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Prevalence and Determinants of Subclinical Atherosclerosis in Familial Hypercholesterolemia

Sanjay Kulkarni[1], Meenal Joshi[1]*, Rohan Patil[2], Aniket Deshmukh[2]

[1]Department of Translational Clinical Research, Savitribai Phule Pune University, Pune, India.
[2]Department of Medical Innovation and Biomedical Systems, IIT Bombay Medical Sciences Unit, Mumbai, India.

Abstract

Familial hypercholesterolemia (FH) represents a genetic lipid disorder that frequently triggers premature cardiovascular complications. Subclinical atherosclerosis is characterized by the silent accumulation of fatty plaques within arterial walls well before any overt clinical symptoms emerge. This study explored the prevalence of subclinical atherosclerosis, its arterial distribution, and which factors independently predict its occurrence in people with FH. We conducted a single-center, prospective cross-sectional investigation. The analysis included 215 patients identified as having FH from an initial group of 1145 individuals evaluated using the Dutch Lipid Clinical Network (DLCN) criteria. To detect subclinical atherosclerosis, participants underwent carotid and femoral artery ultrasound and coronary artery calcium scoring. Levels of apolipoprotein A-I, apolipoprotein B, and lipoprotein (a) were quantified through nephelometry. Among the participants, 136 were women (63%), and the average age was 54 years (interquartile range 43–62 years). Visible physical signs of FH (stigmata) appeared in 18% of the group. At the time of screening, only 32% were taking statins, and just 8 patients (4%) had successfully lowered their LDL-C to < 70 mg/dL. Overall, subclinical atherosclerosis was present in 148 individuals (69%). Site-specific involvement included 48% in the coronary arteries, 47.5% at the carotid bifurcation, and 40.5% at the femoral bifurcation. Key independent predictors were older age, male gender, higher pre-treatment LDL-C concentrations, diabetes, and a lower ApoA-I to ApoB ratio. Elevated Lp(a) at or above 30 mg/dL specifically forecasted coronary involvement, diabetes combined with a low ApoA-I/ApoB ratio predicted carotid disease, and smoking was linked to femoral atherosclerosis. Subclinical atherosclerosis occurs frequently in individuals with FH, while adherence to prescribed lipid-lowering medications remains disappointingly low. Incorporating routine screening for subclinical disease could meaningfully shape clinical decision-making by encouraging doctors to follow treatment guidelines more rigorously and helping patients stay committed to therapy.

Keywords: Familial hypercholesterolemia, Subclinical atherosclerosis, Medication adherence, Dutch Lipid Clinical Network (DLCN)

Corresponding author: Meenal Joshi
E-mail: meenal.joshi@gmail.com

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Introduction

Familial hypercholesterolemia (FH) is an inherited autosomal dominant condition known for causing cardiovascular disease at a young age. The disorder appears in heterozygous and homozygous variants. While genetic testing that uncovers disease-causing mutations often confirms the diagnosis, such mutations are found in only 60%–80% of clinically suspected cases. This gap reduces the reliability of genetic testing for many patients who lack identifiable variants. Moreover, in resource-limited developing nations such as Türkiye, the high cost

of genetic analysis makes universal testing impractical. As a result, well-validated clinical scoring tools become especially useful where genetic confirmation is not widely accessible. Among these tools, the Dutch Lipid Clinical Network (DLCN) score is the most frequently used method for establishing an FH diagnosis.

Even though FH carries a substantially heightened risk of cardiovascular problems, public and medical awareness of the condition remains poor. Consequently, a large share of patients never reach the lipid targets advised by current guidelines. Poor disease control, in turn, drives the elevated occurrence of major adverse cardiovascular events (MACEs) seen in this population. For these reasons, rapid and accurate diagnosis, early recognition of atherosclerotic changes, and swift start of lipid-lowering treatment are crucial steps that can help protect the lives of people living with FH [1-4].

Subclinical atherosclerosis refers to the early buildup of plaque deposits in major arteries—including the coronary, carotid, and iliofemoral vessels—long before patients experience related symptoms. Non-invasive imaging methods can reveal this hidden atherosclerotic load, which acts as an important warning sign of future risk. Over time and with advancing age, plaques undergo continuous changes in size, composition, and stability, and these processes differ markedly between various arterial regions. In some vessels, plaques may harden and stabilize, while in others they remain prone to rupture and sudden events. Because of this regional variation, a comprehensive evaluation encompassing multiple vascular territories is necessary to capture the true progression and overall risk posed by the disease. Evaluating multiple sites simultaneously also provides a clearer picture of the total atherosclerotic burden an individual carries [5-9].

Improving patient adherence to therapy and raising general awareness of FH are, therefore, vital goals. Although the medical literature contains a handful of reports on subclinical atherosclerosis in FH, most have examined relatively small patient groups, and none have yet been conducted in Türkiye. Accordingly, the present study aimed to assess the frequency, anatomical distribution, and independent risk factors of subclinical atherosclerosis in patients classified as having FH by the DLCN scoring system.

Materials and Methods

The study followed a single-center, prospective, and cross-sectional design. All participants were recruited from the lipid outpatient department of our institution after initial evaluation with the DLCN scoring system. Only adults aged 18 years and above who met the clinical criteria for FH according to DLCN were enrolled. Based on their scores, patients were assigned to one of two categories: those with scores above 8 were labeled as definite FH cases, while those with scores between 6 and 8 were classified as probable FH.

Participants were excluded if they were younger than 18 years, had any secondary condition capable of causing elevated cholesterol levels (for example, hypothyroidism, nephrotic syndrome, current pregnancy, Cushing's syndrome, or ongoing treatment with corticosteroids or immunosuppressive drugs), possessed a prior history of atherosclerotic cardiovascular disease (ASCVD) requiring secondary prevention, or suffered from severe organ dysfunction or cancer. In this context, ASCVD encompassed previous myocardial infarction, coronary stent placement or angioplasty, coronary artery bypass grafting (CABG), confirmed carotid artery stenosis on imaging after a stroke or transient ischemic attack, or established peripheral arterial disease.

