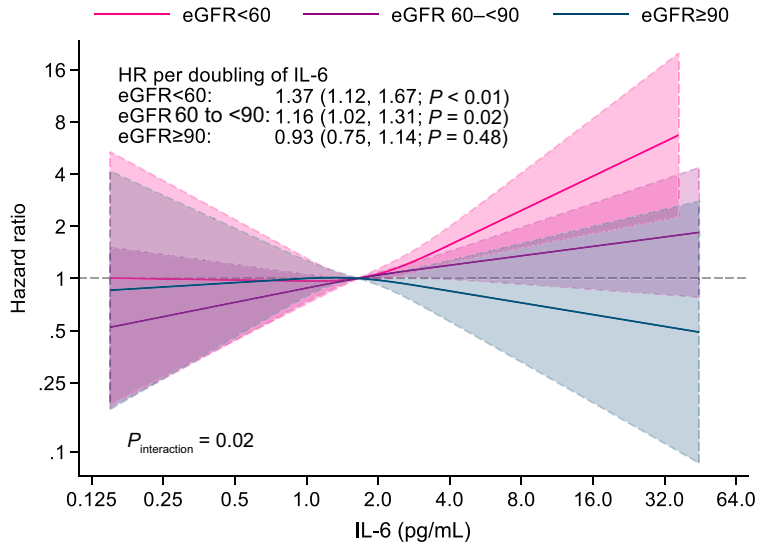

Figure 2—Associations between baseline plasma IL-6 and risk for CV outcome in the fully adjusted model according to eGFR subgroups. CV outcome is a composite of nonfatal myocardial infarction, nonfatal stroke, or CV death. The numbers of patients are 494, 1,902, and 1,105 in the subgroups with eGFR <60, 60 to <90, and ≥90 mL/min/1.73 m^2, respectively. The restricted cubic spline model included adjustment for the following covariates: age, sex, race, randomized treatment, history of CV disease, current smoking, systolic and diastolic BP, HbA_{1c}, BMI, LDL cholesterol, eGFR, and log-transformed UACR. The median IL-6 value, 1.6 pg/mL, was used as reference value in the model.

of ≥23.7% (Supplementary Fig. 6). The change in IL-6 at year 1 did not correlate with other CV risk markers or their change during the first year, except for baseline IL-6 value (Supplementary Table 7). A lower IL-6 at 1 year was associated with subsequent lower risk for all outcomes after accounting for baseline characteristics and their change during the first year (Supplementary Table 8). Specifically, each 25% lower level of IL-6 was independently associated with a 7% lower risk of the CV outcome (95% CI 1, 12; P = 0.01), an 11% lower risk of CV death or HHF (95% CI 4, 16; $P < 0.01$), a 15% lower risk of HHF (95% CI 6, 23; $P < 0.01$), and a 14% lower risk of the composite kidney outcome (95% CI 5, 22; $P < 0.01$) (Fig. 4 and Supplementary Table 8). No significant interaction was observed between the change in IL-6 at year 1 and treatment assignment for each outcome (Supplementary Fig. 7).

