Determination of Trace Elements (Copper, Zinc and Iron) and some Biochemical Variables in Patients with Kidney Failure Using a Spectrophotometer

Huda Ahmed Abd[1], Abdul Majeed Khorsheed Ahmed[2]
[1]Department of Chemistry, College of Science, University of Kirkuk, Kirkuk, Iraq
[2]Department of Chemistry, College of Education for Pure Sciences, University of Kirkuk, Kirkuk, Iraq

***Abstract*—Background: A disorder known as chronic Kidney Disease (CKD) causes a significant loss of many kidney function units, nephrons, which negatively affects kidney functions and leads to their decline. It also results in a decrease in kidney mass, which in turn leads to hypertrophy of the remaining nephrons.**

Aims: The study's objective was to ascertain the kidney failure patients' trace element and biochemical parameter concentrations.

Methods and Material: The study included drawing blood samples from 40 healthy individuals who did not suffer from any chronic diseases, aged between 20-40 years. After drawing the blood was placed in anticoagulant glass tubes and left at temperature (25°C). Then were separated in a centrifuge. Blood serum was gathered using a precise pipette, and the samples were subsequently preserved at 20°C until needed. The spectrophotometer was used to estimate the concentrations of trace elements and some biochemical parameters.

Results: Research indicates that CKD patients have a critical trace element imbalance, and treatment centered around these elements is a promising direction for the years to come. Comprehending trace element balance is essential to extending the illness's course and improving the prognosis of individuals with chronic kidney disease.

Conclusions: The study revealed a decrease in trace element concentrations (zinc, iron) and an increase in copper in kidney failure patients, with no significant difference in male and female genders. Biochemical variables showed an increase in urea and creatinine concentrations, but no significant differences in sexes.

***Index Terms*— Chronic kidney disease, Copper, Iron, Spectrophotometer.**

I. INTRODUCTION

One significant organ that environmental pollutants target is the kidney. As well as being environmental pollutants, trace metals like Cd, Pb, and as have nephrotoxic effects1,2. These trace elements primarily enter the human body through polluted food and drinking water 3. Trace elements have an important influence on metabolism in the body and any alterations in the trace element levels have been found as either the cause for or the result of disorders like diabetes mellitus (DM)4. The two primary causes of chronic kidney disease (CKD), diabetes and hypertension, are widely treated; yet, the prevalence of CKD is still rising quickly globally5,6. End-stage renal disease, a risk for cardiovascular disease, and an early death are the results of chronic kidney disease 7-9. Consequently, the growing number of CKD patients is seen as a serious public health issue 10,11.
Decreased kidney function, damage to the kidneys and diminished performance that lasts longer than three months, and kidney damage and their structural damage were the criteria used to identify chronic kidney disease 12,13. Chronic kidney disease (CKD) is a serious public health problem with a global prevalence of 13.4% and a mortality rate of 1.2 million (approximately) per year. To understand the etiology of CKD, physical and chemical causative factors such as soil, water, food, heat stress, pesticides and environmental samples have been analyzed by different research groups, but they have failed to provide clues about the exact causative factors 14.
Nephrons are both the functional and structural components of the kidney. Nephrons affect blood plasma variations by secretion, reabsorption, filtration, and excretion. The kidneys regulate fluid volume, osmolality, electrolyte concentrations, and acid-base balance through various processes15,16. The kidneys perform additional functions in the balance of calcium and the manufacture of erythropoietin and renin, two hormones, in addition to the activity of nephrons. The kidneys' capacity to efficiently regulate the level of electrolytes and fluids and preserve homeostasis can be impacted by both acute damage and chronic kidney failure 17,18.

