Biochemical Role of Blood Electrolytes in Old Iraqi Patients with COVID-19

Rasha Zuhair Jasim[1] , Samira Mouyad Yaseen[2] and Hanan Adnan Shaker[3]

[1,3] **Department of chemistry, College of education for pure science (Ibn Al Haitham) , University of Baghdad , Baghdad , Iraq**

[2]**Department of Biology /College of Education For Pure Science Ibn Al-Haitham , University of Baghdad / Baghdad / Iraq.**

**Corresponding Author rasha.z.j@ihcoedu.uobaghdad.edu.iq*

ABSTRACT:

Background: Age progression is regarded as a critical risk factor in morbidity and mortality because of a weakened immune system. Although various studies have dealt with electrolyte imbalance in COVID-19 patients, the outcomes of these studies were partially understood. Objective: The current study aims to determine some biochemical parameters in old Iraqi COVID-19 patients and highlight the outcomes according to the aging role in the development of COVID-19 by suggesting new mechanisms. Materials and methods: forty COVID-19 patients were enrolled in the current study and divided into two groups: Gm includes (20) men, and Gf includes (20) women. The parameters (Na+, K+, Cl-, LDH, and Hb) were determined in sera of patients and control groups, G1: healthy men and G2: healthy women. Results: The results reported that the levels of sodium, chloride, and (hemoglobin for men) were highly significantly decreased. In contrast, potassium level was highly significantly increased in Gm and Gf compared to G1 and G2, respectively, and hemoglobin level in women was decreased in Gf compared with G2. LDH activity did not significantly increase in Gm compared with G1, while it increased dramatically in Gf compared with G2. The difference between Gm and Gf was non-significant for sodium, potassium, chloride, and hemoglobin, but it was highly significant for lactate dehydrogenase. Conclusions: The present study proposed definite mechanisms to elucidate hyponatremia, hyperkalemia, and hypochloremia in old COVID-19 patients by highlighting both COVID-19 complications and risk factors linked to age progression. At the same time, it revealed an interesting biochemical relationship between higher activity of LDH, hyponatremia, and hypochloremia in the same patients .

KEYWORDS : *Aging , COVID-19 , Hypochloremia , Hyponatremia , Hyperkalemia.*

INTRODUCTION

The new 2019 novel corona virus (2019-nCoV) called COVID-19 infection invades the immune system so it is featured by a number of symptoms including fever, difficult breathing , headache in addition to different complications leading to uncontrolled cases leading to death (1) COVID-19 is the most recent global pandemic(2). This virus is due to severe acute respiratory syndrome 2 (SARS-CoV-2) or the new coronavirus (2019-novel) virus that is named by the World Health Organization as coronavirus disease 19 or COVID-19 (3). The most common mechanism of COVID-19 infection revealed that SARS-CoV-2 invades human tissues via binding its spikes to angiotensin I converting enzyme 2 (ACE_2) on the cell membrane , ACE_2 plays a key role in controlling electrolyte balance (4). ACE_2 is expressed in both kidneys and the lungs but much more in the kidneys ,with about four times the percent in the renal proximal tubule cells as seen in the type II pneumocytes (5). Electrolyte imbalance is a prevalent complication of COVID-19 (6). Globally , electrolyte imbalance is defined as abnormalities in the levels of body fluids and electrolytes due to different mechanisms responsible for electrolyte imbalance , such as excessive or inadequate administration , impaired absorption and distribution , hormonal changes , renal defects and altered excretion via gastrointestinal (4). COVID-19 infection invades the immune system so it features by a number of symptoms including fever, difficultly breathing and headache , in addition to different complications leading to uncontrolled cases leading to death (5). Sodium, potassium and chloride are three of the most common electrolytes included in the different metabolic and homeostatic functions involving hormonal functions , neurotransmission and also enzymatic / biochemical reactions (3). Blood electrolytes have a key influence in controlling blood clotting , acid - base balance , body fluid , and muscles contractions ; this is why serum electrolyte levels are clinically used as biochemical indicators to assess the clinical status (6). Noticeably , minimal changes in blood electrolytes levels and body fluids in COVID-19 patients may predict the disease severity (7). Metabolic acidosis is also regarded as a main complication of COVID-19 , which is caused by the generation of lactic acid which is secondary to hypoxia. Higher levels of lactic acid in complicated cases of COVID-19 may cause unexpected death (4). The current study aims to determine some biochemical parameters in old Iraqi COVID-19 patients and highlight the outcomes according to the aging role in the development of COVID-19 by suggesting new mechanisms

