Effect of ApoE4 Genotype on the Association Between Metabolic Phenotype and Subclinical Atherosclerosis in Postmenopausal Women

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Metabolic profile and ApoE4 genotype have effects on coronary heart disease. We examined the interaction between these factors on subclinical atherosclerosis in postmenopausal women from the Early versus Late Intervention Trial with Estradiol (n = 497). Based on nine metabolic biomarkers (fasting blood glucose, insulin sensitivity, ketones, triglycerides, high-density lipoprotein, low-density lipoprotein, hemoglobin A1c, and blood pressure), K-means clustering categorized women into three distinct phenotypes: healthy, high blood pressure, and poor metabolic. ApoE4 genotype was classified as either ApoE4+ or ApoE4−. General linear models tested whether the cross-sectional association between metabolic phenotypes and common carotid intima media thickness (CIMT) differed by ApoE4 genotype. Mixed effects linear models evaluated the modifying role of ApoE4 genotype on the association of metabolic phenotype with CIMT progression over a median follow-up of 4.8 years. In cross-sectional analysis, ApoE4+ women with poor metabolic phenotype had the highest CIMT compared with all other groups. In ApoE4− women, CIMT was significantly lower in those classified as healthy compared with high blood pressure phenotype (p = 0.004). In ApoE4+ women, CIMT was significantly higher in those with poor metabolic phenotype compared with healthy (p = 0.0003) and high blood pressure (p = 0.001) phenotypes. These results indicate that metabolic phenotype had a negative effect on CIMT in women with ApoE4+ but not ApoE4− (interaction p = 0.001). These effects were not observed on CIMT progression in longitudinal analysis. In conclusion, ApoE4+ women are more likely to have higher levels of subclinical atherosclerosis if their metabolic phenotype is poor compared with ApoE4+ women without poor metabolic profile and ApoE4− women. **(Am J Cardiol 2019;124:1031−1037)**

Metabolic syndrome, characterized by high blood pressure, hyperglycemia, elevated triglycerides, decreased high-density lipoprotein cholesterol (HDL-C), and abdominal obesity,[1] is one example of a metabolic phenotype, a cluster of multiple risk factors for cardiovascular disease (CVD). Individuals with metabolic syndrome have a 2.02-fold (95% confidence interval, 1.42 to 2.89) greater risk of CVD-related mortality,[2] and higher levels of subclinical atherosclerosis measured as carotid artery intima media thickness (CIMT).[3] Apolipoprotein E (ApoE) is a key protein in lipoprotein metabolism. Homozygous or heterozygous carriers of the ApoE4 allele are at increased risk of coronary heart disease (CHD) and demonstrate higher levels of subclinical atherosclerosis.[4−7] Although metabolic phenotype and ApoE4 genotype have been independently linked with subclinical atherosclerosis, their joint effect on atherosclerosis is largely unknown. We previously reported a significant interaction between ApoE4 genotype and metabolic phenotype on cognition.[8] In this study, we tested the hypothesis that the association of metabolic phenotype with subclinical atherosclerosis is modified by ApoE4 genotype.

Methods

This was a post hoc analysis in postmenopausal women participating in the Early versus Late Intervention Trial with Estradiol (ELITE). Details of ELITE methods and primary results have been published.[9,10] Briefly, ELITE was a single-center, randomized, double-blinded, placebo-controlled trial of hormone therapy (HT) in postmenopausal women, stratified within 6 years of menopause (early postmenopause) and

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10 years or more after menopause (late postmenopause). ELITE was specifically designed to test the HT timing hypothesis, that is, whether the effects of HT vary according to the timing of HT initiation in relation to menopause. Eligible participants were postmenopausal women with no clinical history of CVD or diabetes. A total of 643 postmenopausal women were randomized to receive either HT or placebo according to time since menopause using a 1:1 ratio of stratified blocked randomization. The HT regimen was oral micronized 17-beta-estradiol 1 mg/day with (in women with uterus) or without 4% vaginal micronized progesterone gel 45 mg/day for 10 days each month. After randomization, women attended study clinic visits every month for the first 6 months and every other month thereafter until trial completion. ELITE was conducted from July 2005 to February 2013 with a median duration of follow-up of 4.8 (range 0.5 to 6.7) years. The primary results of ELITE showed that HT reduced progression of subclinical atherosclerosis (measured as rate of change in CIMT) compared with placebo when therapy was initiated in early, but not in late postmenopausal women.[10] The ELITE trial was registered on ClinicalTrials.gov (NCT00114517), funded by the National Institute on Aging, National Institutes of Health (R01AG-024154) and was approved by the Institutional Review Board of the University of Southern California.

