Supplemental Data

Dickkopf-3 (DKK3) in urine identifies patients with short-term risk of eGFR loss

Table of contents

Supplemental Table 1

Baseline characteristics of the I LIKE HOMe participants.

	All participants (N=481)
Age (years)	48.3±8.3
Gender (% male)	32.7
Body mass index (kg/m^2)	26.2±4.6
Systolic blood pressure (mmHg)	136±18
Diastolic blood pressure (mmHg)	88±11
Smoking (%)	26.5
Diabetes (%)	4.8
Plasma creatinine (mg/dL)	0.8±0.1
eGFR (ml/min/1.73 m^2)	93±13
Urine DKK3/creatinine (pg/mg)	33 (126)
Log urine DKK3/creatinine (pg/mg)	3.8±1.7
Albuminuria (mg/g)	4.2 (3.0)
Log albuminuria (mg/g)	1.6±0.6
Serum C-reactive protein (mg/L)	1.5 (2.2)
Log serum C-reactive protein (mg/L)	0.4±1.0

eGFR, estimated glomerular filtration rate

Supplemental Table 2

Causes of chronic kidney diseases (CKD) in participants of the CARE FOR HOMe study.

Cause of CKD	**N**
CKD after acute kidney injury	8
Congenital kidney diseases	9
Cardiorenal syndrome	13
Cystic kidney diseases	23
Tubulointerstitial diseases	35
Glomerular diseases	79
Diabetic nephropathy	80
Diabetic/hypertensive kidney injury	104
Hypertensive/vascular diseases	202
Other	22

Supplemental Table 3

Baseline characteristics of the CARE FOR HOMe participants according to categories of urinary DKK3/creatinine concentrations.

	All participants (N=575)	**DKK3/creatinine ≤200 pg/mg (N=167)**	**DKK3/creatinine 201-1,000 pg/mg (N=213)**	**DKK3/creatinine 1,001-4,000 pg/mg (N=126)**	**DKK3/creatinine >4,000 pg/mg (N=69)**	***P***
Age (years)	65.5±12.1	64.2±12.3	65.6±11.4	65.9±12.7	66.2±14.0	0.583
Gender (% male)	41.3	43.0	49.8	30.3	30.8	0.002
Body mass index (kg/m^2)	30.4±5.5	31.2±6.0	30.1±5.2	30.6±5.6	28.9±4.9	0.035
Systolic blood pressure (mmHg)	152±24	150±25	152±23	152±22	159±26	0.094
Diastolic blood pressure (mmHg)	85±13	85±12	85±12	86±13	86±14	0.614
Smoking (%)	10.5	11.4	7.5	13.4	12.3	0.330
ACE inhibitor (%)	35.6	33.4	37.6	40.8	34.7	0.032
AT1 receptor blocker (%)	50.5	56.6	49.1	49.2	53.1	0.007
Diabetes (%)	39.2	33.5	42.8	38.7	43.1	0.304
Plasma creatinine (mg/dL)	1.6±0.6	1.4±0.4	1.4±0.5	1.7±0.5	2.3±0.8	<0.001
eGFR (ml/min/1.73 m^2)	48.6±18.3	53.6±15.8	52.4±17.8	44.4±16.7	32.1±17.1	<0.001
Urine DKK3/creatinine (pg/mg)	431 (1,388)	95 (87)	388 (339)	1,721 (1,207)	7,738 (10,422)	<0.001
Log urine DKK3/creatinine (pg/mg)	6.1±1.7	4.3±0.8	6.0±0,5	7.5±0.4	9.2±0.7	<0.001
Albuminuria (mg/g)	322 (1,930)	115 (390)	272 (1,140)	1,089 (3,620)	3,702 (17,650)	<0.001
Log albuminuria (mg/g)	3.8±1.9	3.1±1.6	3.6±1.8	4.4±1.8	5.7±1.9	<0.001
Serum C-reactive protein (mg/L)	2.7 (4.0)	2.4 (4.3)	2.7 (3.7)	2.9 (6.4)	2.5 (2.9)	0.476
Log serum C-reactive protein (mg/L)	0.9±1.1	0.9±1.0	0.9±1.0	0.9±1.0	0.9±1.0	0.476

eGFR, estimated glomerular filtration rate

Supplemental Table 4

Multivariate adjusted means of DKK3/creatinine in urine in the general population (I LIKE HOMe study) and in CKD patients (CARE FOR HOMe study). Adjusted for age, gender, estimated glomerular filtration rate, presence of diabetes, systolic blood pressure, smoking, body mass index and albuminuria.