II. MATERIALS AND METHODS

This research was conducted in the city of Kirkuk during the period that began in August 2023 until the end of December 2023. Blood samples were drawn from 80 patients with chronic kidney failure after they were diagnosed by specialized doctors, and their ages ranged between 20-70 years from the Al Amal Dialysis Center at Kirkuk Teaching Hospital and the dialysis unit at hawija general hospital. The study also included drawing blood samples from 40 healthy individuals who did not suffer from any chronic diseases, aged between 20-40 years. After puncturing a vein and drawing 5 ml of blood, the blood was placed in anticoagulant glass tubes and left at room temperature (25°C). Then were separated in a centrifuge for ten minutes at 3000 rpm. Blood serum was gathered using a precise pipette, and the samples were subsequently preserved at 20°C until needed. Optical absorption spectroscopy was employed to

DOI: http://doi.org/10.24086/biohs2024/paper.1248

estimate the concentrations of trace elements such as copper, zinc, and iron. The research revealed that the concentration of zinc in the blood of individuals with kidney failure was lower compared to the healthy group. Additionally, the study demonstrated a heightened concentration of copper and a diminished concentration of iron in the blood of kidney failure patients when compared to the healthy group. Biochemical parameters, namely urea and creatinine, were also assessed using an optical absorption spectrometer. The results of the experiment showed that kidney failure patients had greater blood concentrations of creatinine and urea than people in the healthy population. The patient samples were then divided into 30 male and 50 female groups. The study found no significant alterations in the levels of copper, zinc, and iron among these gender groups. Regarding biochemical variables, there was no notable change in urea concentration between males and females, whereas a significant change in creatinine concentration was observed between the two sexes.

A. Statistical Analysis Used

Statistical analysis was performed using GraphPad Prism v8.0 (GraphPad Software, San Diego, Ca, USA), and mean standard deviation (SD) was used to express the results. The comparison of mean ±SD was performed using the ANOVA test, and statistical Significance was defined as P≤0.05 and the correlation between the parameters.

III. RESULTS

The results showed that the concentration of zinc in the blood serum of patients with kidney failure was reduced compared to healthy people, and the statistical significance was (P<0.0001) as shown in Table 1, and after dividing the patient samples into males and females, that there was no significant change in zinc concentration and the statistical function was (P<0.4762) as in Table 2. The findings demonstrated that kidney failure patients had lower blood serum concentrations of copper than healthy individuals, with a statistical significance of (P< 0.0085) as indicated in Table 1. Additionally, there was no significant change in copper concentration after dividing patient samples into male and female groups, with a statistical function of (P< 0.0085) as indicated in Table 2. According to Table (1), the results indicated that the blood serum concentration of iron in kidney failure patients was lower than that of healthy individuals, with a statistical significance of (P< 0.0085). Additionally, after separating patient samples into groups of men and women, Table (2) demonstrated that the iron concentration did not significantly vary, with a statistical function of (P<0.2046). The research revealed that the blood serum of kidney failure patients had higher concentrations of creatinine than that of healthy individuals. This difference was statistically significant (P< 0.0001), as Table 3 illustrates, while there is no difference in urea concentration between males and females. As shown in Table 4.

Table 1: Concentrations of trace elements in two groups (subjects with chronic kidney disease and healthy subjects) and their significance values

Elements	Mean± S.D Control (G1)	Mean ± S.D Patients (G2)	*p*-value
Zinc (µg/dl)	126.3 ± 14.88	102.9 ± 7.464	0.0001
Copper(mg/dl)	94.14 ± 14.54	111.7 ± 40.03	0.0085
Iron(mg/dl)	75.15 ± 19.91	16.98 ± 11.09	0.0001

Table 2: Concentration of trace elements in two groups of patients with Chronic kidney disease (males and females) and their significant values

Elements	Group	Mean± S.D patient	*p*-value
Zinc(µg/dl)	Males Females	103.7 ± 7.705 102.0 ± 6.786	0.4762
Copper(mg/dl)	Males Females	103.3 ± 35.75 119.7 ± 42.22	0.1724
Iron(mg/dl)	Males Females	13.93 ± 6.063 16.06 ± 5.654	0.2046