Patient demographics and key clinical data were systematically collected. Blood tests were performed on samples obtained within the last month, following an overnight fast of at least 12 hours. For individuals already receiving cholesterol-lowering drugs, both baseline (pre-treatment) and current (post-treatment) lipid values were documented. The overall participant selection process is summarized in **Figure 1**.

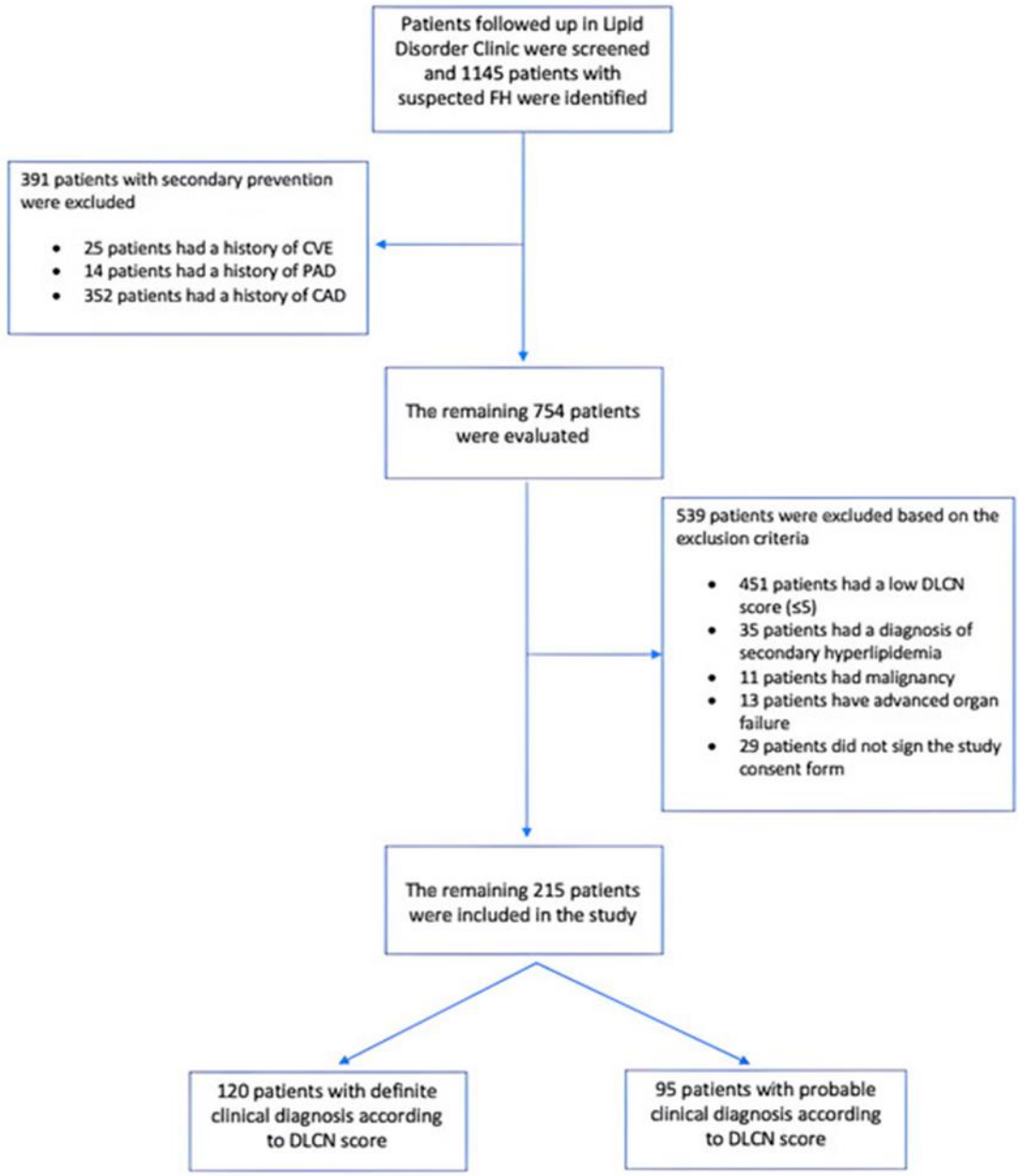

Figure 1. Flow chart of this study.

Carotid ultrasonography (USG)

Examination of the carotid arteries was performed on a Samsung RS85 Prestige ultrasound machine (Samsung Electronics Health & Medical Equipment and Samsung Medison, Seoul, Republic of Korea) equipped with a high-frequency linear transducer (LA2-14A model). Subjects lay flat on their backs with the head rotated approximately 45 degrees away from the side being scanned. Scans included the distal 10 mm of the common carotid artery just before the split, the bulb area itself, and the first 10 mm of both the internal and external carotid arteries. Every segment was thoroughly imaged from multiple angles in both transverse and longitudinal orientations.

All scans and measurements were performed separately by two qualified radiologists who had no access to the patients' medical histories. Plaque identification followed the standards set by the European Mannheim Consensus [10]. A plaque was recorded when there was a localized protrusion of at least 0.5 mm into the vessel lumen, when the wall thickening reached 50% or more of the nearby intima-media thickness (IMT), or when the IMT itself measured more than 1.5 mm. Maximum plaque thickness was calculated by identifying the greatest thickness across all detected lesions on both sides. Plaque appearance was graded according to the Gray–Weale system, which compares echogenicity with that of surrounding tissue. The four original categories were: type 1 (uniformly hypoechoic), type 2 (mostly hypoechoic), type 3 (mostly hyperechoic), and type 4 (uniformly hyperechoic). Due to the small number of purely type 2 and type 3 lesions, these were grouped as mixed plaques [11].

