Relationship of Retinal Vascular Caliber With Diabetes and Retinopathy

The Multi-Ethnic Study of Atherosclerosis (MESA)

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OBJECTIVE — To examine the relationship of retinal vascular caliber with diabetes, glycemia, and diabetic retinopathy.

RESEARCH DESIGN AND METHODS — Population-based study using data from the Multi-Ethnic Study of Atherosclerosis (MESA), comprising 5,976 individuals (whites, blacks, Hispanics, and Chinese) residing in six U.S. communities who were free of clinical cardiovascular disease at baseline. Retinal vascular caliber was measured from digital retinal photographs.

RESULTS — There were 4,585 individuals with normal fasting glucose (NFG), 499 with impaired fasting glucose (IFG), 165 with diabetes with retinopathy signs, and 727 with diabetes without retinopathy signs. After multivariate analysis, retinal arteriolar caliber increased from 143.8 μm in subjects with NFG to 144.5 μm in IFG and 146.1 μm in diabetes ($P < 0.001$ for trend). Retinal venular caliber increased from 214.4 μm in NFG to 216.7 μm in IFG and 218.0 μm in diabetes ($P < 0.001$ for trend). Retinal venular caliber was significantly larger with increasing levels of fasting glucose and A1C. In a subgroup analysis by ethnicity, the association between wider arteriolar caliber and diabetes was evident in whites only, whereas wider venular caliber and diabetes was evident in Hispanics and Chinese only. In people with diabetes, eyes with retinopathy had larger retinal venular but not arteriolar caliber.

CONCLUSIONS — Retinal arteriolar and venular calibers are larger in individuals with diabetes, but the pattern of associations appears to vary by ethnicity. Retinal venular caliber is additionally associated with retinopathy signs. These findings add further to the concept that variations in retinal vascular caliber may reflect early diabetic microvascular damage.

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The retinal blood vessels are accessible to direct noninvasive visualization. There is increasing evidence that changes in retinal vascular caliber may be markers of early microvascular dysfunction associated with diabetes, prediabetes, and diabetes complications (1,2). However, although there have been a number of studies reporting various associations of retinal vascular caliber, the specific changes in arteriolar and venular caliber size with glycemic levels remain unclear. Early studies, for example, evaluated associations with the ratio of the retinal arteriolar to venular caliber (AV ratio), initially thought to reflect smaller arteriolar caliber. These studies showed that a smaller AV ratio was related to the development of type 2 diabetes (3,4). However, other studies subsequently found that these associations are driven by venular caliber, and one study demonstrated that larger retinal venular caliber was associated with the incidence of impaired fasting glucose and possibly diabetes (5). Associations of retinal arteriolar and venular caliber with diabetic microvascular complications, such as retinopathy, have also been inconsistent (6,7). Furthermore, it is now recognized that because arteriolar and venular calibers are highly correlated, statistical analysis of retinal vascular caliber should account for this correlation (8).

To address these issues, we assessed the associations of retinal arteriolar and venular caliber with the full spectrum of glycemic-related disorders and complications, ranging from impaired fasting glucose and clinically diagnosed diabetes to the presence of diabetic retinopathy in a multiethnic population-based cohort.

RESEARCH DESIGN AND METHODS

— The Multi-Ethnic Study of Atherosclerosis (MESA) is a prospective cohort study of men and women aged 45–84 years comprising four racial/ethnic groups (whites, blacks, Hispanics, and Chinese). Participants have no history of clinical cardiovascular disease at baseline and are residents of six U.S. communities (9). Tenets of the Declaration of Helsinki were followed, and institutional review board approval was granted at each study site. Written informed consent was obtained from each participant.

At the first examination, there were 6,814 participants. Retinal photography was done at the second examination, which immediately followed the baseline examination, from August 2002 to January 2004 (10). At the second examination, 6,237 returned, 6,147 had retinal photographs for grading retinopathy, and 5,976 (97.3%) had photographs that

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Abbreviations: CRAE, central retinal artery equivalent; CRP, C-reactive protein; CRVE, central retinal vein equivalent; MESA, Multi-Ethnic Study of Atherosclerosis; NFG, normal fasting glucose; WESDR, Wisconsin Epidemiological Study of Diabetic Retinopathy.

