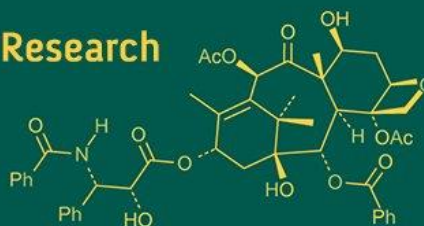


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Serum fetuin: A and PCSK9 are independently elevated in newly diagnosed, drug-naïve type 2 diabetes mellitus with atherogenic dyslipidemia: A case-control study

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Abstract

Background: Type 2 diabetes mellitus (T2DM) is frequently accompanied by atherogenic dyslipidemia from the point of diagnosis, yet the upstream hepatic drivers of this phenotype in drug-naïve patients remain poorly characterised. Fetuin-A and proprotein convertase subtilisin/kexin type 9 (PCSK9) are two hepatokines with distinct mechanistic links to insulin resistance and hepatic lipoprotein metabolism. Whether their circulating concentrations move together or independently in early atherogenic dyslipidemia has not been examined in a treatment-naïve cohort.

Methods: In a cross-sectional case-control design, 120 adults aged 30-65 years with newly diagnosed, drug-naïve T2DM were enrolled at Al-Imamain Al-Kadhmain Medical City between July and December 2025. Sixty patients with atherogenic dyslipidemia (fasting triglycerides ≥ 150 mg/dL and HDL-C < 40 mg/dL in men or < 50 mg/dL in women) were compared with 60 normolipidemic controls. None had received glucose-lowering, lipid-lowering, or antihypertensive treatment. Serum fetuin-A and PCSK9 were quantified by sandwich ELISA. HOMA-IR, a full lipid panel including apolipoprotein B and the TG/HDL ratio, ALT, GGT, and hsCRP were measured concurrently. Between-group differences were tested by independent-samples *t*-tests; ROC analysis identified diagnostic cut-offs by the Youden index.

Results: Both hepatokines were higher in the atherogenic group: fetuin-A (410 ± 88 vs 306 ± 80 $\mu\text{g/mL}$; $p < 0.0001$) and PCSK9 (372 ± 98 vs 274 ± 83 ng/mL; $p < 0.0001$). The two analytes did not correlate with each other in either subgroup ($r = 0.053-0.067$; $p > 0.6$). ROC performance was comparable: fetuin-A reached an AUC of 0.877 at a cut-off of > 353.97 $\mu\text{g/mL}$ (sensitivity 78.3%, specificity 81.7%); PCSK9 reached 0.861 at > 325.99 ng/mL (83.3%, 80.0%). Within the atherogenic subgroup, fetuin-A tracked positively with HOMA-IR ($r = 0.282$), triglycerides ($r = 0.311$), and hsCRP ($r = 0.264$). PCSK9 correlated inversely with LDL-C in the same subgroup ($r = -0.361$; $P = 0.005$), a counterintuitive direction consistent with SREBP-1c-driven rerouting of hepatic lipid flux toward VLDL export rather than LDL accumulation.

Conclusion: Fetuin-A and PCSK9 are both elevated in drug-naïve T2DM with atherogenic dyslipidemia, but their elevations are statistically independent of one another. The dissociation suggests two distinct hepatic axes, TLR4-driven inflammatory insulin resistance and SREBP-1c-driven lipogenic transcription, converging on the same atherogenic lipid phenotype. Both markers need more evaluation as complementary biomarkers of cardiometabolic risk in the newly diagnosed T2DM patients.

Keywords: Type 2 diabetes mellitus, atherogenic dyslipidemia, fetuin-A, PCSK9, hepatokines, insulin resistance, HOMA-IR; SREBP-1c

Introduction

Type 2 diabetes mellitus affects over 537 million people globally, a figure projected to approach 783 million by 2045 [1, 2]. Glycaemic derangement, however, is only part of the problem. Although not all diabetic patients show all manifestations, Data indicate that roughly 60-70% of patients with newly diagnosed T2DM show at least some lipid abnormality [3] — not a coincidental finding, but one that reflects the tight pathophysiological coupling between insulin resistance and hepatic lipid metabolism [4]. A majority of newly diagnosed patients, in other words, carry elevated atherosclerotic cardiovascular risk from the outset.

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Nevertheless, the atherogenic dyslipidemia phenotype itself rests on an uncertain diagnostic foundation. Most centers and researchers define atherogenic dyslipidemia as a triad of hypertriglyceridaemia, low HDL-C, and a presumed excess of small dense LDL particles, yet in practice the construct is almost always inferred from conventional lipid panels using fixed thresholds (TG ≥ 150 mg/dL; HDL-C $< 40/50$ mg/dL) and surrogate indices such as AIP or the TG/HDL-C ratio rather than from direct lipoprotein subfraction quantification. How reliably AIP and TG/HDL-C perform across different ethnicities, metabolic phenotypes, and early-stage T2DM has not been systematically benchmarked against NMR spectroscopy or ultracentrifugation. Patients with borderline triglyceride values or relatively preserved HDL-C may therefore harbour qualitative lipoprotein abnormalities that these indices were never designed to detect.

Timing poses a separate difficulty. Knowing that most studies in the field are cross-sectional, which leaves unresolved whether atherogenic lipid perturbations precede overt dysglycaemia or emerge as early consequences of insulin resistance. Longitudinal datasets tracking HOMA-IR trajectories alongside lipoprotein remodeling are scarce, and the causal relationship remains, to date, an open question.

What the diagnostic framework excludes matters as much as it includes. LDL-C is deliberately deemphasised in definitions of atherogenic dyslipidemia, yet neither apolipoprotein B (apoB) particle number nor remnant cholesterol has been incorporated into standard criteria. For patients whose LDL-C and apoB levels are discordant, this omission carries direct consequences for risk stratification; remnant lipoproteins, in particular, have been repeatedly implicated as mediators of residual cardiovascular risk in T2DM^[5] risk that the conventional lipid indices, by design, do not capture.