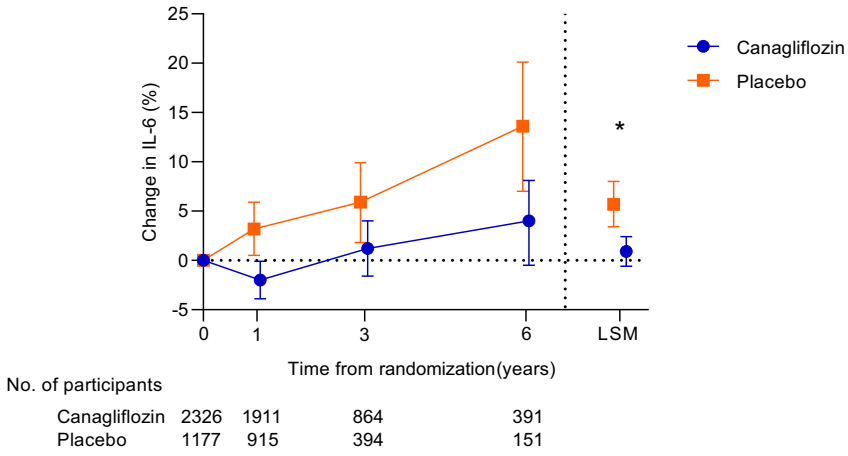

Figure 3—Percent change in plasma IL-6 levels over time in the canagliflozin and placebo groups. The least squares mean (LSM) change in IL-6 in the placebo group was 5.8% (95% CI 3.4, 8.3), and the LSM change in the canagliflozin group was 0.9% (95% CI −0.6, 2.5), resulting in a between-group difference of 4.4% (95% CI 1.3, 9.9; P = 0.01). *$P < 0.01$. The bars indicate 95% CI at each visit.

CONCLUSIONS

We observed that in individuals with type 2 diabetes at high CV risk enrolled in CANVAS, higher baseline IL-6 was independently associated with a higher risk of CV and kidney outcomes. Canagliflozin treatment significantly attenuated the increase in plasma IL-6 over time observed in the placebo group. A lower level of IL-6 at year 1 compared with baseline was associated with a lower risk of subsequent CV and kidney outcomes. These results support the prognostic role of IL-6 for CV and kidney outcomes in patients with type 2 diabetes.

Higher plasma IL-6 concentrations have been associated with adverse CV outcomes in previous observational studies in patients with established kidney or CV disease of whom only few had type 2 diabetes (11,12). The current study confirms the prognostic value of IL-6 for CV outcomes in a large global cohort of patients with type 2 diabetes. Subgroup analyses suggested a stronger association between baseline IL-6 and CV outcomes in participants with worse kidney function. We cannot rule out chance as a factor due to multiple subgroup analyses and because associations between IL-6 and diabetes complications were consistent by baseline eGFR for the other outcomes. However, it is interesting to note that in a recent study in patients with chronic coronary syndrome investigators reported similar results with a stronger association between IL-6 and CV outcomes among those with baseline eGFR <60 mL/min/1.73 m^2 (11).

Associations between baseline IL-6 and kidney outcomes were previously reported in patients with preserved kidney function without known CV disease (26) and in patients with CKD with and without type 2 diabetes (15). A small study of 70 patients with type 2 diabetes and CKD demonstrated an association between IL-6 and progression of kidney disease (17). We confirm these initial findings and demonstrate higher IL-6 levels in participants with more severe kidney disease—higher UACR or lower eGFR—and show that IL-6 levels are associated with the development and progression of CKD in a broad population of

