Retinal arteriolar caliber and urine albumin excretion: the Multi-Ethnic Study of Atherosclerosis

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Abstract

Background. Changes in retinal microvascular caliber, which occur prior to onset of retinopathy, may indicate presence of kidney damage.

Methods. This study examined the association between retinal arteriolar [central retinal artery equivalent (CRAE)] and venular caliber [central retinal venule equivalent (CRVE)] and presence of albuminuria (micro- or macroalbuminuria) among participants of the Multi-Ethnic Study of Atherosclerosis (MESA), a cohort of adults aged 45–84 years without baseline clinical cardiovascular disease. During the second MESA exam, digital fundus photography was completed in 5897 participants who provided spot urine specimens. Albuminuria was defined by spot urine albumin/creatinine ratios ≥30 mg/g. Multivariable adjusted odds of albuminuria by quintiles of CRAE and CRVE were determined using logistic regression. Analyses were repeated after stratifying by presence of type 2 diabetes.

Results. Albuminuria was noted in 11.5% (n = 675) and included 584 subjects with microalbuminuria and 91 with macroalbuminuria. A significant U-shaped pattern was seen with higher prevalence of albuminuria across quintile extremes in CRAE (15.7, 8.8 and 10.6% in CRAE Quintiles 1, 3 and 5, respectively; P < 0.0001). After adjustment for covariates, both narrower CRAE [odds ratios (OR) 1.55; 95% confidence interval (CI) 1.17–2.04, Quintile 1 versus 3] and wider CRAE (OR 1.44; 95% CI 1.07–1.93, Quintile 5 versus 3) were significantly associated with albuminuria. Associations appeared substantially stronger in adults with than without type 2 diabetes but the interaction term for diabetes and CRAE on presence of albuminuria did not meet statistical significance (P = 0.3). No association was noted between CRVE quintiles and albuminuria.

Conclusions. Albuminuria is associated with narrower and wider arteriolar caliber. Future studies should determine whether variation in arteriolar caliber predicts incident albuminuria and whether associations are mediated by hypertension and diabetes. Such information could further clarify early microvascular processes in the pathogenesis of kidney disease.

Keywords: albuminuria; diabetic retinopathy; MESA (Multi-Ethnic Study of Atherosclerosis); retinal arteriolar; retinal venular

Introduction

Fundoscopic examination of the retina constitutes part of the recommended routine physical examination of any adult with newly diagnosed hypertension or type 2 diabetes [1, 2]. Pathologic changes (e.g. retinopathy, disc edema) may be observed with clinical fundoscopic examination of a dilated pupil, but more subtle changes will likely go undetected. Glomerular and retinal arterioles are small vessels, which may be mutually affected by systemic diseases including diabetes and hypertension. Severe retinopathy has been well described in individuals with kidney disease and type 1 diabetes [3, 4] but retinopathy is not universally present in adults with kidney disease in the absence of type 1 diabetes [5–9].

Digital fundus photography now allows for the early recognition of retinal microvascular changes, which may occur before the onset of retinopathy. Measurement of retinal microvascular caliber is emerging as an important marker of many vascular conditions, including coronary heart disease [10], stroke [11], diabetes [12] and retinopathy [13]. However, the association between retinal microvascular caliber and early kidney dysfunction or presence of micro- or macroalbuminuria has not been well established.
The direct noninvasive visualization of the retinal microvasculature offers the opportunity to further study the relationship between systemic microvascular disease and early glomerular damage as assessed by presence of increased urine albumin excretion. In this study, we examined the association between retinal microvascular caliber and albuminuria (micro-/macroalbuminuria) in an adult population without clinical cardiovascular disease (CVD) at baseline. We hypothesized that narrower retinal arteriolar and venular calibers are associated with the presence of albuminuria. Because kidney disease and retinal microvascular changes frequently cosegregate in adults with type 2 diabetes, we explored whether the association between retinal arteriolar and venular caliber and albuminuria differs by presence of diabetes.

Methods

Population

The Multi-Ethnic Study of Atherosclerosis (MESA) is a prospective cohort study of 6814 men and women aged 45–84 years (mean age 63.2 years), without clinical CVD, recruited from 6 US communities (Baltimore, MD; Chicago, IL; Forsyth County, NC; Los Angeles County, CA; northern Manhattan, NY and St. Paul, MN). MESA is designed to determine the characteristics of subclinical CVD and its progression. Adults with symptoms or history of medical or surgical treatment for CVD were excluded. Information on the sampling frame and study design has been previously reported [14].

Participants who self-reported their race/ethnicity group as Caucasian or white, African-American or black, Chinese or Spanish/Hispanic/Latino were asked to participate and were enrolled between July 2000 and August 2002. Institutional review board approval was obtained at all MESA sites and informed consent was obtained from all participants.