Table 1—*Characteristics of participants in MESA*

	Glucose status (n = 5,976)				Diabetes (n = 892)		
	NFG	IFG	Diabetes	P*	No retinopathy	Retinopathy	P†
	n = 4,585	n = 499	n = 892		n = 727	n = 165	
Sex (male)	2,110 (46.0)	287 (57.5)	465 (52.1)	<0.001	382 (52.3)	83 (51.6)	0.87
Alcohol consumption	2,476 (54.2)	270 (54.3)	328 (37.0)	<0.001	271 (37.3)	57 (35.6)	0.70
Current cigarette smoker	553 (12.1)	43 (8.6)	97 (10.9)	0.06	86 (11.8)	11 (6.8)	0.07
Hypertension	1,947 (42.5)	300 (60.1)	654 (73.3)	<0.001	524 (71.7)	130 (80.7)	0.02
Oral diabetic medication	—	—	562 (64.8)	—	449 (63.2)	113 (72.0)	0.04
Insulin	—	—	97 (11.2)	—	58 (8.2)	39 (24.8)	<0.001
Age (years)	62.7 ± 10.1	63.9 ± 9.5	65.2 ± 9.2	<0.001	65.5 ± 9.2	63.8 ± 9.0	0.04
Diabetes duration (years)	—	—	5.3 ± 7.7	—	4.2 ± 6.8	10.3 ± 9.6	<0.001
Serum glucose (mg/dl)	94.6 ± 8.3	115.4 ± 4.4	154.2 ± 54.3	<0.001	149.9 ± 50.4	173.7 ± 66.0	<0.001
Systolic blood pressure (mmHg)	122.4 ± 20.2	127.7 ± 21.3	130.5 ± 21.4	<0.001	129.7 ± 20.2	134.1 ± 25.9	0.02
Diastolic blood pressure (mmHg)	70.3 ± 10.0	72.1 ± 10.1	70.6 ± 10.4	0.001	70.8 ± 10.2	69.5 ± 11.2	0.13
BMI (kg/m^2)	27.7 ± 5.2	30.5 ± 5.6	30.9 ± 6.0	<0.001	30.8 ± 5.9	31.2 ± 6.4	0.46
Total cholesterol (mg/dl)	193.3 ± 34.9	191.4 ± 38.1	181.8 ± 37.3	<0.001	181.7 ± 36.4	182.5 ± 41.3	0.81
HDL cholesterol, mg/dl	53.3 ± 15.4	48.0 ± 13.4	46.5 ± 12.8	<0.001	46.5 ± 13.0	46.5 ± 11.8	0.98
LDL cholesterol (mg/dl)	115.3 ± 31.8	113.9 ± 33.6	105.0 ± 32.0	<0.001	104.5 ± 31.1	107.1 ± 35.9	0.35
Triglycerides (mg/dl)	124.8 ± 72.2	148.6 ± 90.2	158.4 ± 114.4	<0.001	160.0 ± 114.3	151.2 ± 114.7	0.38
CRP (mg/dl)‡	1.70 ± 3.18	2.31 ± 3.67	2.57 ± 4.67	<0.001	2.58 ± 3.26	2.53 ± 4.64	0.89

Data are n (%) or means ± SD and are shown as median and interquartile range. *P based on χ^2 (categorical), ANOVA (quantitative and normal), or Kruskal-Wallis test (quantitative and skewed), comparing NFG, impaired fasting glucose (IFG), and diabetes. †P based on χ^2 (categorical), ANOVA (quantitative and normal), or Kruskal-Wallis test (quantitative and skewed), comparing diabetes with and without retinopathy. ‡Results are shown as median and interquartile range.

were suitable for measurement of retinal vascular caliber.

Measurement of retinal vascular caliber

Retinal photography was performed using a standardized protocol (11). Both eyes of each participant were photographed using a 45-degree 6.3-megapixel digital nonmydriatic camera. Two photographic fields (optic disc and macula) were taken of each eye. Images were sent from the six field centers to the University of Wisconsin, Madison, for measurement of retinal vascular caliber and assessment of other retinal pathology.

Retinal vascular caliber was measured using a computer-based program by trained graders who were masked to participant characteristics, based on a detailed protocol (11). Photographs in the right eye were selected for measurement; the left eye was chosen if measurements could not be performed in the right eye. For each image, all arterioles and venules coursing through an area one-half to one-disc diameter from the optic disc margin were measured and summarized as the central retinal artery equivalent (CRAE) and central retinal vein equivalent (CRVE) (10,11). These equivalents are projected calibers for the central retinal vessels, measured away from the optic disc. Reproducibility of these measurements has been reported, with intra- and intergrader intraclass correlation coefficients ranging from 0.78 to 0.99 (10).

