EFFECT OF OXIDIZED LDL ALTER THE LIPOPROTEIN BETWEEN PROTEINURIA AND STAGE 3A CKD PATIENTS IN DYSLIPIDEMIA

E. Keerthika[1], Dr. GSR Kedari [2]

[1,2] Department of Biochemistry, Saveetha Medical college and Hospital, Saveetha Institute of Medical and Technical Science, Chennai, Tamil Nadu, India-602105.

Corresponding author:

E. Keerthika,
Tutor,
Department of Biochemistry
Saveetha Medical College and Hospital
Saveetha Institute of Medical and Technical Science
Chennai, Tamil Nadu, India – 602105

ABSTRACT:

CKD stage 3a represents a critical phase where proteinuria, dyslipidaemia, and oxidative stress accelerate renal decline and cardiovascular risk, with oxidized LDL (Ox-LDL)—a proatherogenic marker not reflected in routine lipid profiles—and reduced antioxidant activity contributing to early vascular and renal injury. This study aimed to evaluate serum Ox-LDL levels in CKD stage 3a patients with and without dyslipidaemia and to examine their demographic and biochemical associations. In this cross-sectional study of patients aged 30–60 years, renal, lipid, and oxidative markers were assessed using standard biochemical assays, UPCR was used to evaluate proteinuria, and Ox-LDL was quantified by ELISA; statistical analysis included ANOVA and correlation tests ($p < 0.05$). Dyslipidaemia patients showed significantly elevated Ox-LDL levels (3.171 ± 0.345 mg/dL) compared to non-dyslipidaemia patients (0.105 ± 0.012 mg/dL; $p = 0.01$), with Ox-LDL demonstrating significant negative correlations with antioxidant activity in both groups, while controls exhibited a nonsignificant positive correlation. Ox-LDL was also significantly associated with GFR, urea, uric acid, ACR, triglycerides, total cholesterol, VLDL, and LDL. Overall, CKD stage 3a is marked by a disrupted oxidative–antioxidant balance, especially in dyslipidaemia, with elevated Ox-LDL reflecting early lipoprotein dysfunction, highlighting the potential utility of oxidative biomarkers and advanced lipid profiling for early risk stratification and targeted therapeutic interventions.

Keywords:
Chronic Kidney Disease, Oxidized Low-Density Lipoprotein (Ox-LDL), Oxidative Stress, Lipid Peroxidation, Cardiovascular disease.

INDRODUCTION

CKD is a progressive disorder that increases renal and cardiovascular risk, with stage 3a (eGFR 45–59 mL/min/1.73 m²) being a critical point for early intervention. Proteinuria signals glomerular injury and predicts disease progression. Dyslipidaemia, particularly oxidative modification of LDL to Ox-LDL, worsens vascular and renal damage by promoting endothelial dysfunction, inflammation, and cytotoxicity, contributing to atherosclerosis and renal injury even without overt lipid abnormalities. Ox-LDL thus acts as a key proatherogenic and proinflammatory factor in CKD, fundamentally altering lipoprotein function and exacerbating organ injury. [1][2].

In CKD, oxidative stress from uremic toxins and weakened antioxidant defenses promotes lipoprotein oxidation undetectable by standard lipid tests. Even early-stage, non-dyslipidemic patients may exhibit a proatherogenic profile with altered particle composition, increased small dense LDL, and elevated oxidized lipoproteins. [3][4]. Oxidative lipoprotein modifications contribute to proteinuria and progressive renal decline, causing glomerular and tubular injury independently of standard lipid levels. Additionally, Ox-LDL and Lp(a) interact to carry oxidized phospholipids, amplifying inflammation and vascular damage. [5][6]. The effect of Ox-LDL on lipoprotein profile alterations in stage 3a CKD patients with proteinuria but without dyslipidaemia is crucial.