Participants completed exams approximately every 18 months after the baseline exam and fundus photography was performed during Exam 2. Among the 6814 participants from the baseline exam, 6233 participants (91.5%) participated in Exam 2 and 5979 had adequate measurements on retinal vascular caliber. We subsequently excluded MESA participants with missing information on spot urine albumin/creatinine ratio (ACR) (n = 82) at Exam 2. Accordingly, the analysis was limited to the 5897 MESA participants with complete information on retinal vascular caliber from fundus photography and spot urine ACR.

Albuminuria

During Exam 2, participants provided a spot urine specimen generally immediately after arriving in the morning at the MESA clinic site. Urine albumin and creatinine were measured at the Clinical Chemistry Laboratory at Fletcher Allen Health Care (Burlington, VT) by nephelometry and the Jaffe reaction, respectively. A spot urine ACR in milligrams/grams was then calculated for all participants. To account for sex differences in urine creatinine excretion, we multiplied the ACR in men by 1.47 [15]. Albuminuria was then defined as an ACR ≥ 30 mg/g for men and women. This definition includes both microalbuminuria (ACR ≥ 30–299 mg/g) and macroalbuminuria (ACR ≥ 300 mg/g), which were grouped together due to the small number of individuals with macroalbuminuria.

Retinal photography

Retinal photography was performed using a standardized protocol [16, 17]. Using a 45-degree 6.3-megapixel digital nonmydriatic camera, photographic fields of optic disc and macula of both eyes of each participant were photographed. These photographs were sent from all six centers to a central site at the University of Wisconsin-Madison, for measurement of retinal vascular caliber and evaluation of other retinal pathology. Trained graders at these centralized sites were blinded to participant characteristics [18, 19]. For each image, all arterioles and venules coursing through an area one-half to one-disc diameter from the optic disc margin were measured using a computer-based program. Retinal arteriolar caliber was summarized as the central retinal artery equivalent (CRAE), while retinal venular caliber was summarized as central retinal vein equivalent (CRVE).

[18, 20]. The CRAE and CRVE equivalents are the projected caliber for the central retinal artery/vein, measured away from the optic disc. The CRAE and CRVE were measured in the right eye, but these measurements were made in the left eye when the right eye could not be assessed. Retinopathy was defined as the presence of any of the following lesions graded as definite or probable: microaneurysms, retinal hemorrhages, soft or hard exudates or vitreous hemorrhage.

Covariates

All MESA participants completed self-administered questionnaires and were interviewed and examined by trained research staff at each exam. Fasting blood specimens and spot urine specimens were collected in the morning when the participant first arrived for each MESA exam. Blood pressure was measured three times at 1-min intervals using a Dinamap PRO 100 automated oscillometric device. The average of the second and third blood pressure measurements was used for this analysis. Presence of diabetes at Exam 2 was defined as self-reported physician diagnosis, use of insulin or oral hypoglycemic agents or fasting glucose 126 mg/dL. Medication use for high blood pressure was defined as self-reported treatment with one of six common classes of antihypertensive medications [thiazide diuretics, beta blockers, calcium channel blockers, angiotensin-converting enzyme inhibitors (ACE), angiotensin-2 receptor blockers (ARB) or other (alpha blockers or peripheral vasodilators)]. Current smoking status was based on self-report. Waist circumference was measured to the nearest 0.1 cm at the high point of the iliac crest while participants were standing. Information on socioeconomic factors including highest degree or level of school completed and health insurance (private health insurance, Health Maintenance Organization, Medicaid, Medicare, veteran’s health care or none) was collected using questionnaires.

Statistical analysis

Means and SDs were used to summarize the characteristics of the study sample by CRAE quintiles. Continuous and categorical variables were compared across quintiles of CRAE using analysis of variance (ANOVA) and chi-square tests, respectively. If the ANOVA test was significant, upper CRAE Quintiles 2–5 were compared to CRAE Quintile 1. The level for statistical significance was set at P < 0.01 (0.05/4) to account for multiple comparisons (Quintile 1 versus Quintiles 2–5).

A nonlinear association was noted between CRAE and albuminuria using a second order fractional polynomial approach [21]. In a logistic regression model with albuminuria as the dependent variable, CRAE as a continuous variable (P = 0.004) and the quadratic term for CRAE (P = 0.005) were both significantly associated with albuminuria. Similar findings were noted for CRVE. To examine the nonlinear association between albuminuria and CRAE and CRVE, both CRAE and CRVE were divided into quintiles and odds ratios (OR) of albuminuria by CRAE and CRVE quintiles were calculated using logistic regression while simultaneously adjusting for confounders. The association between CRAE quintiles and albuminuria was U-shaped and the middle (third) quintile was selected as the referent group.