At baseline and at every 6-month follow-up visit, blood pressure was recorded and fasting blood was drawn. Fasting glucose, insulin, ketones, total cholesterol, triglycerides, HDL-cholesterol, low-density lipoprotein cholesterol (LDL-C), and hemoglobin A1c (HbA1c) levels were measured as previously described.[11,12] The homeostatic model assessment (HOMA), a measure of insulin resistance, was measured as [glucose mmol/l x insulin]/22.5. ApoE genotype was determined with standard methodology[13] as described in our previous studies.[8] Briefly, 3 isoforms (E2, E3, and E4) of the apolipoprotein E gene were determined according to 2 nonsynonymous single nucleotide polymorphisms (rs429358 and rs7412) encoding arginine for cysteine amino acid variants at codon positions 112 and 158, respectively (TaqMan Assay-on-Demand Genotyping Service; Applied Biosystems, Foster City, California). ApoE4 genotype was defined as being positive for 1 or 2 copies of the E4 allele (E2/E4, E3/E4, E4/E4) and was otherwise defined as negative (E2/E2, E2/E3, E3/E3).

The metabolic risk phenotypes within the ELITE population were determined using 9 biomarkers: glucose, the HOMA score, ketones, triglycerides, HDL-C, LDL-C, HbA1c, and systolic and diastolic blood pressure.[14] A K-means clustering algorithm was used to identify 3 clusters; healthy, high blood pressure and poor metabolic phenotypes, that were descriptively identified based on their biomarker profile. Predefining the number of clusters as 3 (based on a previous factor analysis of the 9 biomarkers), the K-means algorithm classified each participant observation into a specific cluster, based on the similarity (measured as Euclidean distance) of the participant's biomarker profile to the cluster mean (or centroid). The labeling of each cluster was defined by the mean cluster biomarker profile. Although the clustering was performed in the total ELITE sample (n = 643), the cohort in this study was restricted to women with ApoE genotype data (n = 497): healthy (n = 208, 41.9%), high blood pressure (n = 190, 38.2%), and poor metabolic (n = 99, 19.9%). As previously reported, the majority of the metabolic biomarker means were within their respective normal ranges, consistent with recruitment of a healthy population of postmenopausal women in the study. Therefore, metabolic indicator means in the poor metabolic phenotype were at the margins of clinically healthy values.[8] Our previous studies showed that these metabolic phenotypes were significantly associated with cognitive function[14]; this association was significantly modified by ApoE4 genotype.[8]

CIMT of the far wall of the right distal common carotid artery was assessed by computer image processing of B-mode ultrasonograms that were obtained at baseline and at every 6-month follow-up visit. Serial CIMT imaging and measurement methodology were specifically developed for longitudinal assessment of change in atherosclerosis; the coefficient of variation for baseline CIMT measurement was 0.69%.[15,16]

Baseline characteristics of the total cohort are reported as mean (SD) for continuous variables and as frequency (%) for categorical variables. Comparison of baseline characteristics including CIMT in the 3 metabolic phenotypes and in 6 groups defined by the combination of metabolic phenotype and ApoE4 genotype were conducted with analysis of variance and chi-square tests, which were reported elsewhere.[8]

A general linear model was used to test whether the cross-sectional association between metabolic phenotypes and baseline CIMT differed by ApoE4 genotype after adjusting for age. The analysis was conducted in the total cohort and stratified by time-since-menopause strata (early and late postmenopause). The modifying effect of ApoE4 genotype on the association between metabolic phenotype and baseline CIMT was tested by including a product term of ApoE4 genotype*metabolic phenotype in the model.

