AUTHOR QUERY FORM

ELSEVIER	**Journal:** JAC **Article Number:** 29442	**Please e-mail your responses and any corrections to:** **E-mail:** R.Hastings@Elsevier.com

Dear Author,

Please check your proof carefully and mark all corrections at the appropriate place in the proof (e.g., by using on-screen annotation in the PDF file) or compile them in a separate list. **It is crucial that you NOT make direct edits to the PDF using the editing tools as doing so could lead us to overlook your desired changes.** Note: if you opt to annotate the file with software other than Adobe Reader then please also highlight the appropriate place in the PDF file. To ensure fast publication of your paper please return your corrections within 48 hours.

For correction or revision of any artwork, please consult http://www.elsevier.com/artworkinstructions.

Any queries or remarks that have arisen during the processing of your manuscript are listed below and highlighted by flags in the proof.

Location in article	Query / Remark: Click on the Q link to find the query's location in text Please insert your reply or correction at the corresponding line in the proof
	NOTE: The DOI of your paper is located on the proof (in most cases at the bottom of the first page, or near the author correspondence information).
Q1	If there are any drug dosages in your article, please verify them and indicate that you have done so by initialing this query. Also, please review/confirm the spelling of all drug names in figures, tables, and text.
Q2	Please confirm the relationship with industry information for each author is correct, accurate, and complete.
Q3	The Central Illustration may have been altered per the Editors and has been redrawn into journal style using the journal's color palette. Please review it carefully to ensure it is correct as is.
Q4	Please note: figures are redrawn to adhere to journal style; please review each figure carefully to ensure all data representation is correct and that all colors represented in the figure are represented correctly in the figure legend.
Q5	Is shortened title ok as set?
Q6	Darlinghurst, New South Wales, correct location for St Vincent's Clinical School?
Q7	Mean values with SD constructed as # ± #, per journal style.
Q8	Gender refers to social, environmental, cultural, and behavioral factors that influence a person's self-identity. Sex is a biological classification as male or female. Please ensure that if either/both of these terms are used in your paper, they are used accurately throughout.
Q9	Is the sentence that begins "Fruit and vegetable consumption…" ok as changed with information added to clarify the Fuster-BEWAT?
Q10	Is "Student" *t* test ok as added?

(*continued on next page*)

Q11	For the sentence that begins "Other covariants…," reference 11 lists Doménech as the head author but sentence indicates “Kleinbaum and colleagues.” All okay here?
Q12	Is reference 8 from an online source? If so, please supply the URL and the date the material was accessed for us in this paper per journal style. Otherwise, please supply complete publishing information including book or journal title and volume and page numbers.
Q13	Please provide complete publishing information for reference 11. It is unclear who the authors are and where the information is sourced (ie, online, printed, etc).
Q14	Please provide complete information for reference 23.
Q15	Central Illustration: please confirm colors called out in legend match colors as used in the image.
Q16	Please confirm that given names and surnames have been identified correctly and are presented in the desired order and please carefully verify the spelling of all authors' names.
	Please check this box or indicate your approval if you have no corrections to make to the PDF file ☐

Thank you for your assistance.

JOURNAL OF THE AMERICAN COLLEGE OF CARDIOLOGY VOL. ■, NO. ■, 2022
© 2022 BY THE AMERICAN COLLEGE OF CARDIOLOGY FOUNDATION
PUBLISHED BY ELSEVIER

Subclinical Atherosclerosis in Young, Socioeconomically Vulnerable Hispanic and Non-Hispanic Black Adults

Josep Iglesies-Grau, MD,[a] Rodrigo Fernandez-Jimenez, MD, PhD,[b,c,d] Raquel Diaz-Munoz, NP, PhD,[e] Risa Jaslow, MS, RDN,[f] Amaya de Cos-Gandoy, MS,[b,g] Gloria Santos-Beneit, PhD,[f,g] Christopher A. Hill, BA,[f] Alexandra Turco, BS,[f] Daniella Kadian-Dodov, MD,[f] Jason C. Kovacic, MD, PhD,[f,h,i] Zahi A. Fayad, PhD,[f,j] Valentin Fuster, MD, PhD[b,f]

ABSTRACT

BACKGROUND Non-Hispanic Black persons are at greater risk of cardiovascular (CV) events than other racial/ethnic groups; however, their differential vulnerability to early subclinical atherosclerosis is poorly understood.

OBJECTIVES This work aims to study the impact of race/ethnicity on early subclinical atherosclerosis in young socioeconomically disadvantaged adults.

METHODS Bilateral carotid and femoral 3-dimensional vascular ultrasound examinations were performed on 436 adults (parents/caregivers and staff) with a mean age of 38.0 ± 11.1 years, 82.3% female, 66% self-reported as Hispanic, 34% self-reported as non-Hispanic Black, and no history of CV disease recruited in the FAMILIA (~~Family-based Approach to Promotion of Health–FAMILIA [Project 2]~~) trial from 15 Head Start preschools in Harlem (neighborhood in New York, New York, USA). The 10-year Framingham CV risk score was calculated, and the relationship between race/ethnicity and the presence and extent of subclinical atherosclerosis was analyzed with multivariable logistic and linear regression models.

RESULTS The mean 10-year Framingham CV risk was 4.0%, with no differences by racial/ethnic category. The overall prevalence of subclinical atherosclerosis was significantly higher in the non-Hispanic Black (12.9%) than in the Hispanic subpopulation (6.6%). After adjusting for 10-year Framingham CV risk score, body mass index, fruit and vegetable consumption, physical activity, and employment status, non-Hispanic Black individuals were more likely than Hispanic individuals to have subclinical atherosclerosis (OR: 3.45; 95% CI: 1.44-8.29; $P = 0.006$) and multiterritorial disease ($P = 0.026$).

CONCLUSIONS After adjustment for classic CV risk, lifestyle, and socioeconomic factors, non-Hispanic Black younger adults seem more vulnerable to early subclinical atherosclerosis than their Hispanic peers, suggesting that the existence of emerging or undiscovered CV factors underlying the residual excess risk (~~Family-based Approach to Promotion of Health~~–FAMILIA (Project 2) ~~[FAMILIA]~~; NCT02481401). (J Am Coll Cardiol 2022;■:■–■) © 2022 by the American College of Cardiology Foundation.

From the [a]Institut de Cardiologie de Montréal, Montréal, Quebec, Canada; [b]Centro Nacional de Investigaciones Cardiovasculares (CNIC), Madrid, Spain; [c]Hospital Universitario Clínico San Carlos, Madrid, Spain; [d]CIBER de Enfermedades Cardiovasculares (CIBERCV), Madrid, Spain; [e]Centro Nacional de Epidemiología (CNE), Instituto de Salud Carlos III, Madrid, Spain; [f]The Zena and Michael A. Wiener Cardiovascular Institute, Icahn School of Medicine at Mount Sinai, New York, New York, USA; [g]Foundation for Science, Health and Education (SHE), Barcelona, Spain; [h]Victor Chang Cardiac Research Institute, Sydney, New South Wales, Australia; [i]St Vincent's Clinical School, University of New South Wales, ~~Darlinghurst~~, New South Wales, Australia; and the [j]BioMedical Engineering and Imaging Institute, Icahn School of Medicine at Mount Sinai, New York, New York, USA.

Sawan Jalnapurkar, MD, MBA, served as Guest Associate Editor for this paper. Athena Poppas, MD, served as Guest Editor-in-Chief for this paper.

The authors attest they are in compliance with human studies committees and animal welfare regulations of the authors' institutions and Food and Drug Administration guidelines, including patient consent where appropriate. For more information, visit the Author Center.

Manuscript received January 10, 2022; revised manuscript received April 6, 2022, accepted April 18, 2022.

