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Correlation of serum iron and chronic kidney disease stages: A cross-sectional study

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ABSTRACT

Introduction: Chronic kidney disease (CKD) is a progressive disorder characterized by a gradual loss of renal function and frequent disturbances in iron metabolism. These disturbances often lead to iron deficiency and anemia. Serum iron and ferritin are fundamental biomarkers for evaluating iron status and can provide valuable insights into disease severity and progression. The aim of this study was to determine if there is an association between CKD stage and serum iron concentration, focusing on the dynamics of iron homeostasis and CKD progression.

Methods: At the Clinical Center of the University of Sarajevo, a cross-sectional study was conducted, including 115 participants diagnosed with CKD in moderate to advanced stages (3-5). Patients with acute infections, other causes of anemia, or acute kidney injury were excluded to avoid factors that could confound iron metabolism. Laboratory analyses included measurements of serum iron, ferritin, urea, and creatinine. Estimated glomerular filtration rate (eGFR) was calculated using the modification of diet in renal disease equation to determine disease stage. Descriptive statistics and correlation analyses were used to examine associations between serum iron, ferritin, and CKD stage, with statistical significance set at $p < 0.05$.

Results: The study included 64% male and 36% female participants, with 17% in stage 3, 22% in stage 4, and 61% in stage 5 of the disease. As CKD progressed, serum ferritin levels increased, while serum iron levels gradually declined. Correlation analysis revealed a significant positive association between eGFR and serum iron ($p < 0.01$). The results also indicated that abnormal iron values were more frequent in males than in females, which is consistent with previous reports.

Conclusion: The severity of CKD is associated with disturbances in iron metabolism, pointing to the importance of monitoring serum iron and ferritin. These findings support early detection, individualized patient management, and timely interventions to prevent iron deficiency and anemia in patients with CKD.

Keywords: Chronic kidney disease; serum iron; ferritin; estimated glomerular filtration rate; anemia

INTRODUCTION

Chronic kidney disease (CKD) is defined as persistent kidney damage or a reduction in glomerular filtration rate (GFR <60 mL/min/1.73 m^2) lasting longer than 3 months (1). The pathophysiology of CKD is associated with disturbances in the excretory, endocrine, and metabolic functions of the kidneys. These disturbances often lead to the accumulation of uremic toxins, imbalance of fluids and electrolytes, reduced synthesis of erythropoietin and active vitamin D, and the development of hypertension and acid-base disorders (2). CKD represents one of the major global public health challenges, with its incidence and prevalence showing a global upward trend (3,4). According to the 2017 Global Burden of Disease study, around 1.2 million deaths were linked to CKD, and predictions suggest that this number could reach up to 4 million annually by 2040, placing CKD among the leading causes of death worldwide (5). The evaluation of CKD includes a comprehensive medical history, clinical examination, laboratory tests, and diagnostic imaging. Serum creatinine and creatinine clearance analyses are still used as standard indicators of kidney function, although they can be affected

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TABLE 1. Differences in biochemical markers according to stage of renal insufficiency

Parameter	CKD	Mean	SD	Median	IQR (25-75%)	Chi-square	*p*-value
Urea (mmol/L)	CKD 5	22.880	5.9248	23.200	18.475-25.650	16.036	<0.001
	CKD 4	19.808	8.3153	19.400	16.350-22.400		
	CKD 3	16.155	6.4831	14.300	11.650-20.475		
Creatinine (umol/L)	CKD 5	777.81	215.608	768.50	610.75-939.00	84.802	<0.001
	CKD 4	303.76	49.591	306.00	280.00-343.50		
	CKD 3	139.90	40.030	140.50	102.00-156.00		
eGFR (mL/min/1.73 m2)	CKD 5	7.04	1.592	7.00	6.00-8.00	87.583	<0.001
	CKD 4	18.32	3.461	17.00	16.00-20.50		
	CKD 3	44.55	11.560	49.00	31.25-53.00		
Iron (umol/L)	CKD 5	7.403	2.1546	7.450	5.925-9.150	52.591	<0.001
	CKD 4	9.388	2.6076	8.200	7.450-11.050		
	CKD 3	16.505	3.7489	16.300	13.500-20.475		
Acidum uricum (umol/L)	CKD 5	395.77	70.471	389.50	352.00-416.25	4.730	0.094
	CKD 4	372.12	104.365	351.00	287.50-450.50		
	CKD 3	360.55	52.797	382.00	333.50-391.75		
CRP (mg/L)	CKD 5	4.479	4.3868	2.550	1.375-6.600	1.269	0.530
	CKD 4	3.648	4.2319	2.400	0.950-4.950		
	CKD 3	3.525	3.4489	2.250	0.775-6.075		
Feritin (ng/mL)	CKD 5	392.962	238.3852	374.315	161.018-551.875	11.929	0.003
	CKD 4	317.764	192.2305	249.200	178.595-470.310		
	CKD 3	217.151	247.9965	135.010	42.080-296.670		

