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Short Communication

Biochemistry Section

Serum Biochemical Changes among Chronic Kidney Disease Patients in a Rural Cohort of Odisha, India: A Cross-sectional Study

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ABSTRACT

Introduction: Chronic Kidney Disease (CKD) is one of the fastest-growing causes of death worldwide. Monitoring biochemical parameters such as liver function tests, kidney function tests, and lipid profiles is crucial for early identification of health complications, prevention, and prompt management in CKD patients.

Aim: To investigate the association of sociodemographic factors and serum biochemical indicators with Glomerular Filtration Rate (GFR) among CKD patients in rural Odisha, India.

Materials and Methods: A cross-sectional study was conducted in a rural cohort of the Model Rural Health Research Unit (MRHRU) established in Cuttack district, Odisha, India, from March 2021 to April 2021. A total of 530 registered CKD patients were enrolled in the study. Sociodemographic data and blood samples were collected for lipid profile, kidney function, and liver function analysis. The data were analysed using STATA 15.1 and RStudio 2021.09.0+351.

Results: The mean age of the study participants was 51.2±14.1 years, with 330 (62.26%) being male. A total of 74% of the participants showed abnormal Estimated Glomerular Filtration Rate (eGFR). Abnormal eGFR was found to be more common among the older age group (>45 years), those who were literate, and those in private jobs, compared to their counterparts. Both lipid profile and liver function abnormalities were significantly associated with individuals having normal eGFR.

Conclusion: The positive association of eGFR with lipid profile and liver function enzymes highlights the importance of regular screening of these parameters among CKD patients for early prevention and control of future complications.

Keywords: Glomerular filtration rate, Kidney function test, Lipid profile, Liver function test

INTRODUCTION

Chronic Kidney Disease (CKD) is one of the most important public health problems in the world, with a prevalence of 13.4% (11.7-15.1%) [1]. CKD is defined as a GFR of less than 60 mL/min/1.73 m^2 for three months or more, irrespective of the cause [2]. If not treated in time, it may lead to kidney failure, cardiovascular disease, and premature death [2]. Regular laboratory follow-up is crucial, especially in high-risk groups such as people with diabetes or hypertension, for preventing any adverse conditions [2].

From a diagnostic perspective, several biochemical markers exist in the serum (serum urea and creatinine concentration) and urine (albumin-to-creatinine ratio), which can be used as markers for renal dysfunction or injury. Most of these biomarkers are associated with their own advantages and disadvantages that should be considered before utilising them for regular monitoring and diagnosis of CKD. Although serum creatinine has been used as an endogenous biomarker for renal function, its dependence on muscle mass stands as a limitation for its use [3]. Similarly, despite guidelines recommending testing urine albumin-to-creatinine ratio to analyse kidney dysfunction, its lack of sensitivity and specificity for progressive decline in eGFR has been widely reported [4].

The GFR is defined as the flow of plasma from the glomerulus into Bowman's space over a specified period and is the most widely used parameter to measure kidney function. The eGFR depends on the serum creatinine level in patients with kidney disease [5]. The level of liver enzymes in the serum is suggestive of aggression against hepatocytes [6]. Interestingly, patients with CKD on haemodialysis have been found to have lower levels of liver enzymes in the serum than those with normal renal function [7].

An association has been observed between one liver enzyme, Alkaline Phosphatase (ALP), and increased mortality in CKD patients before dialysis and on dialysis as well [8]. Due to continuous changes in dietary habits and lifestyle, risk factors for CKD are increasing. Some research studies have reported that the intake of a high-fat and high-sugar diet increases the incidence of CKD [9,10]. The prevalence of dyslipidaemia was found to be 78.67% among non diabetic CKD patients admitted to a hospital in India, and it was accompanied by decreased High-density Lipoprotein (HDL) and Triglycerides (TG) levels in the serum [11].

However, the role of TG, cholesterol, Low-density Lipoprotein (LDL), and HDL in the development and progression of CKD is still unclear. Further studies are inevitable to understand whether these are independent risk factors for CKD or if they associate with eGFR, and which type of lipid has the greatest impact on CKD. In India, the rural health system is unable to manage the rising burden of CKD due to a shortage of qualified doctors, specifically nephrologists, leading to under-diagnosis and under-treatment, resulting in a high rate of adverse outcomes among CKD patients [12].