Finally, there is a translational gap between research definitions and clinical guidelines. Although the described indices are frequently used in epidemiological studies, they have not been formally integrated into major guideline algorithms, resulting in under-recognition of atherogenic dyslipidemia at the point of T2DM diagnosis. Finding the upstream regulators of this lipid profile is crucial, particularly in the early stages of the disease when therapeutic intervention has the best chance of enhancing long-term outcomes. Hepatokines, have helped explain how the metabolically challenged liver affects peripheral organs^[6, 7] Fetuin-A ($\alpha 2$ -Heremans-Schmid glycoprotein, AHSG) offers the strongest evidence linking hepatic steatosis to insulin resistance. Skeletal muscle and adipose tissue signal transduction is reduced by the protein's direct inhibition of insulin receptor tyrosine kinase autophosphorylation^[8, 9].

In 2012, Pal *et al.* found that fetuin-A acts as an adaptor molecule, presenting saturated free fatty acids to TLR4 on adipocytes and macrophages, triggering an NF- κ B-driven inflammatory cascade that reinforces insulin resistance^[10]. Prospective cohort analyses, including the EPIC-Potsdam and Cardiovascular Health Study cohorts, have consistently demonstrated that elevated circulating fetuin-A is associated with an increased risk of incident type 2 diabetes mellitus^[11, 12].

PCSK9, a 72-kDa secreted serine protease, works differently. Following the discovery of gain-of-function mutations in familial hypercholesterolemia^[13], it binds the

epidermal growth factor-like repeat A (EGF-A) domain of the LDL receptor (LDLR) on hepatocyte surfaces to redirect the receptor-PCSK9 complex from recycling endosomes to lysosomes for degradation^[14, 15]. Falling hepatic LDLR density translates directly into slower clearance of circulating LDL particles. Under hyperinsulinaemic, lipogenically active conditions, SREBP-1c drives transcription of hepatic PCSK9; the secreted convertase then binds surface LDLRs and shunts them toward lysosomal degradation, deepening the receptor deficit^[16]. In a well-characterized group of drug-naïve, newly diagnosed T2DM patients, no study has assessed serum fetuin-A and PCSK9 concurrently, stratified by atherogenic dyslipidemia phenotype. The gap is significant. Two hepatokines elevated separately and through different ways may provide complementing biological information that a single biomarker cannot capture. We compared fetuin-A and PCSK9 concentrations in T2DM patients with and without atherogenic dyslipidemia, assessed each biomarker's diagnostic performance, and examined their intercorrelation to test this idea.

Materials and Methods

Study Design and Setting

This cross-sectional case-control study recruited consecutive patients from July to December 2025 at the Endocrinology and Diabetes Clinic at Al-Imamain Al-Kadhimain Medical City, Baghdad, Iraq. Following the Declaration of Helsinki (2013 revision), all participants gave written informed consent to the study protocol submitted to the institutional review board of Al-Nahrain University/College of Medicine.

Participants and Group Stratification

Eligible participants were aged 30-65 years and had newly diagnosed T2DM (first diagnosis within the preceding 3 months, confirmed by fasting plasma glucose ≥ 126 mg/dL or HbA1c $\geq 6.5\%$ on two occasions per ADA 2024 criteria). None had received antidiabetic, lipid-lowering, or antihypertensive medications at the time of enrolment.

Stratification was based on the following operational definitions:

Case group (atherogenic dyslipidemia, $n = 60$): fasting triglycerides ≥ 150 mg/dL and HDL cholesterol < 40 mg/dL (males) or < 50 mg/dL (females).

Control group (normolipidemic, $n = 60$): fasting triglycerides < 150 mg/dL and HDL cholesterol ≥ 40 mg/dL (males) or ≥ 50 mg/dL (females).

Exclusion criteria encompassed known liver disease (viral hepatitis B/C, cirrhosis), estimated glomerular filtration rate < 60 mL/min/1.73 m² (CKD stage ≥ 3), thyroid dysfunction (TSH outside 0.4-4.0 mIU/L), concurrent use of corticosteroids or immunosuppressants, pregnancy, active infection, and known malignancy.

Sample Size

An a priori calculation guided enrolment. Using published estimates, we assumed a between-group mean difference in serum fetuin-A of 40 μ g/mL with a pooled SD of 65 μ g/mL — giving an effect size (Cohen's d) of about 0.62. At $\alpha = 0.05$ and 80% power, roughly 42 participants per arm were required. We recruited 60 per group.

Specimen Collection and Biochemical Assays

Fasting venous blood (≥ 10 h overnight fast) was drawn by antecubital venipuncture into serum separator tubes. Serum was separated by centrifugation at $1,200 \times g$ for 10 min at room temperature and stored at -80°C until analysis. No aliquot was subjected to more than a single freeze-thaw cycle.

Serum fetuin-A was assayed by a commercial sandwich ELISA (Human Fetuin-A; cat. no. RD191037100, BioVendor, Brno, Czech Republic). Microplate wells are pre-coated with a monoclonal anti-human fetuin-A antibody; after sample incubation, captured fetuin-A is detected through a biotin-labelled secondary antibody followed by streptavidin-HRP. Per the manufacturer, analytical sensitivity is 0.35 ng/mL, intra-assay CV $<5.5\%$, and inter-assay CV $<6.9\%$.

PCSK9 was measured with a separate sandwich ELISA from the same vendor (cat. no. RD191473200R; BioVendor). The capture reagent here is a polyclonal anti-human PCSK9 antibody, with detection by a biotin-conjugated polyclonal antibody and streptavidin-HRP. Reported assay performance: analytical sensitivity 0.16 ng/mL, intra-assay CV $<3.9\%$, and inter-assay CV $<7.6\%$.

Routine biochemistry

(FBG, total cholesterol, triglycerides, HDL-C, ALT, GGT, HbA1c, and hsCRP) was performed on a Cobas c311 automated analyzer (Roche Diagnostics). LDL-C was estimated by the Friedewald equation, and non-HDL-C was derived as total cholesterol minus HDL-C. Apolipoprotein B was quantified by immunoturbidimetry. Fasting insulin was measured by chemiluminescent immunoassay, and HOMA-IR was calculated as $[\text{fasting glucose (mg/dL)} \times \text{fasting insulin } (\mu\text{IU/mL})]/405$.

