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Original Research Paper

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ANEMIA IN CHRONIC KIDNEY DISEASE AND CORRELATION OF SERUM CREATININE WITH SERUM FERRITIN LEVELS IN THE SAME GROUP.

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ABSTRACT

Purpose: Anemia is a common complication associated with CKD (Chronic Kidney disease) that limits the quality of life and affects the morbidity and mortality in CKD patients. Mainstays for treatment of anemia are erythropoiesis stimulating agents (ESA) and Iron therapy. Serum ferritin, frequently used as a marker of iron stores in dialysis patients, is also an acute phase reactant, which could be used as a marker of morbidity and mortality in CKD patients. Thus, our aim is to study anemia in CKD patients who were on dialysis and receiving ESA and Iron therapy and to study the correlation between serum creatinine levels with serum ferritin levels in the same group.

Methods & Results: The study was conducted on 150 indoor CKD patients and 150 control group. The following parameters were observed in the cases with mean values of: haemoglobin (9.11 ± 2.54 g/dl), MCV (79.26 ±14.23fl), MCH (26.36 ± 7.16pg), MCHC (31.32 ± 2.73g/dl), serum iron (45.02 ± 50.40u/dl), TIBC (251.68 ± 102.20u/dl), transferrin saturation (17.47 ± 29.85%), serum ferritin (452.78 ± 433.45ng/dl), serum creatinine (13.77±8.54mg/dl) and serum urea (105.25±55.98mg/dl).

Conclusion: To conclude, the data analysis showed persistence of renal anemia with significant p value (<0.01) and there was a positive correlation of serum creatinine and serum ferritin levels (r value 0.98). Ferritin, being an acute phase reactant can thus be used as a marker of morbidity in CKD patients.

KEYWORDS : chronic kidney disease, renal anaemia, ferritin, serum creatinine, transferrin saturation.

INTRODUCTION

CKD is a prevalent, worldwide condition; and the number of patients affected continues to increase with overall prevalence of 14% in general population [1]. Prevalence of End stage renal disease (ESRD) in U.S. in 2017 showed a 2.5% increase since 2016 with its ranking among the highest in the world [2].Anemia is a major factor that limits the quality of life in CKD patients and may affect their morbidity and mortality rates. Insufficient production of erythropoietin from the failed kidneys is the major cause of anemia in this population. Several other factors such as, deficiencies of iron, folic acid, and vitamin B12; blood loss, and reduced erythrocyte survival may contribute to the causes of anemia in CKD [3-6].Anemia, as defined by the NKF, is haemoglobin (Hb) concentration < 12 g/dl for women and < 13.5 g/dl for men [7]. In patients receiving dialysis, recommended Hb target value is ≥ 11 g/dl in women and ≥ 12 g/dl for men.

In addition to well known symptoms of anemia like fatigue, dizziness, and shortness of breath, it has been associated with more severe adverse outcomes, such as cardiovascular complications including left ventricular hypertrophy and congestive heart failure [8]. Correction of anemia has been shown to improve cardiac function possibly by reducing exercise-induced myocardial ischemia [9]. Treatment of anemia associated with CKD has also been shown to result in improvements in exercise capacity, physical performance features such as endurance, energy, and physical mobility [10]. Finally, treatment of anemia has been shown to reduce hospitalization and mortality rates [11]. ESAs and iron are the mainstays of treatment for anemia associated with CKD. This standard protocol was also followed in our hospital.

Most commonly used parameters for monitoring iron stores are serum ferritin and transferrin saturation (TSAT). Serum ferritin concentrations are also elevated in infections and inflammation and transferrin saturation is altered in states of hypoalbuminemia and chronic disease, resulting in false TSAT values [3]. In this study patients were on haemodialysis receiving iron therapy and ESA hence, had sufficient iron stores. In spite of the above therapy, anemia persisted. Hence, to reduce morbidity related to anemia, earlier mentioned causes should be given equal importance.We also studied the correlation between raised serum creatinine with serum ferritin levels in CKD patients. Hence, the aims and objectives of our study were to study anemia in CKD patients and identify the correlation between serum creatinine with serum ferritin in the same group.

MATERIALS AND METHODS

This prospective study was performed after approval from the institute's ethical committee. The study included 150 CKD indoor patients undergoing regular haemodialysis at Nephrology Unit in MGM Medical College, Aurangabad. It was an analytical study conducted from January 2019-December 2019. Control group was chosen with matched age, sex and BMI with non-anaemic, non-CKD and had no major medical illness. Patients included were CKD patients with stage IV (7/150) and V (143/150) as defined by The National Kidney Foundation (NKF), Kidney Disease Outcomes Quality Initiative (KDOQI) [13]. The patients were on haemodialysis and receiving iron therapy and ESA. We excluded patients with liver disease, non-alcoholic hepatic steatosis, chronic alcoholism, chronic inflammatory conditions, rheumatoid arthritis, inflammatory bowel disease, bacterial infection, haematological malignancy and thyrotoxicosis.