Furthermore, most researchers focus on urban settings, and research on CKD patients residing in rural areas remains in the background. The present study focused on CKD patients in a rural area that is known to have a large number of CKD patients and aims to understand the sociodemographic as well as serum biochemical risk factors among them. Additionally, no similar study has been conducted in a rural cohort from the eastern region of India.

The MRUHU, Tigiria, is a unit of Indian Council of Medical Research-Regional Medical Research Centre (ICMR-RMRC), Bhubaneswar, situated in a rural area of Odisha, an eastern state of India. The catchment area of this MRHRU harbours some blocks that are known to be CKD hotspots (according to secondary data collected from hospitals in respective places). The present study was conducted to study the sociodemographic parameters and evaluate different biochemical indicators (serum creatinine, liver function

enzymes: total and direct bilirubin, Serum Glutamic-oxaloacetic Transaminase (SGOT); Serum Glutamic-pyruvic Transaminase (SGPT); lipid profile: Total Cholesterol (TC); TG; HDL; LDL and Very Low-density Lipoprotein (VLDL) in CKD patients and also to check their association with eGFR.

MATERIALS AND METHODS

A cross-sectional study was conducted in the rural catchment area (three blocks: Tigiria, Badamba and Narasjnghapur) of MRHRU, Cuttack, Odisha, during March 2021-April 2021. The study was approved by the Institutional Ethical Committee (IEC) of ICMR-RMRC, Bhubaneswar (ICMR-RMRC/IHEC-2021/89).

Inclusion criteria: Registered CKD patients with all grades, aged 18 years and above, who gave written informed consent were included in the study.

Exclusion criteria: Pregnant women and individuals with cognitive impairment were excluded from the study.

Sample size calculation: The minimum sample size for the finite population was calculated as 564, using the following formula:

$$n=(deff*N*p*q)/[(d^2/Z^2)*(N-1)+p*q]$$

where n=sample size; deff=design effect; N=population size; p=estimated proportion; q=1-p; d=desired precision or absolute level of precision. Assuming a prevalence of dyslipidaemia as 78.6% [11], an absolute precision of 3.93%, and a non response rate of 10%, for a finite population of 322,056.

Study Procedure

Data of registered CKD patients were collected from the hospital CKD register of respective areas. From a total of 2,481 registered CKD patients, 570 were randomly chosen and approached for their participation, and 545 patients consented to participate. On the day of sample collection, a total of 530 patients came to the facility, and using a validated structured proforma, sociodemographic data (age, gender, education, caste, occupation, etc.) were recorded through the Open Data Kit (ODK) smartphone application.

A 3 mL blood sample was collected using aseptic techniques under careful supervision and immediately sent to the Model Rural Health Research Unit (MRHRU) laboratory for serum separation by the centrifugation method (3,000 rpm for three minutes at room temperature). Out of all, 37 samples could not be processed for liver function tests, and one sample could not be analysed for SGOT, SGPT, and ALP due to an insufficient amount of sample volume.

Laboratory procedure: All the serum biochemical tests were done at the MRHRU laboratory, Tigiria, using the Erba Chem 5x semi-auto analyser. Biochemical test methods and reference ranges for all the laboratory parameters are presented in [Table/Fig-1] [13,14].

Parameters	Biochemical test method	Reference range
Serum creatinine	Jaffe's method, initial rate	Male: 0.7-1.4 mg/dL
		Female: 0.6-1.2 mg/dL
Total cholesterol	Dynamic extended stability CHOD-PAP method with LCF, end point	<200 mg/dL
Triglyceride (TG)	Dynamic extended stability with lipid clearing agent GPO-Trinder method, end point	<161 mg/dL
High-density Lipoprotein (HDL)	End point trinder reaction	Male: 35.3-79.5 mg/dL
		Female: 42-88 mg/dL
Low-density Lipoprotein (LDL)	TC (HDL+VLDL)	<150 mg/dL
Very Low-density Lipoprotein (VLDL)	TG/5	2-35 mg/dL
Total bilirubin	Diazo method, end point	0-2.0 mg/dL
Direct bilirubin	Diazo method, end point	0-0.2 mg/dL
Serum Glutamic-oxaloacetic Transaminase (SGOT)	IFCC method, kinetic	Male: 35 IU/L
		Female: 31 IU/L
Serum Glutamic-pyruvic Transaminase (SGPT)	IFCC method, kinetic	Male: 45 IU/L
		Female: 34 IU/L
Alkaline Phosphatase (ALP)	Lowry method	<15 years: 54-369 IU/L
		<50 years: 42-128 IU/L
		≥50 years: 53-141 IU/L