Femoral ultrasonography

The femoral arteries were evaluated using the identical Samsung RS85 Prestige ultrasound equipment (Samsung Electronics Health & Medical Equipment and Samsung Medison, Seoul, Republic of Korea) and the same LA2-14A linear probe. Assessment focused on a 20 mm zone around the bifurcation point where the common femoral artery divides into the superficial and profunda branches. Complete imaging was performed from both cross-sectional and longitudinal directions. Plaque detection, description, and thickness measurement in the femoral region followed the same approach and definitions as those applied during carotid scanning. The two radiologists, kept unaware of any clinical information, conducted all femoral examinations independently.

Coronary artery calcium (CAC) score

Calcium scoring of the coronary arteries was performed on a 128-slice CT scanner (Philips Ingenuity 128 Circular Edition; Philips Medical Systems, Best, Amsterdam, The Netherlands) without intravenous contrast. Technical settings included a slice thickness of 3 mm, a tube current of 80-200 mAs at 120 kV, and a rotation speed of 0.3 seconds. After a preliminary topogram image with the patient lying supine, a short-breath-hold acquisition of about 8–10 seconds covered the entire heart, beginning 1 cm below the tracheal bifurcation.

Cardiac images were interpreted jointly by two experienced radiologists who remained blinded to the patients' clinical profiles, using Philips Intellispace Portal software version 11. Quantification relied on the Agatston method. Calcified lesions were defined as areas showing density above 130 Hounsfield Units (HU), covering more than 1 mm^2, and involving at least two or three neighboring pixels. Software automatically calculated lesion area, density, and individual calcium scores on each axial slice. The final CAC score was obtained by adding the scores from the left main coronary artery (LMCA), left anterior descending artery (LAD), circumflex artery (Cx), and right coronary artery (RCA). Presence of coronary atherosclerosis was indicated by any total CAC score of 1 or higher.

Measurement of laboratory parameters

Fasting blood samples were drawn from all participants after at least 12 hours without food to quantify ApoA-I, ApoB, and lipoprotein (a) [Lp(a)] concentrations. The collected specimens were spun at 4000 rpm for 15 minutes. Following separation, the plasma and serum fractions were immediately frozen and stored at −80 °C for subsequent testing. A nephelometric assay was used to measure ApoA-I, ApoB, and Lp(a) levels. Previously frozen serum samples were thawed before processing. Once the Siemens BNProspec® analyzer (Siemens Healthineers, Erlangen, Germany) had been properly calibrated and quality controls completed, the prepared sera were loaded for analysis.

The study protocol received formal ethical clearance from the Istanbul University-Cerrahpasa Ethics Committee on 19 September 2023, under reference number E-83045809-604.01.01-789808. All procedures were performed in accordance with the ethical standards of the Declaration of Helsinki, and every participant provided written informed consent before enrollment.

Statistical analysis

Statistical processing was conducted using SPSS software version 23 (SPSS Inc., Chicago, IL, USA). Normality of continuous variables was assessed with the Kolmogorov–Smirnov and Shapiro–Wilk tests. Variables showing normal distribution were compared using the Student's t-test or one-way ANOVA and reported as mean values with their standard deviations. Non-normally distributed data were evaluated using the Mann–Whitney U test or the Kruskal–Wallis test, and results were reported as the median with interquartile range (25th–75th percentiles). For categorical variables, the chi-square test or Fisher's exact test was applied, with results shown as counts and percentages.

Associations between subclinical atherosclerosis (including coronary, carotid, and femoral components) and other study variables were investigated using multivariable logistic regression. Model adequacy was checked using the Hosmer–Lemeshow goodness-of-fit test. Correlations involving the extent of subclinical atherosclerosis and continuous parameters were examined with Pearson's or Spearman's correlation coefficients. Receiver operating characteristic (ROC) curve analysis was performed to evaluate the discriminatory power of relevant numerical variables and to establish optimal cut-off thresholds. A p-value below 0.05 was accepted as the threshold for statistical significance across all tests.

Results and Discussion

Initial screening identified 1145 individuals with possible FH. After application of the study's inclusion and exclusion criteria, 215 patients without prior ASCVD were enrolled **(Figure 1)**. Within this group, 120 were categorized as definite FH and 95 as probable FH based on DLCN scores. The cohort consisted of 136 women (63%) and 79 men (37%), with an average age of 54 years (43–62). Hypertension affected 41% of participants, while diabetes affected 13.5%. Physical signs of FH (stigmata) were observed in 38 cases (18%). Genetic testing was performed in 46 patients, identifying LDL-R mutations in 42 and ApoB mutations in 4.

Medication use at enrollment included aspirin (17%), statins (32%), and high-intensity statins (19.5%; rosuvastatin 10%, atorvastatin 9.5%). Ezetimibe was taken

by 3.5% and PCSK-9 inhibitors by 1%. A comprehensive overview of baseline clinical features and laboratory results for the entire group and the two FH subgroups is presented in **Table 1**.

Table 1. Clinical characteristics and laboratory profiles of the study population.

