High Apolipoprotein E Levels Predict Adverse Limb Events in Patients with Peripheral Artery Disease Due to Peripheral Artery Disease Undergoing Endovascular Treatment and On-Statin Treatment

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Summary

Little is known about the association between limb prognosis in peripheral artery disease and apolipoprotein E (apoE). We evaluated the long-term impact of apoE on adverse limb events in patients with intermittent claudication receiving statin treatment.

A total of 218 consecutive patients (mean age, 73 ± 8 years; 81% men) with intermittent claudication who underwent their first intervention between 2009 and 2020 were included in this study. All patients had achieved LDL-C < 100 mg/dL on statin treatment and were divided into two groups based on the apoE value (≥ 4.7 or < 4.7 mg/dL). We evaluated the incidence of major adverse limb events (MALEs), including vessel revascularization and limb ischemia development.

A total of 39 and 179 patients were allocated to the higher and lower apoE groups, respectively. Compared to the lower apoE group, the higher apoE group had a significantly higher total cholesterol level, triglyceride level, and non-high-density lipoprotein cholesterol level. During the median follow-up period of 3.6 years, 30 patients (13.8%) developed MALEs. Kaplan-Meier analysis revealed that the cumulative incidence of MALEs in the higher apoE group was significantly higher than that in the lower apoE group (44.0% versus 21.6%, log-rank test, $P = 0.002$). During multivariable Cox hazard analysis, higher apoE level (≥ 4.7 mg/dL) (hazard ratio, 2.61; 95% confidence interval, 1.18–5.70, $P = 0.019$) was the only strong independent predictor of MALEs.

ApoE levels could be a strong predictor and residual risk for long-term limb prognosis in patients with intermittent claudication and achieving LDL-C < 100 mg/dL with statin treatment.

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Patients with intermittent claudication due to peripheral artery disease (PAD) experience a slow disease progression; however, the frequency of revascularization, development of critical limb ischemia, and the risk of amputation increases without proper prevention and treatment. In addition, intermittent claudication as a manifestation of PAD is a strong marker for high morbidity and mortality due to generalized atherosclerosis, including cardiovascular and cerebrovascular diseases. Thus, symptoms and quality of life in patients with PAD have to improve and the risk of cardiovascular and limb events, such as supervised exercise training, smoking cessation, blood pressure management, lipid-lowering, glycemic control, antiplatelet therapy, and anticoagulants therapy such

as direct oral anticoagulants should be reduced.^{1–7} Many studies focusing on lipid-lowering therapies have reported that lowering low-density lipoprotein cholesterol (LDL-C) in patients with intermittent claudication prevents cardiovascular morbidity and mortality^{8,9} and improves claudication symptoms and exercise tolerance.¹⁰ The American College of Cardiology/American Heart Association guideline provides a class I indication for statin treatment in all patients with intermittent claudication due to PAD.¹¹ However, few studies have examined the effectiveness of lowering LDL-C in preventing ischemic limb events compared to cardiovascular events. In addition, little is known about the association between risk of adverse limb events and lipid profile (except for LDL-C); particularly, nothing

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is known about the role of apoE in the incidence of adverse limb events. In spite of the fact that apoE strongly contributes to arteriosclerosis including cardiovascular disease,¹²⁾ the related report is only between the apoE genotype and prevalence of PAD.¹³⁾ Thus, we aimed to evaluate the long-term impact of apoE on adverse limb events in patients with intermittent claudication due to PAD who underwent endovascular treatment and achieved lowering LDL-C on-statin treatment.