This approach can uncover early mechanistic links between oxidative lipoprotein modifications and renal injury. It may also enable the identification of novel biomarkers and therapeutic targets aimed at attenuating oxidative stress, thereby potentially slowing CKD progression and reducing cardiovascular risk before manifest dyslipidaemia or overt clinical complications arise [7][8].Collectively, this framework underscores the proteinuria stage 3a CKD patients with dyslipidaemia, oxidized LDL significantly alters lipoprotein profiles by generating highly atherogenic small dense LDL particles and dysfunctional HDL, thereby amplifying oxidative stress, inflammation, and vascular injury. This intricate interplay contributes to the accelerated progression of cardiovascular and renal disease seen in this population[9]. A comprehensive understanding of Ox-LDL driven lipoprotein alterations may unlock novel biomarkers and therapeutic targets beyond conventional lipid lowering, aiming to mitigate residual risk associated with oxidative lipoprotein dysfunction in CKD.

MATERIALS AND METHODS:

Participants:

Inclusion criteria: CKD stage 3a patients aged 30–60 years, with diagnosed dyslipidaemia per NKF KDOQI guidelines and comorbid conditions like type 2 diabetes mellitus, obesity, or hypertension.

Exclusion criteria: End-stage renal disease, acute infections, inflammatory diseases.

Sample Collection:

Blood Samples will be collected from chronic kidney disease 2 patients from Saveetha Medical College and Hospital.10 ml of blood will be collected from the anti-cubital vein under aseptic conditions. Serum is separated after centrifugation. Handle and store specimens in stoppered containers to avoid contamination and evaporation. Refrigerated at - 20 ˚ C (long term storage)

Biochemical Analysis:

Renal function (urea, creatinine) and lipid markers were measured using the Vitros 5600 dry chemistry analyzer. Total cholesterol (CHOD–PAP), triglycerides (GPO–PAP), and HDL cholesterol (precipitation method) were determined directly, while LDL and VLDL were calculated using the Friedewald formula for triglycerides <400 mg/dL, following standard protocols and quality control. Proteinuria was assessed via the Spot Urine Protein-to-Creatinine Ratio (UPCR), calculated as UPCR (mg/g) = (Urine Protein / Urine Creatinine) × 1000, using first-morning spot urine. Protein was measured by Biuret or turbidimetric methods and creatinine by Jaffe's or enzymatic assays. Serum Ox-LDL was quantified by sandwich ELISA, involving sequential incubation with standards/samples, biotinylated antibody, Streptavidin-HRP, and TMB substrate, with absorbance read at 450 nm and concentrations determined from a standard curve.

Statistical Analysis:

One way ANOVA, Conducted using SPSS v15.0.P-values < 0.05 considered statistically significant.

Ethical clearance:005/04/2024/IEC/SMCH

RESULT :

Table 1: Frequency and percentage distribution of levels of Serum Oxidized LDL among CKD stage 3a patients without dyslipidaemia.

Serums	Status of Dyslipidaemia	
	With Dyslipidaemia CKD stage 3a	Without Dyslipidaemia CKD stage 3a
Ox-LDL		
<0.12: Low oxidative stress	41 (91.11%)	40 (88.89%)
0.12–1.48: Normal	4 (8.89%)	5 (11.11%)
>1.48: High oxidative stress	0 (0.00%)	0 (0.00%)

Table 1, In CKD stage 3a patients, serum Ox-LDL levels showed a similar pattern in both dyslipidaemia and non-dyslipidaemia groups, with the majority exhibiting low oxidative stress. Low Ox-LDL levels (<0.12) were observed in 91.11% of dyslipidaemia and 88.89% of non-dyslipidaemia individuals, while only a small proportion in each group fell within the normal range, and none showed high oxidative stress. Overall, oxidative stress remained low regardless of dyslipidaemia status in this population.

Figure 1: Percentage distribution of levels of Serum Oxidized LDL among CKD stage 3a patients with and without dyslipidaemia