The parameters studied in the patients and control group were; blood urea, serum creatinine levels, complete blood counts with indices, iron studies and serum ferritin (Table 1). However the RBC count and the peripheral blood smear findings were not included in the study. Data management and statistical analysis were conducted by SPSS version 21.

Table 1: Showing methods used for assessment of various parameters.

Assessment	Methods
Assessment of Renal Function • Serum Creatinine • Blood urea	Fully automated biochemistry analyzer VITROS 5600 Integrated System by using a modification of the kinetic Jaffe´ reaction and bichromatic endpoint technique, respectively
Hemoglobin with blood indices	Siemens ADVIA 2120 i and peripheral smears were examined accordingly
Iron Studies	Fully automated biochemistry analyzer VITROS 5600 Integrated System Dade Behring RxL Max
Serum Ferritin	VITROS 5600 Integrated System using Intellicheck Technology (immunometric technique)

RESULTS

In the present study out of 150 patients, 140 had raised blood urea levels and 123 patients had raised serum creatinine level. Among the study group and controls the mean age was 65 ±15 years, out of which 85 were men and 65 were women. We found that out of 150 CKD patients, 34 patients were diabetic and 66 were hypertensive and 50 had both. Control group had normal kidney function test and no major illness. It was observed that the mean value in the study group (n=150) for haemoglobin was 9.11 ± 2.54g/dl, MCV was 79.26 ±14.23fl, MCH was 26.36 ± 7.16 pg, MCHC was 31.32 ± 2.73g/dl. The mean values for serum iron was 45.02 ± 50.40μg/dl, TIBC was 251.68 ± 102.20μg/dl, transferrin saturation was 17.47 ± 29.85% and serum ferritin was 452.78 ± 433.45ng/dl. The mean values for serum creatinine was 13.77±8.54mg/dl and serum urea was 105.25±55.98mg/dl. Control group had mean values of hemoglobin as 13.46 ± 1.99g/dl, MCV was 89.06 ± 6.45fl, MCH was 29.66 ± 2.53pg, MCHC was found to be 33.14 ± 1.17g/dl. Mean value for iron studies were 92.67 ±20.86μg/dl for serum iron, 294.96±44.17μg/dl for TIBC, 32.99±11.68% for transferrin saturation and 203.73±68.88ng/dl for serum ferritin. Mean values of serum creatinine was 0.47±0.26mg/dl and serum urea was 11.31±4.39mg/dl (Table 2).

Table 2. Descriptive statistics in CKD patients and controls.

Parameters	Controls	CKD patients
Number	150	150
Age (years)	65 ± 15	65 ± 15
Haemoglobin (g/dl)	13.46 ± 1.99	9.11 ± 2.54
MCV (ft)	89.06 ± 6.45	79.26 ± 14.23
MCH (pg)	29.66 ± 2.53	26.36 ± 7.16
MCHC (g/dl)	33.14 ± 1.17	31.32 ± 2.73
Serum iron (μg/dl)	92.67 ±20.86	45.02 ± 50.40
TIBC(μg/dl)	294.96 ± 44.17	251.68 ± 102.20
Transferrin saturation (%)	32.99 ± 11.68	17.47 ± 29.85
Serum ferritin (ng/ml)	203.73 ± 68.88	452.78 ± 433.45
Blood urea(mg/dl)	11.31± 4.39	105.25 ± 55.98
Serum Creatinine(mg/dl)	0.47± 0.26	13.77 ±8.54

Haemoglobin values were found to be low in 76% patients as compared to the control group, indicating that they had anemia. It was observed that MCV was reduced in 96.6%, MCH was reduced in 97.3 %, and MCHC was reduced in 97.3% of CKD patients, depicting it as microcytic hypochromic anaemia, considering haemoglobin and blood indices. Iron studies showed that serum iron was reduced in 95.3 %, TIBC was normal in 96%, iron saturation was reduced in 84.6% and serum ferritin levels were increased in 77.3% of patients (Table 3).