Anthropometric and Hemodynamic Measurements

BMI was taken as weight (kg) divided by height squared (m^2). Waist circumference was recorded at the midpoint between the lowest rib and the iliac crest with participants standing and breathing gently. Seated blood pressure was obtained with a validated oscillometric device; two readings taken five minutes apart were averaged for both systolic and diastolic values.

Statistical Analysis

Normality of continuous variables was examined with the Shapiro-Wilk test, normally distributed variables are presented as Mean $\pm$ SD. independent-samples t-tests was used for between-group comparisons. Pearson correlation coefficients were computed to examine bivariate relationships among continuous variables. ROC curves, AUCs, and 95% confidence intervals were produced. The Youden Index determined optimal cut-off values. Statistics were done using GraphPad Prism 10.4.1 and SPSS 28.0. Statistical significance was defined as a two-sided P value <0.05 .

Ethical Considerations

Al-Nahrain University/Medical College's institutional review board reviewed this study protocol. Each subject gave written informed permission. The study follows the

Declaration of Helsinki (2013 revision) and local laws.

Results

Demographic and Anthropometric Characteristics Table 1 and figure 1 shows dyslipidemia phenotype-specific demographic, anthropometric, glycaemic, lipid, hepatic, and biomarker data. Age did not differ between groups (atherogenic 50.4 ± 7.60 vs normolipidemic 48.5 ± 7.60 years; $t = 1.89$, $df = 118$, $P = 0.0611$), indicating sufficient confounding variable adjustment. The atherogenic group had higher BMI (31.5 ± 4.33 vs 29.2 ± 4.33 kg/m^2 ; $t = 4.03$, $p < 0.0001$) and waist circumference (103 ± 9.08 vs 97.8 ± 9.08 cm; $t = 4.26$, $p < 0.0001$), supporting the link between visceral adiposity and atherogenic lipids.

Systolic and diastolic blood pressures were both elevated in the atherogenic group (SBP: 135 ± 16.9 vs 128 ± 16.9 mmHg, $t = 2.99$, $P = 0.0034$; DBP: 85.9 ± 11.9 vs 81.2 ± 11.9 mmHg, $t = 3.06$, $P = 0.0027$). Elevated blood pressure and atherogenic dyslipidemia in drug-naïve individuals indicate a clustering of cardiometabolic abnormalities early in T2DM's natural course.

Table 1: Comparison of demographic, anthropometric, glycemic, lipid, hepatic, and biomarker parameters between atherogenic dyslipidemia and normolipidemic groups.

Parameter	Atherogenic (n = 60)	Normolipidemic (n = 60)	t	P
Age (years)	50.4 $\pm$ 7.60	50.9 $\pm$ 7.18	0.42	0.67
BMI (kg/m^2)	30.1 $\pm$ 4.90	27.8 $\pm$ 4.90	3.54	0.0006
Waist (cm)	106 $\pm$ 13.3	95.8 $\pm$ 13.3	5.66	<0.0001
SBP (mmHg)	135 $\pm$ 16.9	128 $\pm$ 16.9	2.99	0.0034
DBP (mmHg)	85.9 $\pm$ 11.9	81.2 $\pm$ 11.9	3.10	0.0024
FBG (mg/dL)	172 $\pm$ 36.7	164 $\pm$ 36.7	1.51	0.1337
HbA1c (%)	8.29 $\pm$ 1.56	7.89 $\pm$ 1.56	1.95	0.0530
Fasting insulin ($\mu\text{IU/mL}$)	18.9 $\pm$ 7.98	13.0 $\pm$ 7.98	5.71	<0.0001
HOMA-IR	8.31 $\pm$ 3.57	5.01 $\pm$ 3.57	7.16	<0.0001
TC (mg/dL)	221 $\pm$ 44.3	183 $\pm$ 44.3	6.72	<0.0001
TG (mg/dL)	254 $\pm$ 66.0	118 $\pm$ 66.0	15.9	<0.0001
HDL-C (mg/dL)	34.9 $\pm$ 9.14	50.0 $\pm$ 9.14	12.8	<0.0001
LDL-C (mg/dL)	130 $\pm$ 41.1	111 $\pm$ 41.1	3.48	0.0007
Non-HDL-C (mg/dL)	189 $\pm$ 47.1	134 $\pm$ 47.1	9.06	<0.0001
ApoB (mg/dL)	119 $\pm$ 31.2	90.9 $\pm$ 31.2	6.98	<0.0001
TG/HDL ratio	7.34 $\pm$ 2.41	2.29 $\pm$ 2.41	16.2	<0.0001
ALT (U/L)	43.7 $\pm$ 18.1	29.1 $\pm$ 18.1	6.27	<0.0001
GGT (U/L)	48.5 $\pm$ 23.2	33.9 $\pm$ 23.2	4.88	<0.0001
hsCRP (mg/L)	4.52 $\pm$ 2.63	2.74 $\pm$ 2.63	5.28	<0.0001
Fetuin-A ($\mu\text{g/mL}$)	410 $\pm$ 88.3	306 $\pm$ 88.3	9.16	<0.0001
PCSK9 (ng/mL)	372 $\pm$ 98.4	274 $\pm$ 98.4	7.73	<0.0001

Values are Mean $\pm$ SD. All comparisons by independent-samples t-test ($df = 118$). Abbreviations: ALT, alanine aminotransferase; ApoB, apolipoprotein B; BMI, body mass index; DBP, diastolic blood pressure; FBG, fasting blood glucose; GGT, gamma-glutamyl transferase; HDL-C, high-density lipoprotein cholesterol; HOMA-IR, Homeostasis Model Assessment of Insulin Resistance; hsCRP, high-sensitivity C-reactive protein; LDL-C, low-density lipoprotein cholesterol; SBP, systolic blood pressure; TC, total cholesterol; TG, triglycerides.

