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ORIGINAL ARTICLE

Serum Apolipoprotein C3: A Link Between Diabetic Nephropathy and Subclinical Atherosclerosis in Children with Diabetes Mellitus

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ABSTRACT

Background: Children with diabetic nephropathy (DN) are at significant risk for morbidity and mortality, mostly from cardiovascular disease. Apolipoprotein C3 (APOC3), a crucial regulator of plasma triglycerides, has developed lately as a potential predictor of cardiovascular events in individuals with diabetes. Therefore, the aim of this study was to use serum human APOC3 for early prediction of subclinical atherosclerosis and diabetic nephropathy in children with type 1 diabetes.

Methods: A case control study included 177 children who were divided into 3 groups, 59 each group. Group I, the health control group; group II, diabetic children without nephropathy; and group III, diabetic with nephropathy, provided clinical and biochemical data. Serum levels of human apolipoprotein C3 and ultrasound of carotid intima media thickness (CIMT) were measured in children.

Results: There is a statistically significant difference between the studied groups regarding APOC3 (the difference is significant between each two groups) being the highest level in group III with a p value <0.001. There is a statistically significant positive correlation between APOC3 and mean CIMT. The best cutoff of APOC3 in prediction of atheromatous plaque among children with diabetic nephropathy is ≥33.6 with area under curve 0.829, with 88.6% sensitivity and 66.7% specificity and a statistically high significance (p value ≤0.001).

Conclusions: APOC3 is a sensitive marker that not only reflects impaired lipid metabolism but also predicts the development of nephropathy and atherosclerosis in children with T1DM.

Keywords: Apolipoprotein C3; Diabetic Nephropathy; Children; Subclinical Atherosclerosis.

INTRODUCTION

Diabetic nephropathy (DN) is one of the most prevalent issues related to small blood vessels in diabetes mellitus. Between 25% and 40% of those with type 1 diabetes (T1D) are affected by it [1]. Modifications to the kidney's glomeruli, which result in albuminuria, hypertension, and a progressive loss of renal function are the characteristics of diabetic nephropathy. Chronic kidney disease (CKD) is a major public health concern since it is the primary cause of chronic renal illness, which increases the need for dialysis and the mortality rate [2].

One important risk factor for atherosclerotic cardiovascular disease is diabetes mellitus. In comparison to non-diabetics, those with diabetes have a doubled risk of stroke and coronary heart

disease. The risk of cardiovascular disease is higher in those with diabetic nephropathy, as cardiovascular disease and microalbuminuria may have a common pathophysiology, including endothelial dysfunction and low-grade inflammation [3].

Triglyceride-rich lipoproteins (TRLs) are largely composed of the 79-amino acid glycoprotein known as apolipoprotein C3 (APOC3). It plays a crucial part in lipoprotein metabolism by blocking lipoprotein lipase, which reduces the rate at which VLDL is converted to LDL and triglyceride hydrolysis in TRLs. Additionally, APOC3 prevents the liver from clearing TRLs through LDL receptors [4].

By examining the effects of APOC3 antisense oligonucleotides (ASOs) on lipoprotein metabolism and atherosclerosis in diabetic mice, ApoC3 ASO therapy reversed the rise in hepatic ApoC3 expression, lowering TRL levels without compromising glucose regulation [5]. APOC3 inhibition in T1DM reduces atherosclerosis by lowering (triglyceride-rich lipoproteins) TRLs, preventing foam cell formation, and halting arterial lipoprotein buildup. In a T1DM mouse model, relative insulin shortage causes elevated levels of TRLs and APOC3, which accelerate atherosclerosis [6].

APOC3 is regarded as a risk factor for DN and regulates serum TG levels significantly. The enzyme lipoprotein lipase (LPL), which helps eliminate TG from the bloodstream by breaking it down in VLDL, is inhibited by APOC3. Furthermore, through interfering with VLDL and LDL's interaction with hepatic lipoprotein receptors, APOC3 obstructs their plasma clearance. This inhibition may result in increased amounts of TG and LDL, which could accelerate the development of DN [7].