Urinary DKK3 (pg/mg)				
General population I LIKE HOMe study (N=481)		**CKD patients CARE FOR HOMe study (N=575)**		
Mean	**SEM**	**Mean**	**SEM**	**P**
336.9	375.8	2260.6	343.8	<0.0001

Supplemental Table 5

Bivariate correlations between urinary DKK3/creatinine and clinical parameters in subjects form the I LIKE HOMe study.

Parameter	**Correlation coefficient (ρ)**	***P***
Age (years)	0.013	0.790
Gender	0.068	0.152
Estimated glomerular filtration rate (ml/min/1.73m²)	-0.200	0.677
Log albuminuria (mg/g creatinine)	0.111	0.051
Body mass index (kg/m^2)	-0.003	0.954
Diabetes	0.045	0.341
Systolic blood pressure (mmHg)	0.012	0.799
Smoking	-0.073	0.157
Log serum C-reactive protein (mg/l)	-0.038	0.432

ρ, correlation coefficient

Supplemental Table 6

Bivariate correlations between urinary DKK3/creatinine and clinical parameters in subjects with CKD form the CARE FOR HOMe study.

Parameter	**Correlation coefficient (ρ)**	***P***
Age (years)	-0.016	0.391
Gender	-0.046	0.016
Estimated glomerular filtration rate (ml/min/1.73m²)	-0.247	<0.001
Log albuminuria (mg/g creatinine)	-0.258	<0.001
Body mass index (kg/m^2)	-0.041	0.033
Diabetes	0.014	0.478
Systolic blood pressure (mmHg)	0.021	0.272
Smoking	-0.016	0.389
Log serum C-reactive protein (mg/l)	-0.018	0.341

ρ, correlation coefficient

Supplemental Table 7

Estimated marginal means of annual change of estimated glomerular filtration rate (eGFR) according to tertiles (1: ≤219 pg/mg; 2: 220-864 pg/mg; 3: >864 pg/mg) or quartiles (1: ≤155 pg/mg; 2: 156-405 pg/mg; 3: 406-1,329 pg/mg; 4: >1,329 pg/mg) of urinary DKK3/creatinine concentrations in participants of the CARE FOR HOMe cohort.

Tertile of urinary DKK3/creatinine	Annual change of eGFR (%)	95 % CI	*P*	Quartile of urinary DKK3/creatinine	Annual change of eGFR (%)	95 % CI	*P*
1	0.8	-1.0 – 2.6	Ref.	1	1.1	-1.0 – 3.1	Ref.
2	-2.0	-3.4 – -0.5	0.006	2	-1.3	-3.2 – 0.5	0.045
3	-3.5	-5.3 – -1.6	<0.001	3	-1.2	-3.0 – 0.6	0.067
				4	-5.0	-7.1 – -2,9	<0.001

Adjusted for age, gender, body mass index, systolic blood pressure, diabetes, smoking, eGFR, and log albuminuria

Supplemental Table 8

Estimated marginal means of annual change of estimated glomerular filtration rate (eGFR) according to categories of DKK3/creatinine concentrations in urine in participants of the CARE FOR HOMe cohort according to therapy with ACE inhibitors or AT1 receptor blockers.

Urinary DKK3/creatinine (pg/mg)	**Annual change of eGFR (%)**	**95 % CI**	***P***	**Annual change of eGFR (%)**	**95 % CI**	***P***
	No ACE inhibitor therapy			**ACE inhibitor therapy**		
≤ 200	-0.3	-2.7–2.0	Reference	2.2	-1.2–5.6	Reference
201 – 1,000	-2.0	-4.1–0.1	0.192	-2.0	-5.2–1.3	0.281
1,001 – 4,000	-3.0	-5.8– -0.2	0.099	-3.3	-7.1–0.4	0.019
> 4,000	-8.5	-12.5– -4.4	<0.001	-11.6	-17.4– -5.8	0.008
	No AT1 receptor blocker therapy			**AT1 receptor blocker therapy**		
≤ 200	1.3	-1.5–4.0	Reference	0.8	-1.9–3.6	Reference
201 – 1,000	-2.0	-4.4–0.5	0.030	-1.4	-4.0–1.2	0.114
1,001 – 4,000	-2.2	-5.3–0.9	0.053	-3.6	-6.9– -0.4	0.013
> 4,000	-8.4	-13.1– -3.8	<0.001	-11.5	-16.1 – -6.8	<0.001