Linear mixed effects models were used to evaluate whether the longitudinal association between metabolic phenotypes and CIMT progression differed by ApoE4 genotype over a median follow-up of 4.8 years controlling for age. Random effects were specified for participant-specific intercept (baseline CIMT) and slope (CIMT progression); time since randomization (years) was included as a continuous time variable in the model. A product term of phenotype-by-time was used to estimate and test for differences in CIMT progression in phenotypes. The analysis was conducted in the total cohort and stratified by time-since-menopause stratum (early and late postmenopause) and randomized intervention stratum (HT and placebo). The modifying effect of ApoE4 genotype on the association between metabolic phenotype and CIMT progression was tested by including a 3-level product term (ApoE4 genotype*metabolic phenotype*time) in the model.

Results

A total of 497 postmenopausal women from ELITE with metabolic phenotype and ApoE genotype data contributed to the current analysis. Women were on average (SD) 59.3 (7.0) years old and were postmenopausal for an average duration of 10.4 (7.8) years. Forty-three percent of the women were stratified as early menopause, whereas

57% were late menopause. Most women were non-Hispanic white. The ApoE allele frequency (%) was: E2/E2 n = 2 (0.40%); E2/E4 n = 6 (1.21%); E4/E4 n = 20 (4.02%); E2/E3 n = 53 (10.66%); E3/E4 n = 128 (25.75%); and E3/E3 n = 288 (57.95%). Sixty-nine percent of women were ApoE4− and 31% were ApoE4+ genotype (Table 1).

In the 3 metabolic phenotypes, the ApoE4 genotype distribution was comparable; race/ethnicity was significantly different, with a larger proportion of Hispanic women in the poor metabolic phenotype. The phenotype differences in metabolic markers were similar between ApoE4+ and ApoE4− women except for LDL-C and diastolic blood pressure, which had significant interaction between metabolic phenotype and ApoE4 genotype.[8] ApoE4+ women with poor metabolic phenotype had higher LDL-C and diastolic blood pressure level.

At baseline, mean CIMT significantly differed in the metabolic phenotypes (p <0.0001). Women in the poor metabolic phenotype had the highest mean CIMT, followed by women in the high blood pressure and healthy phenotypes. ApoE4+ women had significantly higher CIMT compared with ApoE4− women (p <0.0001). These associations were significant in both early and late postmenopausal women. (Table 2)

In the total cohort, ApoE4+ carriers with poor metabolic phenotype had the highest baseline CIMT. In ApoE4+ carriers, CIMT was statistically significantly higher in the poor metabolic phenotype compared with the other 2 phenotypes. In ApoE4− women, CIMT significantly differed between the healthy and high blood pressure phenotypes. Overall, adjusted for age, ApoE4 genotype significantly modified the effect of metabolic phenotypes on CIMT (metabolic phenotype*ApoE4 genotype interaction p = 0.001). ApoE4 carriers had an apparent negative association with atherosclerosis from having poor metabolic phenotype, whereas this negative association was not evident in ApoE4 noncarriers. The modifying effect of ApoE4 genotype was equally evident in early and late postmenopausal women (metabolic phenotype*ApoE4 genotype*time-since-menopause interaction p = 0.59; Table 2, Figure 1).

In longitudinal analysis adjusting for age, the annual mean CIMT progression rate was highest in ApoE4+ women in the poor metabolic phenotype and high blood pressure phenotype (Table 3). Neither metabolic phenotype (p = 0.83) nor ApoE4 genotype (p = 0.14) were associated with CIMT progression rate. The difference in CIMT progression rate between ApoE4+ and ApoE4− carriers increased from healthy, high blood pressure, and poor metabolic phenotypes, respectively. However, ApoE4 genotype did not significantly modify the effect of metabolic phenotype on CIMT progression rate (metabolic phenotype*ApoE4 genotype interaction p = 0.79) in the total sample (Table 3).