We hypothesized that measuring APOC3 levels in diabetic children could reflect endothelial function and predict future risks of cardiovascular and kidney diseases. Interest in APOC3 has grown recently due to its role as a predictor of cardiovascular events in diabetic patients. To our knowledge, no previous studies have explored the relationship between APOC3, the onset of nephropathy, and carotid intima-media thickness (CIMT) as an indicator of atherosclerosis in diabetic children. Therefore, we investigated the association between subclinical atherosclerosis, nephropathy progression, and APOC3 levels in this population.

METHODS

This case-control study was conducted at Pediatrics Endocrinology and Nephrology Units, the Children's Hospital, and the Diabetes and Endocrinology Unit in the adult internal medicine department at Zagazig University, Alahrar Hospital, and other hospitals in the Sharqia government. The Board of Institutional Review (IRB number 55/24, Jan-2024) of Zagazig University's Faculty of Medicine approved the study. Written, informed consent was given by the parents.

Class I: 59 healthy control group (age and sex match with case groups)

Class II: 59 diabetic children without nephropathy

Class III: 59 diabetic children with nephropathy

Children diagnosed with type 1 diabetes (T1DM) can be categorized into T1DM groups using the 2022 ADA criteria, including an HbA1C≥6.5% or fasting plasma glucose (FPG) of 126 mg/dl (7.0 mmol/l) or a 2-hour plasma glucose (PG) of 200 mg/dl (11.1 mmol/l). Additionally, classic symptoms of hyperglycemia or a hyperglycemic crisis, along with a random plasma glucose of ≥200 mg/dl (11.1 mmol/l), were considered in individuals aged 1 to 18 years.

Both genders (male and female), ages 1 to 18 years, had at least five years of diabetes history. Participation in the study required frequent clinic visits as well as regular insulin administration. The presence of any clinical or laboratory evidence of chronic infection, symptomatic heart disease, cardiovascular disease not related to diabetes, hypertension, obesity, familial hypercholesterolemia, history of allergy, rheumatoid arthritis, surgery, and liver dysfunction were exclusion criteria.

Blood Sampling:

Laboratory testing includes the C-reactive protein, urine albumin-to-creatinine ratio, HbA1c, CBC, fasting blood glucose, liver and kidney function tests, fasting lipid profile, and estimated glomerular filtration rate (eGFR) calculated based on Schwartz formula [8].

Urine Sampling:

Urinary albumin excretion, a marker of nephropathy, was measured using the albumin-to-creatinine ratio in an early morning urine sample. An albumin-to-creatinine ratio of 30 to 300 mg/g (corresponding to microalbuminuria) or more than 300 mg/g (corresponding to macroalbuminuria) was used to identify diabetic nephropathy. [9].

Biochemical Analysis:

By enzymatic method-based kit (Spinreact, Girona, Spain), FBG and 2-h postprandial plasma glucose levels were calculated.By cation-exchange resin-based assay kit (Stanbio Laboratory, Boerne, TX), HBA1C was calculated. Total cholesterol, high-density lipoprotein (HDL) cholesterol, and triglyceride levels were determined by colorimetric commercial kits (Spinreact). For calculation of low-density lipoprotein (LDL) cholesterol level using the Friedewald formula.

Immunochemical Assays:

Serum level of human apolipoprotein C3 was evaluated by enzyme-linked immunosorbent assay (ELISA) using the DLR-APOC3-HU Human Apolipoprotein C3 (APOC3) ELISA kit (DEVELOP), a company made in China. The ELISA process was applied according to the manufacturer's instructions.

Doppler ultrasound for carotid intima media thickness (CIMT):

A 7–12MHz linear array transducer and a LoGIQ P7 were used for all studies.Every study used anatomically standardized measurement points by first locating the proximal portion of the carotid bulb and then utilizing a variety of interrogation angles to locate the bulb's starting in the common carotid artery. We conducted IMT measurements on 177 children (59 in the healthy control group, 59 in the diabetic group without nephropathy, and 59 in the diabetic group with nephropathy) using 15 distinct interrogation angles that covered approximately 120° of the carotids far wall circumference in order to support the use of these two interrogation angles.