ISSN 0735-1097/$36.00 https://doi.org/10.1016/j.jacc.2022.04.054

ABBREVIATIONS AND ACRONYMS

3DVUS = 3-dimensional vascular ultrasound

CV = cardiovascular

FAMILIA = Family-Based Approach in a Minority Community Integrating Systems-Biology for Promotion of Health

PESA = Progression and Early detection of Subclinical Atherosclerosis

Cardiovascular (CV) disease tends to affect non-Hispanic Black persons earlier in life and to a greater extent than is the case with other racial and ethnic groups.[1] This may be attributable in part to an elevated prevalence of CV risk factors such as hypertension, diabetes, peripheral artery disease, and chronic kidney disease, or to a more frequent clustering of multiple risk factors; however, other factors could also play a critical role in generating these disparities.

Recently developed CV risk scores that consider race and ethnicity together with well-established CV risk factors have proved to be useful tools for predicting CV events and establishing prevention strategies.[2] Nevertheless, a substantial proportion of events occur in individuals classified at low or moderate risk. Because CV disease has a slowly progressing asymptomatic phase, interest has grown in the preventive potential of noninvasive imaging techniques used to detect direct signs of early subclinical CV disease. Progress in this area has been achieved with the recent establishment of 3-dimensional vascular ultrasound (3DVUS) as a safe, inexpensive, and reliable tool for detecting the presence and quantifying the extent or burden of subclinical atherosclerosis.[3] The observed racial and ethnic disparities in CV disease may reflect differential vulnerability to early atherosclerosis; however, few studies have addressed this relationship.

The randomized interventional trial FAMILIA (Family-Based Approach in a Minority Community Integrating Systems-Biology for Promotion of Health) enrolled young families (preschool children and their parents/caregivers) and school staff from a low-income area in the neighborhood of Harlem, New York, New York, USA.[4] A high proportion of FAMILIA participants are of African-American and Hispanic descent. The main goal of the trial was to test the efficacy of a family-based approach to CV health promotion across the lifespan that integrates behavioral and imaging strategies.[5,6] At enrollment, adults (parents/caregivers and staff) underwent a comprehensive assessment of lifestyle and CV health, including bilateral carotid and femoral 3DVUS. Here, we assessed the impact of race and ethnicity on the presence, extent, and distribution of 3DVUS-detected subclinical atherosclerosis through a cross-sectional analysis of the information collected at baseline in adult participants enrolled in the FAMILIA study.

METHODS

STUDY DESIGN AND POPULATION. The FAMILIA trial rationale and design have been described elsewhere.[4] The FAMILIA study recruited a total of 635 adult caregivers and school staff from 15 Head Start preschools in the neighborhood of Harlem, New York, New York, USA. These adult trial participants underwent a complete clinical evaluation at baseline, including lifestyle questionnaires supervised by trained personnel and point-of-care testing to determine blood glucose and lipid profile. All questionnaires were available in English and Spanish, and health counselors were fluent in both languages to accommodate participant language preferences. In addition, all participants were invited to sign a separate consent form to undergo noninvasive 3DVUS to examine for the presence and burden of atherosclerosis in the carotid and femoral arteries.

Non-Hispanic White, Asian, and Native American racial and ethnic groups each formed a small proportion of participants (2.3%, 2.3%, and 0.3%, respectively), and people in these groups were excluded from the present analysis. Also excluded were adults with a history of heart disease or stroke. The Icahn School of Medicine at Mount Sinai Institutional Review Board approved the study (HS#:14-01054), which was conducted in accordance with institutional and federal guidelines involving human participants. The study is registered on ClinicalTrials.gov, identifier number NCT02481401.

BASELINE CHARACTERISTICS. The following characteristics were assessed at baseline before initiation of the FAMILIA trial intervention.[5] Ethnic and racial background was self-reported and classified as Hispanic, non-Hispanic Black, or others (including non-Hispanic White, Asian, and Native American). Data were collected on well-established socioeconomic determinants of health, including self-reported employment status and average annual household income, as well as self-reported history of hypertension, diabetes, and dyslipidemia. Family history of CV disease was defined as a self-reported diagnosis of heart attack or stroke in a full parent or sibling by the age of 60 years. In addition, several health metrics (blood pressure, fasting blood glucose, total cholesterol, low-density lipoprotein-cholesterol, high-density lipoprotein-cholesterol, triglycerides, body mass index (BMI), fruit and vegetable consumption, smoking habits, and physical activity) were measured as detailed in the Supplemental Methods.

UNCORRECTED PROOF

PREDICTED 10-YEAR CV RISK AND CATEGORIZATION OF HEALTH METRICS. The predicted 10-year CV disease risk for each participant at enrollment was calculated with the Framingham Heart Study CV risk score equation using the user-written Stata command "framingham."[7,8] The equation includes the following parameters: age (in years), sex, measured systolic blood pressure (mm Hg), self-reported antihypertensive medication, smoking status, self-reported diabetes status, and measured total cholesterol and high-density lipoprotein-cholesterol (mg/dL). Based on Framingham CV risk scores, 10-year CV disease risk was classified as low (<10%), moderate (≥10-<20%), or high (≥20%).

Fruit and vegetable consumption and moderate/vigorous physical activity were categorized according to thresholds described in the Fuster-BEWAT score (a health metric that includes 5 factors: blood pressure, exercise, weight, alimentation, and tobacco).[9] This risk scale has a demonstrated accuracy for predicting subclinical atherosclerosis in relatively low-risk individuals similar to that of other American Heart Association-approved risk scales, but without the need for laboratory results.[10]

3DVUS IMAGING PROTOCOL/ANALYSIS AND DEFINITION OF ATHEROSCLEROSIS. Imaging protocol. Vascular ultrasound imaging to quantify the presence and extent of atherosclerosis in carotid and femoral arteries was performed with the Philips EPIQ-7G ultrasonography system (Philips Healthcare). Transducers used in this study were the 2-dimensional (2D) 9L-D linear array (9-3.1 MHz) and C1-6 curved array (6-1 MHz) transducers and the 3D VL 13-5 linear volume array transducer (13-5 MHz). The imaging protocol was adapted from the one used in the PESA (Progression and Early detection of Subclinical Atherosclerosis) study.[3] The procedure was performed by experienced registered vascular technologists who completed additional specialized training with the Philips ultrasonography system. The scanning protocol included standard imaging of the left and right carotid artery bifurcation and its branches (internal and external carotid arteries) and of the left and right common femoral artery bifurcation and its branches (superficial and deep femoral arteries). These 4 territories were scanned in cross-section with a 2D linear or curved array transducer to detect the presence of plaques and to sum plaque burden. The same vessels were examined by 3D ultrasound with the VL 13-5 linear array volume transducer, which performs a mechanical automated sweep in cross-section, allowing the assessment of plaque volume and estimation of total atherosclerotic burden.

Image analysis. All ultrasound recordings and digital images were analyzed at the Zena and Michael A. Wiener Cardiovascular Institute at Icahn School of Medicine at Mount Sinai by 2 observers (raters). Three-dimensional images were analyzed with dedicated software specially engineered by Philips (QStation-VPQ [vascular plaque quantification] 3.5). Each carotid and femoral 2D cross-sectional image was assessed for the presence of plaque. When a plaque was identified, QStation was used to assess the 3D dataset (displayed as multiple transverse slices) and to sum plaque volume in each vascular bed (carotid or femoral) on each side (left or right), as well as the total plaque volume in all vascular beds (bilateral carotid and femoral). Maximum percent stenosis was also estimated.

Definition of atherosclerosis. Plaque was defined according to the Mannheim consensus criteria as a focal structure encroaching into the arterial lumen and measuring ≥0.5 mm or >50% of the surrounding intima-media thickness or having a diffuse thickness ≥1.5 mm measured from the media-adventitia to the intima-lumen interface in any of the territories.[3]

The extent of atherosclerosis was defined according to the number of regions with presence of plaque (disease-free, 0 vascular sites affected; focal disease, 1 territory affected; multiterritorial disease, >1 territory affected). Plaque burden was defined as the plaque volume (mm^3), and global plaque burden corresponded to the sum of all plaque areas from all images showing plaque, including both carotid and femoral arteries.