Table 1
Baseline characteristics by metabolic phenotype and ApoE4 genotype

	Metabolic phenotype and ApoE4 genotype						
	Healthy		High blood pressure		Poor metabolic		
Characteristics	ApoE4-	ApoE4+	ApoE4-	ApoE4+	ApoE4-	ApoE4+	Total
Sample size	147	61	129	61	67	32	497
Age (years)	59.7 (7.5)	58.7 (6.8)	60.4 (6.9)	59.7 (6.6)	60.3 (6.1)	61.1 (7.4)	59.3 (7.0)
Time since menopause (years)	10.4 (7.8)	8.6 (6.3)	10.1 (7.5)	11.2 (8.2)	11.5 (9.0)	11.6 (7.8)	10.4 (7.8)
Time-since-menopause stratum							
Early <6 years)	64 (43.5%)	30 (49.2%)	56 (43.4%)	25 (41.0%)	28 (41.8%)	11 (34.4%)	214 (43.1%)
Late (≥10 years)	83 (56.5%)	31 (50.8%)	73 (56.6%)	36 (59.0%)	39 (58.2%)	21 (65.6%)	283 (56.9%)
Education (years)	16.2 (2.2)	16.7 (2.0%)	16.1 (2.1%)	16.4 (1.9%)	15.6 (2.4%)	15.9 (1.9%)	16.2 (2.2%)
Race/Ethnicity							
White, non-Hispanic	109 (74.1%)	48 (78.7%)	92 (71.3%)	48 (78.7%)	40 (59.7%)	17 (53.1%)	354 (71.2%)
Black	8 (5.4%)	5 (8.2%)	10 (7.8%)	7 (11.5%)	5 (7.5%)	4 (12.5%)	39 (7.9%)
Hispanic	11 (7.5%)	6 (9.8%)	15 (11.6%)	6 (9.8%)	16 (23.8%)	8 (25.0%)	62 (12.5%)
Asian	19 (13.0%)	2 (3.3%)	12 (9.3%)	0 (0%)	6 (9.0%)	3 (9.4%)	42 (8.45%)
Randomized intervention							
Hormone therapy	76 (51.7%)	29 (47.5%)	62 (48.1%)	30 (49.2%)	33 (49.3%)	17 (53.1%)	250 (50.3%)
Placebo	71 (48.3%)	32 (52.5%)	67 (51.9%)	31 (50.8%)	34 (50.7%)	15 (46.9%)	247 (49.7%)
Biomarkers							
Glucose (mg/dl)	80.8 (7.3)	80.3 (8.2%)	80.0 (7.4%)	80.7 (7.6%)	92.1 (10.5%)	90.2 (1.0%)	94.9 (10.3%)
Insulin resistance (HOMA score)	1.0 (0.5)	0.9 (0.4)	1.2 (0.5)	1.2 (0.5)	2.6 (1.2)	2.6 (1.0)	1.4 (0.9)
Ketone (mM)	0.13 (0.07)	0.12 (0.05)	0.10 (0.03)	0.10 (0.03)	0.10 (0.03)	0.11 (0.04)	0.1 (0.05)
High density lipoprotein cholesterol (md/dl)	76.5 (18.4)	71.5 (16.1)	66.6 (16.6)	62.7 (13.3)	52.5 (11.2)	51.5 (10.4)	66.8 (18.0)
Low density lipoprotein cholesterol (md/dl)*	129.7 (29.8)	131.1 (29.4)	135.7 (27.2)	139.2 (32.3)	136.0 (30.0)	161.0 (34.5)	135.4 (30.3)
Triglycerides (mg/dl)	79.8 (27.3)	82.0 (26.5)	97.1 (32.0)	98.2 (36.4)	167.1 (72.4)	164.2 (51.4)	105.2 (53.0)
HbA1c (%)	5.6 (0.4)	5.6 (0.4)	5.5 (0.4)	5.5 (0.4)	5.8 (0.4)	5.7 (0.5)	5.6 (0.4)
Systolic Blood Pressure (mm Hg)	106.2 (8.9)	105.1 (9.0)	125.7 (9.1)	124.3 (12.5)	120.7 (9.7)	122.8 (12.5)	117.5 (87.3)
Diastolic blood pressure (mm Hg)*	68.2 (5.5)	67.4 (5.6)	80.8 (5.7)	80.9 (6.0)	75.3 (7.3)	78.4 (8.4)	74.8 (56.3)

Continuous variables presented as mean (standard variation), categorical variable presented as frequency (percent).