Definition of diabetic retinopathy

Diabetic retinopathy assessment has been previously published. For each eye, a retinopathy severity score was assigned based on modification of the Airlie House Classification system (12). Levels 14–20 were defined as minimal retinopathy, and levels >20 (levels 31–80) were defined as early to severe diabetic retinopathy. A person's retinopathy level was based on the scores in the right eye, as most of the retinal vascular caliber measurements were obtained from this eye. Six eyes with proliferative retinopathy were excluded from analysis because of previous laser treatment, which may have an effect on vascular calibers (6,7).

Assessment of diabetes

Diabetes was defined as fasting glucose ≥7.0 mmol/l (126 mg/dl) or use of insulin or oral hypoglycemic medication (13). No distinction was made between type 1 and type 2 diabetes. Impaired fasting glucose (IFG) was defined as a fasting glucose level of 6.1–6.9 mmol/l (110–125 mg/dl). All other participants were defined as having normal fasting glucose (NFG).

Assessment of other risk factors

A detailed questionnaire was used to obtain participant information, including past medical history, cigarette smoking, and alcohol consumption status (9). Hypertension was defined as systolic blood pressure ≥140 mmHg, diastolic blood pressure ≥90 mmHg, or current use of antihypertensive medications. Height and weight were measured to determine BMI. Fasting (>8 h) blood samples were drawn from participants, and aliquots were prepared for central analysis. The following were analyzed in this report: plasma total and HDL cholesterol, plasma triglycerides, serum glucose, A1C, and C-reactive protein (CRP). LDL cholesterol was calculated with the Friedewald equation.

Statistical analysis

Retinal vascular caliber (CRAE and CRVE) was normally distributed in the population. We used ANCOVA to estimate mean CRAE/CRVE for categories of NFG, IFG, and diabetes and used linear regression to determine the linear relationship of CRAE/CRVE with fasting glucose and A1C levels. We constructed the following models: model 1, adjusted for age, sex, race/ethnicity, and examination center, and model 2, adjusted for variables in model 1 plus systolic blood pressure, BMI, total cholesterol, triglycerides, current smoking, and CRP. In model 3,

Table 2—*Relationship of retinal arteriolar and venular caliber with fasting glucose status: all individuals and by racial/ethnic groups*

	Retinal vascular caliber (μm)			
	NFG	IFG	Diabetes	*P* for trend
n	4,585	499	892	
Retinal arteriolar caliber (CRAE)				
Model 1*	144.0 ± 0.24	143.9 ± 0.63	145.3 ± 0.48†	0.009
Model 2‡				
All individuals	143.8 ± 0.24	144.5 ± 0.62	146.1 ± 0.48†	<0.001
White	141.6 ± 0.35	144.3 ± 1.05†	144.2 ± 1.02†	0.02
Black	144.8 ± 0.44	144.6 ± 1.15	146.0 ± 0.80	0.19
Hispanic	139.7 ± 2.36	141.6 ± 2.65	142.2 ± 2.48†	0.008
Chinese	144.5 ± 3.35	141.3 ± 3.83	148.2 ± 3.61†	0.01
Model 3§				
All individuals	143.7 ± 0.20	143.6 ± 0.54	144.9 ± 0.42†	0.008
White	141.8 ± 0.29	143.5 ± 0.87	144.3 ± 0.85†	0.006
Black	144.9 ± 0.39	142.8 ± 1.01	145.4 ± 0.71	0.57
Hispanic	140.5 ± 2.10	141.7 ± 2.36	141.8 ± 2.212	0.13
Chinese	144.2 ± 2.86	141.9 ± 3.27	144.7 ± 3.09	0.70
Retinal venular caliber (CRVE)				
Model 1*	214.1 ± 0.35	217.1 ± 0.94†	218.0 ± 0.72†	<0.001
Model 2‡				
All individuals	214.4 ± 0.36	216.7 ± 0.95†	218.0 ± 0.73†	<0.001
White	206.0 ± 0.54	208.8 ± 1.58	206.5 ± 1.55	0.74
Black	221.4 ± 0.69	224.2 ± 1.81	224.0 ± 1.27	0.07
Hispanic	214.4 ± 3.66	217.1 ± 4.10	219.4 ± 3.85†	0.001
Chinese	215.9 ± 4.75	213.6 ± 5.42	225.1 ± 5.11†	<0.001
Model 3§				
All individuals	214.7 ± 0.31	216.5 ± 0.82†	216.3 ± 0.63†	0.02
White	206.9 ± 0.44	207.4 ± 1.31	205.2 ± 1.29	0.22
Black	221.8 ± 0.61	225.5 ± 1.59	223.3 ± 1.12	0.23
Hispanic	218.9 ± 3.28	220.3 ± 3.68	221.8 ± 3.44†	0.03
Chinese	214.8 ± 4.05	214.9 ± 4.64	221.0 ± 4.37†	0.001