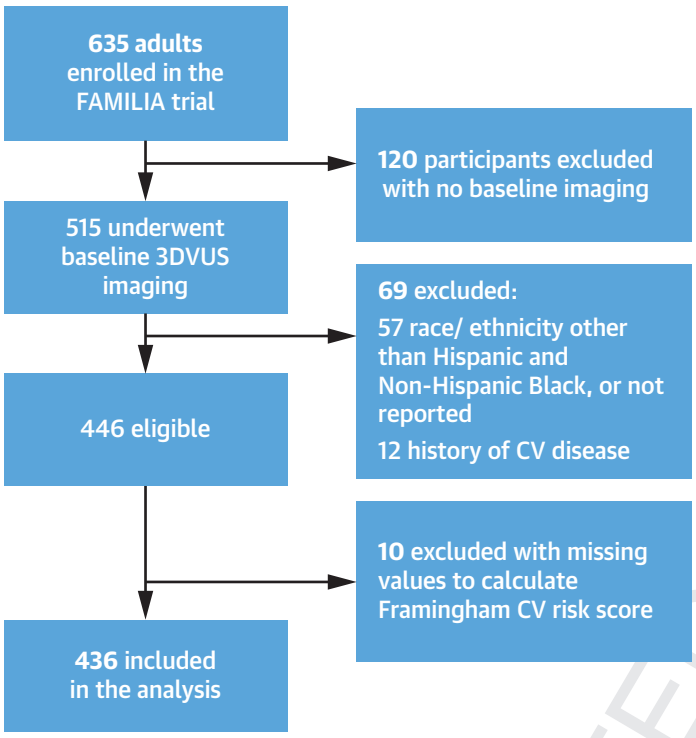
STATISTICAL ANALYSIS. Summary statistics describing baseline characteristics are presented as mean ± SD for continuous variables and as count and frequencies for categorical variables. Unpaired Student's *t*-tests were used to assess crude differences between continuous variables; the chi-square test and Fischer exact test were used to determine crude differences between categorical variables. The Cochran-Mantel-Haenszel test was used to assess crude differences across ordered categorical variables.

To assess the adjusted impact of race and ethnicity on the presence and burden of atherosclerosis, we used multivariable logistic regression models for categorical outcome variables (presence or absence of atherosclerosis) and linear regression models for continuous outcomes (atherosclerosis burden in mm^3). Participant race and ethnicity were included in all models as the main independent variable. Other covariates were selected according to their reported

UNCORRECTED PROOF

print & web 4C/FPO

FIGURE 1 Participant Flow Chart of FAMILIA Participants Included in This Study



The FAMILIA (~~Family-based Approach to Promotion of Health–FAMILIA [Project 2]~~) trial enrolled 635 adults, of whom 515 consented to and underwent 3-dimensional vascular ultrasound (3DVUS) imaging. After exclusion of participants with a history of cardiovascular (CV) disease (n = 12), those self-reporting a race and ethnicity other than Hispanic or non-Hispanic Black (n = 57), and those with missing data for calculating the Framingham CV risk score (n = 10), a total of 436 adults were included in this study.

association with atherosclerosis (clinical plausibility) or as potential confounders according to the rules proposed by Kleinbaum and colleagues using the user-written Stata command "confound."[11] The covariates included in the multivariable models were as follows: participant 10-year CV Framingham risk score (categorical variable: low, moderate, high), participant employment status (categorical variable: employed, unemployed, other status, unknown), BMI (continuous variable), physical activity (categorical variable: <10, 10-<75, ≥75-<150, and ≥150 minutes/week of moderate-to-vigorous exercise) and fruit and vegetable consumption (categorical variable: <1, 1-2, 3-4, >4 daily servings). Practical and clinical interpretations are presented as measures of association (OR) and estimated mean differences with 95% CIs. Statistical significance was assigned at $P < 0.05$.

Prevalence-adjusted and bias-adjusted kappa coefficients were calculated in a subsample of FAMILIA participants to assess intraobserver and interobserver reproducibility for plaque detection. For intraobserver and interobserver reproducibility analysis of atherosclerosis burden (plaque volume quantification), the intraclass correlation coefficient and Bland-Altman plots were used for all plaque-positive FAMILIA participants. Methodological details of agreement and reproducibility analysis are presented in the Supplemental Methods. Statistical analyses were performed with STATA (2017, Stata Statistical Software: Release 15; StataCorp LLC).

RESULTS

The FAMILIA trial enrolled 635 adults, of whom 515 consented to and underwent 3DVUS imaging. Participants with a history of CV disease were excluded, as were those self-reporting a race and ethnicity other than Hispanic or non-Hispanic Black and those with missing data for calculating the Framingham CV risk score. A final total of 436 adults were included in this study (Figure 1).

BASELINE CHARACTERISTICS, HEALTH METRICS, AND PREDICTED CV RISK. Participants self-reporting as Hispanic or non-Hispanic Black accounted for approximately two-thirds (n = 289, 66.3%) and one-third (n = 147, 33.7%) of the study population, respectively. The mean age was 38.0 ± 11.1 years, and 82.3% were women. Age and sex profiles did not differ significantly between racial and ethnic groups. Baseline characteristics of adults included in this study are summarized in Table 1, whereas baseline characteristics of all individuals enrolled in the FAMILIA trial grouping subjects by availability of 3DVUS information are presented in Supplemental Table 1.

Compared with their Hispanic counterparts, non-Hispanic Black participants were ~3.5 times more likely to be hypertensive (OR: 3.54; 95% CI: 2.14-5.87; $P < 0.001$). Non-Hispanic Black participants similarly were 3 times more likely to be active smokers (OR: 3.15; 95% CI: 1.83-5.41; $P < 0.001$), and also had a higher BMI (mean between-group difference = 1.45 kg/m^2; 95% CI: 0.17-2.74; $P = 0.027$) and reported higher consumption of fruits and vegetables ($P < 0.001$). There was no between-group difference in the prevalence of self-reported diabetes ($P = 0.735$) or determined fasting glucose ($P = 0.402$) and total cholesterol ($P = 0.873$). The mean 10-year Framingham CV risk score for the whole study population was 4.0% ± 5.6%, and there were no significant differences between racial and ethnic groups ($P = 0.104$). Most participants (89%) were classified at low risk, with ~9% and 2% classified at

UNCORRECTED PROOF

TABLE 1 Baseline Characteristics of Enrolled Adults in the FAMILIA Trial Included in This Study

	Overall (N = 436)	Hispanic (n = 289 [66.3%])	Non-Hispanic Black (n = 147 [33.7%])	*P* Value
Age	38.0 ± 11.1	37.5 ± 11.0	39.0 ± 11.3	0.191
Female	359 (82.3)	232 (80.3)	127 (86.4)	0.113
Employment status				
Employed	276 (63.3)	168 (58.1)	108 (73.5)	0.018
Unemployed/unable to work/homemaker	123 (28.2)	92 (31.8)	31 (21.1)	
Other (student, retired)	23 (5.3)	18 (6.2)	5 (3.4)	
Unknown	14 (3.2)	11 (3.8)	3 (2.0)	
Annual household income				
<$25,000	214 (59.8)	151 (67.4)	63 (47.0)	<0.001
≥$25,000	144 (40.2)	73 (32.6)	71 (53.0)	
Unknown	78 (17.9)	65 (22.5)	13 (8.8)	
Cardiovascular risk factors				
Self-reported hypertension	77 (17.7)	32 (11.1)	45 (30.6)	<0.001
Active smoking	63 (14.5)	27 (9.3)	36 (24.5)	<0.001
Body mass index, kg/m^2				
Low, <18.5	4 (0.9)	2 (0.7)	2 (1.4)	0.322
Normal, 18.5-<25	74 (17.0)	49 (17.0)	25 (17.0)	
Overweight, 25-<30	156 (35.8)	113 (39.1)	43 (29.3)	
Obese, ≥30	202 (46.3)	125 (43.3)	77 (52.4)	
Self-reported diabetes	56 (12.8)	36 (12.5)	20 (13.6)	0.735
Self-reported dyslipidemia	104 (23.9)	77 (26.6)	27 (18.4)	0.055
Family history of CV disease	30 (7.7)	14 (5.5)	16 (11.9)	0.026
Mean 10-y Framingham CV risk score	4.0 ± 5.6	3.6 ± 5.1	4.6 ± 6.4	0.104
Categorized 10-y Framingham CV risk				
Low risk, <10%	388 (89.0)	261 (90.3)	127 (86.4)	0.279
Moderate risk, 10%-<20%	40 (9.2)	23 (8.0)	17 (11.6)	
High risk, ≥20%	8 (1.8)	5 (1.7)	3 (2.0)	
Physical activity, mod/vig activity, min/wk				
<10	83 (19.9)	58 (21.2)	25 (17.4)	0.753
10-<75	60 (14.4)	33 (12.0)	27 (18.8)	
75-<150	85 (20.3)	54 (19.7)	31 (21.5)	
≥150	190 (45.5)	129 (47.1)	61 (42.4)	
Fruit/vegetable consumption, n daily servings				
<1	22 (5.1)	13 (4.5)	9 (6.1)	<0.001
1-2	239 (54.8)	173 (59.9)	66 (44.9)	
3-4	139 (31.9)	86 (29.8)	53 (36.1)	
>4	36 (8.3)	17 (5.9)	19 (12.9)	

