DYSLIPIDAEMIA DRIVEN CHANGES IN PON1 ACTIVITY AND LIPID METABOLISM IN CKD STAGE 3A

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ABSTRACT

Chronic kidney disease (CKD) is associated with early dyslipidaemia changes that impair lipoprotein quality and promote oxidative stress. Paraoxonase-1 (PON1), an HDL-bound antioxidant enzyme, plays a critical role in preventing lipid peroxidation, yet its early alterations in CKD stage 3a remain under-explored. This study evaluated serum PON1 activity and its relationship with dyslipidaemia and oxidized LDL (Ox-LDL) in CKD stage 3a patients. Blood samples were analyzed for renal function, lipid profile, Ox-LDL, and PON1 using standardized biochemical assays and ELISA. CKD stage 3a patients with dyslipidaemia showed markedly reduced PON1 levels (100% low) compared to predominantly normal levels in non-dyslipidaemia patients (93.33%). Patients with dyslipidaemia exhibited significantly lower PON1 values ($p = 0.01$). A strong negative correlation between PON1 and Ox-LDL was observed, most pronounced in non-dyslipidaemia CKD ($r = -0.856$). These findings indicate that dyslipidaemia in early CKD is closely linked to reduced PON1 activity and heightened oxidative stress, reflecting early HDL dysfunction not captured by routine lipid panels. Assessing PON1 may improve cardiovascular risk stratification and highlight potential therapeutic targets in moderate CKD.

Keywords:
Chronic Kidney Disease, Paraoxonase-1 (PON1), Oxidative Stress, Lipid Peroxidation, Cardiovascular disease.

INTRODUCTION

CKD is characterized by a distinct dyslipidaemia pattern that includes hypertriglyceridemia, accumulation of triglyceride-rich remnants, reduced HDL cholesterol, and compositional and size changes in LDL and HDL particles. These abnormalities arise from impaired lipoprotein catabolism, insulin resistance, chronic inflammation, and oxidative stress, as well as altered activity of lipases and transfer proteins [1]. CKD profoundly remodels HDL structure and cargo, leading to “dysfunctional HDL” with reduced antioxidant, anti-inflammatory, and cholesterol efflux capacities. Lipidomic analyses in CKD cohorts have shown that specific changes in HDL sphingolipids and ceramides are independently associated with mortality, underscoring the prognostic relevance of HDL quality rather than HDL-C concentration alone [2].

Paraoxonase 1 (PON1) is a liver-derived esterase/lactonase that circulates almost exclusively bound to HDL and is considered a key determinant of HDL’s anti-oxidative function [3]. By hydrolysing oxidized cholesteryl esters and phospholipids on lipoproteins and cell membranes, PON1 attenuates lipid peroxidation, limits LDL oxidation, and modulates multiple atheroprotective processes, including endothelial function and reverse cholesterol transport [4]. Experimental and clinical data consistently link low systemic PON1 activity to heightened oxidative stress and increased incidence of major cardiovascular events, supporting its role as an important mediator—and biomarker—of residual cardiovascular risk [5],[6],[7]. PON1 function is influenced not only by genetic polymorphisms but also by the surrounding lipoprotein milieu; in particular, triglyceride-rich VLDL and alterations in HDL composition can directly inhibit or displace PON1 from HDL, thereby compromising its enzymatic activity and biological effects [8],[9].

In CKD, reduced PON1 activity and concentration have been reported, especially in advanced stages and dialysis populations, and have been associated with higher oxidative stress, vascular dysfunction, and atherosclerotic disease burden [4]. In particular, it remains unclear whether specific dyslipidaemia patterns are already accompanied by measurable reductions in PON1-dependent antioxidative capacity at this early stage, and how these changes relate to renal function, inflammation, and markers of lipoprotein quality. Addressing these gaps is clinically relevant because stage 3a often coincides with the window in which cardiovascular risk-modifying therapies are first considered, yet current risk stratification relies largely on traditional lipid measurements that may fail to capture early lipoprotein dysfunction. A better understanding of the interplay between dyslipidaemia, PON1 activity, and lipid metabolism in CKD stage 3a could help identify high-risk individuals before overt cardiovascular events occur, refine risk assessment beyond conventional lipid parameters, and inform the development of targeted strategies aimed at preserving HDL function and enhancing PON1 activity as a means to mitigate cardiovascular risk.