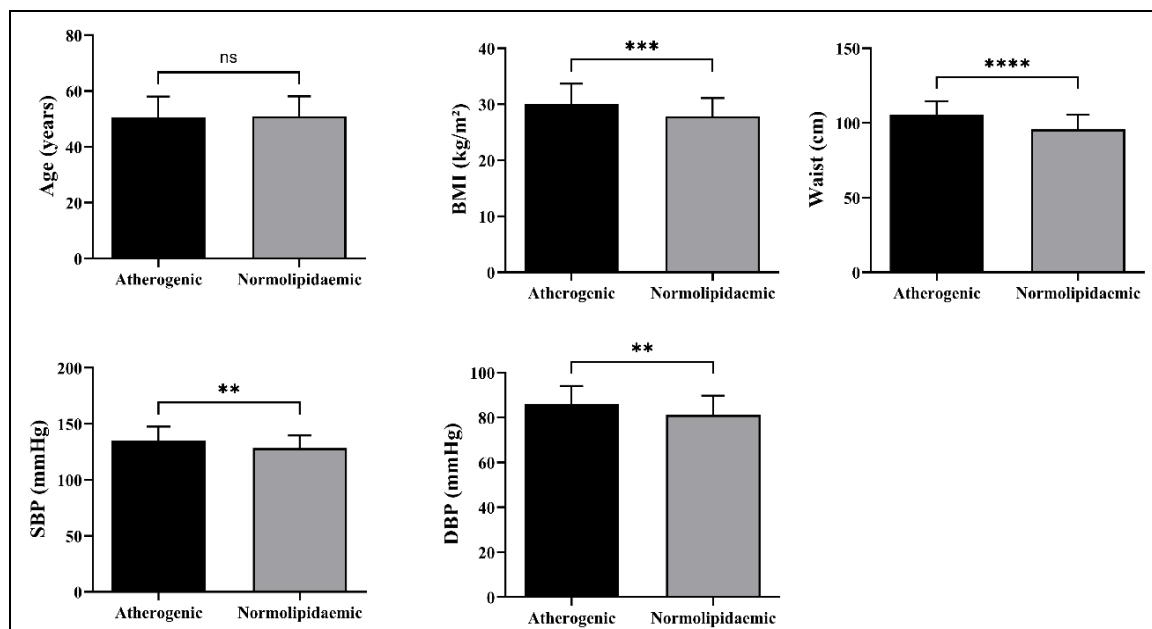


Fig 1: Comparison of demographic and clinical parameters between atherogenic dyslipidemia and normolipidaemic groups. Bar charts show Mean $\pm$ SD for age, BMI, waist circumference, systolic blood pressure, and diastolic blood pressure. Asterisks denote statistical significance (* p < 0.05, ** p < 0.01, *** p < 0.001, **** p < 0.0001; ns, not significant).

Glycemic Parameters and Insulin Resistance

No significant difference in fasting blood glucose was observed across groups (atherogenic 172 ± 36.7 vs normolipidaemic 164 ± 36.7 mg/dL; $t = 1.51$, $df = 118$, $P = 0.1337$, $R^2 = 0.019$), as predicted given their shared T2DM diagnosis (Table 1 and Figure 2). HbA1c levels were not significantly different across groups (8.39 ± 1.49 vs $7.93 \pm 1.49\%$; $t = 2.33$, $P = 0.0213$, $R^2 = 0.044$), suggesting a minor link between persistent hyperglycaemia and lipogenic

drive.

Insulin resistance indicators disagreed. The atherogenic group had a 45% higher fasting insulin (18.9 ± 7.98 vs 13.0 ± 7.98 μ IU/mL; $t = 5.71$, $df = 118$, $p < 0.0001$, $R^2 = 0.216$) and a greater effect on HOMA-IR (8.14 ± 3.75 vs 5.34 ± 3.75 ; $t = 5.72$, $p < 0.0001$, $R^2 = 0.217$). These findings suggest that insulin resistance, not glycaemic severity, causes atherogenic dyslipidemia in early T2DM.

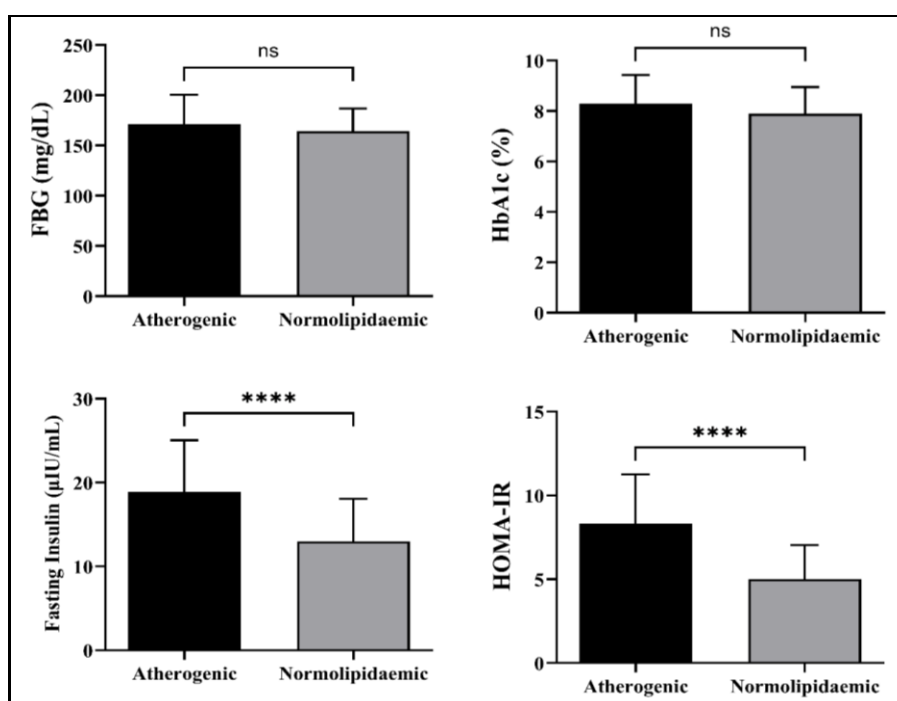


Fig 2: Glycemic parameters and insulin resistance indices in atherogenic dyslipidemia versus normolipidaemic groups. Bar charts depict fasting blood glucose, HbA1c, fasting insulin, and HOMA-IR.

