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Evaluation of the Levels of Erythropoietin and Some Haehematological Parameters in Patients with History of Haemodialysis and Non- haemodialysis.

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Abstract
Background: Kidney diseases are associated with a variety of hematological changes. Anemia is the most common finding among these haematological indicators and its severity increased rise with disease chronicity.
Aim: To determine the association between erythropoietin and hematology parameters with different stages with chronic kidney failure and haemodialysis.
Subjects and methods: A total of 106 patients with chronic renal failure were included in the study and their age range was (20-69 years) and divided in 5groups according to disease staging. The study comprised 40 participants who had never had dialysis before. While there were 66 patients with a dialysis history, 21 had a dialysis history of less than 6 months and 21 had a dialysis history of more than 6 months (45).Erythropoietin were determined in serum of all subjects by using a commercial ELISA Micro wells kit (My Bio Source, USA). Hematology parameters (reticulocytes, red blood cells, haematocrit and hemoglobin) in whole blood of all subjects were determined by using an automated hematological analyzers XT2000i (Sysmex, Japan).
Results: The erythropoietin concentration, percentages of reticulocyte and red blood cells count were with significant differences ($P<0.05$)between groups.
Anemia present in all chronic kidney failure patients and increased deterioration directly associated to the stage of kidney failure.
Conclusion: Erythropoietin, reticulocyte, red blood cells, haematocrit and haemoglobin levels were reduced in patients with chronic kidney disease and reduction was associated with disease stages.
Keywords: Erythropoietin, Chronic kidney disease, RBC, Haematocrit, Haemoglobin.

Introduction

Kidney diseases are associated with a variety of hematological alterations [1]. Anemia is the most common finding among these haematological indicators and its severity increased rise with disease chronicity. Development of anaemia in chronic renal failure patients was attributed to many underlying aetiology, that include B12 or folate deficiency, iron deficiency, functional deficiency of iron due to reduced iron use for erythropoiesis, reduced synthesis of erythropoietin and reduced sensitization of erythroblasts to erythropoietin and the effect of drugs used for treatment. However in this patients the most common cause for anemia in chronic renal disease is reduced production of erythropoietin from the decreased form of functioning kidney tubular cells [2].

Erythropoietin (EPO) controls erythrocyte production. This action is interceded by specific receptors on the surface of the burst-forming unit-erythroid (BFU-E) and colony-forming unit erythroid (CFU-E) cells, where the EPO-receptor complex is responsible for progenitor cell development into mature erythrocyte. Moreover, EPO prevents automatic cell death of BFU-E and CFU-E[3]. Under standard conditions circulating EPO levels are typically little but in response to anemia or tissue hypoxia, these levels are rise several folds [4]. However in chronic kidney disease (CKD) patients as a result of decreased functional mass of kidneys this response is hampered with subsequent inadequate response of hemoglobin level [2]. Thus this study conducted to illustrate the differences in erythropoietin and some haematologic parameters in chronic kidney disease patients exposed to haemodialysis compared to those without history of dialysis.

Subjects and Methods

A total of 106 patients with chronic renal failure were included in the study and their age range was (20-69 years),40 patients without dialysis history and 66 patients with dialysis history. The study population was divided into 5 groups according to disease staging and duration from last haemodialysis performance. Group one included patients with stage III (12 patients; 8 male and 5 female); group 2 included patients with stage IV (13 patients; 8 male and 5 female). Group 3 included patients with stage V chronic renal failure (15 patients; 8 male and 7 female). Group 4 included patients with history of haemodialysis less than six months (21 patients; 16 male and 5 female). While group 5 included patients with dialysis history of more than 6 months of duration (45 patients; 28 male and 17 female).

Erythropoietin were determined in serum of all subjects by using a commercially ELISA Micro wells kit (from My Bio Source, USA). Hematological parameters (reticulocytes, red blood cells, haematocrit and hemoglobin) in whole blood of all subjects were determined by using an automated hematological analyzers XT2000i(from Sysmex, Japan).

Statistical analysis

Statistical analysis was carried out using SPSS (Version 20). The results presented as mean and standard deviation. Differences between groups were analyzed by using ANOVA, then if there are significant differences, they were analyzed by LSD test. The P value of < 0.05 was considered significant.

Results

The results indicated presence of anemia. EPO levels demonstrated a significant differences ($p<0.05$) between groups of stage III, stage IV, and stage V. In addition, a significant differences between patients with history of less and more than 6 months of dialysis, Table.1. While, none significant difference was demonstrated in mean values between those with stage IV and those with history of dialysis of less than 6 months duration, Fig.1.