Adjusted for age, gender, body mass index, systolic blood pressure, diabetes, smoking, eGFR, and log albuminuria

Supplemental Table 9

C-statistics derived from multivariate adjusted logistic regression models of the eight variable kidney failure risk equation and urinary DKK3 in addition to the eight variable kidney failure risk equation in predicting >0 %, >5 % and >10 % decrease of eGFR in participants of the CARE FOR HOMe study.

	AUC Kidney failure risk equation	**95 % CI**	**AUC Kidney failure risk equation +DKK3**	**95 % CI**	***P***
>0 % decrease of eGFR	0.56	0.54-0.58	0.60	0.58-0.63	0.098
>2 % decrease of eGFR	0.55	0.52-0.58	0.59	0.56-0.62	0.015
>5 % decrease of eGFR	0.56	0.54-0.59	0.59	0.56-0.61	0.015
>10 % decrease of eGFR	0.61	0.59-0.64	0.63	0.61-0.66	0.023

Supplemental Table 10

Estimated marginal means of annual change of estimated glomerular filtration rate (eGFR) according to suPAR plasma concentrations in participants of the CARE FOR HOMe cohort.

Plasma suPAR (pg/mL)	**Annual change of eGFR (%)**	**95 % CI**	***P***
Crude			
≤ 3,040	-2.78	-5.69–0.13	Ref.
> 3,040	-2.78	-5.28–-0.27	0.998
Model 1			
≤ 3,040	-3.70	-7.67–0.27	Ref.
> 3,040	-3.25	-6.67–0.16	0.817
Model 2			
≤ 3,040	-2.58	-6.67–1.51	Ref.
> 3,040	-3.70	-7.12–-0.28	0.587
Model 3			
≤ 3,040	-2.51	-6.50–1.48	Ref.
> 3,040	-2.38	-5.77–1.01	0.949

Model 1: adjusted for age, gender, body mass index, systolic blood pressure, diabetes and smoking

Model 2: Model 1 + adjusted for eGFR

Model 3: Model 2 + adjusted for log albuminuria

Supplemental Table 11

Integrated discrimination improvement (IDI) and Net reclassification improvement by suPAR compared to urinary DKK3/creatinine in predicting >0 %, >5 % and >10 % decrease of estimated glomerular filtration rate (eGFR) in participants of the CARE FOR HOMe study.

	IDI	**95 % CI**	***P***	**NRI**	**95 % CI**	***P***
>0 % decrease of eGFR	0.012	0.009 – 0.015	0.227	0.033	0.028 – 0.095	0.301
>2 % decrease of eGFR	0.003	0.001 – 0.006	0.140	0.019	0.052 – 0.089	0.598
>5 % decrease of eGFR	0.006	0,001 – 0.030	0.624	0.067	0.051 – 0.185	0.257

Supplemental Table 12

Baseline characteristics of participants of the kidney biopsies study divided into categories of DKK3/creatinine.

	All participants (N=76)	**DKK3/creatinine ≤200 pg/mg (N=18)**	**DKK3/creatinine 1,001-4,000 pg/mg (N=20)**	**DKK3/creatinine >4,000 pg/mg (N=38)**	***P***
Age (years)	54.0±18.5	51.2±19.7	47.8±18.8	61.0±14.5	0.070
Gender (% male)	71.1	72.2	70.0	71.1	0.989
Diabetes (%)	11.8	11.1	5.0	15.8	0.479
Plasma creatinine (mg/dL)	2.9±1.8	1.8±1.5	2.3±1.5	3.6±1.7	<0.001
eGFR (ml/min/1.73 m^2)	38±31	58±34	48±38	23±16	<0.001
Urine DKK3/creatinine (pg/mg)	3,778 (11,273)	642 (364)	2,806 (1,744)	13,234 (15,246)	<0.001
Log urine DKK3/creatinine (pg/mg)	8.3±1.4	6.3±0.4	7.8±0.4	9.5±0.9	<0.001
Albuminuria (mg/g)	2,725 (4,481)	2,826 (4,838)	1,521 (2,044)	4,079 (4,854)	0.072
Log albuminuria (mg/g)	7.4±1.8	7.0±2.0	6.8±1.8	7.9±1.4	0.085
Tubulointerstitial fibrosis (%)	50.7±20.6	30.9±15.8	47.1±18.0	62.1±15.8	<0.001

eGFR, estimated glomerular filtration rate

Supplemental Table 13

Estimated marginal means of tubulointerstitial fibrosis according to urinary DKK3/creatinine.