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 tests (urea, creatinine) and lipid markers measured via Vitros 5600 dry chemistry analyzer. Lipid profile estimation included total cholesterol (CHOD–PAP method), triglycerides (GPO–PAP method), and HDL cholesterol (indirect precipitation method). LDL and VLDL cholesterol were calculated using the Friedewald formula, excluding samples with triglycerides >400 mg/dL. All assays were performed following the manufacturer's instructions with appropriate quality control, and reference ranges were as per standard clinical laboratory guidelines. Serum PON1 levels assessed using sandwich ELISA. The assay procedure involves running all standards and samples in duplicates or triplicates with a standard curve for each assay. First, 100 µL of standard diluent, standards, and samples are added to the respective wells and incubated for 80 minutes at 37 °C. After washing the plate four times, 100 µL of biotinylated antibody working solution is added and incubated for 50 minutes at 37 °C, followed by another wash. Then, 100 µL of Streptavidin-HRP conjugate is added, incubated for 50 minutes, and washed again. Next, 100 µL of TMB substrate is added and incubated for 10 minutes at 37 °C without shaking. The reaction is stopped with 100 µL of stop solution, changing the colour from blue to yellow. Absorbance is read at 450 nm within 10–15 minutes. The mean absorbance values of standards are plotted against concentrations to generate a standard curve, from which sample concentrations are determined, adjusting for dilution if necessary.

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: Comparison of Biochemical Parameters between CKD Stage 3a Patients and Control Group

Table 1 shows the comparative analysis of renal and lipid profile parameters, including Serum PON1 levels, between CKD Stage 3a patients and healthy controls.

Parameter	Control (Mean ± SD)	CKD Stage 3a (Mean ± SD)	P-value
Urea (mg/dL)	26.27 ± 7.23	72.13 ± 7.36	< 0.01
Creatinine (mg/dL)	0.72 ± 0.35	1.34 ± 0.18	< 0.01
Uric Acid (mg/dL)	4.28 ± 1.33	8.77 ± 1.79	< 0.01
Triglycerides (mg/dL)	87.09 ± 10.33	184.78 ± 2.70	< 0.01
Total Cholesterol (mg/dL)	114.84 ± 17.19	257.00 ± 3.66	< 0.01
HDL Cholesterol (mg/dL)	61.00 ± 6.46	25.16 ± 9.31	< 0.01
VLDL Cholesterol (mg/dL)	18.36 ± 4.82	45.04 ± 7.36	< 0.01
LDL Cholesterol (mg/dL)	73.60 ± 8.94	167.89 ± 3.91	< 0.01
Serum PON1 level (ng/mL)	4.05 ± 0.78	0.55 ± 0.26	< 0.01

Table 2: Frequency and percentage distribution of levels of Serum Paraoxonase 1 among CKD stage 3a patients without dyslipidaemia.

Serums	Status of Dyslipidaemia	
	With Dyslipidaemia CKD stage 3a	Without Dyslipidaemia CKD stage 3a
Serum Peroxonase 1 (PON1)		
<3.1: Low	45 (100.00%)	3 (6.67%)
3.1–9.7: Normal	0 (0.00%)	42 (93.33%)
>9.7: Elevated	0 (0.00%)	0 (0.00%)

Table 2, CKD stage 3a patients with dyslipidaemia showed low PON1 levels (100%), while those without dyslipidaemia predominantly had normal PON1 levels (93.33%). This indicates that dyslipidaemia is strongly associated with reduced antioxidant enzyme activity (PON1) in CKD 3a patients