The percentages of RET were with significant differences ($p<0.05$) among patients with stage III, IV, V, HD < 6 months, and HD > 6 months. While, no significant differences ($p>0.05$) in HD more than 6monthsgroupwhen compared with stage III and haemodialysis less than 6months, Fig.2.

RBCs count showed significant differences($p<0.05$) among groups III, IV, V, HD less than 6 months and HD more than 6 months period. While no significant differences ($p>0.05$)in HD more 6 months in stage V and HD more 6months, Fig.3.The percentage of HCT and Hb concentration showed significant differences ($p<0.05$) between stage III, stage IV, stage V, HD<6months and HD>6monthsduration, Table.1, Figs.4 and 5.

Table (1). Mean serum value of biomarkers in patients with chronic renal failure

Biomarker	Stage3 (N=12) M±SE	Stage4 (N=13) M±SE	Stage5 (N=15) M±SE	HD<6 months (N=21) M±SE	HD>6 months (N=45) M±SE
EPO pg/ml	337.01±16.30	276.32±8.78	196.17±8.35	138.00±16.42	182.36±19.20
RET %	1.24±0.02	1.00±0.09	1.09±0.05	1.39±0.04	1.31±0.07
RBC x 10^{12}/l	4.16±0.05	3.97±0.10	3.45±0.03	3.24±0.05	3.14±0.08
HCT %	33.16±0.43	31.03±0.24 b	27.92±0.29 c	25.89±0.39 d	27.33±0.65 c
Hb g/dl	10.88±0.14	9.78±0.10	9.06±0.08	8.36±0.13	9.02±0.22
N =Number, M =Mean, SE = Stander Error, EPO= Erythropoietin, RET = Reticulocyte, RBC= Red Blood Cells, HCT= Haematocrit and Hb Hemoglobin.					

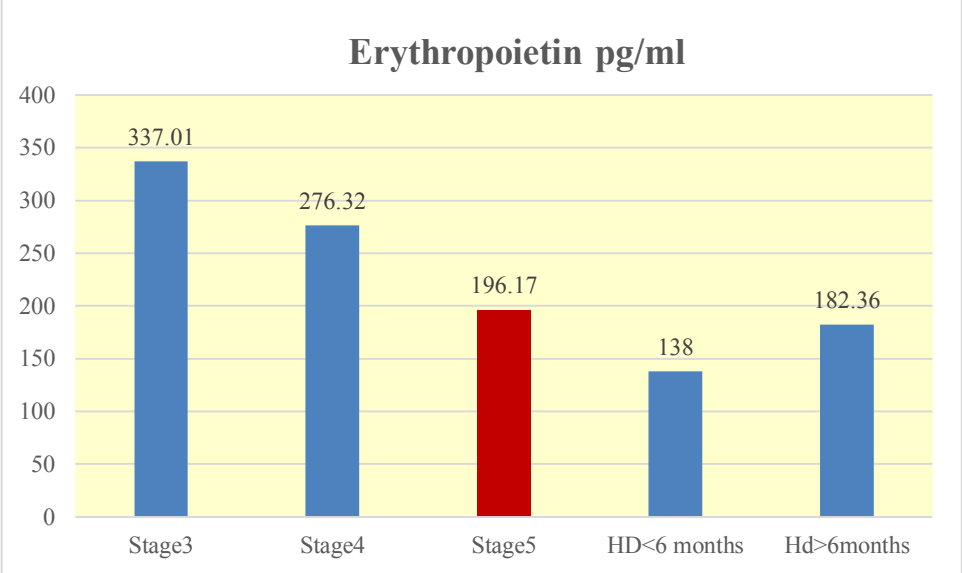

Figure (1) Erythropoietin levels in all groups.