Urinary DKK3/creatinine (pg/mg)	Tubulo-interstitial fibrosis (%)	95 % CI	*P*
Crude			
≤ 1,000	20.8	14.6–27.0	Ref.
1,001-4,000	35.0	29.2–40.9	0.001
> 4,000	46.1	41.8–50.3	<0.001
Model 1			
≤ 1,000	19.9	13.5–26.2	Ref.
1,001-4,000	34.2	28.1–40.2	0.001
> 4,000	45.2	40.6–49.7	<0.001
Model 2			
≤ 1,000	23.4	17.2–29.6	Ref.
1,001-4,000	35.7	30.0–41.4	0.002
> 4,000	42.9	38.4–47.3	<0.001
Model 3			
≤ 1,000	22.2	15.2–29-3	Ref.
1,001-4,000	36.2	30.1–42.2	0.002
> 4,000	44.3	39.1–49.5	<0.001
Model 4			
≤ 1,000	20.6	13.5–27.7	Ref.
1,001-4,000	35.2	29.4–41.0	0.001
> 4,000	45.7	40.9–50.5	<0.001

Model 1: adjusted for age, gender

Model 2: Model 1 + adjusted for estimated glomerular filtration rate

Model 3: Model 2 + adjusted for albuminuria

Model 4: Model 3 + adjusted for cause of kidney disease

Supplemental Table 14

Baseline characteristics of participants of the STOP-IgAN trial divided into two groups at median of DKK3/creatinine levels in urine.

	All participants (N=96)	**DKK3/creatinine ≤779 pg/mg (N=48)**	**DKK3/creatinine >779 pg/mg (N=48)**	***P***
Age (years)	43.6±12.7	41.2±13.7	46.0±11.3	0.071
Gender (% male)	22.8	11.8	34.0	0.009
Body mass index (kg/m^2)	27.4±4.3	27.9±4.3	27.0±4.2	0.545
Systolic blood pressure (mmHg)	131±14	132±17	130±11	0.394
Smoking (%)	41.6	45.1	38.0	0.546
ACE inhibitor therapy (%)	75.2	82.3	68.0	0.111
Angiotensin-receptor-blocker therapy (%)	41.6	47.1	36.0	0.314
Aldosterone antagonist therapy (%)	1.0	0	2.0	0.495
Statin therapy (%)	57.4	60.8	54.0	0.549
Plasma creatinine (mg/dL)	1.5±0.5	1.4±0.5	1.6±0.6	0.074
Estimated glomerular filtration rate (ml/min/1.73 m^2)	63±27	70±27	56±26	<0.001
Albuminuria (mg/g creatinine)	1,214 (1161)	930 (875)	1,424 (1291)	0.010
Log albuminuria (mg/g creatinine)	7.0±0.7	6.8±0.7	7.3±0.6	0.010
Urine DKK3/creatinine (pg/mg)	781 (1,233)	389 (398)	1,601 (1,236)	<0.001
Log urine DKK3/creatinine (pg/mg)	6.4±1.3	5.5±1.1	7.4±0.6	<0.001
Total cholesterol (mg/dL)	216±49	208±55	224±42	0.121

Supplemental Table 15

Baseline characteristics of participants of the STOP-IgAN trial divided into two groups at DKK3/creatinine in urine of 1,000 pg/mg.