Statistical Analysis

Version 28 of SPSS (Statistical Package for the Social Sciences) was used to examine the data. ROC curve, one-way ANOVA, Mann Whitney test, independent sample t test, chi square test, Pearson correlation coefficient, Kruskal-Walli's test, and Fisher exact tests were among the tests that were employed.

RESULTS

There is a statistically significant difference between the studied groups regarding systolic and diastolic blood pressure and disease duration (significantly higher among the DN group), while there is a statistically non-significant difference between the studied groups regarding gender, age, weight, BMI, or height **(Table 1).**

There is a statistically significant disparity in HbA1c, triglycerides, total cholesterol, and fasting blood sugar across the studied groups, albumin/creatinine ratio, eGFR, LDL, and apolipoprotein C3 across the studied groups (the difference is significant between each two groups).

Also, there is a statistically significant dissimilarity in HDL, ALT, and AST between the studied groups (difference between patients with DN and each other group) **(Table 2).**

There is a statistically significant difference between the studied groups regarding the presence of atheromatous plaques and IMT of right and left carotid arteries and mean CIMT (difference between patients with DN and each other group) **(Table 3).**

The optimal cutoff level of apolipoprotein C3 for predicting nephropathy in diabetic children is ≥26.5 mg/dl, with an area under the curve (AUC) of 0.971. This threshold demonstrated 93.2% sensitivity and 90.7% specificity, with positive and negative predictive values of 83.3% and 96.4%, respectively, and an overall accuracy of 91.5% ($p < 0.001$, **Table 4, Figure 1).**

Among diabetic patients with microalbuminuria, there is a statistically significant relation between the presence of atheromatous plaques and apolipoprotein C3 (significantly higher among those with plaques) **(Table 5).** The best cutoff of apolipoprotein C3 in detection of atheromatous plaques among DN children is ≥33.6 mg/dl with area under curve 0.829, with 88.6% sensitivity and 66.7% specificity. Positive and negative predictive values were 79.5% and 80%, with overall accuracy of 79.7% with a P value <0.001 **(Table 6, figure 2S).** There is a statistically significant positive correlation between apolipoprotein C3 and mean CIMT (**Figure 1).**

Table (1): Comparison between the studied groups regarding demographic and clinical data:

	Group I N=59 (%)	Group II N=59 (%)	Group III N=59 (%)	χ2	p
Gender: Female Male	24 (40.7%) 35 (59.3%)	25 (42.4%) 34 (57.6%)	24 (40.7%) 35 (59.3%)	0.047	0.977
	Mean ± SD	**Mean ± SD**	**Mean ± SD**	**F**	**p**
Age (year)	13.63 ± 2.56	13.31 ± 2.77	14.09± 1.28	1.75	0.177
Height (cm)	154.95 ± 13.62	154.0 ± 14.58	159.03 ± 6.38	2.886	0.058
Weight (kg)	55.07 ± 13.06	53.39 ± 14.22	58.37 ± 5.39	2.831	0.062
BMI (kg/m2)	22.85 ± 4.9	21.91 ± 2.88	23.06 ± 1.29	1.949	0.164
SBP (mmHg)	109.05 ± 5.81	112.12 ± 8.11	130.1 ± 12.05	93.559	<0.001**
Posthoc	P_1 0.158	P_2 <0.001**	P_3 <0.001**		
DBP (mmHg)	69.14 ± 5.83	71.41 ± 5.83	82.2 ± 6.18	76.678	<0.001**
Posthoc	P_1 0.112	P_2 <0.001**	P_3 <0.001**		
	Median (IQR)	**Median (IQR)**	**Median (IQR)**	Z	p
Duration (year)		6(5.5 – 9)	9(6 – 10)	-4.657	<0.001**

χ^2Chi square test independent sample t test**p≤0.001 is statistically highly significant p1 (differences between group 1 and group 2)
P2 (differences between group 2 and group 3) & p3 (differences between group 1 and group 3)
SBP: systolic blood pressure
DBP: diastolic blood pressure

Table (2): Comparison between the studied groups regarding laboratory data:

	Group I Mean ± SD	Group II Mean ± SD	Group III Mean ± SD	F	p
HbA1c (%)	5.15 ± 0.36	9.08 ± 1.31	10.93 ± 1.72	323.197	<0.001**
Post hoc	P1 <0.001**	P2 <0.001**	P3 <0.001**		
Fasting blood sugar (mg/dl)	94.47 ± 8.46	177.03 ± 26.62	210.41 ± 40.27	248.073	<0.001**
Posthoc	P_1 <0.001**	P_2 <0.001**	P_3 <0.001**		
Cholesterol (mg/dl)	112.9 ± 27.84	180.36 ± 45.04	251.8 ± 6.5	300.135	<0.001**
Posthoc	P_1 <0.001**	P_2 <0.001**	P_3 <0.001**		
LDL (mg/dl)	117.88 ± 10.05	121.54 ± 5.56	145.32 ± 4.75	254.058	<0.001**
Posthoc	P_1 0.017*	P_2 <0.001**	P_3 <0.001**		
HDL (mg/dl)	45.63 ± 6.07	45.88 ± 6.54	43.02 ± 5.01	4.251	0.016*
Posthoc	P_1 0.97	P_2 0.025*	P_3 0.046*		
TG (mg/dl)	91.92 ± 24.25	142.59 ± 35.75	273.97 ± 33.48	523.132	<0.001**
Posthoc	P_1 <0.001**	P_2 <0.001**	P_3 <0.001**		
Apolipoprotein C3 (mg/dl)	11.81 ± 1.86	20.66 ± 5.71	32.89 ± 5.25	311.909	<0.001**
Posthoc	P_1 <0.001**	P_2 <0.001**	P_3 <0.001**		
ALT	27(20 – 33)	21(15 – 40)	30(22 – 50)	7.747¥	0.02*
Pairwise	P_1 0.852	P_2 0.012*	P_3 0.021*		
AST	23(20 – 29)	22(12 – 32)	30(23 – 43)	13.632¥	<0.001**

	Group I Mean ± SD	Group II Mean ± SD	Group III Mean ± SD	F	p
Pairwise	P_1 0.516	P_2 <0.001**	P_3 0.005*		
Creatinine (mg/dl)	0.68 ± 0.06	0.98 ± 0.12	1.2 ± 0.05	545.487	<0.001**
Posthoc	P_1 <0.001**	P_2 <0.001**	P_3 <0.001**		
eGFR (ml/kg/min)	93.13 ± 2.29	64.42 ± 4.65	55.63 ± 2.57	2029.64	<0.001**
Posthoc	P_1 <0.001**	P_2 <0.001**	P_3 <0.001**		
Albumin/creatinine ratio	17(12 – 20)	25(20.13 – 26)	44.36(40.7 – 88.24)	135.816¥	<0.001**
Pairwise	P_1 <0.001**	P_2 <0.001**	P_3 <0.001**		

t independent sample t test *p<0.05 is statistically significant **p≤0.001 is statistically highly significant ¥data is represented as median (interquartile range) and compared by Kruskal Walis test

Table (3): Comparison between the studied groups regarding results of carotid Duplex:

	Group I N=59 (%)	Group II N=59 (%)	Group III N=59 (%)	χ2	p
Atheromatous plaque	0(0%)	0 (0%)	35(59.3%)	Fisher	0.003*
	Mean ± SD	**Mean ± SD**	**Mean ± SD**	**F**	**p**
CIMT right carotid artery (cm)	0.09 ± 0.02	0.09 ± 0.01	1.78 ± 0.42	946.502	<0.001**
Posthoc	P_1 >0.999	P_2 <0.001**	P_3 <0.001**		
CIMT left carotid artery (cm)	0.09 ± 0.02	0.09 ± 0.02	1.91 ± 0.44	1000.503	<0.001**
Posthoc	P_1 >0.999	P_2 <0.001**	P_3 <0.001**		
Mean CIMT (cm)	0.09 ± 0.02	0.09 ± 0.02	1.85 ± 0.42	1004.781	<0.001**
Posthoc	P_1 >0.999	P_2 <0.001**	P_3 <0.001**		

χ^2Chi square test *p<0.05 is statistically significant **p≤0.001 is statistically highly significant
Z Mann Whitney test

Table (4): Performance of apolipoprotein C3 in prediction of nephropathy in diabetic children:

Cutoff	AUC	Sensitivity	Specificity	PPV	NPV	Accuracy	p
≥26.5	0.971	93.2%	90.7%	83.3%	96.4%	91.5%	<0.001**

PPV positive predictive value NPV negative predictive value AUC area under curve**p≤0.001 is statistically highly significant

Table (5): Relation between presence of atheromatous plaques and apolipoprotein C3 among diabetic patients with microalbuminuria:

	No plaques Mean ± SD	Plaques Mean ± SD	t	p
Apolipoprotein C3	30.35 ± 6.67	34.64 ± 3.04	-2.649	0.006*

*p<0.05 is statistically significant Z Mann Whitney test

Table (6): Performance of apolipoprotein C3 in detection of subclinical atherosclerosis in children with diabetic nephropathy:

Cutoff	AUC	Sensitivity	Specificity	PPV	NPV	Accuracy	p
≥33.6	0.829	88.6%	66.7%	79.5%	80%	79.7%	<0.001**

PPV positive predictive value NPV negative predictive value AUC area under curve**p≤0.001 is statistically highly significant

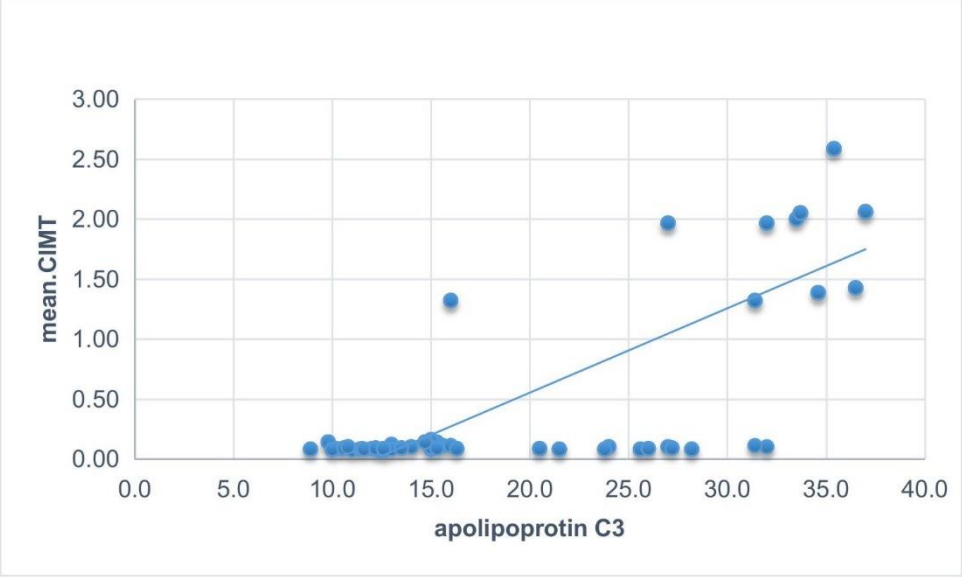

Figure (1): Scatter dot plot shows significant positive correlation between mean CIMT and apolipoprotein C3.

DISCUSSION

Diabetic glomerular lesions, a decrease in glomerular filtration rate (GFR), and excessive amounts of urine albumin excretion are the hallmarks of diabetic nephropathy (DN), sometimes referred to as diabetic kidney disease among individuals with diabetes [10].

Diabetic nephropathy (DN) affects 15-20% of individuals with type 1 diabetes (T1DM), significantly increasing morbidity and premature mortality. While advanced DN or kidney failure is rare in childhood or adolescence, diabetic kidney disease likely begins shortly after T1DM onset in susceptible individuals and may progress rapidly during adolescence, resulting in microalbuminuria or early DN. Therefore, regular monitoring of kidney function and early screening for signs of renal injury are essential for all diabetics [11].

Compared to the general population, accelerated vascular aging and an increase in preclinical indicators of vascular injury are characteristics of type 1 diabetes. This is in line with the disease's higher macrovascular morbidity and mortality rates [3].

Research priorities in the field of T1DM should focus on improving our knowledge of the risk factors for early vascular damage in T1DM, finding new potential predictors in addition to the conventional ones, and figuring out the "residual risk" as the first steps toward better prediction and prevention. Therefore, the purpose of this study was early detection of diabetic nephropathy and its correlation with the development of subclinical atherosclerosis in children with diabetes.