MATERIALS AND METHODS

Study design and Patients' selection

Forty Iraqi COVID-19 subjects participated in the current study ; they were recently diagnosed by polymerase chain reaction (PCR) as COVID-19 patients after selecting from several private clinical institutes laboratories and while sample collections and laboratory tests were performed in the International Center for Research and Development .This study included patients of the two genders (males and females). Remarkably, the practical part of the present study was conducted between January and the end of April 2021. Accordingly, the patients were classified into two groups: The first patients group (males) G_m composed of (20) COVID-19 men with ages ranging between (53-75) years and the second patients group (females) G_f composed of (20) COVID-19 females with ages ranging between (57-61) years. Patients were compared to healthy subjects as control groups: G_1 composed of (20) healthy men and G_2 composed of (20) healthy women. Healthy subjects were approximately in the same ages of patients.

Exclusion Criteria

Blood specimens were taken from all COVID-19 patients without vaccines, who were attended in private clinics and laboratories in Baghdad after getting ethical approval. We divided the samples into two genders. Subjects with other inflammatory or chronic diseases and subjects who were under treatment with drugs that may minimize the accuracy of biochemical measurements were excluded from the study ; this is why the number of subjects participated in the present study was limited because most patients in this range of ages (old subjects) have chronic diseases.

Blood samples collection

Ten : milliliters from venous blood were sampled from each person participated in the current study. Remarkably , blood sera amount obtained from some clinical cases may be lower than the normal value ; this is why an excessive amount of blood were sampled. blood sera were separated from other blood components by the centrifuge for 5 minutes at 4000 rounds per minute, and the resulting serum from each specimen was divided into four small parts (each put within an Eppendorf) and kept under freezing at (-20°C) until getting started with clinical laboratory analysis of blood electrolytes and LDH. Regarding Hb, it was measured directly in red blood cells. Anyway , this amount of blood is too enough if any laboratory analysis needed to be repeated to get more accuracy.

Biochemical analysis

A Mindray BS 800 analyzer (8). measured blood electrolytes (Na, K, and Cl). This novel device used an indirect ion electrode method to determine these electrolytes. A buffer solution was used to dilute samples before they were disposed of in the electrode flow cell. The ion-selective electrode method has been shown to be the new generation of modern electronics and potentiometric devices. This device combines innovation, high performance, and resistance principles. Regarding lactate dehydrogenase (LDH) , it was measured by oxidation method caused by LDH catalyzes lactate oxidation to pyruvate and reduction of NAD^+ to NADH. The rate of NAD^+ reduction was detected as an increase in the absorption at 340 nm , the reaction rate was directly correlated to LDH activity in serum. Regarding Hb , it was measured by the cyanmethemoglobin method , the principle of this test is to convert Hb to HiCN by the addition of potassium cyanide and ferricyanide with absorbtion recorded at 540 nm in a photoelectric colorimeter versus a standard solution. Biochemical laboratory tests were conducted in the international center for research and development (ICRD) in Baghdad.

Statistical analysis

All data were quantified and documented by the Student T test according to probability (p) to compare the difference between data resulting from each group. When p value ≤ 0.05 the difference is known as significant , when p value ≤ 0.001 , the difference is known as highly significant , when p value > 0.05 the difference is known as non- significant. The statistical analysis was performed using (Microsoft excel 2010)

RESULTS

Table 1. Sodium level in sera of COVID-19 patients and control groups.