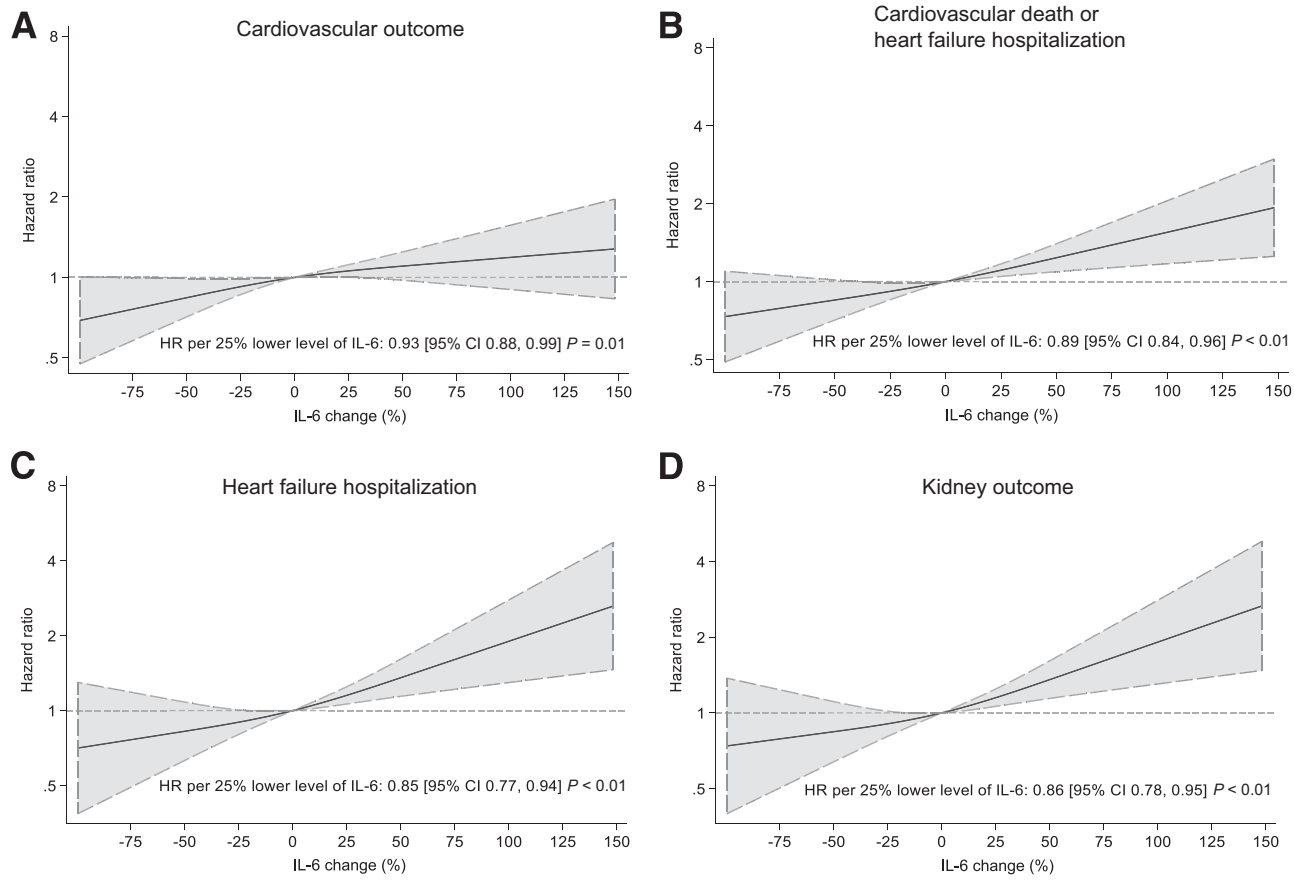

Figure 4—The associations between percent change in plasma IL-6 from baseline to year 1 and subsequent CV and kidney outcomes. CV outcomes were as follows: a composite of nonfatal myocardial infarction, nonfatal stroke, or CV death (*A*); composite of CV death or HHF (*B*); and HHF (*C*). *D*: Kidney outcome was a composite of sustained ≥40% reduction from baseline eGFR, end-stage kidney disease, or kidney-related death. The statistical model to describe restricted cubic splines included adjustment for the following covariates: IL-6 at baseline, age, sex, race, randomized treatment, history of CV disease, current smoking, systolic and diastolic BP, BMI, $HbA_{1c,}$ LDL cholesterol, eGFR, UACR, and change in systolic BP, BMI, HbA_{1c}, LDL cholesterol, eGFR, and UACR from baseline to year 1. The reference value for 1-year percent change in IL-6 is 0%.

patients with type 2 diabetes independent of other risk markers of CKD progression.

Experimental studies support a strong association between IL-6 and CV and kidney outcomes in patients with type 2 diabetes. High glucose concentrations have been shown to trigger IL-6 secretion from a variety of cells including cardiomyocytes, mesangial cells, and tubular epithelial cells (1,3,8,10,27). In the coronary and peripheral vasculature, IL-6 has been implicated in atherosclerotic plaque development and instability through its effect on leukocytes, smooth muscle cells, and endothelial function (28,29). In the kidney, IL-6 secretion activates a cascade of effects, including migration of macrophages, generation of reactive oxygen species, and production of collagen I, which can lead to tubulointerstitial fibrosis (10).