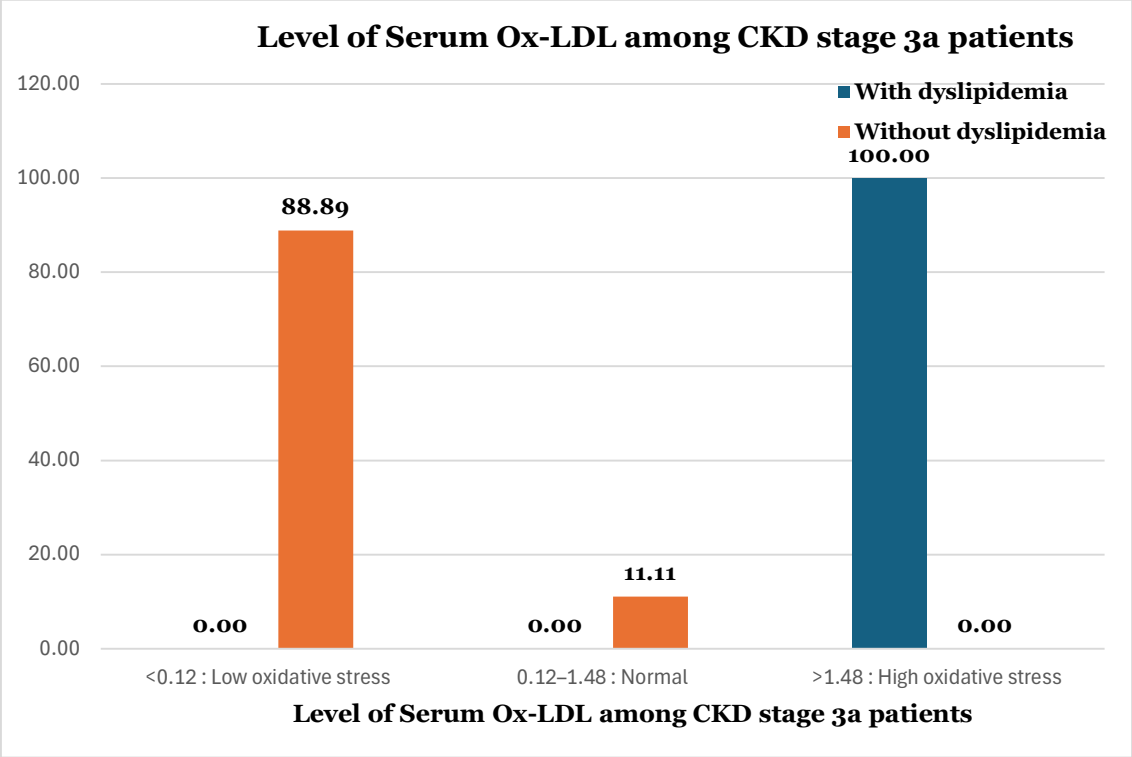

Figure 1,For Ox-LDL levels, all patients with dyslipidaemia (100%) showed high oxidative stress (>1.48 mg/dL), whereas 91.11% of the non-dyslipidaemia group had low oxidative stress (<0.12 mg/dL), and 8.89% displayed normal levels. Notably, none of the non-dyslipidaemia individuals fell into the high oxidative stress category.

Table 2: Level of Serum Oxidized LDL among the CKD patients across stages with and without dyslipidaemia.

Stages	Status of dyslipidaemia	Serum Ox-LDL mg/dl			Mean difference	F-value	p-value
		Mean	S. D	Variance			
Stage3a	**With**	3.171	0.345	0.119	3.066491	705.5917	0.01
	Without	0.105	0.012	0.00016			

Table 2, In CKD stage 3a patients, serum Ox-LDL levels were markedly higher in those with dyslipidaemia compared to those without, with mean values of 3.171 ± 0.345 mg/dL and 0.105 ± 0.012 mg/dL, respectively. This large mean difference was statistically significant (F = 705.59, p = 0.01), indicating a strong association between dyslipidaemia and increased oxidative stress in this group.

Table 3: Correlation of PON1 and Ox-LDL in CKD Stage 3a **With/Without Dyslipidaemia**

CKD Stages	Status of dyslipidaemia	Correlation between serum PON 1 & Ox-LDL		
		Correlation value	Remarks	p value
Stage 3a	With	-0.20301	Negatively correlated	< 0.00001*
	Without	-0.0456	Negatively correlated	< 0.00001*
Control group		0.258271	Positively correlated	1.8033

Table 3, In CKD stage 3a patients, serum PON1 and Ox-LDL showed a significant negative correlation in both dyslipidaemia (r = –0.20301) and non-dyslipidaemia (r = –0.0456) groups, indicating reduced antioxidant activity with increasing oxidative stress. In contrast, the control group showed a nonsignificant positive correlation (r =

0.258271), reflecting a normal oxidative–antioxidant balance. Overall, these findings suggest a disrupted relationship between PON1 and Ox-LDL in CKD stage 3a.

