Capillary Rarefaction Associates with Albuminuria: The Maastricht Study

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ABSTRACT

Albuminuria may be a biomarker of generalized (*i.e.*, microvascular and macrovascular) endothelial dysfunction. According to this concept, endothelial dysfunction of the renal microcirculation causes albuminuria by increasing glomerular capillary wall permeability and intraglomerular pressure, the latter eventually leading to glomerular capillary dropout (rarefaction) and further increases in intraglomerular pressure. However, direct evidence for an association between capillary rarefaction and albuminuria is lacking. Therefore, we examined the cross-sectional association between the recruitment of capillaries after arterial occlusion (capillary density during postocclusive peak reactive hyperemia) and during venous occlusion (venous congestion), as assessed with skin capillaroscopy, and albuminuria in 741 participants of the Maastricht Study, including 211 participants with type 2 diabetes. Overall, 57 participants had albuminuria, which was defined as a urinary albumin excretion ≥30 mg/24 h. After adjustment for potential confounders, participants in the lowest tertile of skin capillary recruitment during postocclusive peak reactive hyperemia had an odds ratio for albuminuria of 2.27 (95% confidence interval, 1.07 to 4.80) compared with those in the highest tertile. Similarly, a comparison between the lowest and the highest tertiles of capillary recruitment during venous congestion yielded an odds ratio of 2.89 (95% confidence interval, 1.27 to 6.61) for participants in the lowest tertile. In conclusion, lower capillary density of the skin microcirculation independently associated with albuminuria, providing direct support for a role of capillary rarefaction in the pathogenesis of albuminuria.

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Albuminuria is strongly associated with cardiovascular disease risk.[1] A leading hypothesis to explain this link is that albuminuria is a biomarker of generalized (*i.e.*, microvascular and macrovascular) endothelial dysfunction.[2,3] According to this concept, endothelial dysfunction of renal arterioles and capillaries (*i.e.*, the renal microcirculation) causes albuminuria by increasing glomerular capillary wall permeability and increasing intraglomerular pressure,[3] the latter eventually leading to glomerular capillary dropout (rarefaction) and further increases in intraglomerular pressure.[4] Concomitantly, endothelial dysfunction in coronary and carotid arteries

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(*i.e.*, the macrocirculation) leads to atherothrombotic cardiovascular disease.[2,3]

Indeed, studies using flow-mediated dilation of the brachial,[5–9] femoral,[10] and left anterior descending coronary artery[11] have provided strong direct evidence for the presence of endothelial dysfunction in the macrocirculation of individuals with albuminuria.

In contrast, evidence for endothelial dysfunction in the microcirculation of individuals with albuminuria is primarily indirect, because it derives from studies using plasma biomarkers,[12–17] the transcapillary escape rate of albumin,[18–21] strain-gauge plethysmography after forearm ischemia,[22] and laser Doppler flowmetry after either iontophoresis of acetylcholine and sodium nitroprusside[23] or arterial occlusion.[24] In addition, evidence for an association between capillary rarefaction and albuminuria is confined to a relatively small study, which showed the frequent co-occurrence of both in individuals with hypertension.[25]

In view of these considerations, we examined in a population–based cohort study the hypothesis that capillary rarefaction is associated with albuminuria. To do this, we used skin capillaroscopy, because capillary rarefaction in the kidney cannot be studied noninvasively in humans. Skin capillaroscopy is a noninvasive technique that allows direct visualization of capillary density in skin by measuring the recruitment of capillaries in response to arterial and venous occlusion, which are thought to be measures of functional and structural capillary density.[26]

RESULTS

Characteristics of the Study Population

For this study on the basis of the first dataset of the Maastricht Study (n=866), four participants with type 1 diabetes were excluded. In the remaining 862 participants, qualitatively satisfactory data on skin capillaroscopy were available in 818 participants. In another 12 participants, the 24-hour urine collections either were collected erroneously (<20 or >28 hours) or were not handed in at all. Of the remaining 806 participants, we additionally excluded participants with missing data on waist circumference (n=3), smoking behavior (n=14), alcohol consumption (n=17), total cholesterol-to-HDL cholesterol ratio (n=8), triglycerides (n=7), eGFR (n=15), office BP (n=2), and/or prior cardiovascular disease status (n=32). These missing data were not mutually exclusive. The study population, therefore, consisted of 741 participants.

Table 1 shows the clinical characteristics of the study population stratified according to tertiles of the percentage recruitment during postocclusive peak reactive hyperemia. Tertile 1 indicates the tertile with the highest level of capillary recruitment.

In general, participants with the lowest recruitment were more often men, were less educated, suffered more often from hypertension and type 2 diabetes, and were more often treated with lipid-modifying or antihypertensive medication. Clinical characteristics according to tertiles of the percentage recruitment during venous congestion are shown in Supplemental Table 1.

Recruitment of Skin Capillaries and the Presence of Albuminuria

Overall, participants with the lowest recruitment more often had albuminuria (Figure 1).

After adjustment for potential confounders and compared with participants with the highest percentage recruitment during postocclusive peak reactive hyperemia (reference category), the odds ratio (OR) and 95% confidence interval (95% CI) for albuminuria for participants in the lowest tertile were OR, 2.27 and 95% CI, 1.07 to 4.80 (Table 2, model 3a). After adjustment for potential confounders and compared with participants with the highest percentage recruitment during venous congestion (reference category), the OR (95% CI) for albuminuria for participants in the lowest tertile was 2.89 (95% CI, 1.27 to 6.61) (Table 2, model 3a).

When we replaced office systolic BP with 24-hour average ambulatory systolic BP (n=665), the OR (95% CI) for albuminuria became 2.19 (95% CI, 0.96 to 5.00) for those with the lowest percentage recruitment during postocclusive peak reactive hyperemia (Table 2, model 3b) and 2.68 (95% CI, 1.10 to 6.52) for those with the lowest percentage recruitment during venous congestion (Table 2, model 3b).

When we replaced the percentage change in capillary density with the absolute numbers of capillaries during postocclusive peak reactive hyperemia as well as during venous congestion, the results were similar (Table 3).

When the variables of models 3a and 3b were added separately to a model adjusted for age and sex, adjustment for type 2 diabetes led to the largest reduction of the OR when the percentage recruitment was the determinant. When the absolute number of capillaries was the determinant, adding additional variables after initial adjustment for age and sex did not materially alter our results (data not shown).

Analyses with interaction terms suggested that the associations between the percentage recruitment during postocclusive peak reactive hyperemia as well as venous congestion and albuminuria were only present in individuals without type 2 diabetes (P value of the interaction term [$P_{\text{interaction}}$] <0.10) (Supplemental Table 2), whereas no such interaction was observed for the absolute number of capillaries under both conditions ($P_{\text{interaction}}$>0.10) (Supplemental Table 3).

Additional Analyses

First, recruitment of capillaries may be expressed as either the absolute or percentage change in capillary density (Concise Methods). When we replaced the percentage change with absolute change in capillary density, results were not materially altered for either postocclusive peak reactive hyperemia or venous congestion (data not shown). Second, mutual adjustment for the percentage recruitment during venous congestion

Table 1. Clinical characteristics of the study population according to tertiles of the percentage recruitment during postocclusive peak reactive hyperemia