[Table/Fig-1]: Details of the methods and reference range of different biochemical tests [13,14].
CHOD-PAP: Cholesterol oxidase/peroxidase aminophena; GPO: Glycerol phosphate oxidase; TC: Total cholesterol; IFCC: International federation of clinical chemistry

STATISTICAL ANALYSIS

Statistical analysis was performed using STATA 15.1 and RStudio 2021.09.0+351. Univariate descriptive analysis was used to determine the prevalence of abnormal eGFR across the sociodemographic characteristics of the population. A multivariate binary logistic regression model was adopted to assess the association of abnormal eGFR with sociodemographic characteristics. Bivariate Chi-square analysis was conducted to examine the categorical association of eGFR status with lipid profile and liver function. The correlation between eGFR values and individual lipid profile and liver function indicators was represented graphically through a correlation plot.

RESULTS

Out of 530 CKD patients, a total of 392 participants had abnormal eGFR. The highest number of patients 219 (55.87%) in the age group of 45-64 years had a significantly higher Adjusted Odds Ratio (AOR) of 3.65 for having abnormal eGFR compared to the age group of 18-44 years, where 71 (18.11%) patients had abnormal eGFR. Among the 330 males and 200 females, 258 (65.82%) males and 134 (34.18%) females had abnormal eGFR, with males having a higher AOR of 2.23 for the risk of abnormal eGFR, although this was not statistically significant. Among the three different caste categories, the majority of patients, 154 (39.23%) in the general category and 153 (39.03%) participants in the Other Backward Caste (OBC) group, had abnormal eGFR. In the present study population, literate individuals, those with co-morbidities, and those in private jobs were found to have significantly higher AOR for abnormal eGFR [Table/Fig-2]. There was no available information for one patient regarding caste, education, and morbidity.

Sociodemographic variables	Sample n (%)	eGFR abnormal (n=392) n (%)	AOR (95% CI)	p-value
Age group (in years)				
18-44	167 (31.51)	71 (18.11)	Ref.	
45-64	256 (48.30)	219 (55.87)	3.65 (2.11-6.28)*	<0.001
65 above	107 (20.19)	102 (26.02)	9.14 (3.11-26.82)*	<0.001
Gender				
Female	200 (37.74)	134 (34.18)	Ref.	
Male	330 (62.26)	258 (65.82)	2.23 (0.65-7.56)	0.198
Caste				
SC/ST	100 (18.90)	85 (21.68)	Ref.	
General	211 (39.89)	154 (39.29)	0.74 (0.33-1.68)	0.480
OBC	218 (41.21)	153 (39.03)	1.02 (0.46-2.26)	0.954
Education				
Illiterate	157 (29.68)	144 (36.73)	Ref.	
Literate	372 (70.32)	248 (63.27)	0.48 (0.23-0.99)**	0.047
Occupation				
Agriculture	67 (12.64)	58 (14.80)	Ref.	
Unemployment	132 (24.91)	128 (32.65)	2.20 (0.55-8.66)	0.259
Private job	116 (21.89)	62 (15.82)	0.36 (0.14-0.93)**	0.036
Government job	37 (6.98)	22 (5.61)	0.73 (0.17-3.09)	0.671

Housewife	158 (29.81)	108 (27.55)	1.26 (0.27-5.80)	0.765
Other	20 (3.77)	14 (3.57)	0.46 (0.10-2.05)	0.314
Any co-morbidity				
No	246 (46.50)	128 (32.74)	Ref.	
Yes	283 (53.50)	263 (67.26)	5.06 (2.77-9.25)*	<0.001