Lipid Profile

Table 1 and figure 3 showed the atherogenic group had significantly higher triglycerides (254 ± 66.0 mg/dL versus 118 ± 66.0 mg/dL; $t = 15.9$, $df = 118$, $p < 0.0001$, $R^2 = 0.683$),

accounting for approximately two-thirds of the difference between groups. Lower HDL cholesterol levels were observed (35.3 ± 7.91 versus 50.8 ± 7.91 mg/dL; $t = 15.3$, $p < 0.0001$, $R^2 = 0.664$).

Total cholesterol was elevated (221 ± 44.3 vs 183 ± 44.3 mg/dL; $t = 6.72$, $p < 0.0001$), LDL cholesterol modestly increased (130 ± 41.1 vs 111 ± 41.1 mg/dL; $t = 3.48$, $P = 0.0007$), and non-HDL cholesterol showed a substantial separation (186 ± 44.5 vs 132 ± 44.5 mg/dL; $t = 9.36$, $p < 0.0001$). The TG/HDL ratio—a validated surrogate for

small dense LDL particle predominance—was approximately 4.5-fold higher in the atherogenic group (7.53 ± 2.67 vs 2.41 ± 2.67 ; $t = 14.9$, $p < 0.0001$). ApoB concentrations were correspondingly elevated (138 ± 34.4 vs 105 ± 34.4 mg/dL; $t = 7.53$, $p < 0.0001$), consistent with an increased number of atherogenic lipoprotein particles.

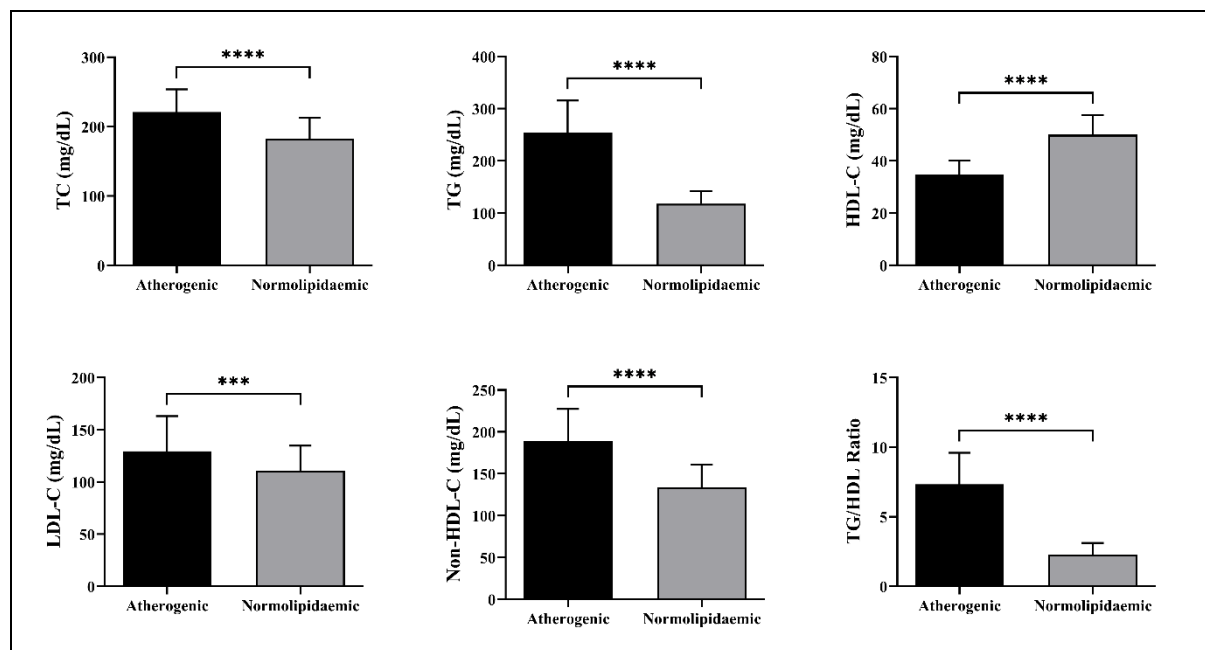


Fig 3: Bar charts of Lipid profile parameters in the study groups. Showing total cholesterol, triglycerides, HDL-C, LDL-C, non-HDL-C, and TG/HDL ratio. The TG/HDL ratio demonstrated the largest between-group effect size ($R^2 = 0.691$).

Hepatic Transaminases and Systemic Inflammation

ALT was elevated in the atherogenic group (43.7 ± 18.1 vs 29.1 ± 18.1 U/L; $t = 6.27$, $df = 118$, $p < 0.0001$, $R^2 = 0.250$), as shown in table 1 and figure 4, suggesting subclinical hepatic metabolic stress despite preserved synthetic function—a pattern that aligns with the hepatic steatosis commonly accompanying insulin-resistant T2DM. GGT followed the same direction (43.7 ± 24.2 vs 28.9 ± 24.2 U/L; $t = 4.74$,

$p < 0.0001$, $R^2 = 0.160$).

High-sensitivity CRP was roughly 60% higher in the atherogenic group (4.69 ± 2.35 vs 2.90 ± 2.35 mg/L; $t = 5.77$, $p < 0.0001$, $R^2 = 0.220$). This finding reinforces the view that atherogenic dyslipidemia in T2DM is not merely a lipid disorder but a state of low-grade systemic inflammation, one in which hepatic-derived acute-phase reactants and adipose tissue-derived cytokines likely converge.