T1DM can manifest at any stage of life; it usually manifests between the ages of 4 and 14. However, age alone is not as significant in influencing the onset of T1DM, but there are other factors such as genetic and environmental variables [12]. Diabetic nephropathy is primarily influenced by factors directly related to diabetes management, such as blood glucose control, duration of diabetes, and the presence of hypertension, rather than demographic factors like age or gender [13].

The current study's findings showed that there was no significant variation in the height, weight, or BMI ($p = 0.058$, 0.062, and 0.164, respectively) of the investigated groups. Similarly, a previous study revealed that there was no significant difference in height, weight, or BMI between children with diabetes and healthy controls [14, 15]. In contrast, 169 individuals aged 9 to 15 took part in a case-control study. The T1DM group's median BMI was 19.2 kg/m2, which was considerably ($P < 0.05$) higher than the control group's BMI of 17.8 kg/m2 [13]. The influence of T1DM on BMI varies based on a number of parameters, such as insulin therapy, glycemic control, and disease stage. [16].

According to our study, the diabetic group with nephropathy had significantly higher systolic and diastolic blood pressure than those of the control group and the diabetic group without nephropathy. ($p<0.001$). Similarly, a study by Zabeen et al. [17] showed that diabetic children with nephropathy were associated with higher systolic blood pressure.

Chronic high blood glucose levels and associated inflammation can cause vascular changes; increasing arterial stiffness and resistance, common in diabetic children, can also contribute to hypertension by promoting sodium retention and increased blood vessel constriction [18].

On contrast, a study by Metwally et al. [15] revealed no significant change in systolic and diastolic blood pressure between diabetic children and healthy controls. The discrepancy in these findings could be the consequence of differences in the research groups' age ranges, disease durations, and T1DM severity. Different results may also arise from variations in the methods used to measure blood pressure, including variations in the time-of-day tests made, measurement protocols, and equipment calibration [19].

Triglyceride levels, LDL, and total cholesterol in our study were greater in the diabetic group without nephropathy compared to the healthy control group and more significantly greater with the DN group (p value <0.001). Similarly, previous studies discovered that, when comparing T1DM to a control, triglyceride, very low-density lipoprotein, and cholesterol levels increased statistically significantly. This is because T1DM is associated with more severe metabolic abnormalities and insulin resistance than control. This finding shows that T1DM is associated with more pronounced dyslipidemia than control [20, 21]. While other studies showed that there was no appreciable variation in LDL, HDL, triglycerides, or total cholesterol between TIDM 1 and control [15, 22].

Elevated blood glucose levels lead to an overabundance of free fatty acids in the blood, which in turn stimulates the liver's production of triglycerides and the release of very-low-density lipoproteins (VLDL), which ultimately transform into low-density lipoproteins (LDL) cholesterol. Further contributing to elevated triglyceride levels is the impairment of lipoprotein lipase's function, an enzyme essential for the breakdown of triglycerides in the blood, caused by insulin shortage or resistance [23]. Elevated triglycerides are also associated with increased production of VLDL, which can induce glomerular damage and promote albuminuria. Dyslipidemia, particularly when characterized by small, dense LDL particles, can exacerbate endothelial dysfunction and accelerate atherosclerosis, further impairing kidney function [24].

Our study demonstrated that eGFR between diabetic groups without nephropathy was lower than the healthy control group and significantly lower with the diabetic group with nephropathy (p<0.001). Likewise, the study by Laursen et al. [25] demonstrated that T1DM patients' median eGFR was lower than controls. GFR is a crucial indicator of renal function in diabetes. GFR may be normal at first, but chronic hyperglycemia can harm the kidneys over time and cause GFR to gradually diminish. GFR declines as the illness worsens, frequently accompanied by a rise in albuminuria, a marker of declining kidney function. GFR can drastically decline in later phases, which may result in end-stage renal illness or chronic kidney disease that calls for dialysis or a kidney transplant. Tracking GFR helps with diabetic nephropathy treatment and kidney function assessment [26].