Na^+ mmol / L			
Group	Mean ± S.D	Group	Mean ± S.D
G_1	145.80±5.70	G_2	146.2±4.97
G_m	136.52±3.69	G_f	134.97±3.47
p G_m/G_1 : < 0.0001 (H.S)		p G_f/G_2 : < 0.0001 (H.S)	
p G_f/G_m : 0.267 (N.S)			

G_1: healthy men (control group) .

G_2: healthy women (control group).

G_m: COVID-19 patients (males).

G_f: COVID-19 patients (females).

S.D: standard deviation.

Table 2. Potassium level in sera of COVID-19 patients and control groups.

K^+ level (mg/dL)			
Group	Mean ± S.D	Group	Mean ± S.D
G_1	3.85±0.28	G_2	3.91±0.31
G_m	4.47±0.46	G_f	4.63±0.28
p G_m/G_1 : < 0.0001 (H.S)		p G_f/G_2 : < 0.0001 (H.S)	
p G_f/G_m : 0.26 (N.S)			

Table 3. Chloride level in sera of COVID-19 patients and control groups.

Cl^- (mg/dL)			
Group	Mean ± S.D	Group	Mean ± S.D
G_1	109.33±3.81	G_2	109.8±3.83
G_m	99.22±4.77	G_f	100.81±4.50

p G_m/G_1 : < 0.0001 (H.S)	p G_f/G_2 : < 0.0001 (H.S)
p G_f/G_m : 0.375(N.S)	

Table 4. The LDH activity in sera of COVID-19 patients and control groups

LDH (U/L)			
Group	Mean ± S.D	Group	Mean ± S.D
G_1	289.6±39.37	G_2	290.6±42.32
G_m	350.6±114.89	G_f	656.3±291.79
p G_m/G_1 : 0.071(N.S)		p G_f/G_2 : < 0.0001 (H.S)	
p G_f/G_m : 0.001(H.S)			

Table 5. Hemoglobin level in sera of COVID-19 patients and control groups.

Hb level (g/dL)			
Group	Mean ± S.D	Group	Mean ± S.D
G_1	13.93±1.23	G_2	12.07±0.49
G_m	11.39±0.55	G_f	10.29±0.89
p G_m/G_1 : < 0.0001 (H.S)		p G_f/G_2 : 0.004 (S)	
p G_f/G_m : 0.541 1(N.S)			

The results of the present study revealed that the levels of Na, Cl, and (Hb for men) significantly decreased. In contrast, K levels increased dramatically in sera of Gm and Gf compared with G1 and G2, respectively, and Hb levels in women significantly decreased in Gf compared to G2. LDH activity did not increase dramatically in Gm compared to G1 and highly considerably increased in Gf compared with G2. Regarding the variation between the two genders related to Na, K, Cl, and Hb, the difference between Gm and Gf was non-significant. At the same time, LDH was highly significant in Gm and Gf.

DISCUSSION

Definitely, older adults are susceptible to fluids overload and dehydration because of lower ability to maintain homeostatic control of body fluids and electrolytes balance caused by immune system depend on proper function of cells, organs and signaling pathways. Hence , the lyphatic system is a part of the immune system which keeps body fluid levels in balance (9). Dehydration and electrolytes dys regulation is accompanied by sodium conserve due to lower activity of antidiuretic hormone (10). On the other hand, electrolytes disorder is regarded as a clinical impact in COVID-19 patients. Generally, association between hypo / hypernatremia (Na decrease / increase) or hypo / hyperkalemia (K decrease / increase) reflects a definite relationship between electrolyte disorder and clinical severity of COVID-19 patients (11). Hyponatremia is defined as Na level is less than 136 mmol/L (12) Accordingly , results of table 1 revealed that G_f were hyponatremic while G_m were on the borderline (pre-hyponatremic). Hence, it has been reported that age is one of the important risk factors which detects the severity and of COVID-19 cases (13). Also, it has been confirmed that hyponatremic is one of the most common electrolyte disorders which occurs in hospitalized patients with a higher risk of mortality. Indeed , the treatment given for COVID-19 patients in hospital such as chloroquinone and hydroxy chloroquinone can lead to this electrolyte imbalance (7). This is why G_m and G_f subjects were pre-hyponatremic and mild-hyponatremic respectively. In this regard, the risk of dehydration plays a key role . Under lower quality of life , older subjects are prone to be dehydrated more than other younger. Hence , the loss of lean mass causes a reduction in the total body water content , the result is a reduced capacity to handle sodium loads. Interestingly, the mechanism involves age -related reduction in glomerular filtration rate which lead to passive re-absorption of fluids with a higher risk of water overload and subsequently to hyponatremia (9). Remarkably , a recent study has reported that hyponatremia is a major complication among hospitalized COVID-19 patients. In particular , older age subjects with severe symptoms compared with young subjects , this may reflected by their weak immunity (4). Recently , It has been noticed that hyponatremia in old subjects may be reflected by firstly the lower physical activity and secondly the unbalanced dietaty style , these factors lead to electrolytes distrabances (6).