Values are mean ± SD or n (%). *P* values are derived from unpaired Student's *t*-tests for continuous variables, chi-square test and Fischer exact test for binary categorical variables, and the Cochran-Mantel-Haenszel tests for ordered categorical variables.

CV = cardiovascular; FAMILIA = ~~Family-based Approach to Promotion of Health–FAMILIA (Project 2);~~ mod/vig = moderate-to-vigorous.

moderate and high risk, respectively. Measured CV risk and health factors of adults included in this study are listed in Supplemental Table 2, whereas those of all individuals enrolled in the FAMILIA trial grouping subjects by availability of 3DVUS information are presented in Supplemental Table 3.

PREVALENCE, EXTENT, AND DISTRIBUTION OF SUBCLINICAL ATHEROSCLEROSIS. The overall prevalence of subclinical atherosclerosis assessed by 3DVUS was 8.7%, and the mean global plaque burden was 5.0 ± 27.9 mm^3 (**Table 2**). The prevalence of subclinical atherosclerosis was higher in non-Hispanic Black participants across all 10-year Framingham CV risk score categories, and this difference was especially prominent in the high-risk category (**Central Illustration**). Overall, the crude odds of having subclinical atherosclerosis were 2 times higher among non-Hispanic Black persons than in the Hispanic subpopulation (OR: 2.11; 95% CI: 1.09-4.08; $P = 0.026$). Non-Hispanic Black participants had a higher mean disease burden (mean difference in total plaque volume = 6.17 mm^3; 95% CI: 0.68-11.66; $P = 0.028$) and a higher prevalence of multiterritorial disease ($P = 0.026$).

TABLE 2 Crude Prevalence, Extent, and Distribution of Subclinical Atherosclerosis as Assessed by 3-Dimensional Vascular Ultrasound

	Overall (N = 436)	Hispanic (n = 289)	Non-Hispanic Black (n = 147)	*P* Value
Evidence of atherosclerosis				
Any territory	38 (8.7)	19 (6.6)	19 (12.9)	0.026
Carotids	33 (7.6)	14 (4.8)	19 (12.9)	0.003
Femorals	18 (4.1)	11 (3.8)	7 (4.8)	0.635
Extent of atherosclerosis				
Disease free	398 (91.3)	270 (93.4)	128 (87.1)	0.026
Focal	15 (3.4)	8 (2.8)	7 (4.8)	
Multiterritorial	23 (5.3)	11 (3.8)	12 (8.2)	
Burden of atherosclerosis, mm^3				
Carotid and femoral	5.0 ± 27.9	2.9 ± 19.6	9.0 ± 38.8	0.028
Carotid	3.7 ± 22.0	1.5 ± 13.0	7.9 ± 32.8	0.004
Femoral	1.3 ± 7.8	1.3 ± 8.0	1.2 ± 7.5	0.819

Values are n (%) or mean ± SD. *P* values are derived from unpaired Student's *t*-test for continuous variables, and chi-square test and Fischer exact test for binary categorical variables.

In the multivariable analysis, race and ethnicity showed an independent association with the presence and extent of subclinical atherosclerosis. Compared with the Hispanic subpopulation, non-Hispanic Black participants had an adjusted OR for having subclinical atherosclerosis of 3.45 (95% CI: 1.44-8.29; $P = 0.006$). The adjusted mean difference between racial and ethnic groups in total plaque volume was 6.94 mm^3 (95% CI: 1.43-12.46 mm^3; $P = 0.014$). Adjustment for individual risk factors instead of Framingham risk score was performed as sensitivity analysis, and associations of race and ethnicity with the presence and extent of subclinical atherosclerosis remained similar.

Separate analyses of carotid and femoral involvement revealed significant differences by race and ethnicity in the carotid territory, with the adjusted odds of finding plaques in the carotids ~6 times higher in non-Hispanic Black participants than in their Hispanic peers (adjusted OR: 5.94; 95% CI: 2.17-16.26; $P = 0.001$). In contrast, no between-group differences were observed in the femoral arteries (adjusted OR: 1.72; 95% CI: 0.53-5.53; $P = 0.364$). Similar results were observed for the comparison of atherosclerosis burden; the mean adjusted difference in total plaque volume between the non-Hispanic Black and Hispanic groups was 7.06 mm^3 (95% CI: 2.65-11.48 mm^3; $P = 0.002$) in the carotid region and -0.12 mm^3 (95% CI: -1.66 to 1.42 mm^3; $P = 0.877$) in the femoral territory.

Intraobserver and interobserver agreement were excellent for the detection of plaque and very good for volume plaque quantification (Supplemental Tables 4 to 6 and Supplemental Figure 1).

DISCUSSION

This cross-sectional study of adult FAMILIA trial participants is one of the first to report the presence of 3DVUS-assessed subclinical atherosclerosis in a young cohort of mainly women, from a socioeconomically disadvantaged community. The study generated a number of key findings, 1) Compared with the Hispanic subcohort, non-Hispanic Black participants had higher rates of hypertension, active smoking, BMI, and self-reported fruit and vegetable consumption. 2) There was an overall low prevalence of subclinical atherosclerosis assessed by carotid and femoral 3DVUS (~9%), and plaques were more frequently found in the carotid than in the femoral arteries. 3) Although 10-year Framingham CV risk scores were similar in the 2 study subcohorts, non-Hispanic Black participants had higher odds of having subclinical atherosclerosis, a higher disease burden, and a higher prevalence of multiterritorial disease. 4) These racial and ethnic differences were mainly driven by differences in the carotid arteries (Figure 2). These findings may help to explain the observed differences in CV-disease prevalence between racial and ethnic groups.

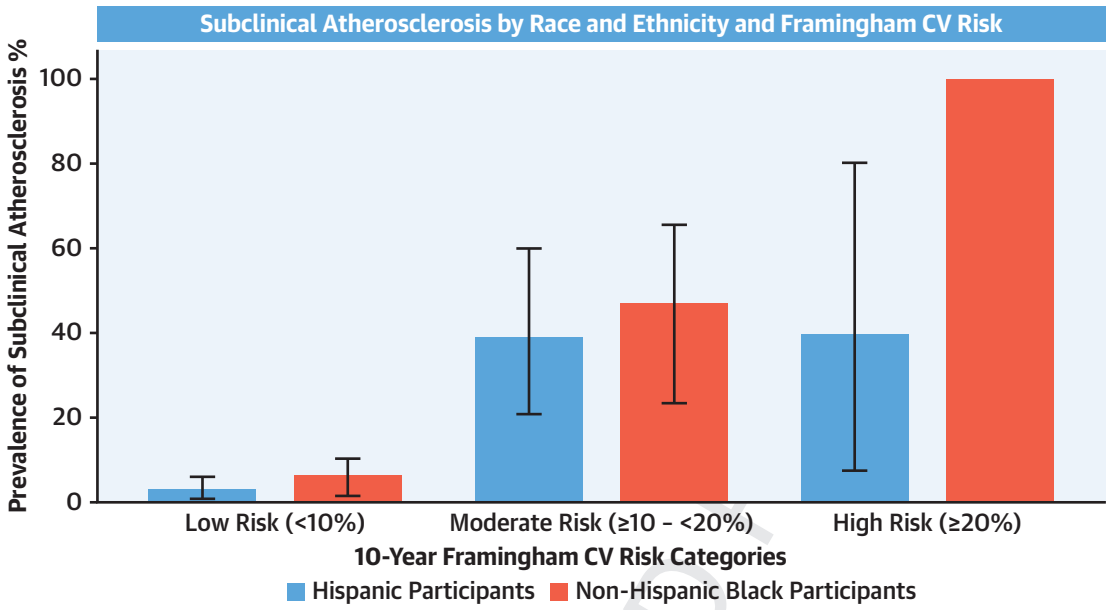
Hispanic and non-Hispanic Black populations in the United States both have a higher prevalence of subclinical atherosclerosis and a higher CV-disease burden than other ethnic groups.[12] Previous studies have also reported an earlier and more diffuse involvement, particularly in the non-Hispanic Black population, which might reflect the presence of more CV risk factors at an earlier age, poorer CV health habits, or other factors such as different genetic predisposition to atherosclerosis.[13,14] However, data on these underrepresented populations are limited, especially in relation to younger age groups.