Methods

Study population: This single-center, observational, retrospective cohort study was conducted at our institution. We enrolled 400 consecutive patients with intermittent claudication due to PAD who underwent their first intervention between January 2009 and March 2020. In patients with an appropriate history and physical examination, we diagnosed PAD when the ankle-brachial index, which is defined as the ratio of the systolic blood pressure measured over the ankle to the systolic blood pressure measured over the brachial artery, was ≤ 0.9 .¹⁴⁾ In addition, we used duplex ultrasonography as needed to identify the location and severity of arterial obstruction, which has 85%-90% sensitivity and $> 95\%$ specificity for detecting stenosis $> 50\%$.¹⁵⁾ Advanced vascular imaging techniques, such as computed tomographic angiography and magnetic resonance angiography, were reserved for patients whose diagnoses remained uncertain following non-invasive testing or in whom intervention was anticipated.^{16,17)} The choice of treatment was based on a multidisciplinary evaluation, as mentioned above. Generally, patients with intermittent claudication were initially treated using pharmacotherapy (some antihypertensive drugs, statins, oral hypoglycemic agents, insulin, antiplatelet agents, etc.), exercise therapy (supervised exercise training, or if it is not feasible unsupervised exercise training), and smoking cessation guidance. However, if these conservative therapies were ineffective, patients were eligible for intervention. The exclusion criteria were as follows: 1) patients not taking statins at the time of the first intervention, 2) patients not achieving the lowering of LDL-C (with a goal of < 100 mg/dL),¹⁸⁾ 3) patients undergoing hemodialysis, and 4) patients with missing data on the lipid profile. Finally, 218 patients were divided into two groups (higher apoE and lower apoE) according to the preprocedural apoE levels based on the optimal cutoff value of apoE using receiver operating characteristic curve (ROC) analysis.

This study was approved by the ethics committee of our institution and was performed following the Declaration of Helsinki. All participants provided written informed consent.

Data collection and definitions: Data on patient characteristics were collected from the institutional database. Blood samples were collected early in the morning after an overnight fast. Baseline plasma samples from the patients were thawed and assayed for each plasma parameter using commercially available analytic systems. Regarding lipid profile, total cholesterol, high-density lipoprotein cholesterol, direct LDL-C, and triglyceride levels were assayed using a LABOSPECT 008 α (Hitachi, Ltd., Tokyo,

Japan) at our institution. The measurement of the following samples was outsourced to an external specialist; lipoprotein (a) was assayed using a latex-enhanced immunonephelometric assay, and apoA-I, apoB, and apoE were simultaneously measured on the same device using an immunoassay. Patients with a blood pressure $> 140/90$ mmHg or those receiving antihypertensive drugs were considered hypertensive. Diabetes mellitus was defined as either a hemoglobin A1c level $\geq 6.5\%$ or the administration of oral hypoglycemic drugs or insulin injections. Chronic kidney disease was defined as an estimated glomerular filtration rate < 60 mL/minute/1.73 m², as calculated by the Diet in Renal Disease equation modified with a Japanese coefficient using the baseline serum creatinine.¹⁹⁾

Study endpoint: The endpoint of this study was major adverse limb events (MALEs), defined as a composite of clinically driven target vessel revascularization and development of limb ischemia. Clinically driven target vessel revascularization was defined as re-intervention at the target vessel when symptoms occurred, and significant stenosis was detected by duplex ultrasonography or when the ankle-brachial index value markedly decreased. In addition, we evaluated the development of limb ischemia when we detected newly developed ischemic rest pain and tissue loss.

Clinical follow-up data were collected from the patients' medical records or by contacting the patients or their families if they had not been followed up in our institution after the intervention. Information about the circumstances and date of death was obtained from the families of patients who died at home, and details of events associated with the cause of death were supplied by staff of other hospitals or clinics in which the patient had been admitted. Investigators blinded to the study collected all the data.

Statistical analysis: Categorical data were presented as numbers and percentages and compared using the chi-squared test. Continuous variables were expressed as mean $\pm$ standard deviation or as median and interquartile range and were compared using one-way analysis of variance or the Kruskal-Wallis test. Univariate analysis of the relationship between the apoE level and variables was performed using Pearson's correlation analysis. A Kaplan-Meier analysis for the cumulative incidence of MALEs was used to compare the two groups based on the cutoff value of apoE using ROC analysis, and differences between the groups were assessed using the log-rank test. The multivariable Cox model was performed by stepwise selection with entry/stay criteria of 0.20/0.20. Regarding MALEs, apoE, total cholesterol, triglyceride, non-high-density lipoprotein cholesterol, and pre-treatment ankle-brachial index were included in the list of candidate covariates.

All probabilities were expressed as two-tailed values, with statistical significance inferred at $P < 0.05$. All confidence intervals (CIs) were computed at the 95% level. All data were analyzed using JMP version 12.0 for Macintosh (SAS Institute, Cary, NC, USA).