Table 3: Concentrations of biochemical variables in two groups (people suffering from Chronic kidney disease and healthy people) and their significant values

Parameters (mg/dl)	Mean± S.D Control (G1)	Mean ± S.D Patients (G2)	*p*-value
Urea	31.15 ± 6.948	116.3 ± 30.15	0.0001
Creatinine	0.8036±0.1997	7.558 ± 2.608	0.0001

Table 4: Concentration of biochemical variables in the two groups of people with chronic kidney disease (males and females) and their significant values

Parameters (mg/dl)	Group	Mean± S.D patient	*p*-value
Urea	Males Females	1114.9 ± 25.72 1129.8 ± 34.01	0.0605
Creatinine	Males Females	8.140 ± 2.490 6.981 ± 1.933	0.0487

IV. DISCUSSION

The findings show that there is a discernible drop in zinc concentration in the blood serum of kidney failure patients when compared to healthy individuals, which is consistent with the findings of the researchers 19,20. Additionally, the findings supported researchers by demonstrating that there is no variation in zinc content between males and females 22. It is a fact that 40% to 78% of individuals with chronic kidney disease have low levels of zinc in the blood 21. Three primary explanations can be given for this outcome: alterations in the body's reserves of this element; low digestive system absorption. a reduction in zinc intake due to the patients' dietary limitations, which prevent them from consuming foods high in zinc 22-26. The dialysate's nature, which permits substantial zinc excretion during the filtering process, is the fourth reason why dialysis patients have zinc insufficiency 27. According to certain other studies, there was a drop in the amount of zinc.

In dialysis patients, it was due to low levels of albumin in the blood of these patients 28, as the zinc present in the serum is combined with plasma proteins, especially with albumin, and proteinuria (Proteinurea) in patients with kidney failure leads to excessive excretion of zinc along with protein. Therefore, persistent urinary loss of zinc may eventually lead to low zinc levels in these patients. This occurs due to the conversion of zinc into red cells under acidic condition 29. Whereas functional iron insufficiency is characterized by sufficient iron stores but insufficient availability for integration into erythroid precursors, absolute iron shortage is characterized by severely

DOI: http://doi.org/10.24086/biohs2024/paper.1248

diminished or missing iron stores. increased hepcidin hormone levels contribute to this lack of functionality 30. Renal disorders can lead to a reduction in systemic iron levels due to increased urinary iron excretion and diminished hepcidin-mediated iron transport, resulting in a decrease in erythropoietic-mediated iron utilization through heightened urinary loss and decreased erythropoietin 31.

Concerning copper, the findings in Table (1) excessive copper levels in the body can lead to organ poisoning, various diseases, and, in rare cases, death. This agrees with researchers 32. Because elevated circulating copper levels have been associated with a rapid deterioration in kidney function and a lower glomerular filtration rate, the study noted that these factors may be contributory risk factors for chronic kidney disease (CKD) 33-35. Overconsumption of copper-containing foods can lead to copper buildup in the kidneys, which can result in nephrotoxicity, which is defined by oxidative stress-induced proximal tubule necrosis and cell damage, ultimately impairing kidney function 36. Copper and kidney disease interact in both directions; in patients with chronic kidney disease (CKD), changes in protein metabolism may be the cause of circulating copper level abnormalities37. Controlling copper levels in CKD patients is essential to avoid problems. Despite the fact that earlier observational research linked elevated copper levels to CKD 38, the causative relationship remains uncertain due to susceptibility to confounding and reverse causality in traditional observational epidemiological studies.

The urea is an inert and relatively non-toxic moleculeurea, however, is a direct and indirect uremic toxin, as demonstrated by numerous investigations. In vitro and in vivo studies have demonstrated that urea alters the phenotypic of smooth muscle cells and promotes the expression of pro-apoptotic family genes, although further research is needed to fully understand the direct toxicity mechanisms of urea. Additional research has demonstrated that urea indirectly damages proteins through protein carbamylation, interfering with their cellular and molecular functions and linked to the onset of chronic kidney disease 39. A byproduct of protein metabolism, urea is frequently utilized in clinical settings as a proxy for the severity of chronic kidney disease and the suitability of dialysis 40.