Several models were created to assess potential confounding. Several different logistic regression models were examined so that changes in the parameter estimate with the addition of demographic and socioeconomic factors could be examined. Covariates for these models were selected based on previously published reports [12, 13, 22, 23]. Model 1 adjusted for age, sex and race/ethnicity. Model 2 added systolic blood pressure and use of antihypertensive medications including ACE/ARB and Model 3 added presence of diabetes (among total participants only). Model 4 then adjusted for all covariates including current smoking, waist circumference, education and access to healthcare. Retinal venular caliber (CRVE) was not included as a covariate in the final model because it did not confound the association between retinal arteriolar caliber and albuminuria. Models were then repeated after stratifying MESA participants by presence of diabetes. In all models, age and systolic blood pressure levels were fitted as continuous variables, whereas current smoking, diabetes and use of antihypertensive medications were included as binary variables. Effect modification by sex, race/ethnicity, diabetes and presence of hypertension on the association between CRAE and albuminuria was examined in the full model using interaction terms.

Results

The mean age of the 5897 MESA participants at Exam 2 was 63.2 (9.8) years and 47.9% were male. Diabetes,
hypertension and current smoking were present in 14.5, 48.7 and 11.1%, respectively. Albuminuria was present in 11.5% (n = 675) and included 584 with microalbuminuria and 91 with macroalbuminuria. The characteristics of the study participants by CRAE quintiles are shown in Table 1. Participants in the narrowest CRAE (Quintile 1) were older, more likely to be male and more likely to be hypertensive compared to the CRAE Quintiles 2–5. Concurrently, participants with the widest CRAE (Quintile 5) were more likely to have diabetes and currently smoke compared to participants in the lower CRAE Quintiles 1–4. Both systolic and diastolic blood pressures were highest in Quintile 1 and lowest in Quintile 5. The prevalence of any retinopathy ranged from ~10–11% across CRAE quintiles while frequency of arteriovenous (AV) nicking ranged from 5.0% in CRAE Quintiles 1–2.8% in CRAE Quintile 4.

Figure 1 shows the prevalence of albuminuria by CRAE quintiles among the MESA participants. Prevalence of albuminuria was highest in CRAE Quintile 1 and lowest in CRAE Quintile 3. Among participants with diabetes, 42.6% of individuals in CRAE Quintile 1 had albuminuria including 35.9% with microalbuminuria and 6.7% with macroalbuminuria. The lowest prevalence of albuminuria was noted in CRAE Quintile 3. Among participants without diabetes, prevalence of albuminuria ranged from 11.3% in CRAE Quintiles 1–6.7% in CRAE Quintile 3. Prevalence of albuminuria was fairly similar across CRVE quintiles ranging from 12.7% in Quintile 2–16.1% in Quintile 5.

Table 2 shows the results of the logistic regression analyses for CRAE quintiles. Among the total MESA group, narrower CRAE (Quintile 1) was associated with a 71% increased odds of albuminuria compared to CRAE Quintile 3 in the model adjusted for age, sex and race (Model 1). Further adjustment for demographic factors, presence of diabetes, systolic blood pressure and use of antihypertensive medications substantially reduced the parameter estimate. In the fully adjusted model (Model 4), narrower CRAE (Quintile 1) was associated with a 55% increased odds of albuminuria compared to CRAE Quintile 3 [95% confidence interval (CI) 1.17–2.04]. Concurrently, wider CRAE (Quintile 5) was also associated with a significantly increased odds of albuminuria compared to CRAE Quintile 3 after adjustment for all covariates (OR 1.44; 95% CI 1.07–1.93). No significant association was noted between CRVE quintiles and presence of albuminuria in any of the models (data not shown).
Interaction terms showed no significant modification by presence of hypertension, sex or race on the association between CRAE and albuminuria. The interaction term for diabetes and CRAE on presence of albuminuria was also not statistically significant (P = 0.3). However, due to the established clinical importance that diabetes holds for risk of albuminuria, we further explored the association between CRAE quintiles and presence of albuminuria in analyses stratified by presence of diabetes. Among participants without diabetes, findings were similar compared to associations noted in the total MESA population with the highest and lowest CRAE quintiles both associated with an increased odds of albuminuria compared to CRAE Quintile 3. (Table 3) However, associations were less robust compared to the total MESA population in the model with all covariates (OR 2.25; 95% CI 1.13–3.81). The association between CRAE quintiles and albuminuria was somewhat less U-shaped in participants with diabetes compared to the total MESA participant group. However, a 64% increased odds of albuminuria was noted in wider CRAE (Quintile 5) compared to CRAE Quintile 3 in this group.