ApoE4 genotype was defined as positive for both heterozygous and homozygous genotypes containing the E4 allele; and was otherwise defined as negative.

* Significant interaction between metabolic phenotype and ApoE4 genotype (p <0.05).

Table 2
Estimated mean CIMT (SE) by metabolic phenotype and ApoE4 genotype (total cohort, by time-since-menopause strata)

Genotype		(1) Healthy	(2) High blood pressure	(3) Poor metabolic	Pairwise p value			ApoE4	Metabolic phenotype	Interaction
		Mean (SE)	Mean (SE)	Mean (SE)	(1) vs (2)	(1) vs (3)	(2) vs (3)	p value	p value	p value
Total		n = 208	n = 190	n = 99						
ApoE4-negative	n = 343	755.65 (8.14)	790.17 (8.69)	770.14 (12.05)	0.004	0.32	0.18	<0.0001	<0.0001	0.001
ApoE4-positive	n = 154	753.68 (12.66)	756.76 (12.63)	832.41 (17.45)	0.86	0.0003	0.001			
Early postmenopause		n = 94	n = 81	n = 39						
ApoE4-negative	n = 148	730.25 (11.10)	772.55 (11.90)	755.23 (16.71)	0.01	0.21	0.40	0.01	0.002	0.05
ApoE4-positive	n = 66	743.29 (16.24)	736.10 (17.71)	805.82 (26.67)	0.76	0.05	0.03			
Late postmenopause		n = 114	n = 109	n = 60						
ApoE4-negative	n = 195	777.68 (11.59)	801.65 (12.36)	782.19 (16.91)	0.16	0.83	0.35	<0.0001	<0.0001	0.02
ApoE4-positive	n = 88	763.72 (18.98)	774.92 (17.61)	851.69 (23.05)	0.67	0.004	0.01			

CIMT reported as μm; Model adjusted for age; Tukey-Kramer method was used to test for pairwise comparisons; metabolic phenotype*ApoE4 genotype*time-since-menopause interaction p value = 0.59; CIMT = carotid artery intima media thickness.

ApoE4 genotype was defined as positive for both heterozygous and homozygous genotypes containing the E4 allele, otherwise, it was defined as negative.

The nonsignificant interaction between metabolic phenotype and ApoE4 genotype was consistent in early and late postmenopausal women (metabolic phenotype*ApoE4 genotype*time-since-menopause interaction p = 0.44) and women who were randomized to HT and placebo (metabolic phenotype*ApoE4 genotype*randomized intervention interaction p = 0.22) (Table 3).

As previous reports linked ApoE2 genotype with decreased subclinical atherosclerosis and risk of developing and dying from CHD,[17–19] we explored the effect of ApoE2 genotype (E2/E2, E2/E3, E2/E4) on subclinical atherosclerosis. At baseline, we found a significant protective association of ApoE2+ on atherosclerosis (p <0.0001). However, there was no significant interaction between ApoE2 genotype and metabolic phenotype on CIMT (p = 0.71). These results were consistent in the total cohort and in sensitivity analyses in ApoE4− women (Table 4, Figure 2).

Discussion

In a sample of 497 healthy postmenopausal women, metabolic phenotype and ApoE4 genotype showed a significant interaction on subclinical atherosclerosis. Consistent with previous reports, we identified an independent association of ApoE4 genotype on level of subclinical atherosclerosis.[18] A pooled analysis of 60,883 individuals from 16 prospective cohort studies showed the highest CIMT and stroke risk in ApoE4+ carriers compared with ApoE4− genotype (p trend = 0.001).[18]

Women classified in the poor metabolic phenotype, where mean values of the 9 biomarkers were close to the preclinical stage of metabolic syndrome, had the highest CIMT compared with women with healthy and high blood pressure phenotypes. These results are thus consistent with the increased risk of atherosclerosis with metabolic syndrome.[3]