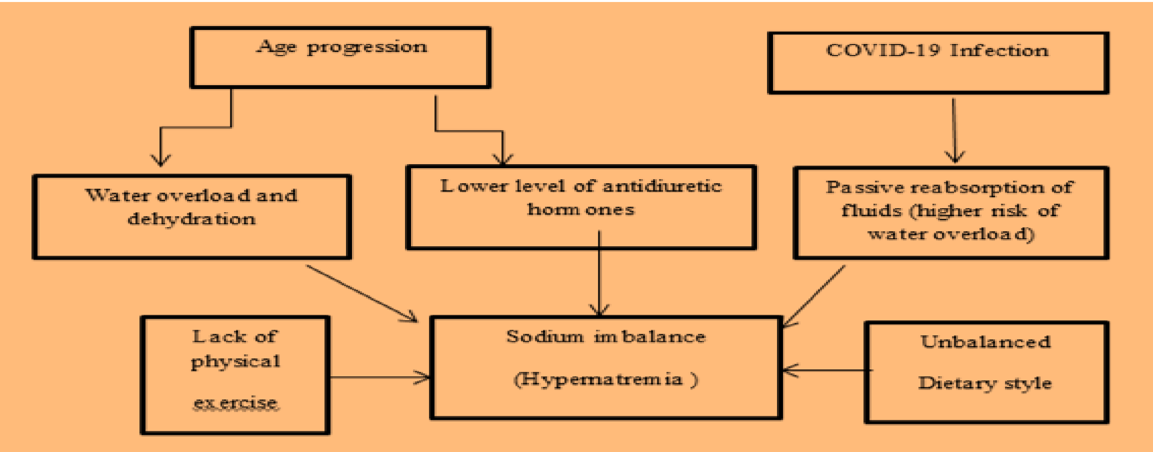

Figure 1. The suggested mechanism for hyponatremia in old Iraqi patients with COVID-19 infection.

Regarding K levels , it has been recently revealed that hyperkalemia (higher levels of potassium) may be associated with clinical scores among COVID-19 patients. At this point , hypertension is the most common complication of COVID-19 patients which subsequently lead to a distrupt in potassium balance and increase in sera K levels (14). Similarly, it has been reported that K levels was significantly higher in the severe COVID-19 patients compared with the non severe cases and the outcomes revealed a positive relationship between the COVID-19 infection and higher levels of potassium in the blood stream (15). The prevalence of hyperkalemia in COVID-19 patients may be reflected firstly by pre-existing clinical conditions that are regarded as risk factors for the development of severe COVID-19 cases , (14) and secondly by the acute kidney injury as a complication of COVID-19 infection because it has been reported that serum urea was increased in severe COVID-19 patients. At this point , it has been reported that hyperkalemia in COVID-19 patients is influenced by impaired glomerular filtration rate. Moreover, COVID-19 patients with higher levels of serum creatinine are characterized by higher levels of serum potassium (15). Hyperkalemia leads to hyperpolarization of the cell membrane and promotes a diminished heart function (10). According to age progression , renal tubular dysfunction and lower levels of aldosterone both and renin when the patient becomes older , play an essential role in the development of hyperkalemia in old patients (16). Interestingly , it has been revealed that hyponatremia and hyperkalemia are apparent features of aging (10,12) and in the same context it has been suggested that hyponatremia and hyperkalemia are remarkable complications of COVID-19 infection (13) the combination of these outcomes agree with the results of table 1 and table 2 . One of these results has shown that hyponatremia is positively correlated with mortality and severity of the disease (13). The present study submits reasonable mechanisms for hyponatremia and hyperkalemia in old COVID-19 patients on the basis of both the disease complications and the aging manifestations