Characteristic	Tertiles of the Percentage Recruitment during Postocclusive Peak Reactive Hyperemia		
	Tertile 1 (High), *n*=247	Tertile 2 (Middle), *n*=247	Tertile 3 (Low), *n*=247
Recruitment during postocclusive peak reactive hyperemia, %	73.7 [55.0–186.7]	38.9 [27.0–54.9]	16.3 [0.0–26.9]
Demographics			
Age, yr	59.2±8.5	60.0±8.6	60.0±8.5
Men	119 (48.2)	143 (57.9)	150 (60.7)
Educational level			
Low	33 (13.4)	37 (15.0)	50 (20.2)
Middle	95 (38.5)	101 (40.9)	110 (44.5)
High	119 (48.2)	109 (44.1)	87 (35.2)
Prior cardiovascular disease	50 (20.2)	36 (14.6)	48 (19.4)
Lifestyle variables			
Smoking behavior			
Never a smoker	80 (32.4)	78 (31.6)	72 (29.1)
Former smoker	128 (51.8)	132 (53.4)	136 (55.1)
Current smoker	39 (15.8)	37 (15.0)	39 (15.8)
Alcohol consumption			
None	34 (13.8)	42 (17.0)	47 (19.0)
Low: women ≤7 glasses per wk; men ≤14 glasses per wk	126 (51.0)	135 (54.7)	131 (53.0)
High: women >7 glasses per wk; men >14 glasses per wk	87 (35.2)	70 (28.3)	69 (27.9)
Metabolic variables			
Body mass index categories			
Normal weight, <25 kg/m^2	81 (32.8)	86 (34.8)	67 (27.1)
Overweight, 25–30 kg/m^2	117 (47.4)	106 (42.9)	122 (49.4)
Obesity, ≥30 kg/m^2	49 (19.8)	55 (22.3)	58 (23.5)
Waist circumference, cm			
Men	100.9±11.7	101.3±11.6	103.2±11.5
Women	89.5±12.1	91.5±13.7	92.4±14.3
Waist-to-hip ratio			
Men	1.00±0.07	1.00±0.06	1.01±0.07
Women	0.87±0.07	0.88±0.07	0.89±0.08
Office systolic BP, mmHg	136.1±18.2	135.9±18.7	140.0±19.9
Office diastolic BP, mmHg	76.5±10.3	76.0±10.8	77.8±10.5
24-h Average ambulatory systolic BP, mmHg[a]	118.3±11.9	119.3±11.9	120.9±12.6
24-h Average ambulatory diastolic BP, mmHg[a]	73.6±7.4	74.3±7.4	74.6±7.1
Hypertension	136 (55.1)	136 (55.1)	158 (64.0)
Glucose metabolism status			
Normal glucose metabolism	151 (61.1)	139 (56.3)	114 (46.2)
Impaired fasting glucose	10 (4.0)	11 (4.5)	19 (7.7)
Impaired glucose tolerance	43 (17.4)	27 (10.9)	16 (6.5)
Type 2 diabetes	43 (17.4)	70 (28.3)	98 (39.7)
Fasting glucose, mmol/L			
Without type 2 diabetes	5.4±0.6	5.4±0.6	5.5±0.6
With type 2 diabetes	7.9±1.7	7.6±1.4	7.7±1.9
HbA1C, %[b]			
Without type 2 diabetes	5.7±0.4	5.6±0.4	5.6±0.3
With type 2 diabetes	6.9±0.8	6.7±0.7	6.9±0.9
Total cholesterol, mmol/L	5.4±1.2	5.2±1.2	5.0±1.1
HDL cholesterol, mmol/L			
Men	1.2±0.5	1.1±0.3	1.1±0.3
Women	1.5±0.4	1.5±0.4	1.5±0.5
LDL cholesterol, mmol/L	3.4±1.1	3.3±1.0	3.1±1.0
Triglycerides, mmol/L	1.20 [0.79–1.75]	1.26 [0.89–1.78]	1.25 [0.89–1.79]
Total cholesterol-to-HDL cholesterol ratio	4.2±1.2	4.3±1.3	4.1±1.2

Table 1. Continued

Characteristic	Tertiles of the Percentage Recruitment during Postocclusive Peak Reactive Hyperemia		
	Tertile 1 (High), *n*=247	Tertile 2 (Middle), *n*=247	Tertile 3 (Low), *n*=247
Kidney function			
eGFR, ml/min per 1.73 m^2	88.7±14.7	87.6±14.5	87.8±16.0
Albumin excretion rate, mg/24 h	8.2 [5.4–11.6]	7.8 [5.5–11.5]	7.8 [5.5–15.7]
Albumin excretion ≥15 mg/24 h	34 (13.8)	37 (15.0)	64 (25.9)
Albumin excretion ≥30 mg/24 h	12 (4.9)	13 (5.3)	32 (13.0)
Medication			
Antihypertensive medication	94 (38.1)	89 (36.0)	113 (45.7)
Renin-angiotensin system inhibitor	75 (30.4)	65 (26.3)	84 (34.0)
Lipid-modifying medication	87 (35.2)	78 (31.6)	106 (42.9)

Data are presented as *n* (%), mean±SD, median [interquartile range], or (only for the percentage recruitment during postocclusive peak reactive hyperemia) median [range].

[a]Twenty-four–hour average ambulatory BP measurements were missing in *n*=76 participants (*n*=27 for tertile 1, *n*=23 for tertile 2, and *n*=26 for tertile 3).

[b]To convert to HbA1c values into millimoles per mole: (10.93× HbA1c [%])−23.5.

or the percentage recruitment during postocclusive peak reactive hyperemia suggested multicollinearity (*i.e.*, a strong increase in the standard error of the regression coefficient of the central determinant; data not shown). Third, results were not materially altered when we excluded participants with an albumin excretion >300 mg/24 h; when we replaced office systolic BP with office diastolic BP, office pulse pressure, office mean arterial pressure, presence of hypertension, 24-hour average ambulatory diastolic BP, 24-hour average ambulatory pulse pressure, or 24-hour average ambulatory mean arterial pressure; when we replaced the use of antihypertensive medication with the use of a renin-angiotensin system inhibitor, the use of a diuretic, or their combined use; when we replaced waist circumference with waist-to-hip ratio or body mass index; when we replaced the total cholesterol-to-HDL cholesterol ratio with LDL and HDL; or when we additionally adjusted for hemoglobin A1c (HbA1c) or an inflammation *Z* score (*n*=738). Fourth, analyses in the subpopulation with two urine collections (*n*=666) and the subset of 24-hour urine collections with measured 24-hour urine creatinine excretion within 30% of expected values (*n*=642) did not indicate nondifferential misclassification caused by biologic variability and inaccurate collection, respectively (data not shown). Fifth, analyses on the basis of quintiles and deciles of the respective capillaroscopy measures were also consistent with a threshold level (Supplemental Tables 4–6). However, these analyses were hampered by a loss of power. Sixth, we defined albuminuria as an albumin excretion ≥15 mg/24 h, in agreement with the fact that an association with (cardiovascular disease) mortality already exists at levels of urinary albumin excretion <30 mg/24 h[1] and to explore whether misclassification of albuminuria status occurred with the clinical cutoff value. With this definition, the associations for the entire study

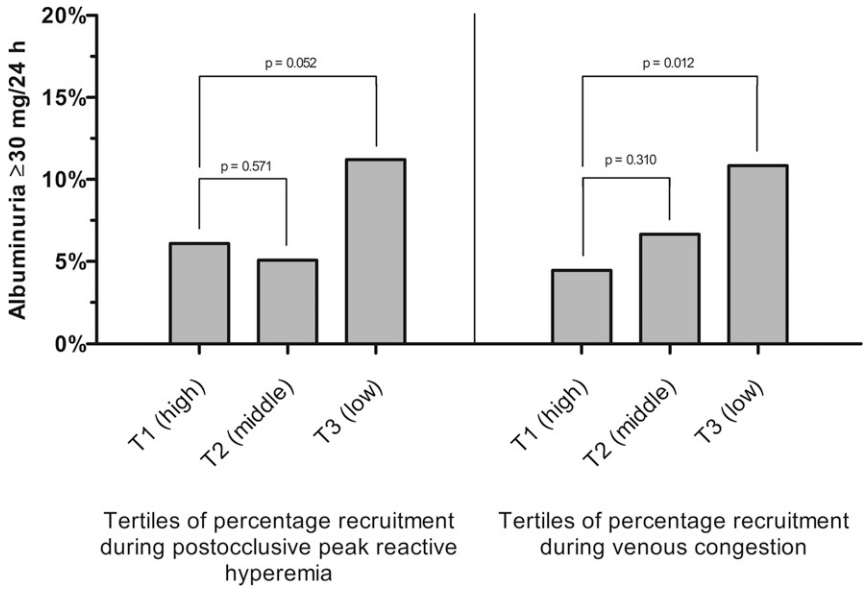

Figure 1. Capillary recruitment is associated with albuminuria. Bar charts showing the association between recruitment during postocclusive peak reactive hyperemia (left panel) as well as during venous congestion (right panel) and the presence of albuminuria ≥30 mg/24 h. Tertiles of the percentage recruitment during postocclusive peak reactive hyperemia ranged from 55.0 to 186.7 (tertile 1 [T1]), from 27.0 to 54.9 (tertile 2 [T2]), and from 0.0 to 26.9 (tertile 3 [T3]). Tertiles of the percentage recruitment during venous congestion ranged from 55.8 to 253.3 (T1), from 27.6 to 55.7 (T2), and from −2.9 to 27.5 (T3). Percentages of participants with albuminuria ≥30 mg/24 h per tertile were adjusted for age, sex, and type 2 diabetes (model 2) by marginal standardization. *P* values were derived from the same models.

Table 2. Association between the percentage recruitment during postocclusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥30 mg/24 h)

Independent Variable	OR (95% CI)	*P* Value
Recruitment during postocclusive peak reactive hyperemia, %		
Model 1		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	1.09 (0.49 to 2.43)	0.84
Tertile 3 (low)	2.92 (1.46 to 5.80)	0.002
Model 2		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	0.82 (0.36 to 1.88)	0.57
Tertile 3 (low)	2.04 (0.99 to 4.19)	0.05
Model 3a		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	0.95 (0.41 to 2.30)	0.95
Tertile 3 (low)	2.27 (1.07 to 4.80)	0.03
Model 3b		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	1.25 (0.50 to 3.13)	0.63
Tertile 3 (low)	2.19 (0.96 to 5.00)	0.06
Recruitment during venous congestion, %		
Model 1		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	1.83 (0.79 to 4.23)	0.16
Tertile 3 (low)	3.94 (1.84 to 8.43)	<0.001
Model 2		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	1.56 (0.66 to 3.67)	0.31
Tertile 3 (low)	2.75 (1.25 to 6.06)	0.01
Model 3a		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	1.75 (0.72 to 4.26)	0.74
Tertile 3 (low)	2.89 (1.27 to 6.61)	0.01
Model 3b		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	1.79 (0.69 to 4.65)	0.23
Tertile 3 (low)	2.68 (1.10 to 6.52)	0.03

ORs represent the odds of having albuminuria (defined as an albumin excretion ≥30 mg/24 h) in the respective tertile of the percentage recruitment during postocclusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference tertile. Model 1 is the unadjusted model. Model 2 is adjusted for age, sex, and type 2 diabetes. Model 3a is model 2 adjusted for waist circumference, total cholesterol-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying drugs, office systolic BP, use of antihypertensive medication, eGFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, and educational level. Model 3b is model 3a adjusted for 24-hour average ambulatory systolic BP instead of office systolic BP (n=665).

population became somewhat weaker (Supplemental Table 7), and there was no statistical interaction with type 2 diabetes ($P_{\text{interaction}}>0.10$). Seventh, multivariable linear regression analyses showed no associations between tertiles of the respective capillaroscopy measures and (inverse square root–transformed) continuous urinary albumin excretion (data not shown).