Table 4: Association of level of Serum Paraoxonase 1 and serum Ox-LDL among the CKD stage 3a patients without dyslipidaemia to their demographic variables.

Demographic variables	Serum Ox-LDL	
	Frequency	Chi square and p value
Age		
30-40	3	0.1899
40-50	6	
50-60	36	
Gender		
Male	33	0.4745
Female	12	
BMI (Obesity)		
Class 1 Obesity	43	0.8426
Class 2 Obesity	2	
Class 3 Obesity	0	
Weight (in kg)		
75 kg - 85 kg	16	0.7406
85 kg - 95 kg	14	
Above 95 kg	15	
GFR		
45–59: Stage 3a	45	0.04
Urea		
7–20: Normal	0	0.05
20–40: Mildly elevated	13	
40–70: Moderately high	8	
>70: High	24	
Creatinine		
0.6–1.3: Normal	24	0.0525
1.2–1.5: Mildly elevated	2	
1.5–2.0: Moderately elevated	16	
2.0–3.0: Severely elevated	3	
Uric Acid		
<2.5: Critically Low	1	0.02266
2.5–3.9: Low	4	
4.0–5.5: Lower-Normal	9	
5.6–7.0: Normal	6	
7.1–8.5: Mildly Elevated	21	
8.6–10.0: Moderately Elevated (Hyperuricemia)	3	

>10.0: Severely Elevated (High Risk)	1	
Albumin-to-Creatinine Ratio		
<30: Normal	0	0.034
30–300: Microalbuminuria	45	
>300: Macroalbuminuria	0	
Triglycerides		
<150: Normal	43	0.0426
150–199: Borderline high	2	
200–499: High	0	
≥500: Very High	0	
Total Cholesterol		
<200: Desirable	45	0.021
200–239: Borderline high	0	
≥240: High	0	
HDL Cholesterol		
<50: Low (risk of heart disease)	0	0.0558
50–59: Better	6	
≥60: Protective	39	
VLDL Cholesterol		
2–30: Normal	25	0.04578
>30: High	20	
LDL Cholesterol (Direct)		
<100: Optimal	39	0.00558
100–159: Borderline to High	6	
≥160: Very High	0	
Total Cholesterol / HDL Ratio		
<3.5: Excellent	11	0.9678
3.5–4.4: Good	0	
4.5–5.9: Borderline	25	
≥6.0: High risk	9	

Table 4, Across CKD stage 3a patients, serum Ox-LDL showed no significant association with age, gender, BMI, weight, or most lipid parameters, as indicated by high p-values. However, significant associations were observed with GFR (p=0.04), urea levels (p=0.05), uric acid (p=0.02266), albumin-to-creatinine ratio (p=0.034), triglycerides (p=0.0426), total cholesterol (p=0.021), VLDL (p=0.04578), and LDL cholesterol (p=0.00558), suggesting that renal function markers and specific lipid abnormalities are more closely linked with oxidative stress status in CKD stage 3a.

DISCUSSION :

In CKD stage 3a, the interaction of Ox-LDL, dyslipidaemia, and proteinuria accelerates renal and cardiovascular damage. Dyslipidaemia patients with proteinuria exhibit elevated Ox-LDL, which is associated with harmful changes in lipoproteins, including increased small dense LDL and impaired HDL function. The formation of oxidizable sdLDL perpetuates oxidative stress, promoting vascular injury and further progression of disease. [10]. Ox-LDL drives lipid peroxidation that damages both LDL and HDL, reducing HDL's cholesterol efflux