Perhaps the most striking finding of our analysis is that the adverse influence of metabolic dysregulation on atherosclerosis was modified by ApoE4 genotype, such that the negative metabolic impact was apparent only in ApoE4+ carriers. The effect of ApoE4 genotype on CHD appears to differ by gender, hyperglycemia, smoking, alcohol consumption, body size, and HT as ApoE4 increases the risk associated with these factors.[20–22] A significant interaction between ApoE4 genotype and diabetes on atherosclerosis was reported from the Framingham Heart Study and suggested higher levels of atherosclerosis in individuals with both ApoE4+ and metabolic syndrome.[23,24] Another study reported differential effects of ApoE4 genotype on lipoprotein particles between middle-aged healthy individuals with and without dyslipidemia.[25] The present study is consistent with previous studies showing a significant positive modifying effect of ApoE4 genotype on the association between metabolic phenotype and subclinical atherosclerosis in postmenopausal women.[20–24]

The modifying role of ApoE4 genotype on metabolic phenotype and subclinical atherosclerosis may occur through the effect of ApoE4 genotype on lipoprotein metabolism, as ApoE genotype mediates the binding of lipoproteins or lipid complexes to their receptors[26] and affects lipid levels especially LDL-C and HDL-C.[18,27–29] Our

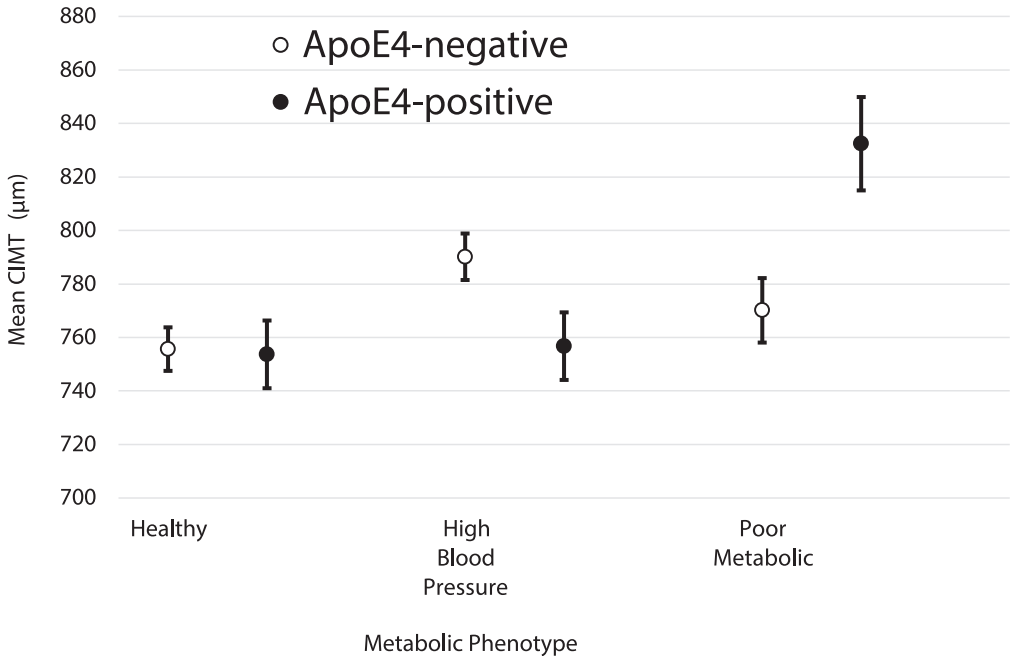

Figure 1. Estimated mean CIMT (SE) by metabolic phenotype and ApoE4 genotype (total cohort).

findings support the possibility that LDL-C level contributes to the effect of ApoE4 genotype on atherosclerosis as ApoE4+ carriers with poor metabolic phenotype had significantly higher LDL-C levels than ApoE4− carriers with poor metabolic phenotype (Table 1). The ApoE4 genotype is also a major risk factor for Alzheimer's disease,[30] and we have similarly shown the adverse impact of metabolic dysregulation on cognition, specifically in ApoE4+ women in the same group of postmenopausal women from ELITE.[8,14]