The most commonly used indicator of renal function, serum creatinine, has a complicated metabolic process. The interpretation of blood creatinine as a marker of renal function should be interpreted with caution because of the numerous physiological and technological difficulties surrounding its measurement, particularly in severe cases or among patients with liver disease or low muscle mass. Serum creatinine can be used to predict clinical outcomes and assess muscle mass in addition to renal function41. Table 5 shows there is a difference between creatinine concentration between males and females that the results obtained are agree with the results obtained by researchers 42.

CONCLUSION

The study found a decrease in the concentration of both trace elements (Zn, Fe), and an increase in the concentration of the trace element (Cu) in patients with kidney failure, and there is no significant difference in the concentration of trace elements in male and female genders of patients with kidney failure. As for biochemical variables, the study found an increase in the concentration of both urea and creatinine in patients with kidney failure. As for the difference in each sex, there is no difference in the concentration of urea in each sex, and there is a difference in the concentration of creatinine between the sexes.

REFERENCES

1. D. Wang et al., "Tubular and glomerular kidney effects in the Chinese general population with low environmental cadmium exposure," Chemosphere, vol. 147, pp. 3–8, 2016.
2. M. T. Hayat, M. Nauman, N. Nazir, S. Ali, and N. Bangash, "Environmental hazards of cadmium: past, present, and future," in Cadmium toxicity and tolerance in plants, Elsevier, 2019, pp. 163–183.
3. L. Järup and A. Åkesson, "Current status of cadmium as an environmental health problem," Toxicol. Appl. Pharmacol., vol. 238, no. 3, pp. 201–208, 2009.
4. A. S. Ahmed and P. H. Tahir, "Physiological effects of some trace elements on some blood parameters in diabetic patients and their correlation with HbA1c.," EurAsian J. Biosci., vol. 14, no. 2, 2020.
5. R. Saran et al., "US renal data system 2016 annual data report: epidemiology of kidney disease in the United States," Am. J. kidney Dis., vol. 69, no. 3, pp. A7–A8, 2017.
6. V. Jha and G. K. Modi, "Getting to know the enemy better—the global burden of chronic kidney disease," Kidney Int., vol. 94, no. 3, pp. 462–464, 2018.
7. A. S. Go, G. M. Chertow, D. Fan, C. E. McCulloch, and C. Hsu, "Chronic kidney disease and the risks of death, cardiovascular events, and hospitalization," N. Engl. J. Med., vol. 351, no. 13, pp. 1296–1305, 2004.
8. D.-C. Jin, "Analysis of mortality risk from Korean hemodialysis registry data 2017," Kidney Res. Clin. Pract., vol. 38, no. 2, p. 169, 2019.
9. K. M. Kim, H. J. Oh, H. Y. Choi, H. Lee, and D.-R. Ryu, "Impact of chronic kidney disease on mortality: a nationwide cohort study," Kidney Res. Clin. Pract., vol. 38, no. 3, p. 382, 2019.
10. A. H. Abdullah, "Evaluation and study the effect of asymmetric dimethylarginine in chronic renal failure patients treated by hemodialysis, "J. Kerbala Univ.S., vol. 9, no. 2, 2011.
11. Y. Xie et al., "Analysis of the Global Burden of Disease study highlights the global, regional, and national trends of chronic kidney disease epidemiology from 1990 to 2016," Kidney Int., vol. 94, no. 3, pp. 567–581, 2018.
12. Y. Kakitapalli, J. Ampolu, S. D. Madasu, and M. L. S. Sai Kumar, "Detailed review of chronic kidney disease," Kidney Dis., vol. 6, no. 2, pp. 85–91, 2020.
13. M. Wickramasinghe, D. M. Perera, and K. Kahandawaarachchi, "Dietary prediction for patients with Chronic Kidney Disease (CKD) by considering blood potassium level using machine learning algorithms," in 2017 IEEE Life Sciences Conference (LSC), IEEE, 2017, pp. 300–303.
14. P. V Parvati Sai Arun, C. Obula Reddy, and Y. Akhter, "Uddanam kidney nephropathy under the light of metagenomics perspective," SN Compr. Clin. Med., vol. 1, pp. 23–25, 2019.
15. H. F. Hassan and A. K. R. A. Barzinji, "Prevalence of ruminants gastro-intestinal parasites in Kirkuk province, Iraq," Kirkuk Univ. Journal-Scientific Stud., vol. 13, no. 3, pp. 96–108, 2018.
16. J. M. Ali, R. Hammed, and A. Raoof, "Evaluationof oxidative stress state in patients with Renal failure ," Kirkuk Univ Journal-Scientific Stud., vol. 12, no. 1, 2017.
17. A. P. S. Munro et al., "Safety and immunogenicity of seven COVID-19 vaccines as a third dose (booster) following two doses of ChAdOx1 nCov-19 or BNT162b2 in the UK (COV-BOOST): a blinded, multicentre, randomised, controlled, phase 2 trial," Lancet, vol. 398, no. 10318, pp. 2258–2276, 2021.
18. H. M. B. Bakr, A. Sevki, and S. H. Korsheed, "Evaluation of some Biochemical parameter on chronic renal failure before and after infection with SARS-COV-2," World Bull. Public Heal., vol. 22, pp. 96–101, 2023.