eGFR: Estimated glomerular filtration rate, Acidum uricum: Uric acid, CRP: C-reactive protein, SD: Standard deviation, IQR: Interquartile range

by various biological and technical factors (6,7). Anemia is the most common and clinically significant complication of CKD, which primarily results from reduced erythropoietin production (8). The prevalence of anemia increases as CKD progresses, affecting about 25% of patients in stage 3 and rising to 75-95% in those with end-stage renal disease. More recent studies, however, report that anemia occurs in 55% of patients in stage 4 and in over 33% of those in stage 5 (9,10). CKD-associated anemia is usually normocytic and normochromic, and it is linked to a higher risk of heart problems, cognitive impairment, and mortality (2,8). Measuring serum iron is an important part of diagnosing and monitoring renal anemia, as it allows for early detection and can help reduce the risk of complications in patients with CKD (11).

METHODS

This cross-sectional study was conducted between June 2023 and June 2024 at the Nephrology Clinic of the Clinical Center of the University of Sarajevo. The study included 115 adult patients (≥18 years) of both genders with CKD, diagnosed according to the kidney disease: Improving global outcomes criteria. Patients with a history of blood transfusions or iron supplementation were not included. Exclusion criteria comprise non-normocytic or non-normochromic anemia, acute infections, conditions that could affect hematological parameters (such as chronic inflammatory diseases and liver disorders), malignancies, acute kidney injury, as well as previous blood transfusions or iron therapy. The study included patients with moderate to advanced stages of CKD (stages 3-5), enabling evaluation of different levels of renal impairment and related hematological changes.

Venous blood samples were collected in serum tubes without anticoagulant and centrifuged at 3,500 rpm for 5 min,

TABLE 2. Prevalence of abnormal serum iron values in the total study population

Iron	Abnormal values n (%)		Reference values n (%)		Chi-square	*p*-value
(umol/L)	84	73.0	31	27.0	24.426	<0.001

while complete blood count (CBC) samples were collected in K2-EDTA tubes. Serum was used to determine concentrations of iron, urea, creatinine, ferritin, uric acid, and C-reactive protein (CRP) on a Cobas 6000 analyzer (Roche Diagnostics, Switzerland) using colorimetric, kinetic, and immunoturbidimetric methods. CBC was analyzed using an automated Sysmex XN-3100 hematology analyzer employing fluorescent flow cytometry, hydrodynamically focused impedance, and the sodium lauryl sulfate method for hemoglobin. Estimated GFR (eGFR) was calculated using the modification of diet in renal disease formula.

The study was approved by the Ethics Committee of the University of Sarajevo - Faculty of Health Studies and the Institute for Scientific Research of the Clinical Center of the University of Sarajevo. Participant's personal data were treated in strict confidence in accordance with the Declaration of Helsinki. As secondary data sources were utilized, individual informed consent was not required.

Statistical analyses were conducted using the IBM Statistical Package for the Social Sciences Version 21. Descriptive statistics, parametric and non-parametric tests, Kolmogorov–Smirnov and Shapiro–Wilk tests for normality, Chi-square and Fisher's exact tests for categorical variables, Mann–Whitney U and Kruskal–Wallis H tests for group comparisons, and Spearman's correlation for association analysis were employed. The results are presented as median (interquartile range) and mean (standard deviation), with statistical significance defined at $p < 0.05$.

TABLE 3. Frequency of abnormal serum iron values (μmol/L) by gender

Gender	Abnormal values (%)	Reference values (%)	Total (%)	Pearson Chi-square	*p*-value
Male	59 (79.7)	15 (20.3)	74 (100)	4.713	0.047
Female	25 (61.0)	16 (39.0)	41 (100)		
Total	84 (73.0)	31 (27)	115 (100)		

RESULTS

A total of 115 patients were included in the study. Men represented 64% of the study population (n = 74), and women 36% (n = 41), resulting in a male-to-female ratio of 1.8. Chi-square testing showed that men were significantly more represented than women ($p = 0.002$). The mean age was 62.26 ± 14.58 years for men and 63.10 ± 13.59 years for women. The Mann-Whitney U test found no significant difference in age between men and women.