Data are ANCOVA models showing mean ± SE of retinal arteriolar and venular caliber (in μm). *Model 1: Adjusted for age, race, sex, and study center. ‡Model 2: Adjusted for age, race (except for race-specific models), sex, study center, systolic blood pressure, BMI, total cholesterol, triglycerides, current smoking, and CRP. §Model 3: Adjusted for variables in model 2 and venular caliber (in models of arteriolar caliber) and arteriolar caliber (in models of venular caliber); †$P < 0.05$ comparing specific group with NFG group.

because CRAE and CRVE are highly correlated (correlation coefficient 0.52), we adjusted for variables in model 2 plus CRVE in models of CRAE (and vice versa). This approach accounts for potential confounding from fellow vascular caliber, which may share common genetic and other influences (8).

We also used ANCOVA to estimate mean CRAE/CRVE for categories of retinopathy (none, minimal, and early-severe) in individuals with diabetes, adjusting for age, race, sex, study center, systolic blood pressure, BMI, total cholesterol, triglycerides, current smoking, CRP, A1C, and duration of diabetes (model 4), and also for CRVE in models of CRAE, and vice versa (model 5). We repeated all analyses in the four racial/ethnic groups separately. All analyses were performed in SPSS version 12.0.1 (SPSS, Chicago, IL).

RESULTS — Selected characteristics for the study sample and for each of the four groups—NFG ($n = 4,585$), IFG ($n = 499$), and diabetes with ($n = 165$) and without ($n = 727$) retinopathy among participants who had retinal photographs ($n = 5,976$)—are shown in Table 1. Compared with NFG, individuals with IFG or diabetes were older, had greater BMI and proportion with hypertension, and higher triglycerides and CRP levels, but lower plasma total, LDL, and HDL cholesterol levels. Individuals with diabetic retinopathy had longer duration of diabetes, greater proportion on oral diabetic medication/insulin, higher serum fasting glucose levels, and systolic blood pressure in comparison with individuals with diabetes but without retinopathy.

In the entire sample, the mean CRAE was 144.1 ± 14.4 μm (mean ± SD) and CRVE was 214.0 ± 22.2 μm. The distribution of CRAE and CRVE by glycemic status is shown in Table 2. After controlling for age, sex, race, and study center, CRAE was significantly larger in individuals with diabetes compared with IFG and NFG ($P = 0.009$ for trend). This trend remained with further adjustment for variables in models 2 and 3. After adjusting for similar variables, CRVE was also significantly larger in individuals with IFG and diabetes compared with NFG (Table 2).

Figure 1 shows distribution of CRVE by levels of fasting plasma glucose and A1C in the whole cohort, adjusting for variables in model 3. Increasing levels of fasting glucose and A1C were associated with larger CRVE. Increasing levels of A1C were not associated with CRAE after adjustment for CRVE (data not shown).

After stratifying by ethnic groups, whites, Hispanics, and Chinese participants with diabetes had significantly larger CRAE than individuals with NFG (model 2, Table 2). However, the association remained significant only in whites in model 3 (after adjusting for CRVE) ($P = 0.03$ for race interaction). In contrast, the association between wider CRVE and diabetes was seen only in Hispanics and Chinese ($P = 0.02$ for race interaction).

Of 892 individuals with diabetes, there were 93 (10.4%) with minimal retinopathy and 72 (8.1%) with early-severe retinopathy (Table 3). There were six (0.7%) eyes with proliferative retinopathy that were excluded from analysis. Larger CRVE was associated with increasing severity level of retinopathy (P for trend 0.003, model 5). This association was largely similar across all four racial/ethnic groups but no longer statistically significant (except in blacks). CRAE was not independently associated with diabetic retinopathy. There was no association between CRVE and retinopathy in nondiabetic participants (data not shown).