[Table/Fig-2]: Multivariable logistic regression model of abnormal eGFR with association of sociodemographic characteristics.
Multivariable logistic regression analysis. p<0.01*, p<0.05** (Denotes statistically significant); eGFR: Estimated glomerular filtration rate; AOR: Adjusted odds ratio; CI: Confidence interval; SC: Schedule caste; ST: Schedule tribes; OBC: Other backward caste.
There was no information available for one patient regarding caste, education and morbidity (N=530)

Out of the 530 CKD patients, 228 (58.16%) patients were found to have both abnormal lipid profiles and abnormal eGFR, and 131 (36.09%) had both abnormal liver function and abnormal eGFR. eGFR showed a statistically significant association with lipid profile (p-value=0.001) and liver function (p-value=0.001), raising concerns for CKD patients [Table/Fig-3].

Biochemical variables		eGFR		Total	Chi-square test	p-value
		Abnormal n (%)	Normal n (%)			
Lipid profile	Abnormal	228 (58.16)	109 (78.99)	530	19.11*	<0.01
	Normal	164 (41.84)	29 (21.01)			
Liver function	Abnormal	131 (36.09)	68 (52.31)	493	10.46*	0.001
	Normal	232 (63.91)	62 (47.69)			

[Table/Fig-3]: Association of estimated Glomerular Filtration Rate (eGFR) with biochemical variables (lipid profile and liver function).
p<0.01 *(Denotes statistically significant)

The mean levels of TG, Cholesterol (CHOL), HDL, VLDL, and LDL among patients with CKD were 142.65±124.32, 170.35±50.42, 51.14±15.53, 28.53±24.86, and 147.73±52.85, respectively. The mean levels of Total bilirubin (T), Direct bilirubin, SGOT, and SGPT were 0.39±0.21, 0.19±0.13, 28.63±10.58, and 22.53±9.17, respectively [Table/Fig-4].

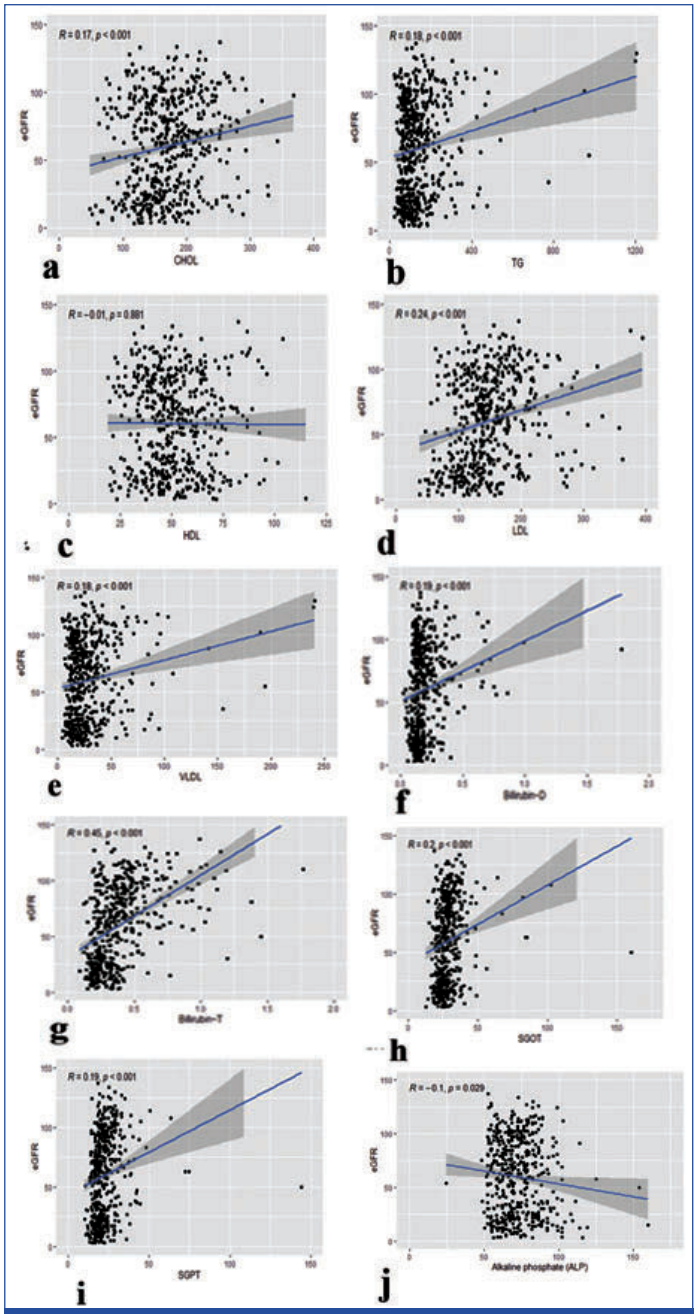
Biochemical variables		Total (N)	Range (min, max)	Mean±SD
Lipid profile test	TG	530	1182.16 (22.84,1205)	142.65±124.32
	CHOL	530	320.51 (47.89,368.4)	170.35±50.42
	HDL	530	96.29 (18.91,115.2)	51.14±15.53
	VLDL	530	236.43 (4.56,241)	28.53±24.86
	LDL	530	356.6 (37.7,394.3)	147.73±52.85
Liver function test	Bilirubin (T)	493	1.68 (0.09, 1.77)	0.39±0.21
	Bilirubin (D)	493	1.76 (0.02, 1.78)	0.19±0.13
	SGOT	492	147.85 (12.5, 160.4)	28.63±10.58
	SGPT	492	134.2 (10, 144.2)	22.53±9.17
	ALP	492	135.5 (24.4, 160.02)	72.22±14.52
Kidney function test	Creatinine	530	17.22 (0.46,17.68)	2.02±2.09
	eGFR	530	134 (3,137)	60.18±34.91