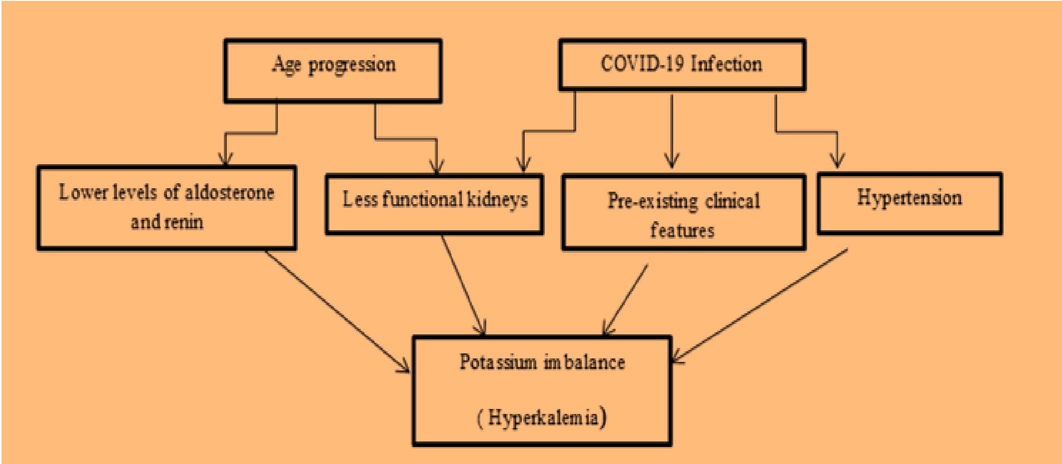

Figure 2. The suggested mechanism for hyperkalemia in old Iraqi patients with COVID-19.

. Regarding Table 3 , various studies agreed with the present outcomes , these studies proposed that hypochloremia (lower levels of chloride electrolyte) is a remarkably common electrolytes disorders in COVID-19 patients (7, 17-19). When chloride balance is disrupted , it can trigger the acute nephritic injury (7, 18). On the other hand, metabolic acidosis in COVID-19 patients due to lactic acid higher production is reflected by higher activity of lactate dehydrogenase which responsible for conversion of pyruvate to lactate, (20) as reported in table 4. At this point , metabolic acidosis can strongly cause hypochloremia because of the loss of too small amount of sodium bicarbonate from the body (7, 20), this is why chloride level was decreased in COVID-19 patients , table 3.

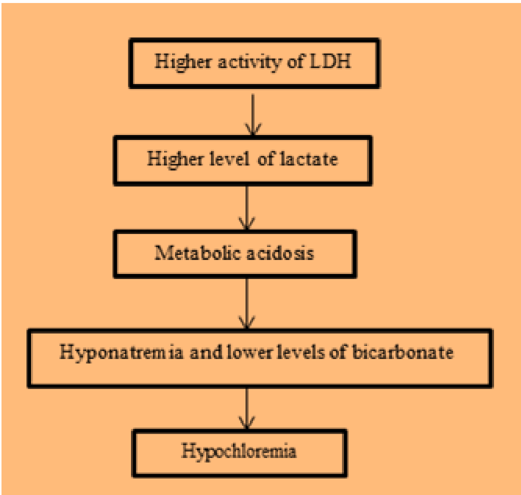

Figure 3. The suggested mechanism for hypochloremia in old Iraqi patients with COVID-19 infection which has revealed an interested relationship between higher activity of LDH , hyponatremia and hypochloremia.