Canagliflozin modestly attenuated the increase in IL-6 observed in the placebo group, suggesting that SGLT2 inhibitors may exert some anti-inflammatory effects. Similar findings were previously reported in experimental and clinical studies. In a mouse model of type 2 diabetes, SGLT2 inhibitors attenuated the increase in plasma inflammatory markers, including IL-6 levels (30). In a double-blinded clinical trial, canagliflozin reduced IL-6 levels compared with glimepiride over a 2-year period despite similar levels of glycemic control during follow-up (21). This observation suggests that any anti-inflammatory properties of canagliflozin are not explained by improved glycemic control, although additional studies are required.

Among the combined canagliflozin and placebo groups, falls in IL-6 levels from baseline to year 1 were associated with lower subsequent risk for the CV and kidney outcomes. The associations remained significant after adjustment for baseline characteristics, including IL-6 levels and 1-year changes in other CV and kidney risk markers, and were consistent in the canagliflozin and placebo groups in separate analyses. Because of the observational nature of these analyses, these findings cannot be interpreted as indicating that lowering IL-6 per se reduces CV or kidney disease. This requires a prospective, randomized, placebo-controlled trial.

The important role of IL-6 in inflammation and its association with CV and kidney disease has practical therapeutic implications (31). Canakinumab, a monoclonal antibody targeting interleukin-1β, improved CV outcomes in patients with previous myocardial infarction and elevated hs-CRP, with the greatest clinical benefit observed among those with the most robust IL-6 response (32,33). Moreover, canakinumab was particularly effective among those with evidence of

impaired kidney function (34), although active treatment did not reduce the incidence of diabetes (35). Ziltivekimab, an IL-6 ligand monoclonal antibody, was developed with the intention to provide CV protection to patients with CV disease and increased inflammation. In patients with CKD with elevated inflammatory markers randomized in the recent phase II RESCUE trial, ziltivekimab reduced hs-CRP levels by >90% and had directionally beneficial effects on several other inflammatory biomarkers, including fibrinogen, serum amyloid A, and secretory phospholipases A2 (36). In the ongoing ZEUS trial (ClinicalTrials.gov identifier NCT05021835) investigators are testing the effect of ziltivekimab on CV, kidney, and safety outcomes compared with placebo in people with CV disease, CKD, and hs-CRP ≥2 mg/L (6,37). The results of this trial will address directly whether IL-6 lowering can prevent CV and kidney events in this population.

This study has limitations. First, because our study was a post hoc analysis, we cannot exclude chance findings. Furthermore, although our statistical analyses were adjusted for various confounders, residual confounding cannot be excluded. Second, most patients originally randomized into CANVAS had plasma IL-6 measurements available at the baseline visit, but fewer patients had blood samples available for IL-6 measurements at subsequent visits. This reduced the study's statistical power to assess the effect of canagliflozin on IL-6 over time. Third, IL-6 physiologically acts through endocrine, paracrine, and autocrine pathways (10). We measured IL-6 in plasma but could not determine IL-6 in target tissues, which would have resulted in more biologically relevant information. Finally, only a small portion of the participants had kidney disease at baseline. Further study is needed to assess the association of canagliflozin, plasma IL-6, and CV and kidney outcomes in patients with type 2 diabetes and impaired kidney function.

In conclusion, plasma IL-6 and its changes over a 1-year period have prognostic value for CV and kidney disease progression in patients with type 2 diabetes at high CV risk. Canagliflozin mitigated the increase of IL-6 over time. These results support attempts to evaluate the CV-protective and kidney-protective properties of anti-inflammatory and specifically IL-6–lowering therapy.

Acknowledgments. The authors thank all patients and CANVAS investigators for their tremendous support in the trial.