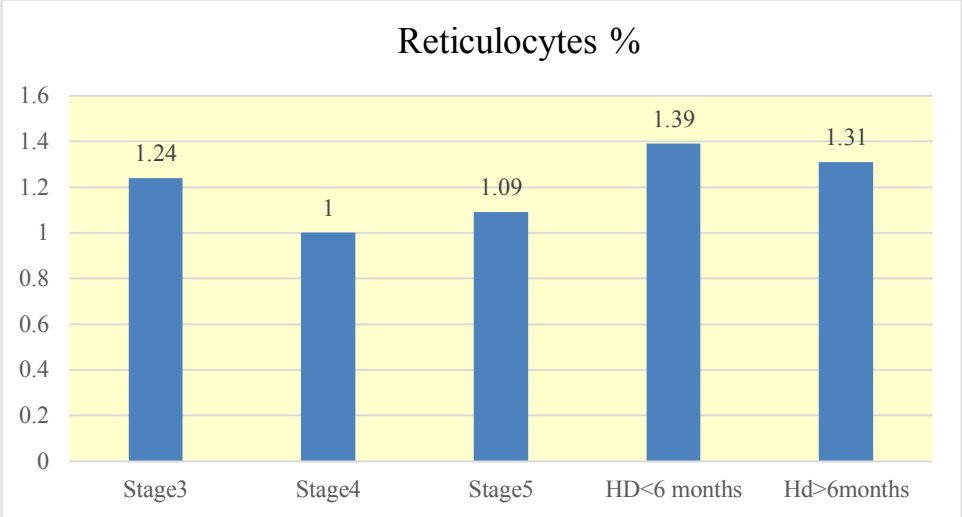

Figure (2). Reticulocytes levels in all groups.

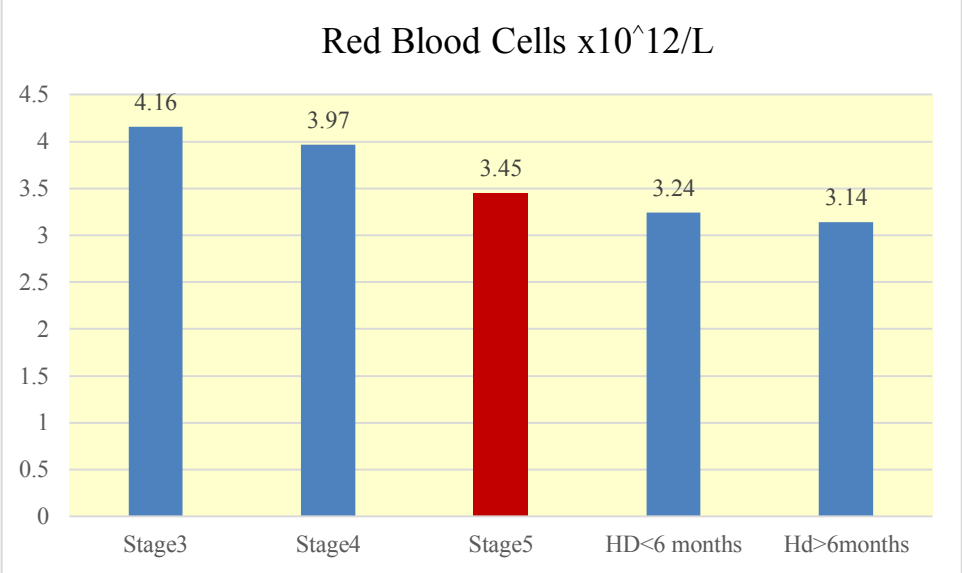

Figure (3). Red Blood Cells levels in all groups.

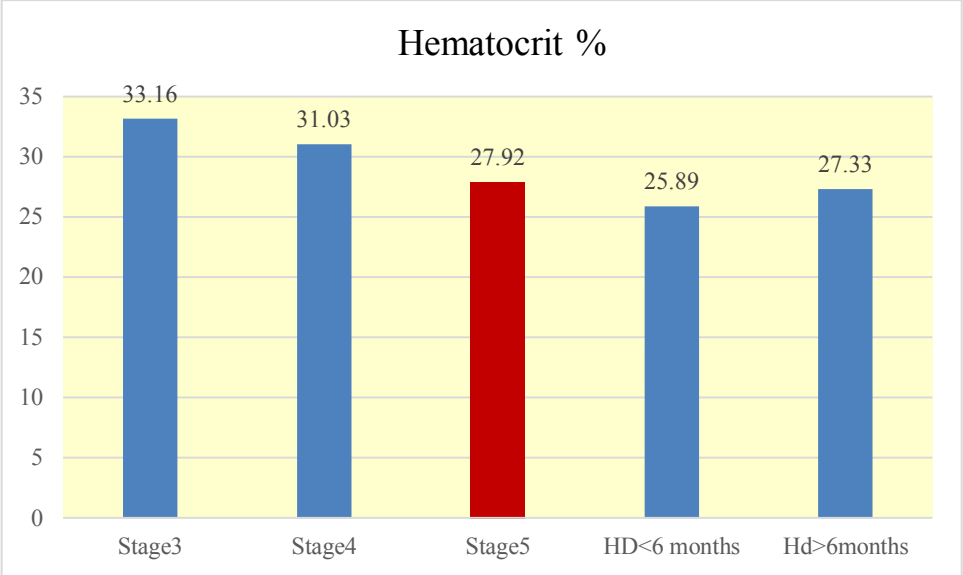

Figure (4). Hematocrit levels in all groups.