We did not find a modifying effect of ApoE4 genotype in the longitudinal association between metabolic phenotype and atherosclerosis progression. This could be due to limited power of longitudinal analysis. The effects of metabolic phenotype, ApoE4 genotype and their interaction may also have been more apparent in cross-sectional versus longitudinal analysis as the baseline cross-sectional analysis reflects long-term exposure and cumulative effects of those risk factors on atherosclerosis. Due to

Table 3
Estimated mean annual CIMT progression rate (SE) by metabolic phenotype and ApoE4 genotype (total cohort, by time-since-menopause strata, randomized intervention strata)

		(1) Healthy	(2) High blood pressure	(3) Poor metabolic	Longitudinal		
Genotype		Mean (SE)	Mean (SE)	Mean (SE)	ApoE4 p value	Metabolic phenotype p value	Interaction p value
Total		n = 208	n = 190	n = 99			
ApoE4-negative	n = 343	7.19 (0.85)	7.45 (0.90)	6.77 (1.25)	0.14	0.83	0.79
ApoE4-positive	n = 154	7.84 (1.31)	8.90 (1.31)	9.24 (1.82)			
Early postmenopause		n = 94	n = 81	N = 39			
ApoE4-negative	n = 148	6.42 (1.33)	5.63 (1.43)	4.16 (2.02)	0.81	0.93	0.52
ApoE4-positive	n = 66	4.33 (1.95)	6.37 (2.14)	6.76 (3.23)			
Late postmenopause		n = 114	n = 109	N = 60			
ApoE4-negative	n = 195	7.79 (1.07)	8.85 (1.14)	8.64 (1.55)	0.06	0.99	0.82
ApoE4-positive	n = 88	11.24 (1.74)	10.65 (1.62)	10.54 (2.12)			
Hormone therapy		n = 103	n = 98	N = 49			
ApoE4-negative	n = 172	8.33 (1.31)	5.74 (1.35)	6.84 (1.89)	0.27	0.73	0.31
ApoE4-positive	n = 78	6.89 (1.95)	8.55 (1.98)	10.76 (2.86)			
Placebo		n = 105	n = 92	n = 50			
ApoE4-negative	n = 171	6.13 (1.08)	9.30 (1.19)	6.69 (1.63)	0.33	0.38	0.63
ApoE4-positive	n = 76	8.90 (1.74)	9.27 (1.71)	7.90 (2.28)			

CIMT progression rate reported as μm per year; Model adjusted for age; metabolic phenotype*ApoE4 genotype*time-since-menopause interaction p value = 0.44; metabolic phenotype*ApoE4 genotype*randomized intervention interaction p value = 0.22; CIMT = carotid artery intima media thickness.

ApoE4 genotype was defined as positive for both heterozygous and homozygous genotypes containing the E4 allele, otherwise, it was defined as negative.