According to the current study, the diabetic group without nephropathy had a higher urine albumin/creatinine ratio than the healthy control group, whereas the diabetic group with nephropathy had a significantly higher ratio (p<0.001). Furthermore, the study conducted by Laursen et al. [25] found that the median urine albumin creatinine ratio was greater in T1DM patients than in controls.

Our study's findings demonstrated that, in comparison to the control group, the diabetic group without nephropathy had considerably higher serum levels of Apolipoprotein C3 and more significantly higher in the diabetic group with nephropathy (mean ± SD 11.81 ± 1.86, 20.66 ± 5.71, 32.98 ± 5.25), respectively, with p<0.001. Similarly, the Ooi et al. [27] study found that plasma APOC3 levels were greater in those with moderate chronic renal disease. This higher level is mostly caused by decreased APOC3 breakdown (catabolism) as opposed to increased APOC3 synthesis. The main cause of the reduced breakdown of APOC3 in moderate CKD seems to be a problem with its fractional catabolism. Giammanco et al. [4] discovered that renal issues are linked to high APOC3 sialylation, which may render TRL particles (triglyceride-rich lipoproteins) that contain APOC3 less amenable to hydrolysis by lipolytic enzymes. Since the kidneys are also involved in the blood's removal of APOC3, moderate CKD may partially account for decreased APOC3 catabolism. [27]

Our study revealed that 59.3% of children with DN had atheromatous plaques with a P value of 0.003. CIMT in the right and left carotid arteries, and mean CIMT among the diabetic group with nephropathy was greater than the control group and diabetic group without nephropathy (p<0.001). This, in

agreement with previous studies, shows that T1DM patients had a noticeably higher CIMT level than controls [28, 29, 30]. Diabetes patients with persistently elevated blood glucose levels have endothelial dysfunction, elevated oxidative stress, and inflammatory responses, all of which hasten the development of atherosclerotic plaques. Furthermore, dyslipidemia—a condition marked by increased levels of LDL, cholesterol, and triglycerides—is frequently linked to type 1 diabetes (T1DM), which accelerates the development of atherosclerosis. These modifications thicken the artery walls, as indicated by increased CIMT readings, and raise the risk of early cardiovascular disease in children with type 1 diabetes [4].

Our study found that among diabetic patients with microalbuminuria, there was a statistically significant relation between the presence of atheromatous plaques and apolipoprotein C3 (significantly higher among those with plaques). In a previous study, APOC3 was identified as a stronger predictor of cardiovascular events than plasma triglycerides in individuals with type 1 diabetes. Their study reinforced that baseline APOC3 levels are predictive of cardiovascular events, but this association diminishes when accounting for remnant cholesterol, raising questions about whether remnant cholesterol might mediate the link between APOC3 and atherosclerosis [31].

In our study, the best cutoff value of apolipoprotein C3 in detection of DN in diabetic children is ≥26.5 mg/dl with an area under curve of 0.971, with 93.2% sensitivity and 90.7% specificity. Overall accuracy for the positive and negative predictive values was 96.4% and 83.3%, respectively. A previous study by Cervantes et al. [32] investigated the potential impact of APOC3 on renal damage in people with type 2 diabetes. The Veterans Affairs Diabetes Trial plasma APOC3 levels were studied. They discovered that a larger loss in renal function was linked to higher baseline APOC3 levels.

Our study found that the best cutoff of apolipoprotein C3 in detection of atheromatous plaques among DN children is ≥33.6 mg/dl with area under curve 0.829, with 88.6% sensitivity and 66.7% specificity. With an overall accuracy of 79.7%, the positive and negative predictive values were 79.5% and 80%. This implies that reducing plasma APOC3 might be an approach to target both atherosclerotic cardiovascular disease and diabetic kidney disease.

APOC3 is a predictor of cardiovascular events and mortality in individuals with type 1 diabetes with albuminuria [33]. indicates that both children with type 1 diabetes and the general population may have residual cardiovascular risk due to APOC3. The study of Sigfrids concluded that there was evidence of a relationship between APOC3 and macrovascular outcomes, with diabetic kidney disease playing a significant role in this interaction [33]. According to this, reducing plasma APOC3 may be helpful in the treatment of diabetic renal disease and atherosclerotic cardiovascular illness.