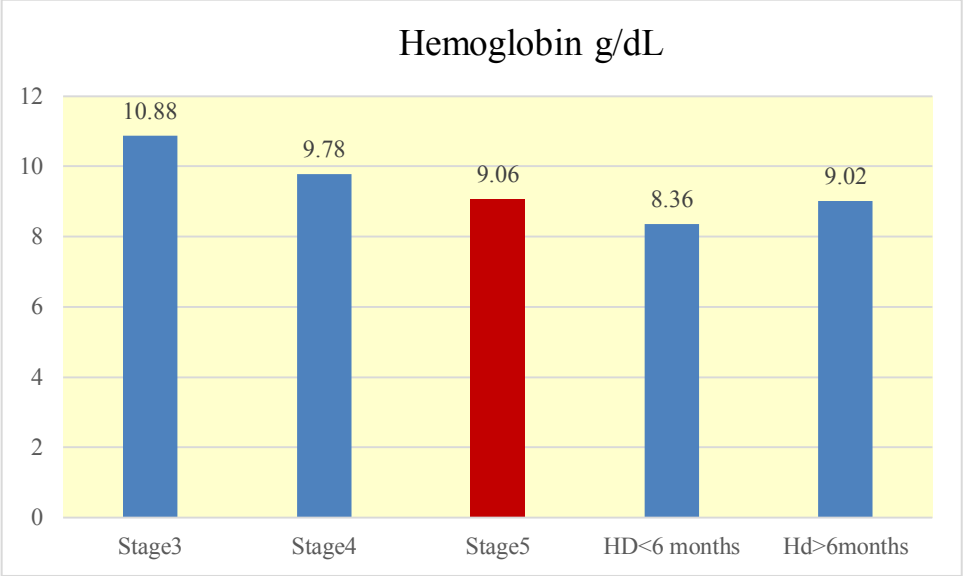

Figure (5). Hemoglobin levels in all groups.

Discussion

Kidney diseases are related with a variety of haemopoietic changes. Anemia linked to the degree of kidney damage and its most significant cause is failure of kidney EPO production. Additional factors include hemolysis, chronic blood loss, and bone marrow suppression via retained uremic causes[5].

Defective renal EPO production is the major cause of renal anemia[6]. RET usually increased in a rate of 2.5% in the response to EPO [7,8]. Sunita*et al*., reported in forty patients (35 males and 5 females) with chronic renal failure at the end stage of the disease and their age ranged between 16-70 years. Patients had anemia with hemoglobin values in the range of 3.3 to 11 gm %. Normocytic norm chromic anemia is the dominant anemia in most

patients [9]. Anemia of the chronic kidney failure is multifactorial, pathogenesis of this type of anemia has been attributed to decreased plasma erythropoietin due to renal damage, inhibitors of erythropoiesis in uremic plasma and decreased hemoglobin oxygen affinity[10]. Asif *et al.,* reported that hemoglobin is the most commonly affected among hematological parameters in patients with chronic renal failure. In individuals with chronic renal failure, erythropoietin plays an active role in boosting hemoglobin levels, and extra EPO (epokine) 4000 IU subcutaneous can be administered if the patient's Hb level falls below 11 g/dl [11].

Michael and Bertil [12] found that RBC survival is reduced in patients with uremia in proportion to the blood urea nitrogen level and improves significantly after extremely haemodialysis. Uremic plasma increases the expression of phosphatidyl serine on the surface of erythrocytes. This increase the recognition of injured erythrocytes by macrophage, leading to their subsequent damage and reduced survival [12]. Hayder *et al.,*[8] also reported that red blood cells, Hemoglobin, mean corpuscular hemoglobin, and packed cell volume, were significantly decreased in patients with CKD on haemodialysis [8].

In this study, EPO level significantly different between groups. EPO deficiency in chronic renal disease could be a functional response to a reduced GFR [13]. The increase in EPO levels may be results due to decrease of the functioning kidney in CKD as the kidney's ability to respond to lower oxygen delivery and EPO secretion was reduced [8]. The reasons for reduced EPO production in the kidneys are explained primarily by destruction of the EPO-producing fibroblasts of the kidney interstitial and the overall decrease in renal mass [13]. Generally the response to EPO is arise in the number of reticulocytes to more than 2.5 % [7].