Variable	P-value	Probable FH (DLCN 6–8) (n = 95)	Definite FH (DLCN > 8) (n = 120)	Overall cohort (n = 215)
Age (years)	0.028	51 (41–62)	55.5 (47.3–62)	54 (43–62)
Female sex, n (%)	0.147	55 (58)	81 (67.5)	136 (63)
Body mass index (kg/m²)	0.595	27.3 (25–30.4)	27.6 (25–30.5)	27.5 (24.8–30.5)
*Obesity, n (%)**	0.954	25 (26.5)	32 (27)	57 (26.5)
Family history of CAD, n (%)	<0.001	43 (45)	103 (86)	146 (68)
Current smoking, n (%)	0.420	47 (49.5)	66 (55)	113 (53)
Physical stigmata, n (%)	<0.001	0 (0)	38 (32)	38 (18)
Hypertension, n (%)	0.138	34 (36)	55 (46)	89 (41.5)
Diabetes mellitus, n (%)	0.019	7 (7.5)	22 (18)	29 (13.5)
Pre-treatment total cholesterol (mg/dL)	<0.001	300 (275–311)	340 (316–378)	319 (290–356)
Pre-treatment LDL-C (mg/dL)	<0.001	211 (200–229)	265 (246–293)	244 (210–277)
Pre-treatment non-HDL-C (mg/dL)	<0.001	239 (220–260)	282 (260–323)	263 (234–300)
Pre-treatment HDL-C (mg/dL)	0.509	50 (46–64)	54 (45–64)	52 (45–64)
Pre-treatment triglycerides (mg/dL)	0.821	170 (129–226)	178 (130–229)	171 (129–227)
Lipoprotein(a), mg/dL	0.003	14 (6–32)	18 (11–45)	16 (9–40)
Apolipoprotein A-I (mg/dL)	0.950	163 ± 25.4	162.8 ± 28.9	162.9 ± 27.4
Apolipoprotein B (mg/dL)	0.016	120.6 ± 25.5	130.7 ± 35.9	126.2 ± 32.1
ApoA-I/ApoB ratio	0.138	1.32 (1.1–1.7)	1.23 (1–1.7)	1.28 (1.1–1.7)
Lp(a) ≥ 30 mg/dL, n (%)	0.285	26 (28)	41 (34)	67 (31)
Lp(a) ≥ 50 mg/dL, n (%)	0.054	13 (14)	29 (24)	42 (19.5)
ApoB ≥ 130 mg/dL, n (%)	0.079	40 (42)	65 (54.2)	105 (49)
Aortic valve disease, n (%)	0.304	2 (2)	7 (6)	9 (4)
Aspirin therapy, n (%)	0.021	10 (10.5)	27 (22.5)	37 (17)
Statin therapy, n (%)	0.001	19 (20)	49 (41)	68 (32)
Ezetimibe therapy, n (%)	0.018	0 (0)	7 (6)	7 (3.5)
≥50% reduction in LDL-C, n (%)	0.099	15 (16)	30 (25)	45 (21)
LDL-C < 70 mg/dL, n (%)	0.306	5 (5)	3 (2.5)	8 (4)

Abbreviations: BMI = body mass index; CAD = coronary artery disease; Lp(a) = lipoprotein (a); Apo = apolipoprotein; LDL-C = low-density lipoprotein-cholesterol; HDL-C = high-density lipoprotein-cholesterol; Stigmata = tendon xanthoma, arcus cornealis, xanthelasma; Obesity = BMI ≥ 30 kg/m². Genetic test results were available for 46 patients (42 with LDL-R gene mutations and 4 with ApoB gene mutations).

Mean pre-treatment lipid values were as follows: LDL-C 252 mg/dL, total cholesterol (TC) 329 mg/dL, triglycerides (TG) 190 mg/dL, HDL-C 55 mg/dL, and non-HDL-C 273 mg/dL. At the time of screening, only 32% of patients were receiving statin therapy. Just 45 individuals (21%) achieved a 50% or greater drop in LDL-C, and only 8 patients (4%) reached an LDL-C level under 70 mg/dL. These observations underline the persistently low rates of both statin prescription and target attainment in this elevated-risk population.

Subjects with definite FH were generally older and accounted for all cases showing physical stigmata. Rates of aspirin, statin, high-intensity statin, and ezetimibe therapy were markedly higher in the definite FH subgroup. Although medication patterns differed significantly, the success rate in reaching recommended LDL-C goals remained similar between the definite and probable FH groups. Pre-treatment lipid concentrations, along with biomarkers such as Lp(a) and ApoB, were elevated in the definite FH category. Overall, female participants displayed higher lipid and biomarker values than males.

No other laboratory measurements revealed meaningful differences between the two FH subgroups.

Subclinical atherosclerosis appeared in 148 patients, accounting for 69% of the cohort. Site-specific involvement included 48% in the coronary arteries, 47.5% at the carotid bifurcation, and 40.5% at the femoral bifurcation. Among the affected individuals, single-vessel involvement occurred in 25%, two-vessel involvement in 27%, and three-vessel involvement in 17%. In both carotid and femoral arteries, type 4 plaques were the most frequent (62% in carotid and 71% in femoral), followed by mixed plaques (20% in carotid and 16% in femoral), and type 1 plaques (18% in carotid and 13% in femoral). Over half the patients exhibited a maximum plaque thickness of 1.5 mm or more in these arterial sites (53% for carotid and 54% for femoral). Of the 188 individuals who underwent CAC scoring, 48% had a score of at least 1, and 11% had a score of 100 or higher. Group comparisons revealed clear differences in the prevalence of femoral and coronary atherosclerosis, whereas carotid atherosclerosis rates did not vary significantly **(Table 2)**.

Table 2. Distribution patterns of subclinical atherosclerosis and plaque characteristics.