[Table/Fig-4]: Descriptive statistics of lipid profile and liver function bio-chemical variables.
SD: Standard deviation; TG: Triglycerides; CHOL: Cholesterol; HDL; High-density lipoprotein; VLDL: Very low-density lipoprotein; LDL: Low-density lipoprotein; (T): Total; (D): Direct; SGOT: Serum glutamic-oxaloacetic transaminase; SGPT: Serum glutamic-pyruvic transaminase; ALP: Alkaline phosphatase; eGFR: Estimated glomerular filtration rate

When the correlation between eGFR and individual parameters of liver function tests and lipid profile was checked using Pearson's correlation coefficient analysis, taking all the parameters as continuous variables, a significantly positive correlation was observed between eGFR and all parameters (TC, TG, LDL, VLDL, direct bilirubin, total bilirubin, SGOT, and SGPT), while eGFR showed a negative correlation with ALP and HDL [Table/Fig-5a-j,6].

DISCUSSION

The CKD has become a worldwide problem, affecting millions of people and its incidence and prevalence are increasing. Timely detection of risk factors and appropriate treatment can prevent



[Table/Fig-5]: Represents association of eGFR with: (a) Total cholesterol; (b) Triglycerides; (c) High-density lipoprotein; (d) Low-density lipoprotein; (e) Very low-density lipoprotein; (f) Bilirubin-direct; (g) Bilirubin total; (h) SGOT; (i) SGPT; and (j) ALP; analysed through bivariate scatter plot and Pearson's correlation coefficient analysis. Value of the correlation coefficient (r) ranges from-1 (negative correlation) to+1 (positive correlation). 0 denotes no correlation; SGOT: Serum glutamic-oxaloacetic transaminase; SGPT: Serum glutamic-pyruvic transaminase; ALP: Alkaline phosphatase.