Figure 1. Study flow chart. Among 400 patients, we excluded the following persons: patients not taking statin, with uncontrolled LDL-C, on hemodialysis, and with an unmeasured lipid profile. We studied 218 patients with controlled LDL-C on statin. The patients were divided into two groups according to the preprocedural apoE levels based on the optimal cutoff value of apoE using ROC analysis (dichotomized to ≥ 4.7 or < 4.7 mg/dL). ApoE indicates apolipoprotein E; and LDL-C, low-density lipoprotein cholesterol.

Results

Baseline characteristics: Overall, we studied 218 patients on statins with LDL-C < 100 mg/dL. (Figure 1). Their baseline clinical characteristics are summarized in Table I. The mean age was 73 ± 8 years, and 81% of the patients were men. The prevalence of hypertension, diabetes mellitus, and smoking was 84%, 56%, and 88%, respectively. Although the prevalence of patients with chronic kidney disease was 37%, the mean estimated glomerular filtration rate of $66.8 \text{ mL/minute/1.73 m}^2$ was within normal limits. All patients took statins, but the use of any other lipid-lowering agents, including ezetimibe, fibrates, and eicosapentaenoic acid, was less than 10%.

Relationships between apoE and risk factors for MALEs: Overall, median and mean apoE levels were 3.6 mg/dL (interquartile range: 3.0, 4.3) and 3.8 mg/dL (standard deviation: 0.6), respectively. When we analyzed the ROC curves of apoE to predict future MALEs, the area under the curve of ROC for apoE levels was 0.64, with the highest discriminating sensitivity and specificity of 0.53 and 0.87, respectively, at apoE of 4.7 mg/dL (Supplemental Figure). Thirty-nine patients (18%) were allocated to the higher-apoE group (apoE level ≥ 4.7 mg/dL), whereas 179 patients (82%) were included in the lower-apoE group (apoE level < 4.7 mg/dL) (Table I). No significant differences were seen in age, sex, hypertension, diabetes mellitus, smoking, chronic kidney disease, or any oral medication between the higher and lower apoE

groups (all $P \geq 0.05$). Compared to the lower-apoE group, the higher-apoE group had significantly higher total cholesterol, triglyceride, non-high-density lipoprotein cholesterol, apoA-I, apoB, and apoE levels (all $P < 0.05$). We summarized the univariate analysis of the relationship between apoE levels and risk factors for MALEs in Table II. As mentioned above, there were significant relationships between apoE and total cholesterol levels, triglyceride levels, and non-high-density lipoprotein cholesterol levels (all $P < 0.001$). By contrast, apoE levels did not correlate with LDL-C levels.

ApoE as a predictive factor for MALEs: The median follow-up duration was 3.6 years (interquartile range, 1.2-6.2 years), and the prognostic data were fully documented during the entire follow-up period. During the follow-up, 30 patients (13.8%) developed MALEs, including clinically driven target vessel revascularization in 30 and development of limb ischemia in 4. The Kaplan-Meier analysis indicated that the cumulative incidence of MALEs in the higher-apoE group was significantly higher than that in the lower-apoE group (44.0% versus 21.6%, log-rank test, $P = 0.002$) (Figure 2). Furthermore, Table III presents results of Cox proportional hazard regression analyses for MALEs. Higher apoE level (≥ 4.7 mg/dL as the cutoff value), total cholesterol (10 mg increase), triglyceride (10 mg increase), non-high-density lipoprotein cholesterol (10 mg increase), and lower ankle-brachial index (< 0.68 as the median value) were applicable as covariates. Multivariable Cox hazard analysis revealed that a