The current study revealed that there was a statistically significant positive correlation between apolipoprotein C3 and mean CIMT. Higher CIMT is linked to T1DM, which supports the idea that conditions like diabetes may interfere with its normal expression and secretion. Recent research, such as that conducted by Cervantes et al. [32], has demonstrated that silencing APOC3 may lessen the atherosclerosis linked to diabetes and that higher baseline APOC3 levels are linked to bigger reductions in renal function. Moreover, the Finnish Diabetic Nephropathy Study provides evidence that APOC3 levels, regardless of sex, the duration of diabetes, and cholesterol levels for an elevated cardiovascular burden in this population, independently predict the course of diabetic kidney disease.

The relatively small sample size of our study was one of our study limitations. However, our findings were obvious and suggestive, leading us to believe that APOC3 may have a potential therapeutic usefulness among children with T1DM and nephropathy. Therefore, research into large samples of children may be able to pinpoint biomarkers more specifically associated with atherosclerosis, which would assist in identifying children with diabetes who are particularly vulnerable to early CVD manifestation and in need of intervention programs.

Conclusion: For children with type 1 diabetes, APOC3 may be a sensitive biomarker for the detection of subclinical atherosclerosis and progression of diabetic kidney disease.

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Conflict of interest: None.

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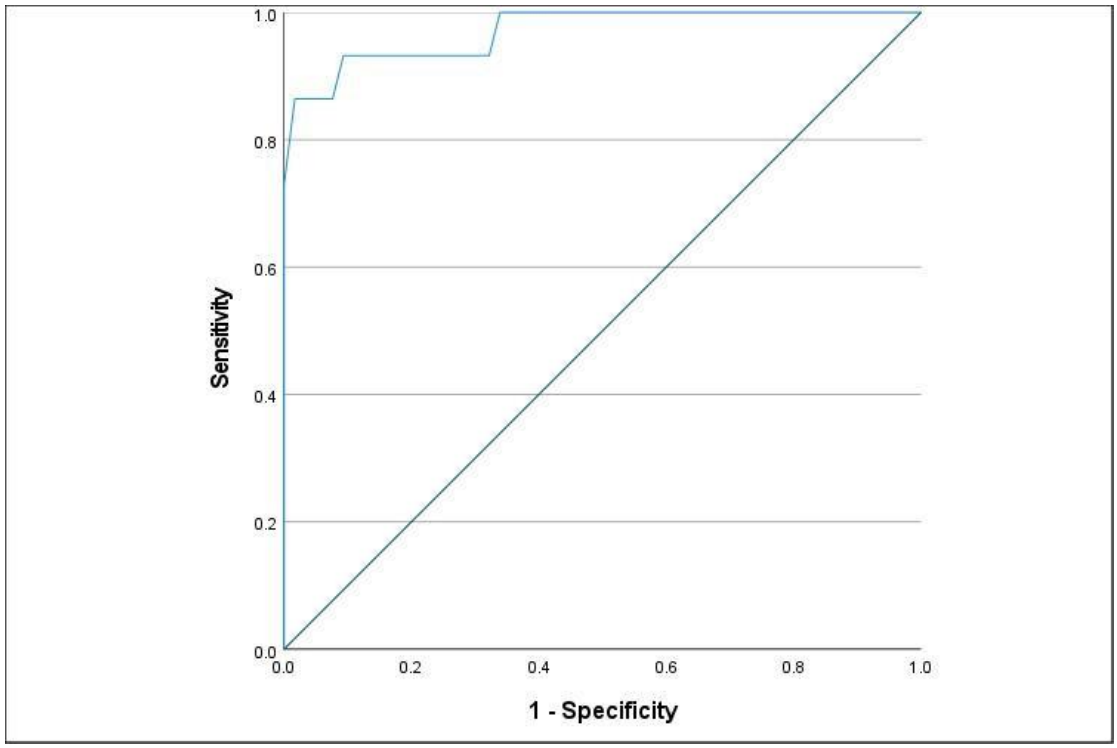

Figure (1S): ROC curve showing performance of apolipoprotein C3 in in prediction of nephropathy in diabetic children