Biochemical variables	Correlation value	p-value
CHOL	0.17	<0.001
TG	0.18	<0.001
HDL	-0.01	0.881
LDL	0.24	<0.001
VLDL	0.18	<0.001
Bilirubin (D)	0.19	<0.001
Bilirubin (T)	0.45	<0.001
SGOT	0.2	<0.001
SGPT	0.19	<0.001
ALP	-0.1	0.029

[Table/Fig-6]: Karl-Pearson's correlation coefficient of estimated Glomerular Filtration Rate (eGFR) values with lipid profile and liver function parameters.
CHOL: Cholesterol; TG: Triglycerides; HDL; High-density lipoprotein; LDL: Low-density lipoprotein; VLDL: Very low-density lipoprotein; (T): Total; (D): Direct; SGOT: Serum glutamic-oxaloacetic transaminase; SGPT: Serum glutamic-pyruvic transaminase; ALP: Alkaline phosphatase

the development or progression of CKD. In the present study, approximately 74% of patients had abnormal eGFR, while the remaining patients had normal eGFR, which could be attributed to their medication practice since their diagnosis. The various functions of the kidney are influenced by the aging process, and the GFR decreases with age, starting around the age of 30 [15]. The present study demonstrates a higher AOR for having abnormal eGFR in older age groups, including those aged 45-64 years and above 65 years, with the highest AOR recorded in people above 65 years. This is supported by the fact that GFR decreases to <60 mL/min/1.73 m^2 in healthy adults over the age of 60-65 years, although this may vary according to gender and age [16]. A study on the Canadian population by Ma I et al., showed that women and elderly individuals were significantly associated with an increased risk of abnormal eGFR, suggesting a higher risk for CKD. Conversely, minority individuals and those with high median household income had a significantly lower risk of CKD [17]. Contradictory to these findings, the current study did not find a significant difference between males and females or among different castes regarding the risk of having abnormal eGFR. Additionally, according to the present study's results, literate individuals and those in private jobs appear to be at a higher risk of having CKD.

The contradictory results between other studies and the present study may be due to different study settings, variations in the health status, and lifestyles of the study participants [17-19]. For example, in the Netherlands, low-level education was found to be associated with higher odds of unfavourable CKD outcomes among all ethnic groups studied [18]. Similarly, in a German CKD cohort, low educational attainment was positively associated with diabetic nephropathy and CKD following acute kidney injury [19]. In the case of individuals in private jobs, their occupation may not directly influence kidney function, but the associated work pressure and poor lifestyle choices may contribute to abnormal kidney function. According to Seligman HK et al., low occupational level is associated with poor food and lifestyle choices, which directly affects the risk of cardiometabolic diseases and CKD [20].

Another common issue in CKD patients is co-morbidity, but its impact on disease progression and outcomes is less studied. The present study showed a significantly higher chance (AOR=5.06) of having abnormal eGFR in people with co-morbidity compared to those without any co-morbid condition. Since co-morbidity is known to be associated with poor renal outcomes in CKD patients, there is an urgent need for tailored treatment strategies for CKD in high-risk groups [21]. Similarly, the association of various biochemical indicators with abnormal eGFR or CKD has also been rarely studied, resulting in ambiguous results.

The present study demonstrates that both lipid profile and liver function are associated with eGFR, highlighting the importance of regularly evaluating these parameters in both healthy individuals and those with kidney disease to prevent aggressive renal conditions. According to Liang X et al., high levels of TG, total TC, and LDL are associated with the occurrence of CKD and indicative of the progression of renal dysfunction and disease [22]. Since hepatic co-morbidities such as hepatitis B and C are common among CKD patients, testing serum liver enzymes is valuable for diagnosing and monitoring liver damage in this group of patients [23]. Furthermore, studies have established a relationship between TC and declined GFR, as well as TC, LDL, and end-stage renal disease in patients with type 1 diabetes and overt nephropathy [24,25]. These findings support the results of the present study and suggest a strong association between lipid profile, liver function enzymes, eGFR, and kidney dysfunction.

Limitation(s)

The present study was conducted in the catchment area of the MRHRU, which includes three specific blocks in a particular geographical area. Therefore, the findings of similar studies conducted in other regions may vary. Additionally, the authors did not compare serum indicators with non CKD cases, which could provide further insights. The small sample size and, in a few cases, the inability to process samples for liver function tests due to insufficient sample volume were major limitations of the present study.

CONCLUSION(S)

The positive association between lipid profile and liver function enzymes with eGFR highlights the importance of regularly examining serum biochemical indicators to detect renal dysfunction at an early stage and ensure proper disease management. Since lipids and liver function enzymes are also associated with other morbidities such as obesity and diabetes, patients with these risk factors should be treated with utmost precaution to prevent further deterioration of their health status.

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