The number of RBCs, HCT, and Hb in CKD and haemodialysis patients decreased as their disease progressed. RBC counts increased in stage III, but decreased in patients who had been on haemodialysis for more than 6 months.RBC survival is reduced in patients with uremia in proportion to the blood urea nitrogen level and, it improves significantly after intensive haemodialysis. Uremic toxin rises the expression of phosphatidyl-serine on the erythrocytes. This improves the recognition of injured erythrocytes via macrophage, leading to their successive damage and reduced survival [12]. Haemoglobin level and haematocrit normally deliver an accurate reflection of the level to which the circulating erythrocytes mass is decreased in chronic renal disease because of reduced erythropoietin secretion, increased damage of erythrocyte, leads to a decrease in erythrocytes count, which decreases the hemoglobin and haematocrit level. [14-16].

Conclusion: This study indicated that anemia was present in all forms of chronic renal failure, and that erythropoietin levels dropped in tandem with disease development and improved when patients continued to receive haemodialysis. Reticulocytes, on the other hand, were unaffected.

ETHICAL APPROVAL: Bilad AlRafidain University College (BARUC) Ethical Committee

CONSENT TO PARTICIPATE: Informed consent was taken from each subject before their enrolment in the study.

HUMAN AND ANIMAL RIGHTS: The study conducted in adherence with Helsinki Ethical standards.

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CONFLICT OF INTEREST: The authors declare no conflict of interest, financial or otherwise.

DATA AVAILABILITY: The data was available in Bilad AlRafidain University College (BARUC), Diyala, information set and available on request.

References

1. Suresh M, Mallikarjuna RN, Singh SM, HariBandi HK,Keerthi SG and Chandrasekhar M. Hematological Changes in Chronic Renal Failure. International Journal of Scientific and Research Publications, 2012;2(9): 1-4.
2. Naghmi A, Sada H and Khalid H. Hmatological Changes in Patients of Chronic Renal Disease and Their Response to Treatment with Erythropoietin.Int. J. Pathol, 2015; 13(1): 14-19.
3. Tong EM and Nissenson AR. Erythropoietin and anaemia. Semin. Nephrol, 2001;21, 190–203.
4. Ge. RL, Witkowski S and Zhang Y. Determinants of erythropoietin release in response to short-term hypobaric hypoxia. J. Appl Physiol. 2002; 92(6):2361-2367.
5. DoddsA and Nicholls M. Haematological aspects of renal disease. Pub Med – index for medline, 1983;11(4): 361-68.
6. Kuo CC, Lee TC, Chuang CH, Y and Chen J.B. Recombinant human erythropoietin independence in chronic hemodialysis patients: clinical features, iron homeostasis and erythropoiesis. Clin. Nephrology, 2005;63:92-97.
7. Bovy C, Tsobo C, Crapanzano L, Rorive G, Beguin Y and Albert A. Factors determining the percentage of hypochromic red blood cells in hemodialysis patients. Kidney Int, 1999;56: 1113-9
8. Hayder S H Zhian, Jamal M Aziz and Mohamad Salih. Jaff. Effect of Dialysis on Erythropoietin and some Hematological Parameters in Patients with Chronic Renal Failure Jaff, Zanco J. Med. Sci., 2009;13(2):14-21.
9. Sunita G. R, Arvind K. Ade and Pravin P. Shekokar. A Study of Haematological Changes in Chronic Renal Failure. Sch. J. App. Med. Sci.,2014;2(4A):1232-1234.
10. Mitchel TR and Pegrum CD. The oxygen affinity of hemoglobin in chronic renal failure. Brit J Haematol.1971;21: 263.
11. Asif N, Hasan S, Hassan K. Hematological Changes in Patients of Chronic Renal Disease and Their Response to Treatment with Erythropoietin. Int J Pathol, 2015;13(1): 14-19.
12. Mostafa F M S , Essam EL , Lamiaa A E M , Mahmoud H Z N. Assessment Of Erythropoietin Efficacy And Dosing In Hemodialysis Patients.European jour.of Molecular& Clin. Med, 2021;8(2):2461-2469.
13. Levy J, Morgan J and Brown E. Oxford handbook of Dialysis, 2nd ed. Oxford University Press,2004.
14. Miguel G. Uriol-Rivera, Aina Obrador-Mulet,SoniaJimenez-Mendoza, Antonio Corral-Baez, Leonor Perianez-Parraga, Angel Garcia-Alvarez, Francisco J. de la Prada. J Hematology. 2021;10(6):255-265.
15. Alsiadi WA, Mahmood MM, Hassan AS. Certain hematological denotations in chronic renal failure patients. Indian J Pub Health Res Develop, 2019;10(6):6-9