DISCUSSION

The main finding of this population-based study is that lower capillary density was associated with the presence of albuminuria, regardless of whether type 2 diabetes was present. This association was independent of cardiovascular disease risk factors, including 24-hour average ambulatory BP and biomarkers of low-grade inflammation. To the best of our knowledge, this is the first population-based study that provides direct support for a role of capillary rarefaction in the pathogenesis of albuminuria.

An association between capillary rarefaction and albuminuria is in agreement with the Brenner hypothesis[4] (*i.e.*, an increase in intraglomerular pressure will lead to glomerular capillary dropout [rarefaction] and further increases in intraglomerular pressure on the one hand and greater permeation of albumin through the glomerular capillary wall on the other hand). Indeed, in individuals with type 2 diabetes, estimated intraglomerular pressure was higher in the presence of albuminuria.[27] Additionally, in individuals who underwent a large reduction in renal mass, remaining kidney mass was inversely associated with urinary albumin excretion.[28] Furthermore, in a smaller study, capillary rarefaction in the skin microcirculation and albuminuria frequently co-occurred in individuals with hypertension.[25] This study extends this knowledge, because it is the first to examine a direct measure of capillary rarefaction in a large population–based sample with adjustment for potential confounders.

A key assumption underlying this study is that skin capillary rarefaction reflects capillary rarefaction of the kidney. Although the skin microcirculation has not been compared directly with that of the kidney, several observations support the view that it is representative for the systemic microcirculation, including the kidney's. First, age-related changes in the skin microcirculation parallel those in the systemic vasculature.[29] Second, the microcirculation of the skin and kidney share associations with salt-sensitive hypertension[30] and low birth weight.[31–34]

Both capillary density during venous occlusion and capillary density after arterial occlusion were used as reproducible[26,35] estimates of maximal skin capillary density.[36] In this study, we could not determine to what extent differences in capillary density were caused by structural (*i.e.*, anatomic) or functional (*i.e.*, nonperfusion) rarefaction. However, the occurrence of multicollinearity after mutual adjustment suggests that both measures assessed the same or at least overlapping construct(s) in this study.

Importantly, misclassification of albuminuria status because of the use of renin-angiotensin system inhibitors may explain why capillary rarefaction in individuals with type 2 diabetes was associated with albuminuria when defined as an albumin excretion ≥15 mg/24 h but not when defined as an albumin excretion ≥30 mg/24 h. Renin-angiotensin system inhibitors reduce urinary albumin excretion by lowering intraglomerular pressure,[37,38] an effect that is enhanced by

Table 3. Association between the absolute number of capillaries during postocclusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥30 mg/24 h)

Independent Variable	OR (95% CI)	*P* Value
Postocclusive peak reactive hyperemia, *n*/mm^2		
Model 1		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	2.51 (1.13 to 5.57)	0.02
Tertile 3 (low)	3.01 (1.38 to 6.55)	<0.01
Model 2		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	2.27 (1.00 to 5.15)	0.05
Tertile 3 (low)	2.38 (1.07 to 5.31)	0.03
Model 3a		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	2.63 (1.10 to 6.30)	0.03
Tertile 3 (low)	2.61 (1.11 to 6.12)	0.03
Model 3b		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	2.09 (0.82 to 5.30)	0.12
Tertile 3 (low)	2.51 (1.04 to 6.06)	0.04
Venous congestion, *n*/mm^2		
Model 1		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	1.15 (0.55 to 2.41)	0.72
Tertile 3 (low)	2.11 (1.08 to 4.13)	0.03
Model 2		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	0.95 (0.44 to 2.04)	0.90
Tertile 3 (low)	1.72 (0.86 to 3.47)	0.13
Model 3a		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	1.12 (0.50 to 2.50)	0.79
Tertile 3 (low)	1.76 (0.84 to 3.69)	0.13
Model 3b		
Tertile 1 (high)	Reference	
Tertile 2 (middle)	1.32 (0.54 to 3.25)	0.54
Tertile 3 (low)	2.32 (1.01 to 5.32)	0.05

ORs represent the odds of having albuminuria (defined as an albumin excretion ≥30 mg/24 h) in the respective tertile of the absolute number of capillaries during postocclusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference tertile. Model 1 is the unadjusted model. Model 2 is adjusted for age, sex, and type 2 diabetes. Model 3a is model 2 adjusted for waist circumference, total cholesterol-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying drugs, office systolic BP, use of antihypertensive medication, eGFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, and educational level. Model 3b is model 3a adjusted for 24-hour average ambulatory systolic BP instead of office systolic BP (*n*=665).

diuretics,[39] and an albumin excretion ≥30 mg/24 h is used as an indication for their use, particularly in individuals with type 2 diabetes.[40] Hence, individuals previously having had an albumin excretion ≥30 mg/24 h could be classified erroneously as having no albuminuria with this definition, thus obscuring an association with capillary rarefaction. Indeed, the frequent use of renin-angiotensin system inhibitors in individuals with type 2 diabetes and a urinary albumin excretion of 15–30 mg/24 h (64.9% versus 19.5% in individuals with a similar urinary albumin excretion but without type 2 diabetes) supports this explanation. Alternatively, the statistically significant interaction between type 2 diabetes and the percentage recruitment during postocclusive peak reactive hyperemia as well as venous congestion may be attributable to the play of chance given the low number of cases.

A lack of power because of the small variation in urinary albumin excretion with only a few individuals with an albumin excretion ≥30 mg/24 h may explain why the results of this study suggest a threshold level for capillary density and why capillary rarefaction was not associated with continuous urinary albumin excretion. In addition, we measured urinary albumin excretion instead of the permeation of albumin through the glomerular capillary wall, and small increases in permeation may be compensated for by tubular reabsorption.[41]

A strength of this study is that participants of the Maastricht Study were well characterized, allowing adjustment for an extensive series of potential confounders, including 24-hour average ambulatory BP and low-grade inflammation. In this regard, the use of office BP only could have underestimated any effect of BP on albuminuria and thereby, overestimated the association between capillary rarefaction and albuminuria. However, some of the variables in our models may also be intermediates in the association between capillary rarefaction and albuminuria, possibly leading to overadjustment bias (*i.e.*, the associations reported are conservative).[42] For instance, capillary rarefaction may be involved in the pathogenesis of type 2 diabetes, which may subsequently lead to albuminuria *via* a hyperglycemia-induced increase in glomerular capillary wall permeability.[3] Similarly, higher BP may be both a cause and a consequence of capillary rarefaction.[43]

From a clinical perspective, both capillary rarefaction itself and the resulting increase in intraglomerular pressure may be a target in the management of albuminuria. Indeed, renin-angiotensin system inhibitors, which reduce intraglomerular pressure,[37,38] form the mainstay of the current management of albuminuria.[40] However, current management is not specifically aimed at regenerating glomeruli. Nonetheless, in a recent study, treatment with a renin-angiotensin system inhibitor led to an increase in kidney vasculature in a rat model of progressive glomerular injury,[44] suggesting that capillary rarefaction itself could be a future therapeutic target.

This study had some limitations. First, no direct measure of capillary rarefaction of the kidney was used. However, at present, capillary rarefaction of the kidney, in contrast to capillary rarefaction of skin, cannot be studied noninvasively in humans. Second, because of the logistics of this large–scale population–based study, participants were not asked to come in fasting. However, to minimize the effects of dietary intake on the microcirculation,[45–47] participants were asked to have a standardized low–fat breakfast (or lunch) and refrain from caffeine-containing beverages and smoking. In addition, results were not materially altered after adjustment for nonadherence to the dietary and smoking restrictions. Third, the

cross-sectional design does not allow us to make strong causal inferences. In addition, a longitudinal design with frequent assessment of urinary albumin excretion and medication use would have avoided misclassification of albuminuria status. Fourth, the study population primarily consisted of white individuals of European descent (99.2%), limiting generalizability to other populations.

In conclusion, lower capillary density of the skin microcirculation was independently associated with the presence of albuminuria, regardless of the presence of type 2 diabetes. Thereby, this is the first population-based study that provides direct support for a role of capillary rarefaction in the pathogenesis of albuminuria.