Table 1. Patients' Baseline Clinical Characteristics

Clinical characteristics	Overall (218 patients)	Higher apoE group (39 patients)	Lower apoE group (179 patients)	P-value
Age, years	73 ± 8	72 ± 9	73 ± 7	0.279
Male sex, <i>n</i> (%)	177 (81)	27 (69)	147 (82)	0.081
BMI, kg/m ²	23.8 ± 3.1	23.9 ± 3.1	22.8 ± 3.1	0.141
SBP, mmHg	142.7 ± 23.4	143.1 ± 20.3	142.6 ± 24.0	0.901
DBP, mmHg	73.2 ± 14.2	73.4 ± 15.5	73.2 ± 13.9	0.943
TC level, mg/dL	149 ± 20	161 ± 20	146 ± 21	< 0.001
TG level, mg/dL	119 ± 54	170 ± 92	109 ± 42	< 0.001
HDL-C level, mg/dL	46 ± 13	47 ± 16	46 ± 13	0.466
Non HDL-C level, mg/dL	103 ± 18	113 ± 21	100 ± 17	< 0.001
LDL-C level, mg/dL	76 ± 14	76 ± 14	77 ± 15	0.656
Lp (a) level, mg/dL	23 (11, 42)	19 (9, 38)	24 (11, 44)	0.710
ApoA-I level, mg/dL	129 ± 26	139 ± 29	127 ± 25	0.013
ApoB level, mg/dL	73 ± 12	82 ± 14	71 ± 12	< 0.001
ApoE level, mg/dL	3.8 ± 0.6	5.3 ± 0.8	3.4 ± 0.6	< 0.001
eGFR, mL/minute/1.73 m ²	66.8 ± 19.8	69.9 ± 18.1	66.2 ± 20.1	0.289
HbA1c level, %	6.5 ± 1.0	6.7 ± 1.1	6.5 ± 0.9	0.243
Hs-CRP level, g/dL	0.1 (0.04, 0.23)	0.1 (0.04, 0.21)	0.1 (0.04, 0.24)	0.888
NT-proBNP level, pg/mL	154.2 (78.5, 526.3)	135.5 (68.0, 452.8)	155.9 (80.9, 545.0)	0.292
Pre-treatment ABI	0.68 ± 0.15	0.69 ± 0.13	0.68 ± 0.16	0.857
Post-treatment ABI	0.94 ± 0.16	0.95 ± 0.16	0.93 ± 0.16	0.559
Comorbidity				
Hypertension, <i>n</i> (%)	184 (84)	36 (92)	148 (83)	0.107
Diabetes mellitus, <i>n</i> (%)	122 (56)	26 (67)	96 (54)	0.133
Chronic kidney disease, <i>n</i> (%)	81 (37)	11 (28)	70 (39)	0.195
Smoking, <i>n</i> (%)	192 (88)	34 (87)	158 (88)	0.851
Coronary artery disease, <i>n</i> (%)	113 (52)	18 (46)	95 (53)	0.433
Cerebrovascular disease, <i>n</i> (%)	40 (17)	5 (13)	35 (20)	0.308
Congestive heart failure, <i>n</i> (%)	10 (5)	3 (8)	7 (4)	0.339
Medication				
Statin, <i>n</i> (%)	218 (100)	39 (100)	179 (100)	
Ezetimibe, <i>n</i> (%)	22 (10)	3 (8)	19 (11)	0.571
Fibrates, <i>n</i> (%)	7 (3)	2 (5)	5 (3)	0.481
EPA, <i>n</i> (%)	17 (8)	2 (5)	15 (8)	0.472
Aspirin, <i>n</i> (%)	177 (81)	33 (85)	144 (80)	0.538
P2Y12 receptor inhibitor, <i>n</i> (%)	153 (70)	24 (62)	129 (72)	0.201
Cilostazol, <i>n</i> (%)	77 (35)	12 (31)	65 (36)	0.508
Prostanoids, <i>n</i> (%)	33 (15)	7 (18)	26 (15)	0.605
Anticoagulants	23 (11)	4 (10)	19 (11)	0.947
β-blocker, <i>n</i> (%)	73 (33)	9 (23)	64 (36)	0.119
CCB, <i>n</i> (%)	120 (55)	22 (56)	98 (55)	0.850
ACE-i/ARB, <i>n</i> (%)	124 (57)	20 (51)	104 (58)	0.437
OHA, <i>n</i> (%)	86 (39)	20 (51)	66 (37)	0.098
Insulin, <i>n</i> (%)	26 (12)	7 (18)	19 (11)	0.222

Higher apoE group, patients with an apoE level ≥ 4.7 mg/dL (*n* = 39). ABI indicates ankle-brachial index; ACE-i, angiotensin-converting enzyme inhibitor; ApoA-I, apolipoprotein A-I; ApoB, apolipoprotein B; ApoE, apolipoprotein E; ARB, angiotensin receptor blocker; BMI, body mass index; CCB, calcium channel blocker; DBP, diastolic blood pressure; eGFR, estimated glomerular filtration rate; EPA, Eicosapentaenoic acid; HbA1c, Hemoglobin A1c; HDL-C, high-density lipoprotein cholesterol; Hs-CRP, high-sensitivity C-reactive protein; LDL-C, low-density lipoprotein cholesterol; Lp (a), lipoprotein (a); NT-proBNP, N-terminal pro-brain natriuretic peptide; OHA, oral hypoglycemic agent; SBP, systolic blood pressure; TC, total cholesterol; and TG, triglyceride.

high apoE level (≥ 4.7 mg/dL) (hazard ratio [HR], 2.61; 95% CI, 1.18-5.70, *P* = 0.019) was the only strong independent predictor of MALEs. Furthermore, continuous variables of the apoE levels were also significantly related to the incidence of MALEs (per log apoE 1 increase, HR, 4.01; 95% CI, 1.00-15.0, *P* = 0.049).