In our cohort, non-Hispanic Black individuals had significantly higher blood pressure and a higher prevalence of self-reported smoking, risk factors known to trigger earlier atherosclerotic-plaque formation and adverse epigenetic pathway activations, and subsequent CV disease.[15,16] Furthermore, both groups had high prevalence rates for other risk factors, including overall diabetes and overweight/obesity prevalence rates of 12.8% and 82.1%, respectively, potentially contributing to earlier vascular aging.[17] These observations are consistent with National Health and Nutrition Examination Survey statistics showing that the non-Hispanic Black population has the lowest prevalence of ≥5 ideal CV health metrics, both in children and adults aged ≥20 years.[12] Together, these findings support the notion

UNCORRECTED PROOF

print & web 4C/FPO

CENTRAL ILLUSTRATION **Prevalence of Subclinical Atherosclerosis Across Framingham Cardiovascular Risk Categories**



Iglesies-Grau J, et al. J Am Coll Cardiol. 2022;■(■):■-■.

Bars (**blue,** Hispanic participants; **red,** Non-Hispanic Black participants) represent the mean prevalence of subclinical atherosclerosis in the ~~Family-based Approach to Promotion of Health–FAMILIA [Project 2]~~ trial adults included in this study stratified by Framingham cardiovascular (CV) risk category. The presence of subclinical atherosclerosis was defined as the detection of any plaque in the explored bilateral carotid and femoral territories by 3-dimensional vascular ultrasound. Error bars corresponding to logit-transformed 95% CIs of the estimated prevalence.

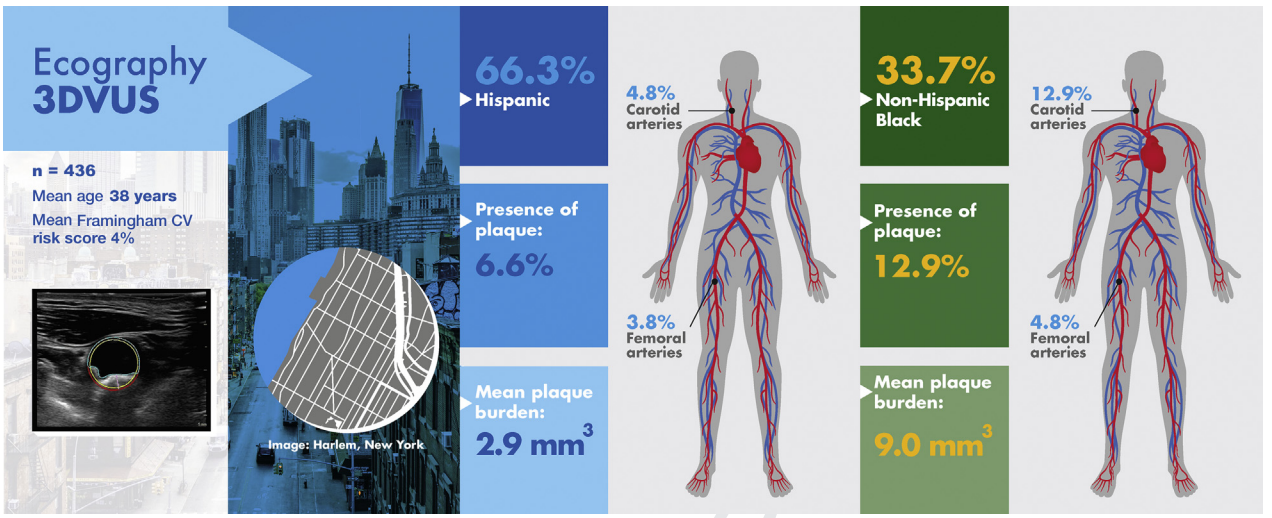
UNCORRECTED

that specific racial and ethnic groups would benefit more from targeted early health promotion programs, CV screening campaigns, and other interventions, such as more intense antismoking health marketing strategies or increased access to cessation treatments, counseling, and medication.[18]

Nevertheless, in our study population, subclinical atherosclerosis prevalence and atherosclerotic burden were both higher in the non-Hispanic Black group than in the Hispanic population, despite both groups having similar 10-year Framingham CV risk scores. These findings were consistent across all Framingham CV risk score categories and suggest that nontraditional or unknown risk factors could potentially work through distinct epigenetic pathways that might explain the earlier and more encroaching progression of subclinical atherosclerosis among the non-Hispanic Black population.[19] The extent to which these differences may be heritable could be addressed through the implementation of genome-wide association study methods to understand the genetic contributions to disease. Nevertheless, evidence of biological differences in disease pathogenesis between racial and ethnic groups remains limited, and previous work has shown that disease development and progression are at least equally influenced by other factors, such as acculturation, socioeconomic status, educational attainment, behavioral and psychological conditions, food environment, access to health care, and other social determinants of health.[20,21] As long ago as 1985, the Task Force on Black and Minority Health reported that noticeable health disparities existed among minority communities and that these communities were underrepresented in health research. This report prompted research initiatives such as the Jackson Heart Study and the creation of the CARDIA (Coronary Artery Risk Development in Young Adults), ARIC (Atherosclerotic Risk In Communities), and MESA (Multi-Ethnic Study of Atherosclerosis) cohorts. These cohorts have since yielded substantial data on the use of noninvasive imaging techniques to assess subclinical atherosclerosis and on the contribution of nonbiological factors to poorer health, including the association between subclinical disease and socioeconomic status.[20,22]

Although the non-Hispanic Black members of our cohort reported higher employment rates and annual

FIGURE 2 **Subclinical Atherosclerosis in Hispanic and Non-Hispanic Black Socioeconomically Vulnerable Young Adults**



This study assessed the impact of race and ethnicity on early subclinical atherosclerosis, as assessed by bilateral carotid and femoral 3DVUS, on 436 young Hispanic and non-Hispanic Black adults (parents/caregivers and staff) recruited in the FAMILIA trial from 15 Head Start preschools in Harlem (New York City). After adjustment for traditional CV risk, lifestyle, and socioeconomic factors, non-Hispanic Black individuals were more likely than Hispanic individuals to have subclinical atherosclerosis and higher burden of disease, suggesting the existence of unknown cardiovascular risk factors underlying such residual excess risk. Early detection of atherosclerosis could play an important role in prevention programs in different populations. Abbreviations as in Figure 1.

household income than their Hispanic counterparts, the unemployment rate in the whole study population was approximately 28%, and almost 2 of 3 participants reported an annual household income <$25,000, just below the $25,465 2018 U.S. Census Bureau Poverty Threshold for a family of 4 members including 2 children.[23] When we additionally included annual household income in multivariable models, the effect of race and ethnicity remained significant for plaque presence and disease burden (total plaque volume). These findings suggest that household income was not the main driver of the racial and ethnic differences observed in this study.