	All participants (N=96)	**DKK3/creatinine ≤1,000 pg/mg (N=61)**	**DKK3/creatinine >1,000 pg/mg (N=35)**	***P***
Age (years)	43.6±12.7	41.9±13.8	46.6±10.2	0.023
Gender (% male)	22.8	17.2	32.4	0.090
Body mass index (kg/m^2)	27.4±4.3	27.4±4.3	27.4±4.3	0.910
Systolic blood pressure (mmHg)	131±14	131±16	130±11	0.090
Smoking (%)	41.6	40.6	43.2	0.836
ACE inhibitor therapy (%)	75.2	75.0	75.7	0.999
Angiotensin-receptor-blocker therapy (%)	41.6	50.0	27.0	0.036
Aldosterone antagonist therapy (%)	1.0	1.6	0	0.999
Statin therapy (%)	57.4	56.3	59.5	0.836
Plasma creatinine (mg/dL)	1.5±0.5	1.4±0.5	1.6±0.6	0.712
Estimated glomerular filtration rate (ml/min/1.73 m^2)	63.3±27.5	68.4±27.4	54.3±25.7	0.389
Albuminuria (mg/g creatinine)	1,408±1,058	1,125±707	1,900±1,361	0.086
Log albuminuria (mg/g creatinine)	7.0±0.7	6.8±0.7	7.4±0.7	0.571
Urine DKK3/creatinine (pg/mg)	1,162±1,254	465±295	2,376±1,362	<0.001
Log urine DKK3/creatinine (pg/mg)	6.4±1.3	5.8±1.1	7.6±0.5	0.001
Total cholesterol (mg/dL)	216±49	209±52	227±41	0.180

Supplemental Table 16

Estimated marginal means of change of estimated glomerular filtration rate (eGFR) during the run-in phase in participants of the STOP-IgAN trial according to DKK3/creatinine in urine divided in two groups at median.

Urinary DKK3/creatinine (pg/mg)	Change of eGFR (%)	95 % CI	*P*
Crude			
≤779 pg/mg	0.0	-4.6 – 4.7	Ref.
>779 pg/mg	-19.5	-23.9 – -15.0	<0.001
Model 1			
≤779 pg/mg	-0.7	-6.2 – 4.8	Ref.
>779 pg/mg	-19.7	-24.4 – -15.0	<0.001
Model 2			
≤779 pg/mg	-0.9	-6.5 – 4.6	Ref.
>779 pg/mg	-19.6	-24.3 – -14.9	<0.001
Model 3			
≤779 pg/mg	-1.3	-7.0 – 4.4	Ref.
>779 pg/mg	-19.1	-24.2 – -14.0	<0.001

Model 1: adjusted for age, gender, body mass index, systolic blood pressure and smoking

Model 2: Model 1 + adjusted for eGFR

Model 3: Model 2 + adjusted for log albuminuria

Supplemental Table 17

Estimated marginal means of change of estimated glomerular filtration rate (eGFR) during the run-in phase in participants of the STOP-IgAN trial according to DKK3/creatinine in urine divided in two groups at 1,000 pg/mg.

Urinary DKK3/creatinine (pg/mg)	Change of eGFR (%)	95 % CI	*P*
Crude			
≤1,000 pg/mg	-3.4	-7.6–0.9	Ref.
>1,000 pg/mg	-21.4	-26.8– -15.9	<0.001
Model 1			
≤1,000 pg/mg	-4.6	-9.5–0.3	Ref.
>1,000 pg/mg	-21.4	-26.9– -15.8	<0.001
Model 2			
≤1,000 pg/mg	-4.8	-9.7–0.2	Ref.
>1,000 pg/mg	-21.2	-26.8– -15.5	<0.001
Model 3			
≤1,000 pg/mg	-4.5	-9.5–0.6	Ref.
>1,000 pg/mg	-21.8	-28.0– -15.7	<0.001

Model 1: adjusted for age, gender, body mass index, systolic blood pressure and smoking

Model 2: Model 1 + adjusted for eGFR

Model 3: Model 2 + adjusted for log albuminuria

Supplemental Table 18

Integrated discrimination improvement (IDI), Net reclassification improvement (NRI) and c-statistics by urinary DKK3/creatinine compared to age, sex, body mass index, systolic blood pressure, estimated glomerular filtration rate (eGFR) and albuminuria (ACR) (model 1) in predicting >0 % and >5 % decrease of eGFR in participants of the STOP-IgAN trial during the run-in phase.