DOI: http://doi.org/10.24086/biohs2024/paper.1248

19. A. Elgenidy, M. A. Amin, A. K. Awad, F. Husain-Syed, and M. G. Aly, "Serum zinc levels in chronic kidney disease patients, hemodialysis patients, and healthy controls: systematic review and meta-analysis," J. Ren. Nutr., vol. 33, no. 1, pp. 103–115, 2023.
20. C. Gómez de Oña, E. Martínez-Morillo, E. Gago Gonzalez, P. Vidau Argüelles, C. Fernández Merayo, and F. V Alvarez Menendez, "Variation of trace element concentrations in patients undergoing hemodialysis in the north of Spain," Scand. J. Clin. Lab. Invest., vol. 76, no. 6, pp. 492–499, 2016.
21. M. Kubota, S. Matsuda, M. Matsuda, K. Yamamoto, and Y. Yoshii, "Association of Serum Zinc Level with severity of chronic kidney disease in diabetic patients: a cross-sectional study," BMC Nephrol., vol. 23, no. 1, p. 407, 2022.
22. S. Lee et al., "Trace Metals' abnormalities in hemodialysis patients: relationship with medications," Artif. Organs, vol. 24, no. 11, pp. 841–844, 2000.
23. C.-Y. Yang et al., "Essential trace element status and clinical outcomes in long-term dialysis patients: a two-year prospective observational cohort study," Clin. Nutr., vol. 31, no. 5, pp. 630–636, 2012.
24. D. Tousoulis et al., "Asymmetric dimethylarginine: clinical significance and novel therapeutic approaches," Curr. Med. Chem., vol. 22, no. 24, pp. 2871–2901, 2015.
25. P. Raimundo, P. Ravasco, V. Proença, and M. Camilo, "Does nutrition play a role in the quality of life of patients under chronic haemodialysis?," Nutr. Hosp., vol. 21, no. 2, pp. 139–144, 2006.
26. S. Hosokawa and O. Yoshida, "Effect of recombinant human erythropoietin (rHuEPO) on protein, zinc (Zn), nickel (Ni) and manganese (Mn) in patients undergoing chronic haemodialysis," Int. Urol. Nephrol., vol. 27, pp. 207–214, 1995.
27. C. C. Pelletier et al., "White adipose tissue overproduces the lipid-mobilizing factor zinc α2-glycoprotein in chronic kidney disease," Kidney Int., vol. 83, no. 5, pp. 878–886, 2013.
28. J. Borawski, K. Pawlak, B. Naumnik, and M. Myśliwiec, "Relations between oxidative stress, hepatocyte growth factor, and liver disease in hemodialysis patients," Ren. Fail., vol. 24, no. 6, pp. 825–837, 2002.
29. A. P. Dsouza, R. Reddy, A. Yadav, and M. Mala, "Effect of Hemodialysis on Trace Elements in Renal Failure Patients," Indian J. Med. Biochem., vol. 23, no. 2, p. 234, 2019.