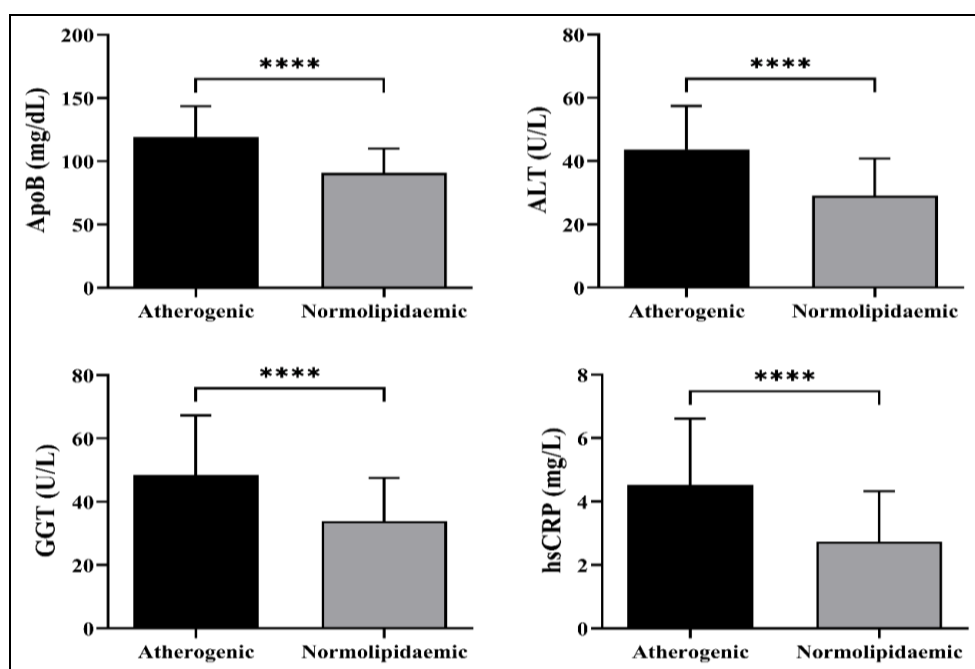


Fig 4: Bar charts for the mean concentration of ALT, GGT, ApoB, and hsCRP in atherogenic dyslipidemia versus normolipidemic groups.

Serum Fetuin-A and PCSK9

Serum fetuin-A was significantly elevated in the atherogenic group (410±88.3 vs 306±80.0 µg/mL; $t = 9.16$, $df = 118$, $p < 0.0001$, $R^2 = 0.416$), as shown in table 1 and figure 5, which comes second after the defining lipid features, the third-largest effect size in the study is the 104 µg/mL difference, or a 34% increase. This increase implies that secretions other than conventional acute-phase reactants are upregulated by hepatic metabolic stress. PCSK9 was likewise increased (372±98.4 vs 274±83.1

ng/mL; $t = 7.73$, $df = 118$, $p < 0.0001$, $R^2 = 0.336$), corresponding to a 98 ng/mL elevation (approximately 36% increase). Both biomarkers did not show any significant correlation with each other independent to atherogenic lipid status ($r = 0.067$, $P = 0.611$ in the atherogenic group; $r = 0.053$, $P = 0.688$ in the normolipidemic controls), despite their significant impact sizes ($R^2 = 0.416$ and 0.336 , respectively). Indicating different upstream mediator mechanisms for their observed differences rather than being a just statistical artifact.

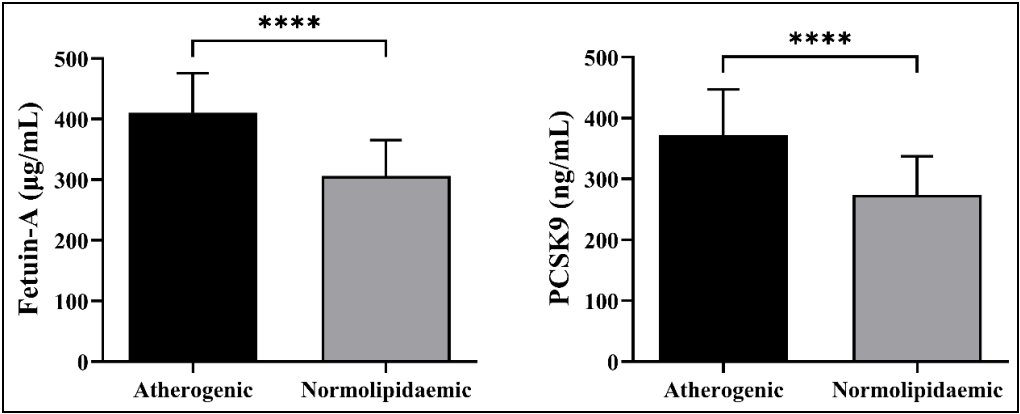


Fig 5: Bar chart of the mean concentrations of Serum fetuin-A and PCSK9 in atherogenic dyslipidemia versus normolipidemic groups.

ROC Analysis

The two hepatokines’ diagnostic performance is summarized in Table 2 and Figure 6. Fetuin-A achieved an AUC of 0.877 (95% CI 0.805-0.930; $p < 0.0001$), with a Youden-optimized cut-off of >353.97 µg/mL yielding 78.33% sensitivity and 81.67% specificity. PCSK9 attained

a comparable AUC of 0.861 (95% CI 0.786-0.917; $p < 0.0001$); its optimal cut-off of >325.99 ng/mL conferred 83.33% sensitivity and 80.00% specificity. Although each hepatokine reflects a distinct pathophysiological channel rather than a downstream lipid measurement, these results are comparable to the diagnostic accuracy of lipid ratios.

Table 2: Receiver operating characteristic analysis for fetuin-A and PCSK9 in discriminating atherogenic dyslipidemia from normolipidemic.

Biomarker	AUC (95% CI)	P	Youden cutoff	Sens (%)	Spec (%)
Fetuin-A	0.877 (0.805-0.930)	<0.0001	>353.97 µg/mL	78.33	83.33
PCSK9	0.861 (0.786-0.917)	<0.0001	>325.99 ng/mL	83.33	80.00

AUC, area under the curve; CI, confidence interval; Sens, sensitivity; Spec, specificity. Optimal cutoff identified by Youden Index.