CONCISE METHODS

The Maastricht Study Population and Design

In this study, we used data from the Maastricht Study, an observational prospective population–based cohort study. The rationale and methodology have been described previously.[48] In brief, the study focuses on the etiology, pathophysiology, complications, and comorbidities of type 2 diabetes mellitus and is characterized by an extensive phenotyping approach. Eligible for participation were all individuals with ages between 40 and 75 years living in the southern part of The Netherlands. Participants were recruited through mass media campaigns and from the municipal registries and the regional Diabetes Patient Registry *via* mailings. Recruitment was stratified according to known type 2 diabetes status for reasons of efficiency. This report includes cross-sectional data from the first 866 participants who completed the baseline survey between November of 2010 and March of 2012. The examinations of each participant were performed within a time window of 3 months. The study has been approved by the institutional medical ethical committee (NL31329.068.10) and the Minister of Health, Welfare and Sports of The Netherlands on the basis of the Health Council's opinion (Permit 131088–105234-PG) and was conducted in accordance with the Declaration of Helsinki. All participants gave written informed consent.

Skin Capillaroscopy

All participants were asked to refrain from smoking and drinking coffee or tea ≥3 hours before the measurements. A light meal (breakfast and/or lunch) low in fat content was allowed before the start of the measurements. Skin capillaroscopy measurements were performed in a quiet, temperature–controlled room (T=24°C) with participants in the supine position as previously described.[49]

Briefly, capillaries were visualized in the dorsal skin of the distal phalanges of the third and fourth finger of the right hand by use of a digital video microscope (Capiscope; KK Technology, Honiton, United Kingdom) with a system magnification of ×100.[49] Capillaries were visualized 4.5 mm proximal to the terminal row of capillaries in the middle of the nailfold. The investigator selected a region of interest of 1-mm^2 skin area. Capillary density (mean of two fields) was measured under three conditions. First, baseline capillary density was measured. Baseline capillary density was defined as the number of continuously erythrocyte–perfused capillaries per 1 mm^2 skin and was counted for 15 seconds. Second, capillary recruitment during postocclusive peak reactive hyperemia was assessed after 4 minutes of arterial occlusion. Arterial occlusion was applied using a miniature cuff at the base of the investigated finger inflated to suprasystolic pressure (260 mmHg) for 4 minutes. Directly after release of the cuff, all (continuously and intermittently) perfused capillaries were counted for 15 seconds. Third, venous congestion was applied, with the cuff inflated to 60 mmHg for 2 minutes, and all (continuously and intermittently) perfused capillaries were counted for 15 seconds. The number of perfused capillaries was counted in the recorded digital raw data with the use of a semiautomatic procedure (CapiAna)[49] by two investigators who were blinded to participants' clinical status. The intra- and interobserver coefficients of variation for the counting procedure were 2.5% and 5.6%, respectively, as described previously.[49]

For the primary analyses, we used recruitment during postocclusive peak reactive hyperemia as well as during venous congestion (expressed as the percentage change in capillary density from baseline) and the absolute number of capillaries during postocclusive peak reactive hyperemia as well as during venous congestion (expressed as capillaries per 1 mm^2).

Kidney Function

GFR was estimated using the Chronic Kidney Disease Epidemiology Collaboration equation on the basis of both serum creatinine and serum cystatin C (Supplemental Material).[50] To assess urinary albumin excretion, participants were requested to collect two 24-hour urine collections (Supplemental Material).

Albuminuria was defined as an albumin excretion ≥30 mg/24 h,[51] which is used in clinical practice to guide cardiovascular disease prevention, particularly in individuals with type 2 diabetes.[40] In an additional analysis, albuminuria was defined as an albumin excretion ≥15 mg/24 h (the upper level of daily albumin excretion in healthy individuals[52]) in agreement with the fact that an association with (cardiovascular disease) mortality already exists at levels of urinary albumin excretion <30 mg/24 h^1 and to explore whether misclassification of albuminuria status occurred with the clinical cutoff. These definitions were preferably on the basis of the average of two (available in 89.9% of the participants) 24-hour urine collections.

Potential Confounders

We assessed glucose metabolism status, body mass index, waist circumference, hip circumference, office BP, 24-hour average ambulatory BP, fasting glucose, HbA1c, total cholesterol, HDL cholesterol, LDL cholesterol, triglycerides, medication use, smoking behavior, alcohol consumption, educational level, and prevalent cardiovascular disease as described previously.[48,53] Definitions of these potential confounders are provided in Supplemental Material. In addition, we assessed the following plasma biomarkers of inflammation: high–sensitivity C–reactive protein, serum amyloid A, IL-6, IL-8, TNF-α, and soluble intercellular adhesion molecule 1.[54]

Statistical Analyses

All analyses were performed with IBM SPSS Statistics, Version 22.0 (IBM SPSS, Chicago, IL) unless stated otherwise.

Participants were divided into tertiles of the percentage recruitment during postocclusive peak reactive hyperemia as well as into tertiles of the percentage recruitment during venous congestion, because the association with albuminuria seemed to be nonlinear. Participants with normal glucose tolerance, impaired fasting glucose, and impaired glucose tolerance were combined into one category (participants without type 2 diabetes) because of the small number of participants with impaired fasting glucose and impaired glucose tolerance.

Associations between tertiles of the percentage recruitment during postocclusive peak reactive hyperemia as well as during venous congestion and the presence of albuminuria were evaluated using multivariable logistic regression analyses. Similarly, associations between tertiles of the absolute number of capillaries during postocclusive peak reactive hyperemia as well as during venous congestion and the presence of albuminuria were evaluated. The tertile with the highest recruitment or the highest absolute number of capillaries (tertile 1) was used as reference category. Next, we adjusted for potential confounders as follows: model 1, unadjusted model; model 2, adjusted for age, sex, and type 2 diabetes; model 3a, model 2 adjusted for waist circumference, total cholesterol-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying medication, office systolic BP, use of antihypertensive medication, eGFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, and educational level; and model 3b, model 3a with replacement of office systolic BP by 24-hour average ambulatory systolic BP.

We used interaction terms to examine whether the associations were modified by the presence or absence of type 2 diabetes. A $P_{\text{interaction}}<0.10$ in model 3a was considered to indicate a statistically significant interaction.

Adjusted percentages of participants with albuminuria per tertile of recruitment were derived from the logistic regression models (model 2) with adjustment for age, sex, and type 2 diabetes by marginal standardization[55] (calculated with Stata Statistical Software, release 11.2SE; StataCorp., College Station, TX).

Several additional analyses were performed, each starting from the models described above. First, we used the absolute change in capillary density from baseline (expressed as capillaries per 1 mm^2) during postocclusive peak reactive hyperemia and during venous congestion to categorize participants. Second, we excluded participants with an albumin excretion >300 mg/24 h (n=8). Third, we replaced office systolic BP with office diastolic BP, office pulse pressure, office mean arterial pressure, presence of hypertension, 24-hour average ambulatory diastolic BP, 24-hour average ambulatory pulse pressure, or 24-hour average ambulatory mean arterial pressure in model 3a, and we replaced the use of antihypertensive medication with the use of a renin-angiotensin system inhibitor, the use of a diuretic, or their combined use. Fourth, we replaced waist circumference with waist-to-hip ratio or body mass index, replaced the total cholesterol-to-HDL cholesterol ratio with LDL and HDL, and additionally, adjusted for HbA1c and a Z score of the inflammation biomarkers[54] in model 3a. Fifth, we repeated the analyses in participants with two urine collections and after exclusion of 24-hour urine collections with a measured 24-hour urine creatinine excretion not within 30% of expected values[56] to explore whether biologic variation and inaccurate collection, respectively, led to nondifferential misclassification with bias toward zero. Sixth, we repeated the analyses with quintiles and deciles of the respective capillaroscopy measures as independent variables. Seventh, we repeated the analyses with albuminuria defined as an albumin excretion ≥15 mg/24 h. Eighth, we performed multivariable linear regression analyses to examine whether the capillaroscopy measures were associated with urinary albumin excretion on a continuous scale. Urinary albumin excretion had to be transformed by taking the inverse square root of urinary albumin excretion to fulfill the normality assumption, because it was highly positively skewed and could not be transformed adequately using common[57] transformations.

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Abstracts on the basis of the results of this study have been published and presented previously at the Dutch Federation of Nephrology Fall Symposium 2014 (Utrecht, The Netherlands; October 10, 2014), the 3rd Joint Meeting of the Dutch Endothelial Biology Society and the Dutch Society for Microcirculation and Vascular Biology (Biezenmortel, The Netherlands; October 30–31, 2014), and the European Renal Association - European Dialysis and Transplant Association 52nd Congress (London, United Kingdom; May 28–31, 2015).

DISCLOSURES

None.

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Online Supplemental Materials

Capillary rarefaction associates with albuminuria: The Maastricht Study

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Contents:

Supplemental Methods

Kidney function

GFR was estimated using the CKD-EPI equation based on both serum creatinine and serum cystatin C.[1] Serum creatinine was measured with a Jaffé method traceable to isotope dilution mass spectrometry (Beckman Synchron LX20, Beckman Coulter Inc., Brea, USA). Serum cystatin C was measured with a particle enhanced immunoturbidimetric assay standardized against ERM-DA471/IFCC reference material (Roche Cobas 8000, F. Hoffman-La Roche Ltd, Basel, Switzerland).