	IDI	95 % CI	P	NRI	95 % CI	P
>0 % decrease of eGFR	0.135	0.070 – 0.200	<0.001	0.450	0.134 – 0.766	0.005
>5 % decrease of eGFR	0.165	0.087 – 0.243	<0.001	0.294	0.072 – 0.516	0.009

	AUC Model 1	95 % CI	AUC Model 1 + DKK3/creatinine	95 % CI	P
>0 % decrease of eGFR	0.62	0.48-0.75	0.79	0.67-0.90	0.016
>5 % decrease of eGFR	0.68	0.57-0.80	0.81	0.71-0.91	0.005

Supplemental Table 19

Association between change of DKK3/creatinine in urine and change of estimated glomerular filtration rate (eGFR in %) during the treatment phase in participants of the STOP-IgAN trial.

	Regression coefficient B	95 % CI	*P*
Crude			
Delta DKK3/creatinine (pg/mg)	-0.008	-0.011 – -0.005	<0.001
Model 1			
Delta DKK3/creatinine (pg/mg)	-0.008	-0.011 – -0.005	<0.001
Treatment	-2.553	-15.457 – 10.352	0.698
Model 2			
Delta DKK3/creatinine (pg/mg)	-0.013	-0.021 – -0.004	0.003
Treatment	-1.539	-14.379 – 11.300	0.814
Interaction term	0.006	-0.003 – 0.015	0.219
Model 3			
Delta DKK3/creatinine (pg/mg)	-0.013	-0.022 – -0.004	0.004
Treatment	-1.254	-14.364 – 11.855	0.851
Interaction term	0.006	-0.003 – 0.016	0.204
Model 4			
Delta DKK3/creatinine (pg/mg)	-0.013	-0.022 – -0.004	0.004
Treatment	-1.584	-14.674 – 11.505	0.812
Interaction term	0.006	-0.004 – 0.015	0.224
eGFR (ml/min/1.73m^2)	0.103	-0.190 – 0.395	0.492
Model 5			
Delta DKK3/creatinine (pg/mg)	-0.013	-0.023 – -0.002	0.016
Treatment	0.224	-13.246 – 13.694	0.974
Interaction term	0.006	-0.005 – 0.017	0.278
eGFR (ml/min/1.73m^2)	0.055	-0.253 – 0.364	0.726
Log albuminuria (mg/g creatinine)	-6.939	-18.142 – 4.264	0.225

Model 1: adjusted for treatment (supportive care alone or supportive care plus immunosuppressive therapy)

Model 2: Model 1 + interaction term of delta DKK3/creatinine and treatment

Model 3: Model 2 + age, gender, body mass index, systolic blood pressure and smoking