Variable	P-value	Probable diagnosis (n = 95)	Definite diagnosis (n = 120)	Total cohort (n = 215)
Patients with subclinical atherosclerosis, n (%)	<0.001	53 (56)	95 (79)	148 (69)
Extent of subclinical atherosclerosis (number of arterial beds), n (%)	0.614	95	120	215
• Involvement of 1 arterial bed		21 (22)[a]	32 (27)[a]	53 (25)
• Involvement of 2 arterial beds		18 (19)[a]	40 (33)[a]	58 (27)
• Involvement of 3 arterial beds		14 (15)[a]	23 (19)[a]	37 (17)
Presence of carotid plaque, n (%)	0.095	39 (41)	63 (52.5)	102 (47.5)
Maximum carotid plaque thickness ≥ 1.5 mm, n (%)	0.581	39 → 22 (56.5)	63 → 32 (51)	102 → 54 (53)
*Carotid plaque composition**	0.359	39	63	102
• Type 1		10 (26)[a]	9 (14)[a]	19 (18)
• Type 2–3 (mixed)		7 (18)[a]	13 (21)[a]	20 (20)
• Type 4		22 (56)[a]	41 (65)[a]	63 (62)
Presence of femoral plaque, n (%)	0.008	29 (31)	58 (48)	87 (40.5)
Maximum femoral plaque thickness ≥ 1.5 mm, n (%)	0.543	29 → 17 (59)	58 → 30 (52)	87 → 47 (54)
*Femoral plaque composition**	0.060	29	58	87
• Type 1		7 (24)[a]	4 (7)[a]	11 (13)
• Type 2–3 (mixed)		3 (10)[a]	11 (19)[a]	14 (16)
• Type 4		19 (66)[a]	43 (74)[a]	62 (71)
Coronary artery calcium (CAC) score ≥ 1, n (%)	0.004	85 → 31 (36.5)	103 → 59 (57)	188 → 90 (48)
Coronary artery calcium (CAC) score ≥ 100, n (%)	0.148	85 → 6 (7)	103 → 14 (13.5)	188 → 20 (11)

Abbreviation: CAC = Coronary artery calcium. a: No significant difference was observed in the subgroup analysis. : Plaque characteristics were classified according to the Gray–Weale classification. The CAC score was calculated over 188 patients.

Patients who had subclinical atherosclerosis were typically older and displayed elevated baseline levels of LDL-C, total cholesterol, non-HDL-C, and ApoB compared with those without detectable disease. Levels of Lp(a), ApoA-I, and the ApoA-I/ApoB ratio showed no notable differences between the groups. Hypertension and diabetes occurred more often in the subclinical atherosclerosis group, but statin therapy usage remained similar across both groups.

Factors associated with the overall burden of subclinical atherosclerosis were examined in terms of the number and location of involved vascular beds. Strong associations emerged with patient age, the maximum thickness of femoral plaques, the maximum thickness of carotid plaques, and the CAC score. In comparison, pre-treatment

LDL-C, non-HDL-C, and DLCN scores showed only modest associations **(Table 3)**.

Table 3. Variables demonstrating strong correlation with the extent of subclinical atherosclerosis.

Variable	Probable diagnosis (n = 95)		Definite diagnosis (n = 120)		Overall cohort (n = 215)	
	r	P-value	r	P-value	r	P-value
Age (years)	0.404	< 0.001	0.389	< 0.001	0.431	< 0.001
Pre-treatment LDL-C (mg/dL)	0.262	0.01	0.227	0.013	0.328	< 0.001
Pre-treatment non-HDL-C (mg/dL)	0.344	0.001	0.283	0.002	0.368	< 0.001
Apolipoprotein A-I (mg/dL)	0.058	0.578	0.220	0.016	0.147	0.031
Apolipoprotein B (mg/dL)	0.103	0.322	0.146	0.111	0.146	0.033
Lipoprotein(a) (mg/dL)	−0.037	0.724	0.092	0.320	0.084	0.218
DLCN score	0.175	0.090	0.115	0.211	0.255	< 0.001
Carotid plaque thickness (mm)	0.747	< 0.001	0.653	< 0.001	0.693	< 0.001
Femoral plaque thickness (mm)	0.740	< 0.001	0.690	< 0.001	0.724	< 0.001
Coronary artery calcium (CAC) score (HU)	0.819	< 0.001	0.754	< 0.001	0.800	< 0.001

Abbreviations: Apo = apolipoprotein; Lp(a) = lipoprotein (a); CAC = coronary artery calcium; DLCN = Dutch Lipid Clinical Network; LDL-C = low-density lipoprotein-cholesterol; HDL = high-density lipoprotein-cholesterol; HU = Hounsfield Unit.

The full study population was divided into definite FH and probable FH categories, and correlation analyses were repeated independently for each. In both subgroups, age, femoral plaque thickness, carotid plaque thickness, and CAC score remained robustly associated with the extent of subclinical atherosclerosis. To further evaluate these relationships, receiver operating characteristic (ROC) analysis was performed to identify optimal cut-off points and calculate sensitivity and specificity. The resulting ROC curve is shown in **Figure 2**, and the specific cut-off thresholds, sensitivities, and specificities are detailed in **Table 4**.