Analysis across CKD stages indicated that 70 patients (61%) were classified as stage 5 CKD with eGFR <15 mL/min/1.73 m^2, 25 patients (22%) as stage 4 CKD with eGFR 15-29 mL/min/1.73 m^2, and 20 patients (17%) as stage 3 CKD (3a or 3b) with eGFR 30-59 mL/min/1.73 m^2. In the stage 5 CKD subgroup, 64.3% were men and 35.7% were women, with a male-to-female ratio of 1.8. Among patients with stage 4 CKD, men represented 76% and women 24% (male-to-female ratio = 3.16). Stage 3 CKD showed an equal distribution of both genders, with 50% men and 50% women (male-to-female ratio = 1.0). Chi-square analysis showed no statistically significant differences in gender distribution across CKD stages.

Table 1 shows the biochemical parameters by CKD stage. Kruskal-Wallis analysis demonstrated statistically significant intergroup differences for urea ($p < 0.001$), creatinine ($p < 0.001$), eGFR ($p < 0.001$), serum iron ($p < 0.001$), and ferritin ($p = 0.003$). eGFR values were lowest in stage 5, intermediate in stage 4, and highest in stage 3, reflecting the expected decline in kidney function. Iron levels also decreased with advancing CKD, with the lowest values in stage 5, intermediate in stage 4, and highest in stage 3. Conversely, ferritin levels showed an opposite trend, being highest in stage 5, moderately lower in stage 4, and lowest in stage 3 patients.

Table 2 presents the frequency of pathological iron values in the total study population (n = 115). Their occurrence was higher compared to cases in which iron levels were within the reference range ($p < 0.001$).

Table 3 presents the observed frequencies of abnormal iron values (μmol/L) stratified by gender. A statistically significant disparity was identified between genders in the prevalence of abnormal versus reference-range iron values ($p = 0.047$), with a higher prevalence observed among male participants.

Table 4 shows a statistically significant difference in the prevalence of abnormal and reference-range iron values across CKD stages ($p < 0.001$). Abnormal iron values were most frequent in the CKD 5 group (90% of cases), moderately frequent in the CKD 4 group (80%), and least frequent in the CKD 3 group (5%).

Table 5 presents the correlation between the investigated parameters. A statistically significant positive correlation was determined using Spearman's correlation test. A significant positive correlation was observed between eGFR and iron levels (rho = 0.523, $R^2 = 0.697$, $p < 0.001$). Higher iron values were associated with a tendency toward higher eGFR values, whereas lower iron values corresponded to a tendency toward lower eGFR values.

TABLE 4. Frequency of abnormal serum iron values (μmol/L) by CKD stage groups

Gender	Abnormal values (%)	Reference values (%)	Total (%)	Pearson Chi-square	*p*-value
CKD 5	63 (90.0)	7 (10.0)	70 (100)	57.864	<0.001
CKD 4	20 (80.0)	5 (20.0)	25 (100)		
CKD 3	1 (5.0)	19 (95)	20 (100)		
Total	84 (73.0)	31 (27)	115 (100)		

CKD: Chronic kidney disease

DISCUSSION

The aim of this study was to determine whether there is a correlation between the CKD stage and serum iron concentration, specifically assessing the dynamics of iron homeostasis in relation to CKD progression. Similar studies have been conducted worldwide in recent years due to the increasing prevalence of CKD and the need for optimized treatment approaches (12,13). Our cohort analysis showed that men represented 64% of participants, while women comprised 36%, indicating a higher occurrence of CKD among men. Conversely, Raji et al. (14) in Nigeria reported a higher prevalence among women (56%), indicating regional differences.

Participants were classified according to CKD stage, based on eGFR (15,16). Most patients were in the terminal stage CKD 5 (61%), followed by CKD 4 (22%) and CKD 3 (17%). These findings differ from Sanni et al. (17), who reported a predominance of CKD 4 (41.6%), but are consistent with Sinomono et al. (18), in which 82.7% of patients were classified as CKD 5.