capacity and diminishing key antioxidative enzymes like PON1 and LCAT, thereby impairing reverse cholesterol transport and its anti-inflammatory functions. [11]. This functional HDL impairment aggravates the pro-atherogenic milieu present in CKD patients with dyslipidaemia and proteinuria. Proteinuria promotes oxidative stress by increasing ROS and inflammation, leading to LDL oxidation and elevated Ox-LDL. Activation of scavenger receptors (LOX-1, CD36) on endothelial and renal cells triggers signaling pathways that drive mesangial proliferation, endothelial dysfunction, and pro-fibrotic cytokine release, exacerbating proteinuria and renal damage.[12]. This bidirectional relationship substantiates Ox-LDL as both a marker and mediator of renal and vascular damage in this patient population. Advanced lipidomics has revealed that LDL particles in CKD undergo compositional remodeling characterized by increased triacyl glycerides and a decrease in protective phospholipids, plasmenyl ethanolamines, and cholesterol sulfate, despite unchanged total cholesterol content. These modifications predispose lipoproteins to enhanced oxidation and dysfunction[13].

Complementarily, proteinuria selectively increases oxidized lipids in very-low-density lipoprotein (VLDL) and HDL fractions rather than LDL, indicating lipoprotein subclass–specific oxidative alterations that complicate the lipid profile in dyslipidemic CKD[14].Lipoprotein subclass profiling further confirms an increased number of small dense LDL and triglyceride-rich VLDL particles in dyslipidemic CKD patients with proteinuria, which are more atherogenic and prone to oxidation. Elevated Ox-LDL to LDL and Ox-LDL to HDL ratios have been proposed as more sensitive biomarkers reflecting the oxidative burden and lipoprotein dysfunction in such clinical settings and are positively correlated with triglyceride levels and inversely with HDL cholesterol[15]. These ratios may outperform traditional lipid measures in cardiovascular risk stratification and renal prognosis. This gene-environment interaction amplifies oxidative lipoprotein remodeling and vascular injury[16].OxLDL promotes foam cell formation, endothelial activation, smooth muscle proliferation, and platelet activation, driving plaque formation and instability. In dyslipidemic, proteinuric CKD patients, these effects are amplified by oxidative stress, inflammation, and lipoprotein remodeling. Ox-LDL also induces endothelial progenitor cell senescence, impairing vascular repair and worsening dysfunction. [17]. Conventional lipid profiles fail to reflect complex lipoprotein alterations in these patients. Including oxidative biomarkers such as Ox-LDL, Ox-LDL /LDL, and Ox-LDL /HDL ratios—along with advanced lipoprotein subclass analysis can improve risk prediction and guide targeted interventions addressing oxidative stress and lipoprotein abnormalities. [18]. The interaction between proteinuria and Ox-LDL mediated oxidative stress intensifies renal and vascular injury. Incorporating oxidative biomarkers and advanced lipoprotein profiling including particle size, number, and oxLDL ratios can enhance risk stratification and guide targeted interventions, as traditional lipid markers underestimate cardiovascular risk in stage 3a CKD. [19]. Ox-LDL triggers the activation of pro-inflammatory genes, promoting monocyte recruitment into the vessel wall and contributing to vascular dysfunction[20]. Atherosclerosis is a condition in which plaque accumulates within the arteries. This plaque is made up of cholesterol, fatty deposits, cellular waste, calcium, and the clot-forming protein fibrin.[21]. These processes narrow or fully block arterial blood flow, resulting in oxygen deprivation of the essential tissues supplied by the affected vessel [22].Elevated Ox-LDL levels potentiate a vicious cycle where in oxidized LDL further promotes formation of sdLDL and dysfunctional HDL, compounding endothelial dysfunction and renal injury even at early CKD stages.

CONCLUSION:

In conclusion, oxidized LDL critically alters lipoprotein quality and function in proteinuric stage 3a CKD patients with dyslipidemia, generating a hidden proatherogenic state that escapes detection by conventional lipid panels but poses significant cardiovascular and renal risk. This pathological nexus involves proteinuria-induced oxidative stress, lipoprotein compositional remodeling, and genetic predispositions, which collectively accelerate vascular injury and CKD progression. Incorporation of oxidative biomarker assessments and detailed lipoprotein characterization into clinical evaluation protocols could improve early risk stratification and guide antioxidant or lipid-modifying interventions aiming to restore lipoprotein homeostasis and mitigate CKD-associated cardiovascular morbidity.

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