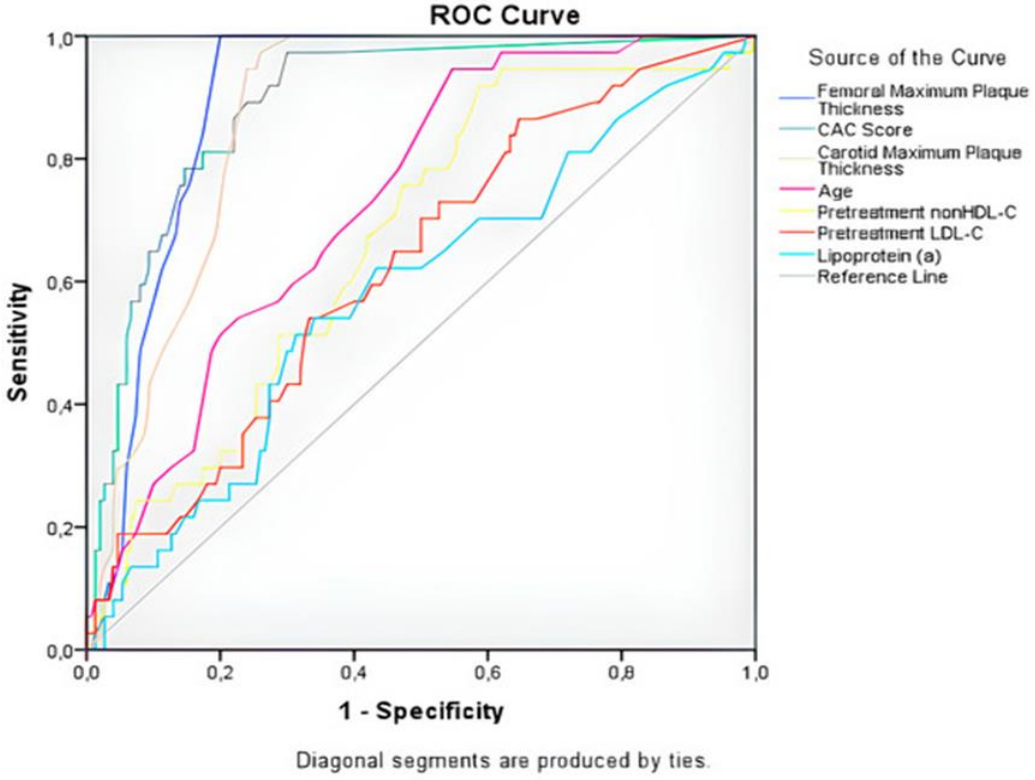

Figure 2. ROC curve analysis.

Table 4. ROC analysis of variables correlated with the extent of subclinical atherosclerosis.

Variable	Optimal cut-off value	AUC (95% CI)	P-value	Specificity	Sensitivity
Age (years)	≥ 55.5	0.732 (0.652–0.812)	< 0.001	65%	65%
Femoral plaque thickness (mm)	≥ 0.75 mm	0.899 (0.856–0.942)	< 0.001	84%	84%
Coronary artery calcium (CAC) score (HU)	≥ 19.5	0.891 (0.838–0.944)	< 0.001	83%	81%
Carotid plaque thickness (mm)	≥ 1.15 mm	0.872 (0.823–0.921)	< 0.001	79.3%	78.4%
Pre-treatment LDL-C (mg/dL)	≥ 246.5	0.625 (0.529–0.726)	0.018	58%	60%
Pre-treatment non-HDL-C (mg/dL)	≥ 263.5	0.665 (0.574–0.757)	0.020	61.3%	60%
Lipoprotein(a) (mg/dL)	≥ 17.5	0.581 (0.478–0.684)	0.127	58%	60%

Abbreviations: LDL = low-density lipoprotein-cholesterol; HDL = high-density lipoprotein-cholesterol; HU = Hounsfield Unit; Lp(a) = lipoprotein (a); CAC = coronary artery calcium.

Multivariable logistic regression analysis pinpointed several independent risk factors for subclinical atherosclerosis: greater age, male gender, elevated pre-treatment LDL-C concentration, lower ApoA-I/ApoB ratio, and coexisting diabetes. Interestingly, male gender and the ApoA-I/ApoB ratio, which lacked significance in initial univariable testing, gained independent predictive power in the final model. Conversely, hypertension and diabetes, which had been significant in univariable analysis, did not retain independent importance. When the analysis focused on individual arterial territories, older age, male gender, and higher pre-treatment LDL-C levels remained consistent predictors across sites. In addition, Lp(a) concentrations above 30 mg/dL specifically forecasted coronary involvement, diabetes combined with a reduced ApoA-I/ApoB ratio predicted carotid involvement, and current smoking predicted femoral involvement **(Table 5)**.

Our investigation of individuals with FH revealed a substantial occurrence of subclinical atherosclerosis, affecting 69% of the cohort. The distribution across vascular territories was comparable, with 48% in the coronary arteries, 48% in the carotid region, and 41% in the femoral region. Key independent factors associated with the development of subclinical atherosclerosis included older age, male gender, elevated pre-treatment LDL-C concentrations, diabetes mellitus, and a decreased ApoA-I/ApoB ratio. Older age, male gender, and higher pre-treatment LDL-C levels consistently predicted disease involvement across all examined vascular beds. Furthermore, Lp(a) concentrations of 30 mg/dL or greater specifically indicated coronary atherosclerosis, diabetes combined with a reduced ApoA-I/ApoB ratio pointed to carotid atherosclerosis, and smoking emerged as a predictor for femoral atherosclerosis. Correlation assessments demonstrated robust links between older age, maximum thicknesses of femoral and carotid plaques, CAC scores, and the overall burden of subclinical atherosclerosis.

Individuals diagnosed with FH face a markedly increased likelihood of ASCVD, as evidenced by large-scale registries and trials such as the CASCADE FH Registry and the SAFEHEART study [12, 13]. In the CASCADE FH Registry, ASCVD events were recorded in 3.6% of participants aged 20 months or older, with older age, male gender, reduced HDL-C, diabetes, and hypertension identified as notable contributors [12]. The SAFEHEART trial, which followed FH patients for a mean of 5.5 years, documented ASCVD events in 5.6% of the cohort. A risk prediction model derived from that study identified older age, male gender, prior ASCVD, elevated blood pressure, increased body mass index (BMI), smoking, higher LDL-C, and elevated Lp(a) as independent determinants of future major adverse cardiovascular event (MACE) occurrence [12]. Familial hypercholesterolemia inherently heightens ASCVD susceptibility, an effect that is further amplified by conventional cardiovascular risk factors. Current clinical guidelines emphasize the value of screening for subclinical atherosclerosis and achieving optimal LDL-C targets to mitigate ASCVD risk in this population effectively [2, 3].

Table 5. Multivariable logistic regression analysis and independent predictors of subclinical atherosclerosis.