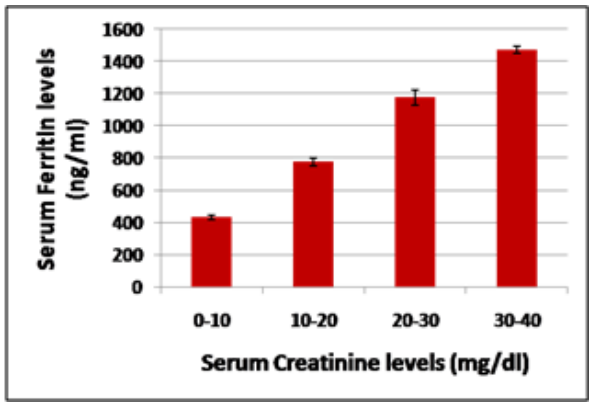
Table 3. Interpretation of parameters in renal anemia

Parameters (Normal range)*	Number of CKD patients	P value
Hemoglobin (12-15 g/dl)	114 (76%) Reduced	<0.05
MCV (80-96 fl)	145(96.6%) Reduced	<0.05
MCH (27-33 pg)	146 (97.3%) Reduced	<0.05
MCHC (33-36 g/dl)	46 (97.3%) Reduced	<0.05
Serum Iron (50-160 μg/dl)	143 (95.3%) Reduced	<0.05
TIBC (250-400μg/dl)	144 (96%) Normal	<0.05
Iron saturation (20-55%)	127 (84.6%) Reduced	<0.05
Serum Ferritin (12-300ng/ml)	116 (77.3%) Increased	<0.05
Blood urea (15-38mg/dl)	140(93.3%) Increased	<0.05
Serum creatinine (0.8-1.3mg/dl)	123 (82%) Increased	<0.05

*Normal ranges adopted from Henry's clinical diagnosis and management by laboratory methods. First South-Asia edition 2016

All the above parameters suggest that renal anemia (microcytic hypochromic) was persisting in patients on dialysis with ESA and iron therapy. Serum creatinine and blood urea was increased in 82 % and 93.3% of patients, respectively. In this study group, with the increasing ferritin levels, there was a rise in serum creatinine levels showing a positive correlation (Figure 1). Hence, serum ferritin can be considered as a marker of morbidity in CKD patients.

Figure 1: Bar diagram showing positive correlation of serum creatinine with serum ferritin levels



DISCUSSION

Approximately, kidneys produce 90% of the hormone erythropoietin. Under normal physiological conditions, hypoxia in the kidney leads to an increase in the production of erythropoietin, which subsequently stimulates erythropoiesis [14]. Hence in patients receiving dialysis and especially those on haemodialysis, chronic blood loss resulting from frequent phlebotomy for laboratory studies and loss of blood in the dialysis tubing and dialyzer after each haemodialysis treatment may also contribute to declining Hb values. Other contributory factors for anemia include malnutrition and deficiencies of iron, folate, and vitamin B12, systemic and chronic inflammatory states, shortened red cell survival by uremic toxins and drugs [15, 16]. According to 2006 NKF KDOQI-recommended target for Hb concentration is ≥ 11–12 g/dl [7].

Therefore, optimal treatment of anemia due to CKD requires appropriate diagnosis, ESA and iron therapy, and close monitoring of response.

ESAs and iron are the mainstays of treatment for anemia associated with CKD. ESAs are used to stimulate erythropoiesis by either directly or indirectly acting on the erythropoietin receptor [8]. Without adequate iron stores, ESAs will not be effective. In patients who do not have CKD, yearly iron loss is ~ 500 mg. Conversely, patients with CKD on haemodialysis, annual iron loss could be around to 2–4 g or more. Patients with CKD may become iron deficient for a number of reasons. In addition to physiological blood loss, patients with CKD often exhibit increased erythropoiesis and iron utilization from occult gastrointestinal blood loss resulting from platelet dysfunction that occurs with uremia [7,15]. The role of iron in oxygen transport makes excess of it toxic by generating free radicals. This iron is provided by recycling from senescent red blood cells which are phagocytosed by reticculoendothelial cells and from dietary absorption or from liver stores [17]. Thus, periodic assessment of serum iron parameters is necessary [7,15]. There is no ideal test for monitoring iron storage. Most commonly, iron status is evaluated by serum ferritin and transferrin saturation (TSAT). However, ferritin is an acute phase reactant and can be elevated for reasons other than sufficient or excessive iron stores. Examples of conditions in which serum ferritin concentrations are elevated despite iron deficiency include infection and inflammation. Similarly, transferrin, and hence transferrin saturation, are altered in states of hypoalbuminemia and chronic disease, resulting in false TSAT values. Ferritin and TSAT are frequently discordant, as they are, respectively, positive and negative acute phase reactants [18]. Consequently in the setting of inflammation or malnutrition, high ferritin and low TSAT are frequently altered. However, as more specific and sensitive tests for the assessment of iron, like content of Hb in reticulocytes (CHr) are not readily available, most clinicians will assess serum ferritin and TSAT. Absolute iron deficiency is defined as a serum ferritin concentration, less than 100ng/ml in non–dialysis-dependent CKD and in patients treated by peritoneal dialysis and a serum ferritin concentration of < 200ng/ml in haemodialysis patients. For all patients with CKD regardless of whether they are receiving dialysis, TSAT concentrations < 20% indicate iron deficiency [7]. In our study the patients were receiving ESA and iron therapy but even then anemia persisted.