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

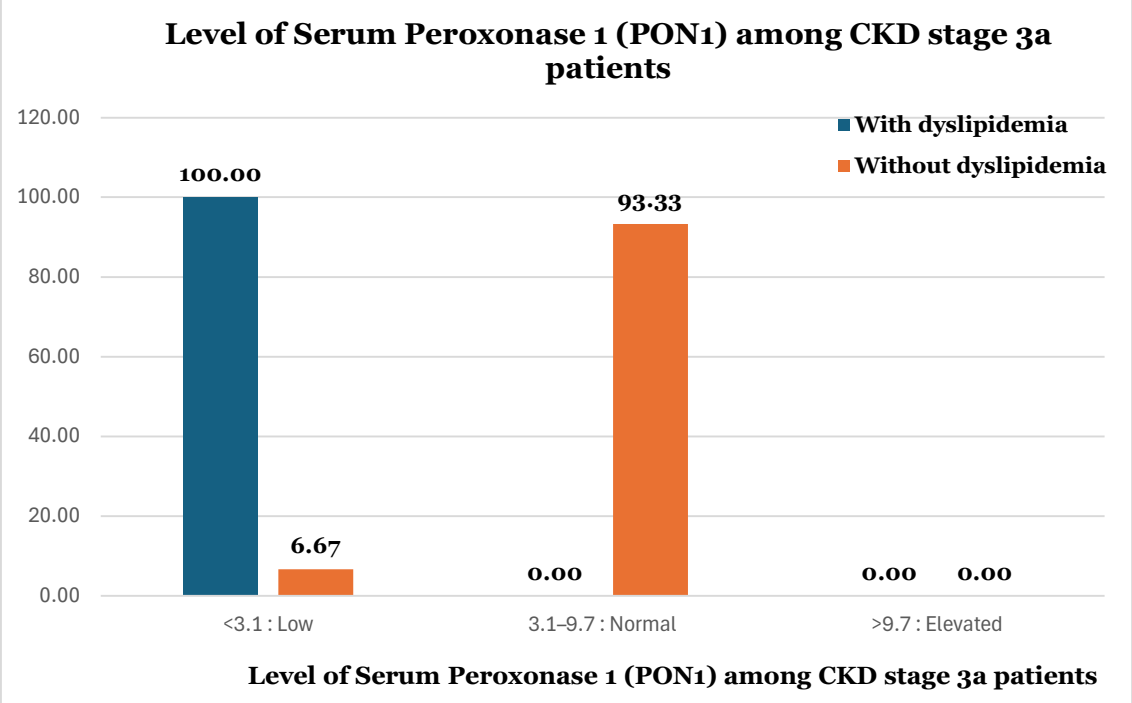

Figure 1, The figure shows that CKD stage 3a patients with dyslipidaemia mostly have low PON1 levels, whereas those without dyslipidaemia mainly have normal PON1 levels, highlighting reduced PON1 activity in dyslipidaemic patients.

Table 3: Level of Serum Paraoxonase 1 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	0.601	0.063	0.004	4.907664	183.8375	0.01

	Without	5.508	1.692	2.862			

Table 3, Stage 3a CKD patients with dyslipidaemia showed significantly lower PON1 levels than those without dyslipidaemia, with a large mean difference and a statistically significant p-value (0.01).

Table 4: 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	-0.0301	Negatively correlated	< 0.00001*
	Without	-0.856	Negatively correlated	< 0.00001*
Control group		0.05271	Positively correlated	< 0.00001*

Table 4, In CKD stage 3a, PON1 and Ox-LDL show a negative correlation in both dyslipidaemia and non-dyslipidaemia patients, with a much stronger negative correlation in those without dyslipidaemia. The control group shows a weak positive correlation. All correlations are statistically significant.

DISCUSSION

Our data show that dyslipidaemia in CKD stage 3a is already accompanied by substantial reductions in PON1 activity and qualitative alterations in lipoproteins, indicating that functional HDL impairment begins early in the course of renal dysfunction and is more tightly linked to the dyslipidaemia milieu than to eGFR alone. The reduction in PON1 activity we observed at eGFR 45–59 mL/min/1.73 m² is consistent with the concept that PON1 dysfunction is an early feature of kidney disease and a plausible contributor to premature cardiovascular risk in CKD.[10] In line with epidemiologic work linking low PON1 activity and specific functional polymorphisms to increased systemic oxidative stress and major adverse cardiovascular events in non-CKD cohorts[11]. our findings suggest that such vulnerability may already be present in moderate CKD, where traditional lipid indices often appear only mildly abnormal. By demonstrating that PON1 activity relates more strongly to triglycerides, remnant cholesterol, and altered HDL traits than to LDL-C or total cholesterol, our results echo the broader shift from focusing on HDL quantity to HDL quality and functionality.[12] Prior lipidomic studies in CKD have shown that specific HDL lipid species, such as ceramides and sphingomyelins, predict mortality independent of bulk HDL-Supporting the idea that compositional remodelling of HDL in CKD has prognostic significance. Our study adds to this picture by linking a key functional enzyme on HDL and PON1 to the dyslipidaemia pattern typical of stage 3a CKD.