Although data on subclinical disease in vulnerable communities remain limited, there are numerous noninvasive imaging tests available to assess early atherosclerosis and their use for CV prevention and health promotion is steadily expanding.[24] Moreover, some of these techniques have a low-cost; thus, they offer economic savings and the opportunity for large-scale implementation in a clinical setting, including screening and follow-up studies. In younger populations, noninvasive imaging also offers opportunities to reclassify risk given that traditional risk scores, mathematically dominated by age and sex, provide insufficient accuracy for the assessment of individual risk. Although 2D imaging has long been used to identify plaques and measure plaque thickness, a major advantage of 3DVUS is that it visualizes and quantifies plaques in the longitudinal and cross-sectional planes, providing a more accurate volumetric assessment of disease than 2D ultrasound.[25] The analysis of global plaque burden in our cohort revealed a higher disease burden among non-Hispanic Black participants (Table 2). When only individuals with plaque were considered, the median plaque volume overall, including both the carotid and the femoral territories, was 40.3 mm^3 (IQR: 13.7-63.0 mm^3), whereas for the non-Hispanic Black group the value was 53.5 mm^3 (IQR: 13.7-71.2 mm^3) and for the Hispanic group 36.8 mm^3 (IQR: 10.3-45.7 mm^3). In the PESA study, 3DVUS was used to quantify subclinical disease burden in 3,860 adults (mean age: 45 years; 63% male; 99.9% Caucasian).[26] This analysis detected extensive atherosclerosis in a considerable number of low-risk individuals. Median global plaque burden was 50.8 mm^3 (IQR: 18.7-121.5 mm^3) among participants with atherosclerosis and 31.2 mm^3 (IQR: 12.7-78.2 mm^3) among those aged 40-44 years. These findings suggest possible applications for noninvasive imaging in the more accurate individual diagnosis, intervention, and prevention.[27]

Our results also show that non-Hispanic Black participants tended to have more extensive

print & web 4C/FPO
UNCORRECTED PROOF

multiterritorial involvement. Nevertheless, separate analyses of the carotid and femoral territories revealed significant between-group differences in atherosclerosis prevalence and disease burden only in the carotid arteries. The disease burden was also generally higher in the carotid than in the femoral arteries. These findings diverge from studies of larger populations such as PESA, in which the iliofemoral territory was more extensively affected.[3] The PESA study cohort consisted of office workers at the Banco de Santander headquarters in Madrid, and the higher femoral involvement may have been driven by the presumably longer working hours per day spent in a sitting posture, which is associated with low oscillatory shear stress, high hydrostatic pressure, and disturbed flow caused by vessel curvature.[28] However, historically, iliofemoral involvement in atherosclerosis has received comparatively little attention, and the differences from the FAMILIA population could be explained by other factors, such as differences in the age profile or in the proportion of male and female participants. Exploration of the impact of socioeconomic status in the PESA population revealed no significant association between the presence of subclinical atherosclerosis and economic status but did find an association with lower education level, mainly related to higher tobacco consumption, a relationship well-described in the literature and also seen in our study.[29,30] These results highlighted once again the critical importance of implementing health promotion and targeted CV prevention strategies, such as smoking cessation and blood pressure control. Efforts in this area should be targeted at the more affected vulnerable communities, where the largest net gains are likely to be made.

STUDY LIMITATIONS. This is a cross-sectional study; therefore, causal inference cannot be evaluated. Most study participants were at low CV risk, and most were women who are known to have a low prevalence of CV disease, particularly at a younger age.[12] These factors explain the overall low presence of identifiable plaques. However, this is one of the first studies to assess the presence of subclinical atherosclerosis by 3DVUS in an underrepresented younger population.

Given the heterogeneity among racial and ethnic groups, assessing associations between self-reported racial or ethnic identity and disease is complex and is vulnerable to confounding due to the effects of socioeconomic inequality, environmental disparity, unequal access to care, and other possible emerging or unknown CV risk factors.[31,32] Although racial and ethnic identity may track the existence of certain alleles, these terms tend to denote superficial physical and sociocultural characteristics, and a more precise categorization of the distinct geographical origins in an individual's lineage can be obtained through analysis of genetic ancestry, especially given the history of genetic admixture and exchange between people of different ancestry. For example, the term Hispanic usually denotes a mixed European, Native American, and African ancestry.[33] Thus, whereas race is more a proxy for socioenvironmental exposure, genetic ancestry examines fixed characteristics in the genome and may help to improve understanding of health disparities and improve precision medicine in the future.

The population included in this study was from a specific area (Harlem, New York City) with known intrinsic health disparities compared with other areas in New York City. This could, to some extent, limit our results' generalizability. Nevertheless, the population studied was predominantly low-income, with participants mainly of Hispanic or non-Hispanic Black origin. Although data on other potential nontraditional risk factors such as glycated hemoglobin or lipoprotein(a) levels was not determined, this study controlled for a battery of risk factors, lifestyle habits, and socioeconomic status for the primary analysis, yielding relevant information on these underrepresented communities.

CONCLUSIONS

For the same predicted CV risk, non-Hispanic Black individuals appear to be more vulnerable than people of Hispanic origin to early subclinical atherosclerosis (particularly in the carotid arteries), potentially placing them at increased risk of clinical CV disease. Despite its limitations, including intrinsic socioenvironmental exposures partially captured by self-reported race and ethnicity, this study contributes to the understanding of higher rates of CV disease observed at an early age in disadvantaged communities. Until underlying biological factors and other undiscovered CV risk factors are better understood and can be addressed by precision medicine, affordable noninvasive imaging techniques such as the 3DVUS can provide valuable information about population disparities and increase the precision of health promotion and prevention programs.

ACKNOWLEDGMENTS The authors thank the children, parents/caregivers, family members, and school staff for their invaluable participation in the FAMILIA study; all volunteers who collaborated in the study; the imaging technicians Latifa Benkader and Christine Graziano-Zill for their invaluable work; Marc Soronellas for support for scientific illustration; and Simon Bartlett for English editing.

UNCORRECTED PROOF

FUNDING SUPPORT AND AUTHOR DISCLOSURES

This study was funded by the American Heart Association under grant No 14SFRN20490315 and the Stephen Gellman Children's Outreach Program. Dr Fernandez-Jimenez is recipient of grant PI19/01704 funded by the Fondo de Investigación Sanitaria- Instituto de Salud Carlos III (ISCIII) and co-funded by the European Regional Development Fund/European Social Fund "A way to make Europe"/"Investing in your future." Dr Santos-Beneit is recipient of grant LCF/PR/MS19/12220001 funded by "la Caixa" Foundation (ID 100010434). The CNIC is supported by the ISCIII, the Ministerio de Ciencia e Innovación (MCIN) and the Pro CNIC Foundation, and is a Severo Ochoa Center of Excellence (grant CEX2020-001041-S funded by MICIN/AEI/10.13039/501100011033). The other authors have reported that they have no relationships relevant to the contents of this paper to disclose.

ADDRESS FOR CORRESPONDENCE: Dr Fernández-Jiménez, Centro Nacional de Investigaciones Cardiovasculares (CNIC), Calle Melchor Fernández Almagro, 3, 28029, Madrid, Spain. E-mail: rfernandez@cnic.es. OR Dr Valentin Fuster, The Zena and Michael A. Wiener Cardiovascular Institute Icahn, School of Medicine at Mount Sinai. 1 Gustave L Levy Pl. New York, NY 10029, USA. E-mail: valentin.fuster@mountsinai.org. Twitter: @rodrigo_fjez, @kovacic_jason, @zahifayad, @CNIC_CARDIO, @MountSinaiHeart, @IcahnMountSinai.

PERSPECTIVES

COMPETENCY IN SYSTEMS-BASED PRACTICE: Atherosclerosis affects non-Hispanic Black individuals earlier in life and more severely than members of other racial and ethnic groups, and may contribute to racial and ethnic disparities in cardiovascular outcomes.

TRANSLATIONAL OUTLOOK: Further research is warranted to identify risk factors that explain the excess risk of cardiovascular disease in certain racial and ethnic groups.