To assess urinary albumin excretion, participants were requested to collect two 24h urine collections. Urinary albumin concentration was measured with a standard immunoturbidimetric assay by an automatic analyzer (Beckman Synchron LX20, Beckman Coulter Inc., Brea, USA) and multiplied by collection volume to obtain the 24h urinary albumin excretion. A urinary albumin concentration below the detection limit of the assay (2 mg/l) was set at 1.5 mg/l before multiplying by collection volume. Only urine collections with a collection time between 20h and 28h were considered valid. If needed, urinary albumin excretion was extrapolated to a 24h excretion.

Albuminuria was defined as an albumin excretion ≥ 30 mg/24h,[2] which is used in clinical practice to guide cardiovascular disease prevention, particularly in individuals with type 2 diabetes.[3] In an additional analysis, albuminuria was defined as an albumin excretion ≥ 15 mg/24h (the upper level of daily albumin excretion in healthy individuals[4]), in agreement with the fact that an association with (cardiovascular) mortality already exists below the clinical cut off value of 30 mg/24h.[5] These definitions were preferably based on the average of two (available in 89.9% of the participants) 24h urine collections.

Potential confounders

We assessed glucose metabolism status, body mass index, waist circumference, hip circumference, office blood pressure, 24h average ambulatory blood pressure, fasting glucose, HbA1c, total cholesterol, HDL cholesterol, LDL cholesterol, triglycerides, medication use, smoking behavior, alcohol consumption, educational level and prevalent cardiovascular disease as described previously.[6,7] Glucose metabolism status was classified according to the World Health Organization 2006 criteria[8] into normal glucose metabolism, impaired fasting glucose, impaired glucose tolerance, and diabetes mellitus. Participants with diabetes mellitus and participants using glucose-lowering medication were considered as having type 2 diabetes if they had no (self-reported) type 1 or other specific type of diabetes. Prevalent cardiovascular disease was defined as a self-reported history of myocardial infarction, and(or) cerebrovascular infarction or hemorrhage, and(or) percutaneous artery angioplasty of, or vascular surgery on, the coronary, abdominal, peripheral or carotid arteries. Alcohol consumption was classified into three categories: non-consumption, low-consumption ($\leq$7 glasses per week for women and $\leq$14 glasses per week for men) and high-consumption (>7 glasses per week for women and >14 glasses per week for men). Waist-to-hip ratio was calculated by dividing waist circumference by hip circumference. Total-to-HDL cholesterol ratio was calculated by dividing total cholesterol by HDL cholesterol. Hypertension was defined as an office systolic blood pressure ≥ 140 mmHg, an office diastolic blood pressure ≥ 90 mmHg and(or) the use of antihypertensive medication. Office pulse pressure was defined as office systolic blood pressure minus office diastolic blood pressure and office mean arterial pressure as office diastolic blood pressure plus 0.412 times office pulse pressure.[9] Similar equations were used to calculate 24h average ambulatory pulse

pressure and 24h average ambulatory mean arterial pressure, respectively. Educational level was assessed during the cognitive assessment and was classified into three groups: low (none, primary, or lower vocational education only), intermediate (intermediate general secondary, intermediate vocational or higher general secondary education) and high (higher vocational education or university level of education).[7,10]

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Supplemental Tables

Supplemental Table 1. Clinical characteristics of the study population according to tertiles of the percentage recruitment during venous congestion

	Tertiles of the percentage recruitment during venous congestion		
	T1 (high) n = 247	**T2 (middle)** n = 247	**T3 (low)** n = 247
Recruitment during venous congestion (%)	75.6 [55.8-253.3]	39.0 [27.6-55.7]	16.2 [-2.9-27.5]
Demographics			
Age (years)	59.4 ±8.5	59.9 ±8.6	59.9 ±8.5
Men	122 (49.4)	131 (53.0)	159 (64.4)
Educational level			
Low	32 (13.0)	29 (11.7)	59 (23.9)
Middle	96 (38.9)	101 (40.9)	109 (44.1)
High	119 (48.2)	117 (47.4)	79 (32.0)
Prior cardiovascular disease	43 (17.4)	41 (16.6)	50 (20.2)
Lifestyle variables			
Smoking behavior			
Never smoker	80 (32.4)	78 (31.6)	72 (29.1)
Former smoker	130 (52.6)	132 (53.4)	134 (54.3)
Current smoker	37 (15.0)	37 (15.0)	41 (16.6)
Alcohol consumption			
Non	30 (12.1)	44 (17.8)	49 (19.8)
Low (women ≤ 7 glasses/week; men ≤ 14 glasses/week)	127 (51.4)	127 (51.4)	138 (55.9)
High (women > 7 glasses/week; men > 14 glasses/week)	90 (36.4)	76 (30.8)	60 (24.3)
Metabolic variables			
BMI categories			
Normal weight (< 25 kg/m^2)	86 (34.8)	86 (34.8)	62 (25.1)
Overweight (25-30 kg/m^2)	115 (46.6)	112 (45.3)	118 (47.8)
Obesity (≥ 30 kg/m^2)	46 (18.6)	49 (19.8)	67 (27.1)
Waist circumference (cm)			
Men	100.0 ±11.9	101.3 ±11.4	103.6 ±11.4
Women	89.4 ±11.6	90.1 ±12.8	94.5 ±15.6
Waist-to-hip-ratio			
Men	0.99 ±0.07	1.00 ±0.06	1.01 ±0.07

Women	0.87 ±0.07	0.88 ±0.07	0.90 ±0.08
Office systolic blood pressure (mmHg)	136.4 ±18.3	136.0 ±18.6	139.6 ±20.0
Office diastolic blood pressure (mmHg)	76.7 ±10.4	75.7 ±10.4	78.1 ±10.7
24h average ambulatory systolic blood pressure (mmHg)*	118.6 ±11.7	119.1 ±12.4	120.8 ±12.3
24h average ambulatory diastolic blood pressure (mmHg)*	73.9 ±7.3	74.1 ±7.4	74.6 ±7.2
Hypertension	135 (54.7)	137 (55.5)	158 (64.0)
Glucose metabolism status			
Normal glucose metabolism	153 (61.9)	144 (58.3)	107 (43.3)
Impaired fasting glucose	7 (2.8)	12 (4.9)	21 (8.5)
Impaired glucose tolerance	44 (17.8)	26 (10.5)	16 (6.5)
Type 2 diabetes	43 (17.4)	65 (26.3)	103 (41.7)
Fasting glucose (mmol/l)			
Without type 2 diabetes	5.4 ±0.5	5.3 ±0.6	5.5 ±0.6
With type 2 diabetes	7.9 ±1.6	7.7 ±1.5	7.6 ±1.9
HbA1C (%)**			
Without type 2 diabetes	5.7 ±0.4	5.6 ±0.4	5.6 ±0.4
With type 2 diabetes	6.9 ±0.8	6.8 ±0.7	6.8 ±0.9
Total cholesterol (mmol/l)	5.5 ±1.2	5.2 ±1.1	5.0 ±1.1
HDL cholesterol (mmol/l)			
Men	1.2 ±0.5	1.2 ±0.3	1.1 ±0.3
Women	1.5 ±0.4	1.5 ±0.4	1.5 ±0.5
LDL cholesterol (mmol/l)	3.5 ±1.1	3.3 ±1.0	3.1 ±1.0
Triglycerides (mmol/l)	1.20 [0.79-1.76]	1.19 [0.86-1.66]	1.31 [0.90-1.92]
Total-to-HDL cholesterol ratio	4.2 ±1.2	4.2 ±1.3	4.3 ±1.2
Kidney function			
Estimated GFR (ml/min/1.73m^2)	89.1 ±14.1	87.3 ±15.4	87.8 ±15.7
Albumin excretion rate (mg/24h)	8.2 [5.4-11.1]	7.4 [5.4-11.4]	7.9 [5.5-16.6]
Albumin excretion ≥ 15 mg/24h	31 (12.6)	38 (15.4)	66 (26.7)
Albumin excretion ≥ 30 mg/24h	9 (3.6)	16 (6.5)	32 (13.0)
Medication			
Antihypertensive medication	91 (36.8)	88 (35.6)	117 (47.4)
Renin-angiotensin system inhibitor	73 (29.6)	59 (23.9)	92 (37.2)
Lipid-modifying medication	85 (34.4)	75 (30.4)	111 (44.9)

Data are presented as n (%), mean ±standard deviation, median [interquartile range] or (only for the percentage recruitment during venous congestion) median [range]. * 24h average ambulatory blood pressure measurements were missing in n = 76 participants (n=23 for T1, n=28 for T2 and n=25 for T3). ** To convert to HbA1c values in mmol/mol: (10.93 * HbA1c [%]) - 23.5.