### Discussion

In this study, we noted a U-shaped distribution of albuminuria prevalence across the range of retinal arteriolar caliber with the higher prevalence of albuminuria consistently seen amongst participants with narrower or wider CRAE. In part, this may be explained because hypertension was more prevalent among individuals with the narrowest retinal arteriolar caliber (CRAE Quintile 1), while diabetes was more prevalent among individuals with the widest retinal arteriolar caliber (CRAE Quintile 5). These findings are supported by previous studies that reported associations between the extreme distributions of retinal vascular caliber and disease risk. For example, retinal arteriolar narrowing has been associated with incident diabetes [12], hypertension [23] and coronary heart disease [24]. Wider retinal arteriolar caliber has been linked with incident retinopathy in adults with diabetes [13]. Retinal arteriolar narrowing is likely a marker of cumulative hypertension exposure [25] and among adults with type 2 diabetes, hypertension burden likely plays a strong role in the progression of glomerular damage, which may begin prior to the onset of diabetes [26].

Hypertensive retinopathy was first described in a patient with both hypertension and kidney disease; the retinopathy in this patient was severe with multiple retinal hemorrhages and optic disc swelling [27]. Such severe changes are strongly associated with kidney failure and cardiovascular mortality, but are uncommon [27]. What remains unclear is whether change in care after detection of early retinal vascular changes will prevent end-organ damage elsewhere. The retinal vasculature is similar to the glomerular vascular bed in that vascular tone increases in response to elevated blood pressure due to auto-regulatory mechanisms [27]. If hypertension persists,
intimal thickening and hyperuricemia of the media wall ensues. At this point, direct visualization of the retinal arteriole may lead to detection of AV nicking, which is classified as mild hypertensive retinopathy. If left untreated, this stage may be followed by development of hemorrhages, microaneurysms and exudates, classified as moderate hypertensive retinopathy [27] that has been associated with decline in glomerular filtration rate [9]. Unlike the retinal vasculature, the glomerular vascular bed cannot be directly visualized. However, albuminuria has been associated with multiple risk factors including increased systolic blood pressure [28–30]. Recent studies indicate that albuminuria not only indicates presence of kidney disease but also a higher risk for cardiovascular events in adults with diabetes or hypertension [31–33] similar to associations noted with narrow retinal arteriolar caliber [24, 34]. Thus, presence of albuminuria may indicate microvascular damage within the glomerulus from cumulative exposure to hypertension and other factors.

In this study, narrow retinal arteriolar caliber showed a stronger association with albuminuria in adults with type 2 diabetes compared to adults without type 2 diabetes. This may be due to the higher overall risk of developing glomerular damage due to the combined effects of multiple systemic processes including hypertension and hyperglycemia among adults with type 2 diabetes. This is supported by the higher overall prevalence of albuminuria among MESA participants with type 2 diabetes compared to those without diabetes. Cross-sectional studies demonstrate a strong correlation between blood pressure and levels of urine albumin excretion in non-diabetic persons [29, 35, 36] including those with prehypertension [30]. In addition, lowering of blood pressure leads to reduction of urine albumin excretion in adults with either hypertension or diabetes [37, 38]. However, the interaction between diabetes and CRAE quintiles on albuminuria was not statistically significant and these findings may be due to chance.

The strengths of this study include the standardized measurement of retinal vascular caliber among MESA participants and the large number of participants from four separate racial/ethnic groups. Other strengths include the use of CRAE and CRVE models separately in the MESA study, as opposed to using the arteriole to venule ratio. These separate models likely provided more unbiased and biological plausible results because risk factors for CRAE (e.g. hypertension) and CRVE (e.g. diabetes and body mass index) are now known to be different [39, 40]. Nevertheless, retinal vessel diameter does not reflect vascular function such as blood flow or endothelial function.

The use of a single spot urine specimen to estimate albuminuria at Exam 2 may have led to misclassification of albuminuria due to substantial intra-individual variability from day to day. However, the misclassification of albuminuria likely does not differ by retinal arteriolar caliber and would have biased the results toward the null. Although MESA is a fairly large cohort, power was limited to detect significant interactions between diabetes and CRAE quintiles on presence of albuminuria. Other limitations include the cross-sectional design which precludes the elucidation of temporal associations. It is possible that kidney disease precedes the development of changes in the CRAE. However, a recent study demonstrated an independent association between baseline CRAE and changes in a summary index of kidney mesangium matrix accumulation over a 5-year period among persons with type 1 diabetes [41]. Future studies should explore the association between retinal arteriolar caliber and progression of urine albumin excretion in adults with and without type 2 diabetes.

In summary, we noted a U-shaped association between retinal arteriolar caliber and prevalence of albuminuria in a large multi-ethnic sample of adults. Although many pathways for the development of albuminuria likely exist, the link between narrow retinal arteriolar caliber and albuminuria suggests that systemic processes such as hypertension may lead to parallel vascular changes in the retinal and glomerular microvasculature, while the relationship between wider retinal arteriolar caliber and albuminuria may reflect diabetes-related pathways. Studying retinal microvascular caliber may therefore further illuminate early microvascular disease processes in the pathogenesis of kidney disease.

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**References**