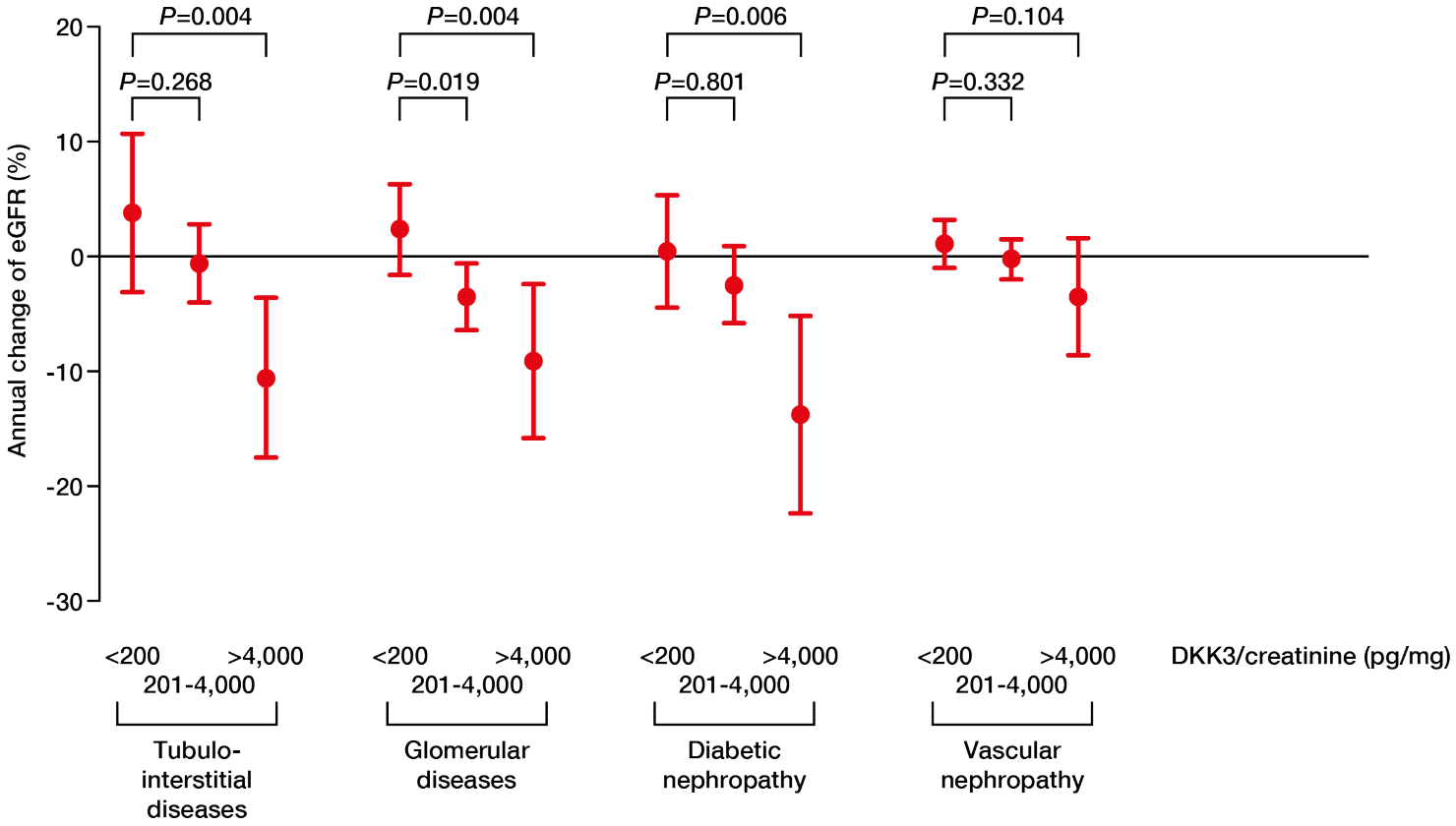
Model 4: Model 3 + adjusted for eGFR

Model 5: Model 4 + adjusted for log albuminuria

Supplemental Figure legends

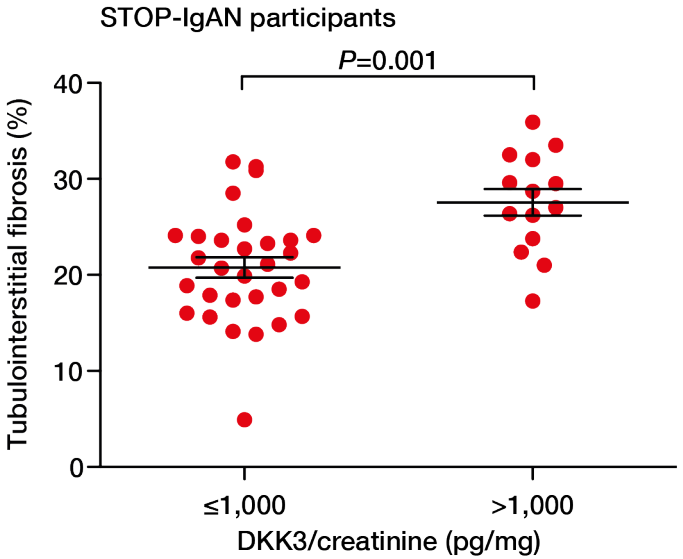
Supplemental Figure 1

Annual change of estimated glomerular filtration rate (eGFR) according to categories of DKK3/creatinine in CKD patients from the CARE FOR HOMe study divided into patients with tubulointerstitial diseases, glomerular diseases, diabetic nephropathy or vascular nephropathy. Analyses were adjusted for age, gender, body mass index, systolic blood pressure, diabetes, smoking status, eGFR and log albuminuria.



Supplemental Figure 2

Tubulointerstitial fibrosis in kidney biopsy specimens of participants of the STOP-IgAN trial according to DKK3/creatinine in urine.



Supplemental Figure 3

Change of DKK3 during early treatment phase in participants of the STOP-IgAN trial according to randomization to supportive care or supportive care plus immunosuppressive therapy.