Supplemental Table 2A. Association between the percentage recruitment during post-occlusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥ 30 mg/24h) in participants without type 2 diabetes

Model	Recruitment during post-occlusive peak reactive hyperemia (%)	OR (95% CI)	P value	Recruitment during venous congestion (%)	OR (95% CI)	P value
1	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	0.92 (0.24; 3.48)	0.903	T2 (middle)	2.68 (0.68; 10.52)	0.158
	T3 (low)	4.13 (1.45; 11.73)	0.008	T3 (low)	6.65 (1.86; 23.78)	0.004
2	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	0.85 (0.22; 3.25)	0.817	T2 (middle)	2.67 (0.68; 10.57)	0.161
	T3 (low)	3.86 (1.35; 11.10)	0.012	T3 (low)	6.16 (1.71; 22.26)	0.006
3a	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	0.97 (0.25; 3.85)	0.970	T2 (middle)	2.69 (0.66; 11.01)	0.169
	T3 (low)	4.49 (1.50; 13.49)	0.007	T3 (low)	6.73 (1.79; 25.34)	0.005
3b	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	1.15 (0.27; 4.96)	0.852	T2 (middle)	3.80 (0.74; 19.66)	0.111
	T3 (low)	4.93 (1.47; 16.51)	0.010	T3 (low)	8.41 (1.75; 40.48)	0.008

Odds ratios represent the odds of having albuminuria (defined as an albumin excretion ≥ 30 mg/24h) in the respective tertile of the percentage recruitment during post-occlusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference tertile. Model 1: unadjusted model; Model 2: age, sex; Model 3a: model 2 + waist circumference, total-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying drugs, office systolic blood pressure, use of antihypertensive medication, estimated GFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, educational level; Model 3b: as model 3a but adjusted for 24h average ambulatory systolic blood pressure instead of office systolic blood pressure (n=477). CI: confidence interval, OR: odds ratio, T: tertile.

Supplemental Table 2B. Association between the percentage recruitment during post-occlusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥ 30 mg/24h) in participants with type 2 diabetes

Model	Recruitment during post-occlusive peak reactive hyperemia (%)	OR (95% CI)	P value	Recruitment during venous congestion (%)	OR (95% CI)	P value
1	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	0.76 (0.26; 2.21)	0.613	T2 (middle)	0.99 (0.33; 3.02)	0.987
	T3 (low)	1.16 (0.44; 3.02)	0.765	T3 (low)	1.40 (0.52; 3.78)	0.513
2	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	0.67 (0.23; 1.99)	0.472	T2 (middle)	0.94 (0.30; 2.91)	0.911
	T3 (low)	1.12 (0.42; 2.97)	0.823	T3 (low)	1.38 (0.50; 3.79)	0.538
3a	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	0.82 (0.26; 2.59)	0.731	T2 (middle)	1.19 (0.36; 3.95)	0.779
	T3 (low)	1.16 (0.41; 3.30)	0.781	T3 (low)	1.41 (0.48; 4.15)	0.530
3b	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	1.16 (0.34; 3.97)	0.817	T2 (middle)	1.04 (0.29; 3.67)	0.957
	T3 (low)	0.92 (0.28; 3.01)	0.889	T3 (low)	1.07 (0.34; 3.40)	0.908

Odds ratios represent the odds of having albuminuria (defined as an albumin excretion ≥ 30 mg/24h) in the respective tertile of the percentage recruitment during post-occlusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference tertile. Model 1: unadjusted model; Model 2: age, sex; Model 3a: model 2 + waist circumference, total-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying drugs, office systolic blood pressure, use of antihypertensive medication, estimated GFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, educational level; Model 3b: as model 3a but adjusted for 24h average ambulatory systolic blood pressure instead of office systolic blood pressure (n=188). CI: confidence interval, OR: odds ratio, T: tertile

Supplemental Table 3A. Association between the absolute number of capillaries during post-occlusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥ 30 mg/24h) in participants without type 2 diabetes

Model	Post-occlusive peak reactive hyperemia (n/mm^2)	OR (95% CI)	P value	Venous congestion (n/mm^2)	OR (95% CI)	P value
1	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	2.71 (0.71; 10.39)	0.145	T2 (middle)	1.82 (0.52; 6.33)	0.346
	T3 (low)	4.71 (1.31; 17.00)	0.018	T3 (low)	3.64 (1.15; 11.52)	0.028
2	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	2.49 (0.65; 9.62)	0.185	T2 (middle)	1.57 (0.45; 5.53)	0.479
	T3 (low)	4.08 (1.12; 14.86)	0.033	T3 (low)	2.96 (0.92; 9.49)	0.068
3a	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	2.48 (0.63; 9.81)	0.196	T2 (middle)	1.84 (0.51; 6.62)	0.352
	T3 (low)	3.85 (1.03; 14.40)	0.045	T3 (low)	2.82 (0.86; 9.28)	0.089
3b	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	2.16 (0.52; 8.86)	0.287	T2 (middle)	2.48 (0.60; 10.29)	0.212
	T3 (low)	4.17 (1.10; 15.78)	0.035	T3 (low)	4.03 (1.06; 15.31)	0.041

Odds ratios represent the odds of having albuminuria (albumin excretion ≥ 30 mg/24h) in the respective tertile of the absolute number of capillaries during post-occlusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference tertile. Model 1: unadjusted model; Model 2: age, sex; Model 3a: model 2 + waist circumference, total-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying drugs, office systolic blood pressure, use of antihypertensive medication, estimated GFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, educational level; Model 3b: as model 3a but adjusted for 24h average ambulatory systolic blood pressure instead of office systolic blood pressure (n=477). CI: confidence interval, OR: odds ratio, T: tertile.

Supplemental Table 3B. Association between the absolute number of capillaries during post-occlusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥ 30 mg/24h) in participants with type 2 diabetes

Model	Post-occlusive peak reactive hyperemia (n/mm^2)	OR (95% CI)	P value	Venous congestion (n/mm^2)	OR (95% CI)	P value
1	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	2.47 (0.88; 6.92)	0.085	T2 (middle)	0.81 (0.31; 2.14)	0.669
	T3 (low)	1.75 (0.63; 4.85)	0.282	T3 (low)	1.22 (0.51; 2.94)	0.653
2	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	2.19 (0.77; 6.23)	0.142	T2 (middle)	0.68 (0.25; 1.84)	0.449
	T3 (low)	1.60 (0.57; 4.51)	0.372	T3 (low)	1.20 (0.49; 2.93)	0.697
3a	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	2.84 (0.91; 8.88)	0.073	T2 (middle)	0.76 (0.26; 2.25)	0.626
	T3 (low)	1.92 (0.63; 5.88)	0.255	T3 (low)	1.24 (0.47; 3.27)	0.668
3b	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	2.16 (0.60; 7.71)	0.237	T2 (middle)	0.78 (0.23; 2.71)	0.700
	T3 (low)	1.60 (0.48; 5.33)	0.442	T3 (low)	1.48 (0.49; 4.50)	0.491

Odds ratios represent the odds of having albuminuria (albumin excretion ≥ 30 mg/24h) in the respective tertile of the absolute number of capillaries during post-occlusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference tertile. Model 1: unadjusted model; Model 2: age, sex; Model 3a: model 2 + waist circumference, total-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying drugs, office systolic blood pressure, use of antihypertensive medication, estimated GFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, educational level; Model 3b: as model 3a but adjusted for 24h average ambulatory systolic blood pressure instead of office systolic blood pressure (n=188). CI: confidence interval, OR: odds ratio, T: tertile

Supplemental Table 4A. Descriptive information of the tertiles of the respective capillaroscopy measures

	Percentage recruitment during post-occlusive peak reactive hyperemia (%)		Percentage recruitment during venous congestion (%)		Absolute number of capillaries during post-occlusive peak reactive hyperemia (n/mm^2)		Absolute number of capillaries during venous congestion (n/mm^2)	
Tertile	Range	≥ 30 mg/24h (n (%))	Range	≥ 30 mg/24h (n (%))	Range	≥ 30 mg/24h (n (%))	Range	≥ 30 mg/24h (n (%))
1	55.0-186.7	12 (4.9%)	55.8-253.3	9 (3.6%)	111.5-161.0	9 (3.7%)	111.5-170.0	14 (5.6%)
2	27.0-54.9	13 (5.3%)	27.6-55.7	16 (6.5%)	97.0-111.0	22 (8.8%)	97.0-111.0	16 (6.4%)
3	0.0-26.9	32 (13.0%)	-2.9-27.5	32 (13.0%)	43.0-96.5	26 (10.4%)	48.0-96.5	27 (11.2%)

Supplemental Table 4B. Descriptive information of the quintiles of the respective capillaroscopy measures

	Percentage recruitment during post-occlusive peak reactive hyperemia (%)		Percentage recruitment during venous congestion (%)		Absolute number of capillaries during post-occlusive peak reactive hyperemia (n/mm^2)		Absolute number of capillaries during venous congestion (n/mm^2)	
Quintile	Range	≥ 30 mg/24h (n (%))	Range	≥ 30 mg/24h (n (%))	Range	≥ 30 mg/24h (n (%))	Range	≥ 30 mg/24h (n (%))
1	68.8-186.7	5 (3.4%)	69.8-253.3	6 (4.1%)	118.5-161.0	7 (4.8%)	119.5-170.0	5 (3.4%)
2	49.1-68.7	11 (7.4%)	47.7-69.6	6 (4.1%)	108.0-118.0	8 (5.2%)	108.5-119.0	12 (7.9%)
3	31.7-49.0	6 (4.0%)	32.3-47.6	12 (8.1%)	99.5-107.5	12 (8.5%)	100.0-108.0	8 (5.5%)
4	18.9-31.4	11 (7.4%)	18.7-32.2	10 (6.8%)	89.5-99.0	16 (10.5%)	91.0-99.5	19 (12.7%)
5	0.0-18.7	24 (16.2%)	-2.9-18.6	23 (15.5%)	43.0-89.0	14 (9.5%)	48.0-90.5	13 (8.8%)