30. A. Gafter-Gvili, A. Schechter, and B. Rozen-Zvi, "Iron deficiency anemia in chronic kidney disease," Acta Haematol., vol. 142, no. 1, pp. 44–50, 2019.
31. R. P. L. van Swelm, J. F. M. Wetzels, and D. W. Swinkels, "The multifaceted role of iron in renal health and disease," Nat. Rev. Nephrol., vol. 16, no. 2, pp. 77–98, 2020.
32. G. Gembillo et al., "Potential role of copper in diabetes and diabetic kidney disease," Metabolites, vol. 13, no. 1, p. 17, 2022.
33. H.-J. Tsai et al., "Associations among heavy metals and proteinuria and chronic kidney disease," Diagnostics, vol. 11, no. 2, p. 282, 2021.
34. A. A. Iyanda, J. Anetor, and F. A. A. Adeniyi, "Altered copper level and renal dysfunction in Nigerian women using skin-whitening agents," Biol. Trace Elem. Res., vol. 143, pp. 1264–1270, 2011.
35. A. A. Iyanda, J. Anetor, and F. A. A. Adeniyi, "Altered copper level and renal dysfunction in Nigerian women using skin-whitening agents," Biol. Trace Elem. Res., vol. 143, pp. 1264–1270, 2011.
36. A. Ay, N. Alkanli, and S. Ustundag, "Investigation of the Relationship Between IL-18 (− 607 C/A), IL-18 (− 137 G/C), and MMP-2 (− 1306 C/T) Gene Variations and Serum Copper and Zinc Levels in Patients Diagnosed with Chronic Renal Failure," Biol. Trace Elem. Res., vol. 200, no. 5, pp. 2040–2052, 2022.
37. A. Ay, N. Alkanli, and S. Ustundag, "Investigation of the Relationship Between IL-18 (− 607 C/A), IL-18 (− 137 G/C), and MMP-2 (− 1306 C/T) Gene Variations and Serum Copper and Zinc Levels in Patients Diagnosed with Chronic Renal Failure," Biol. Trace Elem. Res., vol. 200, no. 5, pp. 2040–2052, 2022.
38. J. H. Sondheimer et al., "Elevated plasma copper in chronic renal failure," Am. J. Clin. Nutr., vol. 47, no. 5, pp. 896–899, 1988.
39. S. M. Laville et al., "Urea levels and cardiovascular disease in patients with chronic kidney disease," Nephrol. Dial. Transplant., vol. 38, no. 1, pp. 184–192, 2023.
40. Z. A. Massy, C. Pietrement, and F. Touré, "Reconsidering the lack of urea toxicity in dialysis patients," in Seminars in dialysis, Wiley Online Library, 2016, pp. 333–337.
41. K. Kashani, M. H. Rosner, and M. Ostermann, "Creatinine: From physiology to clinical application," Eur. J. Intern. Med., vol. 72, pp. 9–14, 2020.
42. X. Zhang, A. D. Rule, C. E. McCulloch, J. C. Lieske, E. Ku, and C. Hsu, "Tubular secretion of creatinine and kidney function: an observational study," BMC Nephrol., vol. 21, pp. 1–9, 2020.

DOI: http://doi.org/10.24086/biohs2024/paper.1248