Variable	Femoral atherosclerosis			Coronary atherosclerosis			Carotid atherosclerosis	Subclinical atherosclerosis			
	OR	95% CI	P-value	OR	95% CI	P-value	OR	OR	95% CI	P-value	95% CI	P-value

Age (years)	1.118	1.071–1.166	< 0.001	1.097	1.051–1.146	< 0.001	1.092	1.134	1.078–1.191	< 0.001	1.051–1.135	< 0.001
Male sex, n (%)	3.250	1.347–7.838	0.009	3.374	1.361–8.360	0.009	2.435	5.225	1.937–14.09	0.001	1.072–5.529	0.033
Body mass index (kg/m²)	0.959	0.883–1.042	0.325	0.986	0.908–1.070	0.730	0.939	0.925	0.848–1.009	0.078	0.867–1.016	0.118
Smoking status, n (%)	2.521	1.261–5.040	0.009	1.495	0.748–2.989	0.255	1.426	1.055	0.512–2.174	0.885	0.746–2.726	0.284
Hypertension, n (%)	1.408	0.681–2.910	0.355	1.675	0.777–3.611	0.188	1.666	1.125	0.469–2.702	0.791	0.823–3.375	0.156
Diabetes mellitus, n (%)	1.285	0.513–3.218	0.593	1.021	0.386–2.698	0.967	3.084	4.079	1.006–16.55	0.049	1.171–8.127	0.023
Pre-treatment LDL-C (mg/dL)	1.014	1.007–1.021	< 0.001	1.011	1.004–1.019	0.004	1.007	1.014	1.006–1.023	0.001	1.002–1.013	0.012
Lp(a) > 30 mg/dL, n (%)	1.149	0.568–2.324	0.698	2.649	1.253–5.599	0.011	1.494	1.308	0.603–2.837	0.497	0.760–2.934	0.244
ApoA-I/ApoB ratio	0.736	0.371–1.463	0.382	0.636	0.311–1.303	0.216	0.420	0.442	0.203–0.965	0.040	0.210–0.842	0.014

Abbreviations: BMI = Body Mass Index, LDL-C = Low-Density Lipoprotein Cholesterol, Apo = Apolipoprotein, Lp(a) = Lipoprotein (a), and Cl = Confidence Interval.

Given their inherently elevated ASCVD risk, patients with FH are anticipated to exhibit a considerable burden of subclinical atherosclerosis. In a study by Mattina *et al*. [14] involving 154 FH patients, subclinical atherosclerosis was detected in 83% of cases. The prevalence rates were 62% for coronary artery calcium, 55% for carotid plaques, and 56% for femoral plaques. No major differences were noted across the three vascular territories overall; however, coronary involvement was more frequent in males, whereas carotid involvement predominated in females. In the present cohort, subclinical atherosclerosis was identified in 69% of participants and occurred most frequently in the coronary arteries, aligning with earlier observations. Unlike the previous report, however, rates of coronary, femoral, and carotid atherosclerosis were numerically higher among females in our series. This pattern may relate to the greater representation of females within the definite FH subgroup and the older mean age of female participants (58 years) compared with males (44 years). The relatively advanced age among women, coupled with the age-associated reduction in estrogen levels that normally confer cardiovascular protection, could partially account for the observed trend toward increased subclinical atherosclerosis in female patients. Additionally, while 33% of participants in the earlier study showed involvement of all three vascular regions, the corresponding figure in our cohort was 25%. Collectively, these observations reinforce the widespread and frequent nature of subclinical atherosclerosis in FH and offer important insights into its distribution within this high-risk group.

Nevertheless, findings regarding the preferential sites of subclinical atherosclerosis have not been uniform across studies. In the Aragon Workers' Health Study (AWHS) reported by Laclaustra *et al*. [15], subclinical atherosclerosis was present in 72% of participants, with the femoral arteries representing the most commonly affected site. Active smoking proved to be the strongest determinant of both femoral and carotid plaque formation. In contrast, hypertension was most closely linked to coronary artery calcium, serving as the primary predictor of coronary disease. In our analysis, hypertension showed an initial association with subclinical and coronary atherosclerosis in univariable testing but did not retain independent significance after multivariable adjustment. Smoking emerged as an independent predictor of femoral atherosclerosis and was not associated with overall subclinical disease. Notably, femoral plaques in the AWHS demonstrated greater sensitivity for predicting coronary artery calcium compared with carotid plaques. Similarly, in our data, femoral plaque burden independently predicted coronary atherosclerosis, whereas carotid plaque burden did not, highlighting the potential prognostic importance of femoral plaque assessment for coronary risk stratification. It should be noted that coronary atherosclerosis was evaluated exclusively

through CAC scoring in both the referenced studies and the current work. Since the CAC score detects only calcified lesions and overlooks non-calcified plaques, it may underestimate the actual extent of coronary atherosclerotic burden.

Predictors of subclinical atherosclerosis have differed somewhat in other reports. Chan *et al*. [16] reported that older age, male gender, hypertension, and elevated pre-treatment LDL-C and Lp(a) levels independently predicted coronary artery disease (CAD). Allard *et al*. [17] found male sex, diabetes, a positive family history of cardiovascular disease, and raised Lp(a) levels to be associated with cardiovascular events (CVE). Consistent with these observations, our multivariable analysis confirmed older age, male gender, higher pre-treatment LDL-C, and Lp(a) levels ≥ 30 mg/dL as independent predictors of coronary atherosclerosis. These results underscore the need to integrate multiple risk factors when evaluating overall ASCVD risk in patients with FH.

Discrepant findings exist in the literature concerning the link between elevated circulating Lp(a) concentrations and CAD among individuals with FH. While certain investigations have identified high Lp(a) as an independent contributor to CAD risk in FH populations, other studies have failed to demonstrate any significant association between Lp(a) levels and CAD development [17-19].

Chan *et al*. [16] divided 390 FH patients into groups based on CAD status. The overall mean Lp(a) concentration reached 42 mg/dL, and levels were markedly elevated among those with a previous CAD diagnosis. Alonso *et al*. [20] examined how Lp(a) concentrations relate to cardiovascular disease (CVD) in both FH patients and their non-affected family members. The mean Lp(a) value was 24 mg/dL, with 30% of participants showing levels above 50 mg/dL. These elevated concentrations were strongly linked to a history of CVD among the FH patients compared with those without such a history. No meaningful difference in Lp(a) levels appeared between men and women. Among FH patients who had experienced CVD, roughly 46% displayed Lp(a) values exceeding 50 mg/dL [20]. Nenseter *et al*. [21] similarly reported no sex-based difference in Lp(a) concentrations.