Supplemental Table 4C. Descriptive information of the deciles of the respective capillaroscopy measures

	Percentage recruitment during post-occlusive peak reactive hyperemia (%)		Percentage recruitment during venous congestion (%)		Absolute number of capillaries during post-occlusive peak reactive hyperemia (n/mm^2)		Absolute number of capillaries during venous congestion (n/mm^2)	
Decile	Range	≥ 30 mg/24h (n (%))	Range	≥ 30 mg/24h (n (%))	Range	≥ 30 mg/24h (n (%))	Range	≥ 30 mg/24h (n (%))
1	88.6-186.7	2 (2.7%)	89.0-253.3	2 (2.7%)	125.5-161.0	3 (3.9%)	126.5-170.0	2 (2.7%)
2	68.8-88.3	3 (4.1%)	69.8-88.3	4 (5.4%)	118.5-125.0	4 (5.6%)	119.5-126.0	3 (4.2%)
3	59.1-68.7	4 (5.4%)	59.2-69.6	2 (2.7%)	113.0-118.0	2 (2.5%)	113.0-119.0	6 (7.4%)
4	49.1-58.8	7 (9.5%)	47.7-59.0	4 (5.4%)	108.0-112.5	6 (8.1%)	108.5-112.5	6 (8.5%)
5	39.5-49.0	2 (2.7%)	39.1-47.6	3 (4.1%)	104.0-107.5	5 (6.8%)	103.5-108.0	1 (1.4%)
6	31.7-39.0	4 (5.3%)	32.3-39.0	9 (12.0%)	99.5-103.5	7 (10.3%)	100.0-103.0	7 (9.3%)
7	24.8-31.4	5 (6.8%)	25.4-32.3	5 (6.8%)	95.5-99.0	10 (12.8%)	96.0-99.5	5 (6.8%)
8	18.9-24.6	6 (8.1%)	18.7-25.3	5 (6.8%)	89.5-95.0	6 (8.1%)	91.0-95.5	14 (18.4%)
9	12.8-18.7	11 (14.9%)	12.7-18.6	14 (18.9%)	82.0-89.0	7 (9.5%)	82.0-90.5	6 (8.2%)
10	0.0-12.7	13 (17.6%)	-2.9-12.7	9 (12.2%)	43.0-81.5	7 (9.6%)	48.0-81.5	7 (9.5%)

Supplemental Table 5A. Association between the percentage recruitment during post-occlusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥ 30 mg/24h)

Model	Recruitment during post-occlusive peak reactive hyperemia (%)	OR (95% CI)	P value	Recruitment during venous congestion (%)	OR (95% CI)	P value
1	Q1	Reference		Q1	Reference	
	Q2	2.30 (0.78; 6.78)	0.132	Q2	1.00 (0.32; 3.18)	1.000
	Q3	1.20 (0.36; 4.02)	0.768	Q3	2.07 (0.76; 5.68)	0.156
	Q4	2.30 (0.78; 6.78)	0.132	Q4	1.72 (0.61; 4.85)	0.309
	Q5	5.54 (2.05; 14.94)	0.001	Q5	4.36 (1.72; 11.04)	0.002
2	Q1	Reference		Q1	Reference	
	Q2	2.03 (0.67; 6.14)	0.209	Q2	0.92 (0.29; 3.00)	0.895
	Q3	0.95 (0.28; 3.24)	0.928	Q3	1.59 (0.56; 4.46)	0.382
	Q4	1.62 (0.53; 4.94)	0.394	Q4	1.33 (0.46; 3.85)	0.601
	Q5	3.51 (1.26; 9.79)	0.016	Q5	2.74 (1.04; 7.20)	0.041
3a	Q1	Reference		Q1	Reference	
	Q2	2.23 (0.70; 7.09)	0.176	Q2	0.98 (0.29; 3.34)	0.974
	Q3	1.20 (0.34; 4.25)	0.782	Q3	1.78 (0.60; 5.26)	0.295
	Q4	1.74 (0.55; 5.52)	0.348	Q4	1.40 (0.46; 4.24)	0.548
	Q5	4.08 (1.40; 11.86)	0.010	Q5	3.11 (1.14; 8.50)	0.027
3b	Q1	Reference		Q1	Reference	
	Q2	1.59 (0.47; 5.44)	0.460	Q2	0.89 (0.24; 3.32)	0.857
	Q3	1.23 (0.34; 4.45)	0.752	Q3	1.64 (0.53; 5.03)	0.391
	Q4	1.49 (0.45; 4.90)	0.514	Q4	1.18 (0.37; 3.80)	0.782
	Q5	3.14 (1.03; 9.52)	0.044	Q5	2.74 (0.96; 7.82)	0.059

Odds ratios represent the odds of having albuminuria (defined as an albumin excretion ≥ 30 mg/24) in the respective quintile of the percentage recruitment during post-occlusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference quintile. Model 1: unadjusted model; Model 2: age, sex, type 2 diabetes; Model 3a: model 2 + waist circumference, total-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying drugs, office systolic blood pressure, use of antihypertensive medication, estimated GFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, educational level; Model 3b: as model 3a but adjusted for 24h average ambulatory systolic blood pressure instead of office systolic blood pressure (n=665). CI: confidence interval, OR: odds ratio, Q: quintile.

Supplemental Table 5B. Association between the absolute number of capillaries during post-occlusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥ 30 mg/24h)

Model	Post-occlusive peak reactive hyperemia (n/mm^2)	OR (95% CI)	P value	Venous congestion (n/mm^2)	OR (95% CI)	P value
1	Q1	Reference		Q1	Reference	
	Q2	1.10 (0.39; 3.10)	0.863	Q2	2.42 (0.83; 7.04)	0.106
	Q3	1.86 (0.71; 4.87)	0.206	Q3	1.64 (0.52; 5.12)	0.399
	Q4	2.35 (0.94; 5.90)	0.068	Q4	4.09 (1.49; 11.27)	0.006
	Q5	2.11 (0.82; 5.38)	0.120	Q5	2.74 (0.95; 7.88)	0.062
2	Q1	Reference		Q1	Reference	
	Q2	0.96 (0.33; 2.79)	0.940	Q2	1.95 (0.65; 5.85)	0.235
	Q3	1.56 (0.58; 4.20)	0.384	Q3	1.18 (0.37; 3.80)	0.785
	Q4	2.20 (0.85; 5.68)	0.105	Q4	3.40 (1.20; 9.69)	0.022
	Q5	1.50 (0.57; 3.95)	0.413	Q5	1.90 (0.64; 5.65)	0.247
3a	Q1	Reference		Q1	Reference	
	Q2	0.86 (0.28; 2.65)	0.799	Q2	1.76 (0.56; 5.54)	0.331
	Q3	1.67 (0.59; 4.74)	0.336	Q3	1.37 (0.41; 4.67)	0.605
	Q4	2.12 (0.78; 5.72)	0.139	Q4	3.23 (1.07; 9.79)	0.038
	Q5	1.55 (0.57; 4.26)	0.392	Q5	1.86 (0.60; 5.81)	0.283
3b	Q1	Reference		Q1	Reference	
	Q2	0.85 (0.26; 2.73)	0.779	Q2	1.45 (0.43; 4.91)	0.552
	Q3	1.23 (0.40; 3.80)	0.715	Q3	1.33 (0.37; 4.78)	0.661
	Q4	1.92 (0.68; 5.42)	0.216	Q4	3.33 (1.04; 10.59)	0.042
	Q5	1.45 (0.51; 4.15)	0.489	Q5	1.98 (0.61; 6.39)	0.252

Odds ratios represent the odds of having albuminuria (defined as an albumin excretion ≥ 30 mg/24h) in the respective quintile of the absolute number of capillaries during post-occlusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference quintile. Model 1: unadjusted model; Model 2: age, sex, type 2 diabetes; Model 3a: model 2 + waist circumference, total-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying drugs, office systolic blood pressure, use of antihypertensive medication, estimated GFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, educational level; Model 3b: as model 3a but adjusted for 24h average ambulatory systolic blood pressure instead of office systolic blood pressure (n=665). CI: confidence interval, OR: odds ratio, Q: quintile.