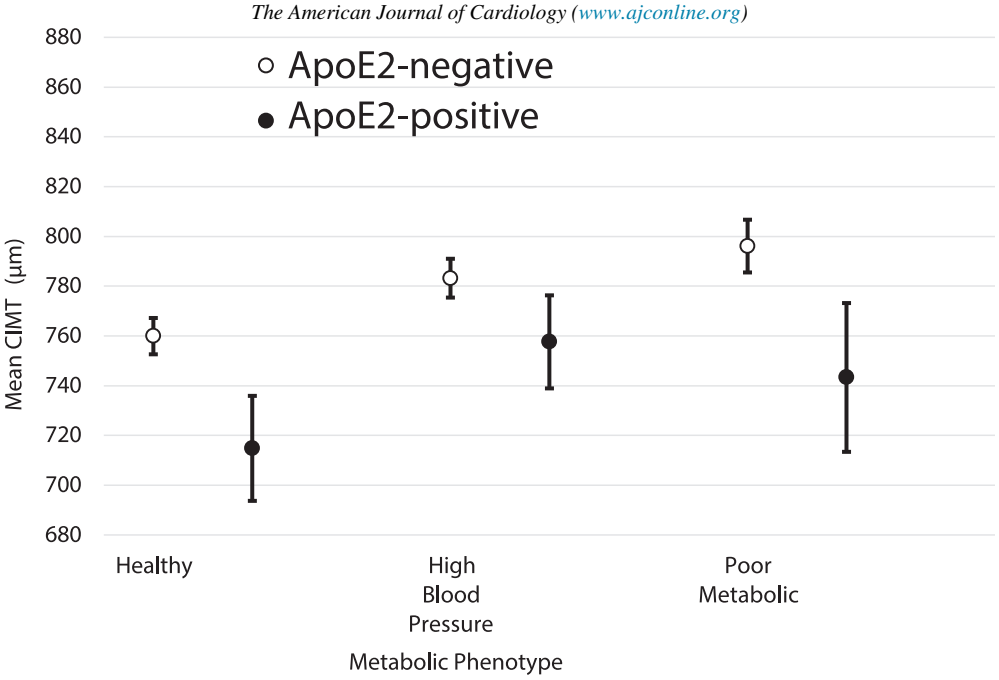

Figure 2. Estimated mean CIMT (SE) by metabolic phenotype and ApoE2 genotype (total cohort).

Table 4
Estimated mean baseline CIMT (SE) by metabolic phenotype and ApoE2 genotype

Genotype		(1) Healthy Mean (SE)	(2) High blood pressure Mean (SE)	(3) Poor metabolic Mean (SE)	ApoE2 p value	Metabolic phenotype p value	Interaction p value
Total		n = 208	n = 190	n = 99			
ApoE2-negative	n = 436	759.90 (7.30)	783.20 (7.80)	796.10 (10.60)	<0.0001	<0.0001	0.71
ApoE2-positive	n = 61	714.80 (21.10)	757.60 (18.70)	743.30 (29.90)			
ApoE4 negative women		n = 147	n = 129	n = 67			
ApoE2-negative	n = 288	762.00 (8.80)	796.00 (9.60)	775.50 (13.10)	<0.0001	<0.0001	0.92
ApoE2-positive	n = 55	717.50 (21.50)	763.60 (20.50)	743.20 (29.70)			

CIMT reported as μm; Model adjusted for age; metabolic phenotype*ApoE2 genotype*time-since-menopause interaction p value = 0.43; metabolic phenotype*ApoE2 genotype*ApoE4 genotype interaction p value = 0.62; ApoE2 genotype*ApoE4 genotype interaction p value = 0.67; CIMT = carotid artery intima media thickness.

ApoE4 genotype was defined as positive for both heterozygous and homozygous genotypes containing the E4 allele; and was otherwise defined as negative.

limited follow-up time in this analysis, it may have been difficult to detect an interaction effect on atherosclerosis progression.

The strength of this study is the randomized prospective design of ELITE, which allowed us to explore both the cross-sectional and longitudinal associations on subclinical atherosclerosis. It is important to note that the cohort in this study was healthy and therefore, the metabolic biomarker levels for the most part were within the normal range. Women in this analysis were categorized into 3 distinct metabolic phenotypes based on 9 metabolic biomarkers identified as healthy, high blood pressure, and poor metabolic using an unsupervised statistical clustering approach. Women with poor metabolic phenotype in the present study who had elevated LDL-C, triglycerides, glucose, HbA1C, HOMA score, and lower HDL-C had higher levels of subclinical atherosclerosis. Limitations of this study include generalizability, as most women in the present study were non-Hispanic white and there was limited power to adequately evaluate the longitudinal effect of metabolic phenotype and ApoE4 genotype on subclinical atherosclerosis progression.

In conclusion, our study showed that the ApoE4 genotype significantly modified the association of metabolic dysregulation on the severity, but not rate of subclinical atherosclerosis progression in postmenopausal women. The highest level of subclinical atherosclerosis was found in ApoE4+ carriers with poor metabolic phenotype. These findings have significant clinical and public health implications as preventive intervention strategies targeted to these high-risk postmenopausal women can potentially reduce the burden of CVD, the leading cause of death in women globally.

Disclosures

The authors have no conflicts of interest to disclose

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