Discussion

The major findings of this study are as follows: (1) Higher apoE levels were significantly associated with a higher incidence of MALEs compared to lower apoE levels in patients on statin with LDL-C < 100 mg/dL; (2) in the multivariable Cox hazard analysis, a high apoE level was the only strong independent predictor of MALEs; and (3) continuous variables of the apoE levels (per log apoE

1 increase) were also significantly related to the incidence of MALEs.

Although the association between apoE alleles and

Table II. Univariate Analysis of the Relationship Between ApoE Level and Variables

Variables	<i>r</i>	<i>P</i> -value
Age, years	-0.09	0.169
BMI, kg/m ²	0.21	0.002
SBP, mmHg	0.09	0.199
DBP, mmHg	0.09	0.198
TC level, mg/dL	0.44	< 0.001
TG level, mg/dL	0.42	< 0.001
HDL-C level, mg/dL	0.10	0.132
Non HDL-C level, mg/dL	0.42	< 0.001
LDL-C level, mg/dL	0.11	0.095
Lp (a) level, mg/dL	0.04	0.570
ApoA-I level, mg/dL	0.26	< 0.001
ApoB level, mg/dL	0.43	< 0.001
eGFR, mL/minute/1.73 m ²	0.01	0.831
HbA1c level, %	0.08	0.231
Hs-CRP level, g/dL	0.01	0.861
NT-proBNP level, pg/mL	-0.04	0.597

ApoA-I indicates apolipoprotein A-I; ApoB, apolipoprotein B; ApoE, apolipoprotein E; BMI, body mass index; DBP, diastolic blood pressure; eGFR, estimated glomerular filtration rate; HbA1c, Hemoglobin A1c; HDL-C, high-density lipoprotein-cholesterol; Hs-CRP, high-sensitivity C-reactive protein; LDL-C, low-density lipoprotein-cholesterol; Lp (a), lipoprotein (a); NT-proBNP, N-terminal pro-brain natriuretic peptide; SBP, systolic blood pressure; TC, total cholesterol; and TG, triglyceride.

Alzheimer's disease has been reported in recent years,²⁰⁾ it is strongly implicated even in cardiovascular disease.¹²⁾ In addition, there is some discussion about the relationship between the apoE genotype and prevalence of PAD,¹³⁾ but the correlation between plasma apoE levels and the incidence of adverse events in patients with PAD has never been reported in real clinical practice. Therefore, this may be the first study to demonstrate clinical significance of systemic blood apoE levels in PAD patients with intermittent claudication during long-term follow-up.

ApoE was discovered as a component of very low-density lipoprotein in the 1970s.^{21,22)} ApoE is one of the major apolipoproteins present as part of several lipoprotein particle classes, including chylomicron remnants, very low-density lipoprotein, intermediate-density lipoprotein, and some high-density lipoprotein; it is a major ligand for LDL receptors and plays a role in cholesterol metabolism.²³⁾ ApoE has structural features of the three isoforms (apoE2, apoE3, and apoE4), of which apoE2 and apoE4 increase the risk for cardiovascular diseases. Some studies have demonstrated that apoE2 is associated with increased plasma triglyceride levels. This mechanism of onset has been ascribed to defective LDL receptor binding of apoE2, which impairs the clearance of triglyceride-rich lipoproteins from the circulation.^{24,25)} In addition, apoE4 increases plasma LDL-C levels and the risk of atherosclerosis.^{23,26,27)} The increase in plasma LDL-C is caused by a down-regulated LDL receptor, when the enrichment of very low-density lipoprotein with apoE4 accelerates plasma cholesterol and lipoprotein clearance from the plasma by receptor-mediated endocytosis in the liver.²⁸⁻³⁰⁾ As mentioned above, plasma apoE levels are closely re-

Figure 2. Kaplan-Meier curves for MALEs between the higher-apoE group and the lower-apoE group. MALEs composite endpoint defined as clinically driven target vessel revascularization and development of limb ischemia. ApoE indicates apolipoprotein E, and MALEs, major adverse limb events.