Finally, we repeated the analyses excluding individuals with CRP >10 mg/dl (since these individuals may have an acute infection), with further adjustments for hypertension and diabetes treatment, and aspirin did not change the relation-

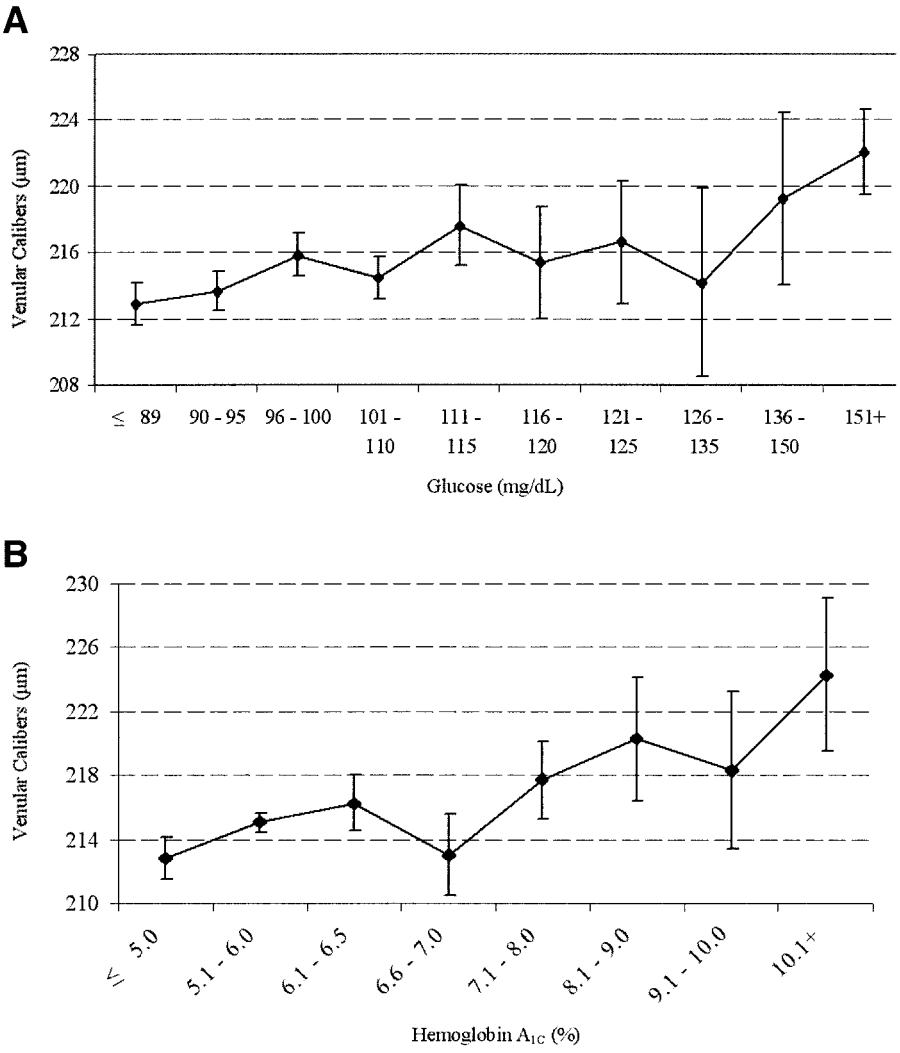

Figure 1—*Distribution of retinal venular caliber (μm) by fasting glucose (A) and A1C (B). Data are means (95% CI), adjusted for variables in model 3 (age, race, sex, study center, systolic blood pressure, BMI, total cholesterol, triglycerides, current smoking, CRP, and arteriolar caliber). The data are for the whole population—those with and without diabetes.*

ships between CRAE/CRVE and glycemic status/retinopathy (data not shown).