Ferritin is the iron storage molecule of the body. It is estimated that almost half of all maintenance haemodialysis patients in the United States have a serum ferritin >500 ng/ml [19]. Extremely high serum ferritin levels, usually >2000 ng/ml are indicative of Iron (Fe) overload, also known as haemosiderosis [20,21]. CKD is associated with oxidative stress and chronic inflammation which links it with cardiovascular diseases. This oxidative stress can increase the inflammatory response and has a synergistic effect on CKD development and progression. Ferritin can thus act as a surrogate marker of oxidative stress and inflammation [22]. The increase in serum ferritin during inflammation, infection, liver disease, malignancies, and other non–Fe-related conditions may hinder the ability to assess the Fe status in CKD under the concurrent presence of foregoing conditions. Serum ferritin is also a marker of malignancy, such as in neuroblastoma, renal cell carcinoma, or Hodgkin's lymphoma [21]. In patients with CKD, hyperferritinemia is paradoxically associated with ESA hypo responsiveness and a more severe anemia [21].

In our study we found microcytic hypochromic picture as the most prevalent anemia with the red cell indices having low mean values of Hb, MCV, MCH and MCHC. This correlated with the study done by Talwar et al in which microcytic hypochromic anemia had a higher prevalence [22]. However the study done by Neha S. et al had normocytic normochromic anemia as the most common finding on peripheral smear followed by microcytic picture, although there was lower mean values of Hb and red cell indices [16]. Our study found low mean serum iron levels with normal TIBC and reduced TSAT in the study group as seen in anemia of chronic disease. The low iron despite of iron therapy can be attributed to other causes of loss of blood in CKD patients. The low TIBC in our study can be explained by hypoalbuminemia due to malnutrition and increased red cell destruction due to oxidative damage in CKD patients. The study done by Csaba P. et al showed low TIBC with high TSAT in patients with raised serum ferritin [23] which was non-concordant with findings of our study.

Our study reported high mean value of serum ferritin in patients with CKD which is in concordance with the study done by Kamyar K.Z. et al in the patients who had CKD with glomerular disease and proteinuria [21]. In another observational study by Kalantar Z. et al, 82 hemodialysis patients with serum ferritin >800ng/ml had significantly higher CRP level and worse malnutrition-inflammation score compared with patients with serum ferritin <800ng/ml [20]. Thus the raised serum ferritin levels in our study can be attributed to inflammation and oxidative stress in CKD. We also tested the hypothesis that high concentration of serum ferritin, a frequently used marker of iron stores in dialysis patients and an acute phase reactant, may be a marker of morbidity in CKD patients. In our study we found the rise in chronicity of disease as predicted by rising serum creatinine and blood urea levels corresponding to the rise in serum ferritin levels. This finding correlated with the study done by Hee-Taik et al wherein higher serum ferritin levels were found to be associated with higher risk of CKD in Korean men [24]. In another study by Yi-Chun et al, the patients of stage 3-5 CKD showed higher levels of ferritin which was associated with rapid renal progression [25]. This correlation of renal progression with raised ferritin can be explained by induction of macrophage accumulation during inflammation by ferritin which increases the formation of reactive oxygen species (ROS) [25]. Ferritin can thus be significantly associated with mortality and cardiovascular outcome in patients with renal failure. Therefore, in setting of uniform iron administration, high serum ferritin level could be a morbidity risk factor and recent increase in serum ferritin may increase risk of death in these patients. The role of ferritin in oxidative damage can serve as the basis of using antioxidants for reducing the risk of cardiovascular events in CKD patients [26].

To conclude, there was persistence of renal anemia in CKD patients with reduced MCV, MCH and MCHC along with low serum iron, normal TIBC, reduced TSAT but raised serum ferritin levels although they were supplemented with ESA therapy and iron. There was coexistence of functional and absolute iron deficiency in our study cases. Hence, other causes for anemia should always be considered in treating anemia in CKD patients along with regular monitoring of iron stores. We also found positive correlation between serum ferritin and serum creatinine levels which, could be used as a marker of morbidity in CKD patients.

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