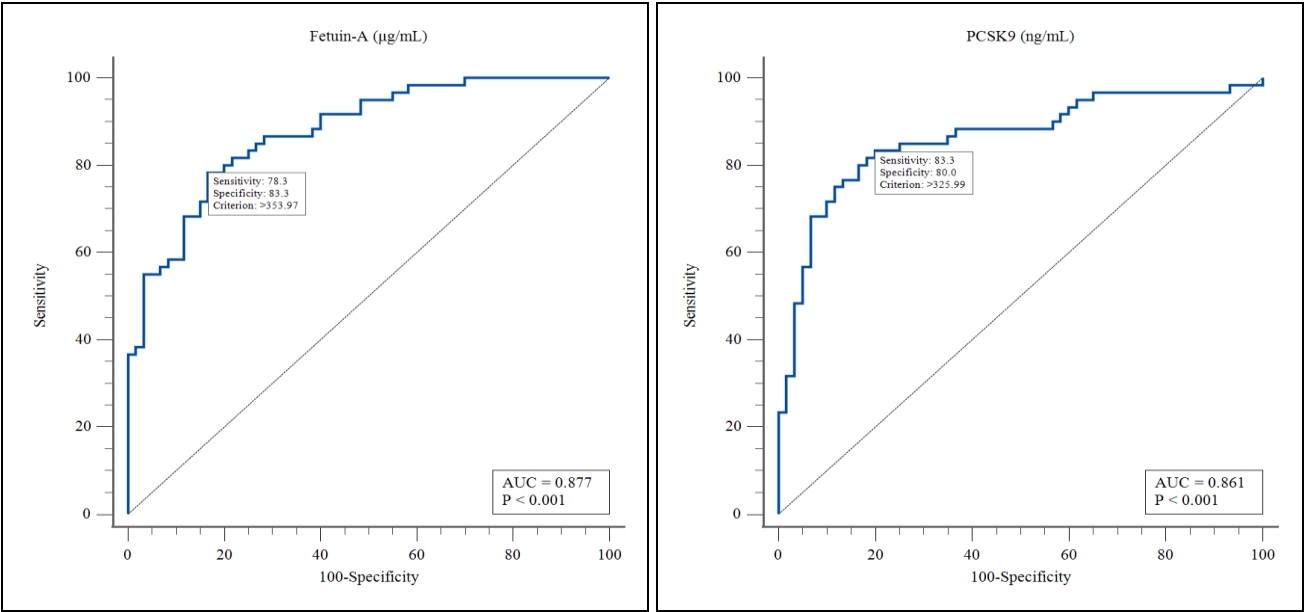


Fig 6: Receiver operating characteristic curve for serum fetuin-A (left) and PCSK9 (Right) in discriminating atherogenic dyslipidemia from normolipidemic group.

Correlation analysis

Significant Pearson coefficients are shown in Table 3. Within the atherogenic subgroup, fetuin-A correlated positively with triglycerides ($r = 0.311$, $P = 0.016$), HOMA-IR ($r = 0.282$, $P = 0.029$), and hsCRP ($r = 0.264$, $P = 0.042$). The pattern is internally coherent: fetuin-A sits at the intersection of hepatic lipid handling, peripheral insulin signalling, and subclinical inflammation, and the correlations followed the expected directions across all three axes.

PCSK9 moved opposite LDL-C in the same subgroup ($r = -0.361$, $P = 0.0045$). This negative relationship deserves closer reading. On a strict receptor model, higher circulating PCSK9 should accelerate LDL-receptor degradation and raise plasma LDL-C. In the setting of hyperinsulinemia, however, SREBP-1c activation redirects hepatic lipid flux into VLDL-triglyceride export rather than allowing LDL-C to accumulate in plasma [16, 17]. This pattern is more consistent with rerouting of lipid flux than with a defect in the receptor pathway itself. In the normolipidemic group, the correlations followed the canonical direction: PCSK9 tracked positively with non-HDL cholesterol ($r = 0.276$, $P = 0.033$) and ApoB ($r = 0.289$, $P = 0.025$), as classical LDL-clearance physiology would predict when insulin signaling is intact.

Table 3: Significant Pearson correlation coefficients within each group.

Correlation pair	r	P
Atherogenic group		
PCSK9 vs LDL-C	-0.361	0.0045
Fetuin-A vs ALT	0.289	0.025
Fetuin-A vs DBP	0.269	0.038
Normolipidemic group		
PCSK9 vs GGT	0.382	0.0026
PCSK9 vs hsCRP	-0.343	0.0073
Fetuin-A vs PCSK9 (pooled)	0.053-0.067	>0.6

Only correlations with $p < 0.05$ are shown in full; Fetuin-A-PCSK9 intercorrelation shown for reference.

Discussion

Drug-naïve, newly diagnosed T2DM patients with atherogenic dyslipidemia carried markedly higher circulating fetuin-A and PCSK9 than their normolipidemic counterparts in this case-control cohort. Two features stand out. First, the magnitude: roughly 34% for fetuin-A and 36% for PCSK9. Second, the weak intercorrelation between the two analytes, which implies that each is driven by its own upstream signal and that the two only meet downstream, at the level of the dyslipidaemic phenotype. Early atherogenic dyslipidemia in T2DM may look like one disease from the clinic, but biochemically it seems to arrive along at least two independent hepatic routes.

ROC performance was solid — AUC 0.861 for PCSK9 and 0.877 for fetuin-A. Those figures are not dramatically better than established lipid indices, but the biomarkers differ in kind: they sit upstream of the dyslipidemia rather than describing its end-products. A composite score is the obvious next step; the statistical independence of the two markers in our data at least argues that a combined index is worth testing prospectively, though a cross-sectional study cannot show it.

Fetuin-A has been studied for about twenty years as a candidate link between hepatic steatosis and systemic insulin resistance [7, 18, 19]. The mechanistic skeleton is, at

this point, fairly well mapped, though the details of how it couples to lipoprotein trafficking remain contested. Fetuin-A reduces signal transmission in skeletal muscle and adipose tissue by inhibiting the autophosphorylation of the insulin receptor and IRS-1 tyrosine kinase [20, 21]. Pal *et al.*, found that fetuin-A acts as an endogenous ligand for TLR4, exposing saturated free fatty acids to TLR4 on adipocytes and macrophages to cause an NF- κ B-dependent inflammatory cascade that hinders insulin signaling [10]. Fetuin-A concentrations follow insulin resistance indices in this study ($r = 0.282$ with HOMA-IR) and in larger population-based cohorts [22, 23, 24], which may be explained by the convergence of direct receptor inhibition and inflammatory amplification.