CONCLUSIONS — In this large population-based sample, we described the relationship of retinal arteriolar and venular caliber across the range of glycemic status from NFG, IFG, and diabetes to the presence of diabetic retinopathy. We also explored the racial/ethnic variations. We found that individuals with diabetes had larger retinal arterioles and venules than individuals with NFG. The association of wider arterioles and diabetes was seen in whites, whereas the association of wider venules and diabetes was more prominent in Hispanics and Chinese. Larger venular caliber, but not arteriolar caliber, was additionally associated with increasing levels of fasting glucose and hemoglobin A1C levels in individuals with and without diabetes. Among individuals with diabetes, larger venular caliber was associated with diabetic retinopathy.

The association of larger venular caliber with both diabetes and IFG is compatible with previous data from the Atherosclerosis Risk In Communities (ARIC) study, which showed that larger venular caliber was associated with IFG (14) and the Rotterdam Study, which demonstrated a prospective association between wider venular caliber and the incidence of IFG in participants <70 years of age (5). The independent association of larger venular caliber with retinopathy signs in individuals with diabetes is also consistent with clinical experience and with findings in the Wisconsin Epidemiological Study of Diabetic Retinopathy (WESDR) among both type 1 and type 2 diabetic subjects (6,7).

The lack of association between retinal arteriolar caliber with diabetic retinopathy in this study is also consistent with WESDR (6,7). The WESDR also reported no association between retinal arteriolar caliber and diabetic retinopathy among participants with type 2 diabetes (7) but showed that retinal arteriolar caliber may be smaller with increasing retinopathy severity among participants with type 1 diabetes (6). It is important to note that the findings of MESA may not be directly comparable to findings from WESDR. Individuals with diabetes in the current MESA population had mean age (65 vs. 67 years) and prevalence of hypertension (74 vs. 75%) similar to those of participants with type 2 diabetes in the WESDR, but the duration of diabetes was shorter in MESA than WESDR (5.5 vs. 12 years) (7), and the overall management of diabetes is likely to be different in the current MESA cohort compared with WESDR, which was conducted 20 years ago. Finally, in our statistical analysis, we accounted for the correlation between arterioles and venules.

We described new racial/ethnic variations; however, these variations should be interpreted cautiously because of reduced sample size in the subanalyses. The association between wider arterioles and diabetes was seen mainly in whites. This is consistent with previous data from predominantly white populations (15,16). In contrast, the association between wider venules and diabetes was only observed in nonwhites (Hispanic and Chinese). This finding has not been reported previously (15,16) and will need confirmation from other studies.

The finding of wider retinal arterioles with diabetes is supported by clinical studies on the effects of diabetes on retinal blood flow and vascular diameters (17,18). These studies postulate that in the diabetic retina, hyperglycemia and hypoxia initiate retinal vasodilation, leading to hyperperfusion. Hyperperfusion interferes with autoregulation, resulting in further vasodilation. However, we cannot offer explanations for the observed ethnic differences in this association.

The biological mechanisms underlying the associations of larger venular caliber with diabetes, IFG, and higher levels of A1C are as yet unknown. We speculate that this retinal vascular feature partly reflects processes related to inflammation and endothelial dysfunction (1). Consis-

Table 3—*Relationship of retinal arteriolar and venular caliber with severity of retinopathy of participants with diabetes (right eyes only), all individuals, and by racial/ethnic groups*

	Retinal vascular caliber (μm)			
	No retinopathy	Minimal retinopathy	Early-severe retinopathy	*P* for trend
n (%)	727 (81.5)	93 (10.4)	72 (8.1)	
Retinal arteriolar caliber (CRAE)				
Model 4*				
All individuals	144.7 ± 0.59	145.8 ± 1.50	145.6 ± 1.79	0.65
White	143.9 ± 1.85	142.7 ± 3.60	136.7 ± 6.22	0.24
Black	146.5 ± 1.29	147.0 ± 2.74	146.0 ± 3.33	0.87
Hispanic	148.2 ± 1.55	153.5 ± 2.95	151.3 ± 3.00	0.28
Chinese	145.8 ± 2.81	142.7 ± 5.81	151.3 ± 5.54	0.35
Model 5†				
All individuals	145.8 ± 0.74	146.4 ± 1.44	144.2 ± 1.69	0.34
White	143.1 ± 1.61	141.9 ± 3.12	134.1 ± 5.40	0.09
Black	145.6 ± 1.14	147.1 ± 2.39	142.0 ± 2.95	0.22
Hispanic	147.9 ± 1.38	150.6 ± 2.65	148.9 ± 2.69	0.70
Chinese	145.8 ± 2.40	139.5 ± 5.02	144.6 ± 4.90	0.82
Retinal venular caliber (CRVE)				
Model 4*				
All individuals	221.8 ± 1.36	224.0 ± 2.66	231.2 ± 3.07	0.002
White	209.2 ± 3.17	209.4 ± 6.17	215.6 ± 10.7	0.54
Black	227.4 ± 2.13	223.6 ± 4.47	239.5 ± 5.34	0.02
Hispanic	222.5 ± 2.51	230.8 ± 4.84	229.1 ± 4.92	0.17
Chinese	221.5 ± 3.90	229.2 ± 8.10	237.9 ± 7.76	0.05
Model 5†				
All individuals	220.6 ± 1.20	221.9 ± 2.34	228.6 ± 2.72	0.003
White	208.2 ± 2.74	209.4 ± 5.32	220.7 ± 9.23	0.16
Black	222.7 ± 3.92	226.4 ± 1.86	237.5 ± 4.77	0.02
Hispanic	221.7 ± 2.31	226.4 ± 4.43	226.3 ± 4.49	0.29
Chinese	220.5 ± 3.32	230.6 ± 6.90	233.1 ± 6.65	0.07