In the RELACS study by Cesaro and co-workers, 774 individuals admitted with acute coronary syndrome (ACS) were categorized into three groups according to their Lp(a) levels. Higher Lp(a) concentrations correlated with an earlier onset of the first coronary event. The findings indicated that elevated Lp(a) accelerates the progression of atherosclerotic cardiovascular disease (ASCVD) and is connected to more complex coronary artery lesions. These results position Lp(a) as a key contributor to atherosclerosis development and support the use of more intensive lipid-lowering approaches even in primary prevention [22].

In the current investigation, the average Lp(a) level was 16 mg/dL. Overall, 32% of participants had Lp(a) values of 30 mg/dL or higher, and 19.5% had levels of 50 mg/dL or above. Patients with subclinical atherosclerosis tended to have higher Lp(a) concentrations, with significantly higher values in women, most likely reflecting the sex distribution within the cohort. Multivariable regression confirmed that an Lp(a) level of 30 mg/dL or higher was an independent predictor of coronary atherosclerosis. As observed in the RELACS study, individuals with raised Lp(a) require more aggressive lipid-lowering interventions [21]. Our data align with this by demonstrating a clear connection between higher Lp(a) and subclinical coronary atherosclerosis. Taken together, these observations highlight the urgent need for intensified lipid management strategies in this high-risk group to lower future cardiovascular events.

Older age represents a major consideration when evaluating ASCVD risk. Research from the SCAPIS study by Bergström *et al*. [23] indicated that coronary atherosclerosis tends to appear roughly 10 years later in women, and that the CAC score reliably reflects the presence of coronary disease. Walus-Miarka *et al*. [24] studied 154 patients classified as definite or possible FH using the Simon–Broome criteria and found that those with detectable carotid plaques were significantly older. In our cohort, individuals with subclinical atherosclerosis had a mean age approximately 10 years higher than those without. This reinforces the role of advanced age as a strong predictor of subclinical atherosclerotic burden.

Female participants in our study generally displayed higher lipid values overall. In particular, Lp(a), ApoA-I, and the ApoA-I/ApoB ratio were notably elevated in women. The protective influence of estrogen is widely recognized and helps explain the reduced cardiovascular disease rates observed in premenopausal women. Estrogen increases LDL receptor activity, which, in turn, lowers circulating total cholesterol and LDL-C concentrations [14]. The relatively higher average age among female subjects in our study, together with hormonal changes, likely contributed to the observed sex differences in lipid and biomarker profiles.

Both national and international research consistently show that, although FH patients are categorized as high-risk for ASCVD, actual medication use and achievement of target lipid levels frequently remain below recommended standards. Ray *et al*. [25] reviewed 4112 individuals on lipid-lowering treatment and noted that about 5% had FH. Target LDL-C levels were reached by only 25% of high-risk primary prevention patients and 11% of very high-risk patients. Kayikcioglu *et al*. [26] evaluated 1071 suspected FH cases and reported a treatment rate of 42%, with goal attainment as low as 2.1%. Additional research has pointed to factors such as lack of motivation, medication side effects, and treatment interruptions as contributors to poor

statin adherence [27]. Data from the U.S. National CASCADE FH Registry indicated that 92.8% of enrolled patients were on lipid-lowering therapy, yet only 73.7% were prescribed statins. The primary reasons for not using statins included intolerance or allergy (77%) and personal preference (10%). Ultimately, just 10.6% of patients achieved the desired LDL-C level below 70 mg/dL [12]. Other reports emphasize that causes of inadequate treatment are complex and involve issues at both the clinician level (such as underprescribing) and the patient level (such as non-adherence) [28]. In the present study, rates of medication use and target attainment were similarly low. However, specific reasons for non-adherence were not explored here. We anticipate that screening for subclinical atherosclerosis could lead to better medication adherence and improved goal achievement in the future. Visualizing the presence of subclinical disease may motivate both patients and physicians to strengthen treatment efforts.

Several limitations should be acknowledged in our study. First, its cross-sectional design and single-center recruitment may limit the generalizability of the results to other populations. Second, the relatively small sample size prevented meaningful subgroup analyses; larger multicenter trials will be required to adequately conduct them. Third, genetic testing results were available for only 21% of participants. The lack of comprehensive genetic data limited our ability to explore genotype–phenotype relationships and better understand individual variations in treatment response and long-term prognosis. Broader access to genetic screening in future research would greatly enhance diagnostic accuracy and risk assessment.

Additionally, the study did not include a control group of non-FH individuals for direct comparison.

Furthermore, with only 32% of participants receiving lipid-lowering medication at the time of evaluation, this low treatment rate may have influenced certain findings. The decision not to use contrast-enhanced coronary CT angiography also meant that non-calcified plaques could have been missed. Future investigations should adopt multicenter, prospective designs to overcome these constraints and yield more comprehensive and dependable conclusions.

Conclusion

The present study demonstrates that subclinical atherosclerosis is common among patients with FH. Timely identification of the condition and immediate commencement of effective lipid-lowering treatment are essential to improve long-term outcomes.

Although individuals with FH carry a substantially elevated cardiovascular risk, adherence to prescribed therapy continues to be inadequate, and few patients reach recommended lipid targets. Routine screening for subclinical atherosclerosis has the potential to enhance patient compliance with medication, encourage adoption of preventive lifestyle measures, and strengthen physicians' commitment to guideline-based treatment. Consequently, identifying subclinical atherosclerosis may play a key role in refining and optimizing management strategies for this high-risk population.

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Informed consent was obtained from all subjects involved in the study. Written informed consent has been obtained from the patient(s) to publish this paper.

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