REFERENCES

1. Carnethon MR, Pu J, Howard G, et al. Cardiovascular health in African Americans: a scientific statement from the American Heart Association. *Circulation*. 2017;136:e393–e423.

2. Arnett DK, Blumenthal RS, Albert MA, et al. 2019 ACC/AHA guideline on the primary prevention of cardiovascular disease: a report of the American College of Cardiology/American Heart Association task force on clinical practice guidelines. *J Am Coll Cardiol*. 2019;74:e177–e232.

3. Fernandez-Friera L, Penalvo JL, Fernandez-Ortiz A, et al. Prevalence, vascular distribution, and multiterritorial extent of subclinical atherosclerosis in a middle-aged cohort: the PESA (Progression of Early Subclinical Atherosclerosis) study. *Circulation*. 2015;131:2104–2113.

4. Bansilal S, Vedanthan R, Kovacic JC, et al. Rationale and design of Family-Based Approach in a Minority Community Integrating Systems-Biology for Promotion of Health (FAMILIA). *Am Heart J*. 2017;187:170–181.

5. Fernandez-Jimenez R, Jaslow R, Bansilal S, et al. Different lifestyle interventions in adults from underserved communities: the FAMILIA trial. *J Am Coll Cardiol*. 2020;75:42–56.

6. Fernandez-Jimenez R, Jaslow R, Bansilal S, et al. Child health promotion in underserved communities: the FAMILIA trial. *J Am Coll Cardiol*. 2019;73:2011–2021.

7. D'Agostino RB Sr, Vasan RS, Pencina MJ, et al. General cardiovascular risk profile for use in primary care: the Framingham Heart Study. *Circulation*. 2008;117:743–753.

8. Linden A. *Framingham: stata module for calculating the Framingham 10-year Cardiovascular Disease Risk Prediction*. 2015.

9. Gomez-Pardo E, Fernandez-Alvira JM, Vilanova M, et al. A comprehensive lifestyle peer group-based intervention on cardiovascular risk factors: the randomized controlled fifty-fifty program. *J Am Coll Cardiol*. 2016;67:476–485.

10. Fernandez-Alvira JM, Fuster V, Pocock S, et al. Predicting subclinical atherosclerosis in low-risk individuals: ideal cardiovascular health score and Fuster-BEWAT score. *J Am Coll Cardiol*. 2017;70:2463–2473.

11. Doménech JMN JB. *Find the best subset for Linear, Logistic and Cox Regression: User-written command confound for Stata [computer program]. V1.1.7*. Bellaterra: Universitat Autònoma de Barcelona; 2020.

12. Virani SS, Alonso A, Aparicio HJ, et al. Heart disease and stroke statistics-2021 update: a report from the American Heart Association. *Circulation*. 2021;143:e254–e743.

13. Hao G, Wang X, Treiber FA, et al. Growth of carotid intima-media thickness in black and white young adults. *J Am Heart Assoc*. 2016;5(12):e004147.

14. Thomas SJ, Booth JN 3rd, Dai C, et al. Cumulative incidence of hypertension by 55 years of age in blacks and whites: the CARDIA study. *J Am Heart Assoc*. 2018;7(14):e007988.

15. Siemelink MA, van der Laan SW, Haitjema S, et al. Smoking is associated to DNA methylation in atherosclerotic carotid lesions. *Circ Genom Precis Med*. 2018;11:e002030.

16. Libby P, Buring JE, Badimon L, et al. *Atherosclerosis. Nat Rev Dis Primers*. 2019;5(1):56.

17. Ryder JR, Northrop E, Rudser KD, et al. Accelerated early vascular aging among adolescents with obesity and/or type 2 diabetes mellitus. *J Am Heart Assoc*. 2020;9:e014891.

18. Turco JV, Inal-Veith A, Fuster V. Cardiovascular health promotion: an issue that can no longer wait. *J Am Coll Cardiol*. 2018;72:908–913.

19. Ammous F, Zhao W, Lin L, et al. Epigenetics of single-site and multi-site atherosclerosis in African Americans from the Genetic Epidemiology Network of Arteriopathy (GENOA). *Clin Epigenetics*. 2022;14:10.

20. Lutsey PL, Diez Roux AV, Jacobs DR Jr, et al. Associations of acculturation and socioeconomic status with subclinical cardiovascular disease in the multi-ethnic study of atherosclerosis. *Am J Public Health*. 2008;98:1963–1970.

21. Wassel CL, Pankow JS, Peralta CA, Choudhry S, Seldin MF, Arnett DK. Genetic ancestry is associated with subclinical cardiovascular disease in African-Americans and Hispanics from the multi-ethnic study of atherosclerosis. *Circ Cardiovasc Genet*. 2009;2:629–636.

22. Gebreab SY, Diez Roux AV, Brenner AB, et al. The impact of lifecourse socioeconomic position on cardiovascular disease events in African Americans: the Jackson Heart Study. *J Am Heart Assoc*. 2015;4:e001553.

23. Bureau USC. Poverty Thresholds.

24. Ahmadi A, Argulian E, Leipsic J, Newby DE, Narula J. From subclinical atherosclerosis to plaque progression and acute coronary events: JACC State-of-the-Art Review. *J Am Coll Cardiol*. 2019;74:1608–1617.

25. Willeit P, Tschiderer L, Allara E, et al. Carotid intima-media thickness progression as surrogate marker for cardiovascular risk: meta-analysis of 119 clinical trials involving 100,667 patients. *Circulation*. 2020;142:621–642.

26. Lopez-Melgar B, Fernandez-Friera L, Oliva B, et al. Subclinical atherosclerosis burden by 3D

UNCORRECTED PROOF

ultrasound in mid-life: the PESA study. *J Am Coll Cardiol*. 2017;70:301-313.

27. Kozakova M, Palombo C. Imaging subclinical atherosclerosis in cardiovascular risk stratification. *Eur J Prev Cardiol*. 2021;28:247-249.

28. Wilkinson MJ, Manoogian ENC, Zadourian A, et al. Ten-hour time-restricted eating reduces weight, blood pressure, and atherogenic lipids in patients with metabolic syndrome. *Cell Metab*. 2020;31:92-104.e5.

29. Redondo-Bravo L, Fernandez-Alvira JM, Gorriz J, et al. Does socioeconomic status influence the risk of subclinical atherosclerosis? A mediation model. *J Am Coll Cardiol*. 2019;74:526-535.

30. Jamal A, Phillips E, Gentzke AS, et al. Current cigarette smoking among adults — United States, 2016. *MMWR Morb Mortal Wkly Rep*. 2018;67:53-59.

31. Burchard EG, Ziv E, Coyle N, et al. The importance of race and ethnic background in biomedical research and clinical practice. *N Engl J Med*. 2003;348:1170-1175.

32. Sinha M, Larkin EK, Elston RC, Redline S. Self-reported race and genetic admixture. *N Engl J Med*. 2006;354:421-422.

33. Borrell LN, Elhawary JR, Fuentes-Afflick E, et al. Race and genetic ancestry in medicine — a time for reckoning with racism. *N Engl J Med*. 2021;384:474-480.

KEY WORDS cardiovascular disease, ethnicity, prevention, race, subclinical atherosclerosis, vascular ultrasound

APPENDIX For supplemental methods, tables, and figures, please see the online version of this paper.