Supplemental Table 6A. Association between the percentage recruitment during post-occlusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥ 30 mg/24h)

Model	Recruitment during post-occlusive peak reactive hyperemia (%)	OR (95% CI)	P value	Recruitment during venous congestion (%)	OR (95% CI)	P value
1	D1	Reference		D1	Reference	
	D2	1.52 (0.25; 9.38)	0.651	D2	2.06 (0.37; 11.59)	0.414
	D3	2.06 (0.37; 11.59)	0.414	D3	1.00 (0.14; 7.29)	1.000
	D4	3.76 (0.76; 18.75)	0.106	D4	2.06 (0.37; 11.59)	0.414
	D5	1.00 (0.14; 7.29)	1.000	D5	1.52 (0.25; 9.38)	0.651
	D6	2.02 (0.36; 11.43)	0.423	D6	4.91 (1.02; 23.55)	0.047
	D7	2.61 (0.49; 13.90)	0.261	D7	2.61 (0.49; 13.90)	0.261
	D8	3.18 (0.62; 16.28)	0.166	D8	2.61 (0.49; 13.90)	0.261
	D9	6.29 (1.34; 29.44)	0.020	D9	8.40 (1.84; 38.43)	0.006
	D10	7.67 (1.67; 35.34)	0.009	D10	4.99 (1.04; 23.82)	0.045
2	D1	Reference		D1	Reference	
	D2	1.46 (0.23; 9.24)	0.689	D2	1.91 (0.33; 11.05)	0.472
	D3	1.98 (0.34; 11.49)	0.445	D3	0.91 (0.12; 6.77)	0.924
	D4	2.96 (0.57; 15.26)	0.195	D4	1.80 (0.31; 10.40)	0.512
	D5	0.75 (0.10; 5.59)	0.776	D5	1.16 (0.18; 7.40)	0.873
	D6	1.16 (0.28; 9.35)	0.594	D6	3.50 (0.71; 17.33)	0.125
	D7	1.64 (0.30; 9.01)	0.572	D7	1.94 (0.35; 10.64)	0.445
	D8	2.45 (0.46; 13.03)	0.293	D8	1.96 (0.36; 10.81)	0.439
	D9	3.92 (0.81; 19.07)	0.091	D9	4.70 (0.98; 22.50)	0.053
	D10	4.75 (1.00; 22.61)	0.050	D10	3.35 (0.68; 16.60)	0.139

Odds ratios represent the odds of having albuminuria (defined as an albumin excretion ≥ 30 mg/24) in the respective decile of the percentage recruitment during post-occlusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference decile. Model 1: unadjusted model; Model 2: age, sex, type 2 diabetes. CI: confidence interval, OR: odds ratio, D: decile.

Supplemental Table 6B. Association between the absolute number of capillaries during post-occlusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥ 30 mg/24h)

Model	Post-occlusive peak reactive hyperemia (n/mm^2)	OR (95% CI)	P value	Venous congestion (n/mm^2)	OR (95% CI)	P value
1	D1	Reference		D1	Reference	
	D2	1.45 (0.31; 6.73)	0.633	D2	1.57 (0.25; 9.66)	0.629
	D3	0.62 (0.10; 3.84)	0.611	D3	2.88 (0.56; 14.74)	0.204
	D4	2.15 (0.52; 8.93)	0.293	D4	3.32 (0.65; 17.05)	0.150
	D5	1.79 (0.41; 7.77)	0.438	D5	0.51 (0.05; 5.80)	0.591
	D6	2.79 (0.69; 11.26)	0.140	D6	3.71 (0.74; 18.47)	0.110
	D7	3.58 (0.95; 13.56)	0.061	D7	2.61 (0.49; 13.90)	0.261
	D8	2.15 (0.52; 8.93)	0.293	D8	8.13 (1.78; 37.17)	0.007
	D9	2.54 (0.63; 10.23)	0.189	D9	3.22 (0.63; 16.53)	0.160
	D10	2.58 (0.64; 10.39)	0.182	D10	3.76 (0.76; 18.75)	0.106
2	D1	Reference		D1	Reference	
	D2	1.83 (0.38; 8.92)	0.452	D2	2.04 (0.32; 13.16)	0.452
	D3	0.59 (0.09; 3.78)	0.581	D3	2.60 (0.49; 13.81)	0.261
	D4	2.16 (0.50; 9.38)	0.304	D4	3.08 (0.58; 16.51)	0.188
	D5	1.49 (0.33; 6.75)	0.606	D5	0.45 (0.04; 5.20)	0.521
	D6	2.96 (0.70; 12.54)	0.142	D6	2.88 (0.56; 14.94)	0.208
	D7	3.92 (0.99; 15.61)	0.053	D7	2.57 (0.46; 14.24)	0.280
	D8	2.12 (0.49; 9.23)	0.316	D8	7.42 (1.56; 35.41)	0.012
	D9	1.98 (0.49; 8.78)	0.349	D9	2.67 (0.50; 14.79)	0.249
	D10	2.08 (0.49; 8.78)	0.319	D10	2.85 (0.55; 14.79)	0.213

Odds ratios represent the odds of having albuminuria (defined as an albumin excretion ≥ 30 mg/24h) in the respective decile of the absolute number of capillaries during post-occlusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference decile. Model 1: unadjusted model; Model 2: age, sex, type 2 diabetes. CI: confidence interval, OR: odds ratio, D: decile.

Supplemental Table 7A. Association between the percentage recruitment during post-occlusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥ 15 mg/24h)

Model	Recruitment during post-occlusive peak reactive hyperemia (%)	OR (95% CI)	P value	Recruitment during venous congestion (%)	OR (95% CI)	P value
1	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	1.10 (0.67; 1.83)	0.700	T2 (middle)	1.27 (0.76; 2.11)	0.364
	T3 (low)	2.19 (1.38; 3.47)	0.001	T3 (low)	2.54 (1.59; 4.07)	< 0.001
2	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	0.86 (0.50; 1.46)	0.571	T2 (middle)	1.09 (0.64; 1.86)	0.751
	T3 (low)	1.62 (0.99; 2.65)	0.055	T3 (low)	1.88 (1.14; 3.10)	0.014
3a	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	0.92 (0.53; 1.60)	0.775	T2 (middle)	1.10 (0.63; 1.91)	0.743
	T3 (low)	1.59 (0.96; 2.64)	0.075	T3 (low)	1.75 (1.05; 2.95)	0.033
3b	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	1.03 (0.57; 1.84)	0.931	T2 (middle)	1.16 (0.65; 2.08)	0.622
	T3 (low)	1.67 (0.97; 2.88)	0.067	T3 (low)	1.79 (1.03; 3.11)	0.040

Odds ratios represent the odds of having albuminuria (defined as an albumin excretion ≥ 15 mg/24) in the respective tertile of the percentage recruitment during post-occlusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference tertile. Model 1: unadjusted model; Model 2: age, sex, type 2 diabetes; Model 3a: model 2 + waist circumference, total-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying drugs, office systolic blood pressure, use of antihypertensive medication, estimated GFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, educational level; Model 3b: as model 3a but adjusted for 24h average ambulatory systolic blood pressure instead of office systolic blood pressure (n=665). CI: confidence interval, OR: odds ratio, T: tertile.

Supplemental Table 7B. Association between the absolute number of capillaries during post-occlusive peak reactive hyperemia as well as venous congestion and the presence of albuminuria (albumin excretion ≥ 15 mg/24h)

Model	Post-occlusive peak reactive hyperemia (n/mm^2)	OR (95% CI)	P value	Venous congestion (n/mm^2)	OR (95% CI)	P value
1	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	1.71 (1.05; 2.79)	0.031	T2 (middle)	1.31 (0.81; 2.14)	0.274
	T3 (low)	1.88 (1.17; 3.04)	0.011	T3 (low)	1.99 (1.25; 3.18)	0.004
2	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	1.57 (0.94; 2.63)	0.084	T2 (middle)	1.15 (0.69; 1.93)	0.594
	T3 (low)	1.54 (0.92; 2.56)	0.098	T3 (low)	1.72 (1.05; 2.83)	0.031
3a	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	1.75 (1.02; 2.99)	0.042	T2 (middle)	1.28 (0.75; 2.18)	0.372
	T3 (low)	1.64 (0.96; 2.80)	0.071	T3 (low)	1.78 (1.06; 2.99)	0.029
3b	T1 (high)	Reference		T1 (high)	Reference	
	T2 (middle)	1.45 (0.82; 2.57)	0.205	T2 (middle)	1.17 (0.66; 2.09)	0.591
	T3 (low)	1.61 (0.92; 2.80)	0.095	T3 (low)	1.92 (1.11; 3.33)	0.019

Odds ratios represent the odds of having albuminuria (defined as an albumin excretion ≥ 15 mg/24h) in the respective tertile of the absolute number of capillaries during post-occlusive peak reactive hyperemia or venous congestion compared with the odds of having albuminuria in the reference tertile. Model 1: unadjusted model; Model 2: age, sex, type 2 diabetes; Model 3a: model 2 + waist circumference, total-to-HDL cholesterol ratio, triglycerides, use of lipid-modifying drugs, office systolic blood pressure, use of antihypertensive medication, estimated GFR, prevalent cardiovascular disease, smoking behavior, alcohol consumption, educational level; Model 3b: as model 3a but adjusted for 24h average ambulatory systolic blood pressure instead of office systolic blood pressure (n=665). CI: confidence interval, OR: odds ratio, T: tertile.