Analysis of gender distribution across CKD stages showed no statistically significant differences ($p > 0.05$), suggesting that gender does not strongly influence stage-specific distribution. Male predominance was observed in CKD 5 and CKD 4 (64.3% and 76%, respectively), while CKD 3 had an almost equal distribution (~50%). These results are inconsistent with Waziri et al. (19) but agree with Shrestha et al. (20).

Biochemical analysis showed significant differences in urea, creatinine, eGFR, serum iron, and ferritin across CKD stages. eGFR values ranged from 5 to 60 mL/min/1.73 m^2, with the lowest in stage 5 and the highest in stage 3, consistent with previous reports by Shrestha et al. (20), Salwaji et al. (21), and Aoun et al. (22).

Serum ferritin increased progressively with advancing CKD, reflecting its role as an acute-phase reactant responding to systemic inflammation and oxidative stress (23,24).

TABLE 5. Correlations between studied parameters

Parameters	Erythrocytes ($\times10^{12}$/L)	MCV (fL)	Platelets ($\times10^{9}$/L)	Urea (mmol/L)	Creatinine (umol/L)	eGFR (mL/min/1.73 m^2)	Iron (umol/L)	Ferittin (ng/mL)
MCV (fL)								
Rho	−0.066	/	/	/	/	/	/	/
p	0.482							
Platelets ($\times10^{9}$/L)								
Rho	0.232	−0.255	/	/	/	/	/	/
p	0.013	0.006						
Urea (mmol/L)								
Rho	−0.235	0.143	−0.186	/	/	/	/	/
p	0.011	0.126	0.047					
Creatinine (umol/L)								
Rho	−0.301	0.302	−0.340	0.456	/	/	/	/
p	0.001	0.001	<0.001	<0.001				
eGFR (mL/min/1.73 m^2)								
Rho	0.295	−0.327	0.258	−0.443	−0.951	/	/	/
p	0.001	<0.001	0.005	<0.001	<0.001			
Iron (umol/L)								
Rho	0.495	−0.031	0.165	−0.321	−0.510	0.523	/	/
p	<0.001	0.744	0.078	<0.001	<0.001	<0.001		
Ferittin (ng/mL)								
Rho	−0.070	0.288	−0.221	−0.002	0.250	−0.299	−0.108	/
p	0.454	0.002	0.017	0.986	0.007	0.001	0.251	
CKD-stage								
Rho	0.350	−0.297	0.318	−0.370	−0.862	0.877	0.639	−0.296
p	<0.001	0.001	0.001	<0.001	<0.001	<0.001	<0.001	0.001

MCV: Mean corpuscular volume of erythrocytes, eGFR: Estimated glomerular filtration rate, CKD-stage: Stage of chronic kidney disease

The highest ferritin levels were observed in stage 5, while the lowest were in stage 3, in line with findings by Alam et al. (25) and Verma et al. (26).

Conversely, serum iron levels decreased as CKD progressed. The lowest levels were observed in stage 5, and the highest in stage 3, consistent with Alam et al. (25), Verma et al. (26), Clementi et al. (27), and Elsayedi and Azab. (28). Gender-specific analysis showed that men had a higher prevalence of abnormal iron levels. Owever, previous studies (22) found that women have more severe iron deficiency, especially in early CKD stages, likely due to a higher risk of anemia. Similar trends have been reported by Minutolo et al. (29) and Fishbane et al. (30), although stage-specific differences were not significant, unlike in our cohort.

Overall, these results confirm a clear association between serum iron, ferritin, erythrocyte parameters, and CKD severity (eGFR/stage). The use of strict exclusion criteria – including patients without acute infection (CRP and leukocyte counts within reference ranges) and the exclusion of other types of anemia (normochromic-normocytic anemia confirmed by mean corpuscular volume and mean corpuscular hemoglobin)-ensured the reliability of the data and the relevance of the findings.

CONCLUSION

The study findings indicate a high prevalence of abnormal serum iron levels, especially among male participants and those with stage 5 CKD. Correlation analysis demonstrated a significant positive association between eGFR and serum iron, indicating that higher eGFR is associated with higher iron levels, while lower eGFR corresponds to reduced iron. A clear connection between iron deficiency and anemia in CKD was observed, and the underlying mechanisms need further investigation in larger studies. Clinically, these results highlight the importance of monitoring for low iron and anemia as common complications in patients with CKD.

Study limitation

The relatively small sample size indicates the need for future larger studies to further validate these findings.

DECLARATION OF INTEREST

Authors declare no conflict of interest.

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