UNCORRECTED PROOF

Subclinical Atherosclerosis in Young, Socioeconomically Vulnerable Hispanic and Non-Hispanic Black Adults

Josep Iglesies-Grau, Rodrigo Fernandez-Jimenez, Raquel Diaz-Munoz, Risa Jaslow, Amaya de Cos-Gandoy, Gloria Santos-Beneit, Christopher A. Hill, Alexandra Turco, Daniella Kadian-Dodov, Jason C. Kovacic, Zahi A. Fayad, Valentin Fuster

The authors assessed the impact of race/ethnicity on early subclinical atherosclerosis in 436 young adults from a vulnerable community enrolled in the FAMILIA (~~Family-based Approach to Promotion of Health–FAMILIA [Project 2]~~) trial who underwent 3-dimensional vascular ultrasound. Subclinical atherosclerosis prevalence was 12.9% among non-Hispanic Black participants and 6.6% among those identifying as Hispanic. After multivariable adjustment, non-Hispanic Black participants were more likely to have subclinical atherosclerosis than Hispanic participants (OR: 3.45; 95% CI: 1.44-8.29). These results suggest the existence of unknown cardiovascular factors underlying such residual excess risk. Early detection of atherosclerosis could play an important role in prevention programs in different populations.

oooo

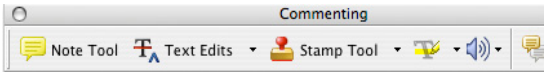
UNCORRECTED PROOF

ELSEVIER

INSTRUCTIONS ON THE ANNOTATION OF PDF FILES

To view, print and annotate your content you will need Adobe Reader version 9 (or higher). This program is freely available for a whole series of platforms that include PC, Mac, and UNIX and can be downloaded from http://get.adobe.com/reader/. The exact system requirements are given at the Adobe site: http://www.adobe.com/products/reader/tech-specs.html.

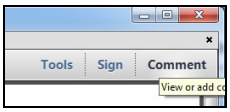
Note: ***Please do NOT make direct edits to the PDF using the editing tools as doing so could lead us to overlook your desired changes.*** *Rather, please request corrections by using the tools in the Comment pane to annotate the PDF and call out the changes you are requesting. If you opt to annotate the file with software other than Adobe Reader then please also highlight the appropriate place in the PDF file.*

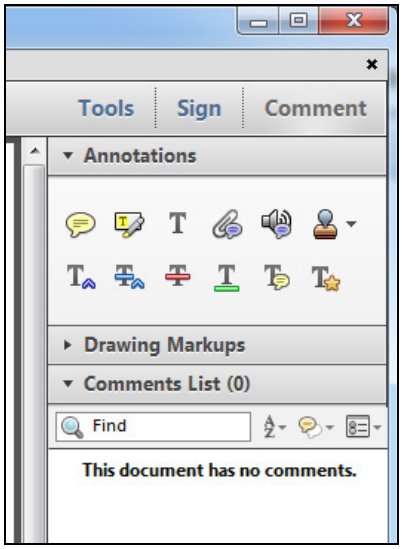
<table>
<tr><th colspan="2">PDF ANNOTATIONS</th></tr>
<tr><th>Adobe Reader version 9</th><th>Adobe Reader version X and XI</th></tr>
<tr><td>When you open the PDF file using Adobe Reader, the Commenting tool bar should be displayed automatically; if not, click on ‘Tools’, select ‘Comment & Markup’, then click on ‘Show Comment & Markup tool bar’ (or ‘Show Commenting bar’ on the Mac). If these options are not available in your Adobe Reader menus then it is possible that your Adobe Acrobat version is lower than 9 or the PDF has not been prepared properly.
 

(Mac)
PDF ANNOTATIONS (Adobe Reader version 9)

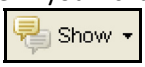
The default for the Commenting tool bar is set to ‘off’ in version 9. To change this setting select ‘Edit | Preferences’, then ‘Documents’ (at left under ‘Categories’), then select the option ‘Never’ for ‘PDF/A View Mode’.
 

(Changing the default setting, Adobe version 9)</td><td>To make annotations in the PDF file, open the PDF file using Adobe Reader XI, click on ‘Comment’.

If this option is not available in your Adobe Reader menus then it is possible that your Adobe Acrobat version is lower than XI or the PDF has not been prepared properly.
 

This opens a task pane and, below that, a list of all Comments in the text. These comments initially show all the changes made by our copyeditor to your file.
 
</td></tr>
</table>

HOW TO...		
Action	**Adobe Reader version 9**	**Adobe Reader version X and XI**
Insert text	Click the 'Text Edits' button  on the Commenting tool bar. Click to set the cursor location in the text and simply start typing. The text will appear in a commenting box. You may also cut-and-paste text from another file into the commenting box. Close the box by clicking on 'x' in the top right-hand corner.	Click the 'Insert Text' icon  on the Comment tool bar. Click to set the cursor location in the text and simply start typing. The text will appear in a commenting box. You may also cut-and-paste text from another file into the commenting box. Close the box by clicking on '_'  in the top right-hand corner.
Replace text	Click the 'Text Edits' button  on the Commenting tool bar. To highlight the text to be replaced, click and drag the cursor over the text. Then simply type in the replacement text. The replacement text will appear in a commenting box. You may also cut-and-paste text from another file into this box. To replace formatted text (an equation for example) please Attach a file (see below).	Click the 'Replace (Ins)' icon  on the Comment tool bar. To highlight the text to be replaced, click and drag the cursor over the text. Then simply type in the replacement text. The replacement text will appear in a commenting box. You may also cut-and-paste text from another file into this box. To replace formatted text (an equation for example) please Attach a file (see below).
Remove text	Click the 'Text Edits' button  on the Commenting tool bar. Click and drag over the text to be deleted. Then press the delete button on your keyboard. The text to be deleted will then be struck through.	Click the 'Strikethrough (Del)' icon  on the Comment tool bar. Click and drag over the text to be deleted. Then press the delete button on your keyboard. The text to be deleted will then be struck through.
Highlight text/ make a comment	Click on the 'Highlight' button  on the Commenting tool bar. Click and drag over the text. To make a comment, double click on the highlighted text and simply start typing.	Click on the 'Highlight Text' icon  on the Comment tool bar. Click and drag over the text. To make a comment, double click on the highlighted text and simply start typing.
Attach a file	Click on the 'Attach a File' button  on the Commenting tool bar. Click on the figure, table or formatted text to be replaced. A window will automatically open allowing you to attach the file. To make a comment, go to 'General' in the 'Properties' window, and then 'Description'. A graphic will appear in the PDF file indicating the insertion of a file.	Click on the 'Attach File' icon  on the Comment tool bar. Click on the figure, table or formatted text to be replaced. A window will automatically open allowing you to attach the file. A graphic will appear indicating the insertion of a file.
Leave a note/ comment	Click on the 'Note Tool' button  on the Commenting tool bar. Click to set the location of the note on the document and simply start typing. Do not use this feature to make text edits.	Click on the 'Add Sticky Note' icon  on the Comment tool bar. Click to set the location of the note on the document and simply start typing. Do not use this feature to make text edits.

HOW TO...		
Action	**Adobe Reader version 9**	**Adobe Reader version X and XI**
Review	To review your changes, click on the 'Show' button  on the Commenting tool bar. Choose 'Show Comments List'. Navigate by clicking on a correction in the list. Alternatively, double click on any mark-up to open the commenting box.	Your changes will appear automatically in a list below the Comment tool bar. Navigate by clicking on a correction in the list. Alternatively, double click on any mark-up to open the commenting box.
Undo/delete change	To undo any changes made, use the right click button on your mouse (for PCs, Ctrl-Click for the Mac). Alternatively click on 'Edit' in the main Adobe menu and then 'Undo'. You can also delete edits using the right click (Ctrl-click on the Mac) and selecting 'Delete'.	To undo any changes made, use the right click button on your mouse (for PCs, Ctrl-Click for the Mac). Alternatively click on 'Edit' in the main Adobe menu and then 'Undo'. You can also delete edits using the right click (Ctrl-click on the Mac) and selecting 'Delete'.

SEND YOUR ANNOTATED PDF FILE BACK TO ELSEVIER

Save the annotations to your file and return as instructed by Elsevier. Before returning, please ensure you have answered any questions raised on the Query Form and that you have inserted all corrections: later inclusion of any subsequent corrections cannot be guaranteed.

FURTHER POINTS

- Any (grey) halftones (photographs, micrographs, etc.) are best viewed on screen, for which they are optimized, and your local printer may not be able to output the greys correctly.
- If the PDF files contain colour images, and if you do have a local colour printer available, then it will be likely that you will not be able to correctly reproduce the colours on it, as local variations can occur.
- If you print the PDF file attached, and notice some 'non-standard' output, please check if the problem is also present on screen. If the correct printer driver for your printer is not installed on your PC, the printed output will be distorted.