Table III. Multivariable Cox Hazard Analysis of Major Adverse Limb Events

Covariate	Univariate		Multivariable	
	HR (95% CI)	P-value	HR (95% CI)	P-value
Higher apoE level	3.27 (1.57–6.74)	0.002	2.61 (1.18–5.70)	0.019
TC, 10 mg increase	1.21 (1.01–1.44)	0.034	1.16 (0.95–1.40)	0.097
TG, 10 mg/dL increase	1.04 (0.99–1.10)	0.117		
Non-HDL-C, 10 mg/dL increase	1.01 (0.99–1.03)	0.181		
Lower pre-treatment ABI	1.64 (0.77–3.47)	0.195	1.93 (0.89–4.19)	0.141

Higher apoE level was defined as apoE level ≥ 4.7 mg/dL (cutoff value); lower pre-treatment ABI was defined as pre-treatment ABI level ≥ 0.68 (median value). ABI indicates the ankle-brachial index; ApoE, apolipoprotein E; HDL-C, high-density lipoprotein cholesterol; MALEs, major adverse limb events; TC, total cholesterol; and TG, triglyceride.

lated to serum triglyceride and serum LDL-C, which are more likely to have increased in the patients with PAD, thereby increasing the risk for exacerbation of ischemic symptoms in the lower limbs.^{31–33)}

Management of LDL-C levels is recommended in patients with PAD; however, consistent and clear reference values of LDL-C levels have not yet been defined in patients with intermittent claudication. The American College of Cardiology/American Heart Association guideline recommends statin treatment in all patients with intermittent claudication. However, no specific LDL-C value is shown (previously, it was recommended that the treatment goal was LDL-C < 100 mg/dL for all patients with PAD using statin).¹¹⁾ In addition, the Trans-Atlantic Inter-Society Consensus for the Management of PAD stipulated in 2007 that recommended goal LDL-C levels were < 100 mg/dL for all patients with PAD, and a more aggressive LDL-C was lowered to < 70 mg/dL for patients with atherosclerosis involving other vascular beds.¹⁸⁾ From the viewpoint of preventing cardiovascular events in patients with PAD, the treatment strategy of lowering LDL-C such as “the lower, the better” is certainly reasonable.^{34–37)} However, such strict treatment may be controversial regarding prevention of adverse limb events. Thus, for evaluating future adverse limb events and previous reports, we decided on LDL-C < 100 mg/dL as the treatment target for lowering LDL-C using statins. In this study, multivariable Cox hazard analysis revealed that apoE was the only strong independent predictor of MALEs in patients achieving LDL-C < 100 mg/dL on statin. The results of this study indicated a significant association between elevated apoE levels and residual risk for poor limb prognosis in patients with intermittent claudication undergoing statin treatment for lowering LDL-C. Furthermore, the higher-apoE group had significantly higher triglyceride levels and non-high-density lipoprotein cholesterol levels. Thus, it is suggested that triglyceride therapy represented by fibrates may lead to reductions in future adverse limb events among individuals with intermittent claudication who have lower LDL-C levels and higher apoE levels.

This study had several limitations that require consideration. First, because of the single-center, retrospective, observational nature of the study, unknown confounding factors might have affected the outcomes regardless of analytical adjustments, and the relatively small number of enrolled patients limited the statistical power of the study. Second, the current study included only Japanese patients.

Thus, it is unclear whether the results can be applied to patients with intermittent claudication in other countries.

Conclusion

Higher preprocedural apoE levels at the time of the first intervention were significantly associated with adverse limb events in patients with intermittent claudication and lower LDL-C < 100 mg/dL on statin treatment. Thus, apoE levels could be a strong predictor for long-term limb prognosis and residual risk. Furthermore, to the best of our knowledge, this is the first report demonstrating the association between apoE and adverse limb events in patients with PAD; thus, the present findings warrant further investigation.

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Disclosure

Conflicts of interest: The authors declare that there are no conflicts of interest.

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Supplemental Files

Supplemental Figure
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