Data are ANCOVA models showing mean ± SE of retinal arteriolar and venular caliber (in μm). *Model 4: Adjusted for age, race (except for race-specific models), sex, study center, systolic blood pressure, BMI, total cholesterol, triglycerides, current smoking, CRP, A1C, and duration of diabetes. †Model 5: Adjusted for variables in model 4 plus venular caliber (in models of arteriolar caliber) and arteriolar caliber (in models of venular caliber).

tent with this concept, larger retinal venular caliber has been found to be associated with raised systemic inflammatory markers in the Atherosclerosis Risk In Communities study and the Rotterdam and Beaver Dam Eye studies (1), including our previous analysis in MESA (10). However, associations were largely unchanged, even when we adjusted for CRP levels. The observed racial/ethnic differences could be analogous to previous reports of different atherosclerotic loading (19) or diabetes complications (20) in different racial/ethnic groups.

There are clearer pathophysiological mechanisms that may explain the relationship of larger venular caliber and diabetic retinopathy. Experimental studies have previously demonstrated that ocular inflammation is associated with larger retinal venules (21), partly reflecting increased nitrous oxide levels (22), and ocular inflammation is a clear pathogenic factor in the development and progression of diabetic retinopathy (23). Larger retinal venular caliber has also been shown to reflect increased blood flow associated with hyperglycemia (24) and retinal hypoxia (25) and may thus be the earliest vascular change of diabetic retinopathy. In contrast, we found no association between venular caliber and retinopathy in nondiabetic participants, which supports the concept that retinopathy signs in individuals without diabetes may be due to other mechanisms, such as hypertension (2).

The strengths of this study include a large population-based sample and the use of quantitative methods to measure retinal vascular caliber, as well as assessment of retinopathy by standardized grading protocols. Limitations of this study should also be noted. First, the cross-sectional nature of the study limits our ability to judge the temporal sequence of the associations reported. Second, we obtained 45-degree nonstereoscopic nonmydriatic photographs to grade diabetic retinopathy, which is less sensitive for detecting retinopathy, as graded from seven fields of stereoscopic fundus photographs (12); thus, we may have underestimated the proportion with diabetic retinopathy in the study population. Third, there is inevitable measurement error in retinal vascular caliber assessment. However, nondifferential error will tend to dilute associations toward the null. Furthermore, there are other factors that were not controlled that influenced retinal vascular caliber such as genetic and ocular factors (e.g., axial length). By adjusting for the other vessel, we are negating some effects of other "nonmeasurable " factors.

In conclusion, we demonstrated subtle variations in retinal vascular caliber across the range of glycemic-related conditions. Our findings add further to the concept that glycemic status has a continuous effect on the vasculature. Although both larger retinal arteriolar and venular

calibers were associated with diabetes, only larger retinal venular caliber was associated with IFG, with increasing levels of fasting glucose and A1C (across individuals with and without diabetes), and with increasing severity of diabetic retinopathy. These associations may vary by race/ethnicity. Variations in retinal vascular caliber are possible detectable markers for pre-diabetes, clinical diabetes, and early diabetic microvascular damage, and their measurements may provide additional clues to understanding the pathophysiology and consequences of impaired glucose metabolism.

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