The increase in fetuin-A in atherogenic dyslipidemia is in line with earlier findings. In 3, 400 Chinese participants from Song *et al.*'s Shanghai Obesity Study, HOMA-IR showed a favorable correlation with circulating fetuin-A [24].

Jensen and colleagues, working within the Cardiovascular Health Study, showed that elevated fetuin-A was an independent predictor of both incident T2DM and downstream cardiovascular events over long-term follow-up [6]. A subsequent systematic review and meta-analysis by Guo and colleagues pooled the available observational data and converged on the same direction of effect, though with the usual caveats about between-study heterogeneity and assay variability [25].

The present study shows that fetuin-A remains elevated specifically in the atherogenic dyslipidemia sub phenotype, even in T2DM patients, suggesting that hepatic metabolic stress is greater in these patients.

PCSK9 uses a distinct molecular route. According to the classic paradigm, which was established by Costet *et al.* (Costet *et al.*, 2006b) and Abifadel *et al.* [16, 26], PCSK9 binds the EGF-A domain of mature LDLR on the hepatocyte surface, rerouting the receptor-ligand complex to lysosomal degradation. Hepatic LDL particle clearance is decreased by decreased hepatocyte surface LDLR density [27, 28]. PCSK9 transcription in T2DM is driven by SREBP-1c, which is triggered by hepatic lipogenic stress and hyperinsulinemia [16]. PCSK9 overproduction and VLDL-triglyceride release in the liver may result from the insulin-resistant condition of atherogenic dyslipidemia.

Extra caution is necessary due to the inverse relationship between PCSK9 and LDL-C in the atherogenic group ($r = -0.361$, $P = 0.0045$). By speeding up the breakdown of LDLR, increased PCSK9 is predicted to raise LDL-C. SREBP-1c-driven PCSK9 overproduction takes place in a metabolic milieu that promotes hepatic triglyceride synthesis and VLDL assembly over LDL-cholesterol buildup, which may account for the counterintuitive reversed direction [16, 17]. Furthermore, through insulin-sensitive mechanisms, hyperinsulinemia may increase residual LDLR recycling, partially counteracting the degradation of PCSK9.

Baass and associates [29] and Ruscica *et al.* [30] reported similarly complex PCSK9-lipid associations in metabolically heterogeneous T2DM cohorts, supporting the hypothesis that PCSK9 biology in the diabetic milieu is not linear with LDL-C.

Yang *et al.*, 2016 [31] and Ibarretxe, *et al.*, 2016 [32] reported a positive correlation between plasma PCSK9 and HbA1c in Chinese T2DM patients, whereas Levenson *et al.* [33] found higher PCSK9 in obese young women with T2DM, suggesting hyperinsulinaemia drives PCSK9 transcription.

In a related cohort, Waluś-Miarka and colleagues linked PCSK9 to both ApoB and pentraxin-3, interpreting the pattern as evidence that circulating PCSK9 reflects more than simple LDL-receptor biology — it seems to carry a signal from the hepatic inflammatory-lipogenic programme as well ^[34]. Against that background, one of the more unexpected features of our own data is the near-absence of any relationship between fetuin-A and PCSK9: correlation coefficients of 0.053-0.067 and P-values comfortably above 0.6 in both subgroups. If the two hepatokines shared a common upstream driver, one would expect at least a modest co-movement; we did not see it.

The metabolically stressed liver in T2DM releases hepatokines through distinct pathways, including fetuin-A via NF-κB and ERK-driven transcription in steatotic hepatocytes and PCSK9 via SREBP-1c-mediated lipogenic transcription. This suggests absence of a shared regulatory axis, similar to the use of multiple cardiac biomarkers in acute coronary syndromes. A combined hepatokine approach may provide a more comprehensive reflection of the underlying pathobiology than either marker alone. Several study limitations should be noted. Whether fetuin-A and PCSK9 increases preceded, accompany, or follow atherogenic dyslipidemia requires prospective follow-up. Imaging (ultrasonography or FibroScan) did not assess hepatic steatosis, so the extent to which hepatokine elevations reflect hepatic fat content versus other metabolic dysfunction is unknown. Longitudinal studies on drug-naïve T2DM patients from diagnosis to treatment initiation could determine if baseline fetuin-A and PCSK9 levels predict cardiovascular events or response to treatment. The statistical independence of the two hepatokines suggests composite scoring methods for risk categorization.

Conclusion

This case-control study demonstrates that serum fetuin-A and PCSK9 are significantly and substantially elevated in drug-naïve, newly diagnosed T2DM patients with atherogenic dyslipidemia compared with normolipidemic counterparts—elevations that are statistically independent of each other.

Fetuin-A elevation appears to reflect hepatic inflammation and TLR4-driven insulin resistance, whilst PCSK9 elevation indicates dysregulated lipogenic transcription and impaired LDLR-mediated LDL clearance. Their dissociated elevations point to distinct mechanistic contributions to the atherogenic phenotype.

These findings suggest that fetuin-A and PCSK9 may serve as complementary biological markers of atherogenic phenotype expression in early T2DM. Prospective validation and investigation of their predictive value for cardiovascular outcomes are warranted before clinical application can be recommended.

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Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability

The datasets generated during this study are available from the corresponding author upon reasonable request.

Author Contributions

Conceptualisation: M.A.M.A., H.H.A.-S.; Data curation: M.A.M.A.; Formal analysis: M.A.M.A.; Investigation: M.A.M.A., H.H.A.-S.; Methodology: M.A.M.A., H.H.A.-S.; Project administration: M.A.M.A.; Resources: H.H.A.-S.; Supervision: H.H.A.-S.; Writing-original draft: M.A.M.A.; Writing-review & editing: H.H.A.-S.

Ethics Statement

This study protocol was submitted to the institutional review board of Al-Nahrain University/Medical College. IRB Ethics approval number IRB/133/202504143. All participants provided written informed consent. The study conforms to the Declaration of Helsinki (2013 revision) and applicable local regulations.

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