Regarding LDH , it has been reported that severe infection causes tissue damage mediated by the cytokines and subsequently the release of LDH . Remarkably, isoenzymes 3 of lactate dehydrogenase is found in lung tissues and has higher activity in the circulation of COVID-19 infection (21). Hence , severe infection can promote macrophage-mediated angiogenesis and result in a high level of lactate in acidic extracellular pH (22). This is why lactate dehydrogenase activity was increased in COVID-19 patients compared with healthy subjects, Table 4 . In the term of age , it has been indicated a positive relationship between lactate dehydrogenase activity and aging (23). Nevertheless, subjects of the control groups in Table 4 have values slightly higher than the borderline. In comparison, COVID-19 patients have higher values than control subjects because they are both old and infected. The normal value of LDH activity is ranged between (140-280) U/L (24). According to table (1), table (2), and table (3), the difference between Gm and Gf was nonsignificant, revealing that hyponatremia, hyperkalemia, and hypochloremia are not influenced by gender. Conversely, the results of Table 4 revealed that LDH activity was significantly increased in Gf compared with Gm, indicating that LDH activity for those patients is influenced by gender. Hence, the role of menopause must be highlighted because all females enrolled are menopausal; LDH activity in G2 is on the borderline because the patients are healthy despite being menopausal, but LDH activity in Gf is very high because the patients are both menopausal and infected by COVID-19.Similarly, it has been shown a rise in blood lactate levels due to alkalosis in menopausal women (25). The Hb level in the blood is influenced by different factors, including sex, human age, nutrition, and diet type (26) because lack of consumption of a diet containing iron results in anemia. Some chronic diseases, in addition to blood loss via menstruation, lead to anemia. During menstruation, females lose a lot amount of blood, and consequently, Hb levels become low (27). Since Hb level is affected by gender, the reference range for females is (11.5-15.2) mg/dL and is (13-17) mg/d/L for males (28). At this point, the results of Table 5 revealed that Gm subjects have anemia while Gf subjects were approximately on the borderline. Hb is a tetrameric protein found exclusively in red blood cells and containing iron; it functions as a transporter for oxygen (by binding with it) from the lungs to the body tissues and also binds with carbon dioxide to return it from the body tissues to the lungs (29). Remarkably, the decreased number of red blood cells (RBC) or hemoglobin levels in human blood below the normal value due to abnormalities in RBC formation or bleeding (even the menstrual cycle) or iron absorption disorders are the major causes of anemia (26). In terms of COVID-19 infection, It has been revealed that Hb levels declined in COVID-19 patients, and the remarkable decrease in Hb levels reflects an obvious clinical progression (30). Interestingly, the mechanism involves the destruction erythrocytes which resulted in anemia. Indeed, the free heme resulting from hemolysis in the blood stream of COVID-19 patients who undergo higher levels of cytokines contributes to endothelial damage and to the remoduling of pulmonary vessels. On the other hand, dysfunctional hemoglobin may influence the oxygen transporting capacity of erythrocytes which subsequently lead to hypoxia and releasing free radicals at the inflammatory sites (31). Remarkably , the difference in hemoglobin level between the two genders is regarded to be caused by a direct stimulatory factor of androgen in males in bone marrow coupled with erythropoietin production in the kidney , and the inhibitory effect estrogen on the bone marrow in women, this action of hormones promotes hemoglobin level in males compared with females (32). Recently , it has been reported that anemia is a major public health issue that affects women (33). Interestingly, a previous study has confirmed that iron deficiency is coupled with the impairment of cell mediated immunity (34). Despite this the difference between G_m and G_f in table (5) was non-significant. This may reflected by dysfunctional androgen and estrogen due to old ages (35). Evidently, the data related to the control groups (G_1 and G_2) in Tables 1 to 5 have confirmed that despite the healthy old subjects are rather stable but they are clinically sensitive and susceptible to any infection when compared with patients. It has been recently highlighted the age role as a risk factor of COVID-19 by confirming that the disease incidence will be more common in men older than 60 years , elderly people are at higher risk of infected with COVID-19 because of a weakened immune system, malnutrition, ongoing illness, elevated ACE-2 expression and organ failure (36). Indeed , the health community in all the world is still research to investigate more conclusions about virus infection and a global support is required to overcome any recently pandemic in faster response (37), so additional clinical management is still required using a larger complete data set containing higher number of COVID-19 cases (38). Also, vaccination is an effective technique and a gold way to hold back the infection and control its breakdown (5).