Our observation that lower PON1 activity clusters with hypertriglyceridemia and remnant-rich dyslipidaemia is mechanistically plausible. VLDL triglycerides can directly inhibit HDL-associated PON1 catalytic activities in a dose-dependent fashion and shift HDL toward a more triglyceride rich, cholesteryl ester–poor phenotype that is less favourable for PON1 stability. The remodelling of HDL by lipases such as endothelial lipase and hepatic lipase further modifies particle size, surface phospholipids, and protein cargo, which in turn affects PON1 binding and displacement from HDL.[13] In CKD stage 3a, where low-grade inflammation and oxidative stress are already present, this adverse lipoprotein environment could both impair hepatic PON1 synthesis and promote inactivation of the circulating enzyme. The strong relationship we observed between PON1 activity and dyslipidaemia indices aligns with the broader literature showing that genetically and biochemically determined PON1 activity tracks with systemic oxidative stress and cardiovascular outcomes. Moreover, experimental augmentation of circulating PON1 in mice improves HDL resistance to oxidation, enhances macrophage cholesterol efflux, and reduces cellular cholesterol accumulation and biosynthesis, providing a biologic rationale for why even modest decrements in PON1 activity in CKD 3a might have disproportionate atherogenic consequences. Our findings of reduced PON1 activity in stage 3a CKD therefore likely reflect early entry into a pathogenic pathway that, if uncorrected, may culminate in structural and functional cardiac damage.

Implications for HDL functionality and risk assessment in CKD 3a contribute to the growing recognition that HDL-C concentration alone is an inadequate surrogate for cardiovascular protection in CKD. Large trials of HDL-C raising strategies have failed to improve outcomes, underscoring the importance of HDL composition and function antioxidant capacity, anti-inflammatory activity, and cholesterol efflux over static cholesterol content.PON1 is now understood as a central determinant of these protective HDL functions, and evolutionary and clinical perspectives suggest that PON1 activity may track atherosclerotic risk at least as well as, and sometimes better than, HDL-C itself.[14]Standard lipid panels may therefore underestimate risk in individuals with low PON1 activity and dysfunctional HDL. This aligns with evidence that alterations in HDL structure and

bioactive lipid cargo, such as enrichment in ceramides or depletion of protective sphingolipids, predict mortality in CKD independently of traditional lipids. Dyslipidaemia is recognized as a key contributing factor in the development of coronary heart disease [15]. Abnormal lipid levels and elevated blood pressure jointly trigger endothelial damage, promote vascular inflammation and remodelling, and ultimately drive atherosclerotic progression [16]. An imbalance in this system may result in cellular damage and inflammatory responses that compromise immune defence [17]. From a clinical standpoint, this suggests that risk stratification in CKD 3a should eventually move beyond LDL-C and HDL-C toward metrics that capture HDL functionality, oxidative stress, and PON1 status. Although routine PON1 testing is not yet established, our data add to the rationale for developing standardized assays and cut-offs that could be integrated into CKD-specific cardiovascular risk models.

CONCLUSION

In stage 3a CKD, dyslipidaemia is already linked to marked reductions in PON1 activity and qualitative disturbances in lipoprotein metabolism, pointing to early HDL dysfunction that is not captured by conventional lipid panels. These findings support a shift from purely quantitative lipid assessment toward functional and qualitative evaluation of lipoproteins in moderate CKD and highlight PON1 as a promising biomarker and potential therapeutic target for preventing cardiovascular complications in this high-risk population.

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