Funding. T.S. is supported by the Biomarker Enterprise to Attack DKD (BEAt-DKD) project. The BEAt-DKD project has received funding from the Innovative Medicines Initiative 2 Joint Undertaking (IMI2 JU) under grant agreement no. 115974. IMI2 JU receives support from the European Union's Horizon 2020 research and innovation program and the European Federation of Pharmaceutical Industries and Associations. K.R.T. is supported by National Institutes of Health research grants R01MD014712, U2CDK114886, UL1TR002319, U54DK083912, U01DK100846, OT2HL161847, and UM1AI109568 and Centers for Disease Control and Prevention contract 75D301-21-P-12254.

Duality of Interest. CANVAS and biomarker measurements were funded by Janssen Research & Development. CANVAS was conducted as a collaboration between the funder, an academic steering committee, and an academic research organization, The George Institute for Global Health. The funder was involved in the study design, data collection, data analysis, data interpretation, and writing of the manuscript. B.L.N. has received fees for travel support, advisory boards, and steering committee roles from AstraZeneca, Bayer, Boehringer Ingelheim, and Janssen, with all honoraria paid to his institution. B.N. has received grant funding, travel reimbursement, and consultancy fees paid to his institution related to his work on CANVAS and Canagliflozin and Renal Events in Diabetes with Established Nephropathy Clinical Evaluation (CREDENCE). C.A. is an employee of The George Institute for Global Health. She has received honoraria from Amgen and AstraZeneca (paid to her institution). V.P. has received fees for advisory boards, steering committee roles, or scientific presentations from AbbVie, Amgen, Astellas, AstraZeneca, Bayer, Baxter, Bristol Myers Squibb, Boehringer Ingelheim, Chinook, Dimerix, Durect, Eli Lilly, Gilead, GSK, Janssen, Merck, Metavant, Mitsubishi Tanabe Pharma, Mundipharma, Novartis, Novo Nordisk, Pfizer, PharmaLink, Relypsa, Retrophin, Sanofi, Servier, Vifor Pharma, Vitae, UpToDate, and Tricida. P.M.R. has received research grant support from and served as a consultant to Bristol Myers Squibb and Pfizer for work unrelated to this project, in addition to unrelated consulting for Corvidia Therapeutics, Novartis, Flame, AGEPHA Pharma, Inflazome, AstraZeneca, Janssen, CiVi Biopharma, SOCAR, Novo Nordisk, Uptton, OMEICOS Therapeutics, and Boehringer Ingelheim. K.R.T. reports other support from Eli Lilly and Gilead; personal fees and other support from Boehringer Ingelheim and AstraZeneca; grants, personal fees, and other support from Bayer AG and Novo Nordisk; grants and other support from Goldfinch Bio; and grants from Travere Therapeutics outside the submitted work. M.K.H. is an employee of Janssen Research & Development, LLC. H.J.L.H. has served as a consultant for AstraZeneca, Bayer, Boehringer Ingelheim, Chinook, CSL Behring, Dimerix, Eli Lilly–Gilead, Goldfinch Bio, Janssen, Merck, Novo Nordisk, and Travere Therapeutics; and has received grant support from AstraZeneca, Boehringer Ingelheim, Novo Nordisk, and Janssen. No other potential conflicts of interest relevant to this article were reported.

Author Contributions. A.K. measured all samples. A.K., T.S., and H.J.L.H. analyzed the data. A.K., M.S., and H.J.L.H. wrote the first draft of the manuscript. B.L.N. and V.P. were involved in data collection. All authors contributed to data interpretation, provided input into subsequent drafts, and approved the final version for submission. All authors reviewed and approved the manuscript. H.J.L.H. made the decision to publish. H.J.L.H. is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Prior Presentation. Parts of this study were presented in abstract form at the European Diabetic Nephropathy Study Group 34th Annual Meeting, 27–28 May 2022, Salzburg, Austria, and at the 82nd Scientific Sessions of the American Diabetes Association, 3–7 June 2022, New Orleans, LA, and will be presented at the 58th Annual Meeting of the European Association for the Study of Diabetes, 19–23 September 2022, Stockholm, Sweden.

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