CONCLUSIONS

The age progression plays a vital role in the incidence of COVID-19 because of a weakened immune system. The present study submits an evident mechanism regarding hyponatremia as a remarkable clinical feature for old Iraqi COVID-19 patients by highlighting the role of water overload resulting from both age progression and COVID-19 infection and also the lower levels of anti-diuretic hormones due to the aging process. The present study also submits a reasonable mechanism regarding hyperkalemia as a remarkable clinical feature for old Iraqi COVID-19 patients by taking together the complications related to both COVID-19 infection including acute kidney injury resulting in low functioning kidneys, pre-existing clinical features as risk factors, hypertension and the risk factors linked with the aging (low functioning kidneys and decreased levels of aldosterone and renin). Moreover, the present study submits a definite mechanism for hypochloremia in old Iraqi patients with COVID-19 infection on the basis of an exciting relationship between higher activity of LDH, hyponatremia, and hypochloremia. The current study confirmed the previous clinical studies regarding the higher activity of LDH and lower levels of Hb (anemia). Finally, it emphasized that hyponatremia, hyperkalemia, hypochloremia, and anemia are not influenced by gender, while LDH activity is influenced by gender in old Iraqi patients with COVID-19 infection.

AKNOWLEDGEMENTS

The authors are grateful for the Bacteriologist Suad Ahmed who helped to find patients cases and also express all thanks and gratitude to Dr. Ali A. Mahdi for help in conducting the laboratory part of this research.

ETHICAL APPROVAL

The study protocol was reviewed and approved by the University of Baghdad and The International Center of Research and Development. All clinical trials were conducted according to the Helsinki Ethical Principles.

AUTHORS CONTRIBUTION

Rasha ZJ proposed the topic of this research , designed the study. Rasha ZJ , Samira and Hanan worked together to collect blood samples. Rasha ZJ and Hanan conducted the laboratory tests , Samira MY made the statistical Analysis , Rasha ZJ discussed the results and wrote the manuscript. Rasha ZJ and Samira worked together to revise the manuscript draft. All authors read and approved the final manuscript.

CONFLICT OF INTEREST

The authors confirm that there are no conflicts of interest.

REFERENCES

[1] Jasim RZ . [2023] : Biochemical action of vaccines in Iraqi patients with COVID-19 infection. Baghdad Sci J. 20(4 special issue), pp.1469-1479.

[2] Polatoğlu I, Oncu-Oner T, Dalman I, Ozdogan S. [2023] : COVID-19 in early 2023: Structure, replication mechanism, variants of SARS-CoV-2, diagnostic tests, and vaccine & drug development studies. MedComm . 8(4), e228.

[3] Hadi JM, Hassan SM , Saeed MM, Hussein BK, Ali BM, Muhamad LE, Abdullah AJ, Ali NN, Rahman HA, Sofihussein HQ, Abdul Aziz JM. [2022] : Estimation of Serum Calcium on the Severity and Mortality in COVID-19 Infections in Sulaymaniyah City, Kurdistan Region of Iraq: A Cross-Sectional Study. Clin Pract. 12(6),1001-1008.

[4] Beyerstedt S , Casaro E , Rangel E. [2021]: COVID-19 : angiotensin – converting enzymes 2 (ACE2) expression and tissue susceptibility to SARS-CoV-2 infection. Eur J Clin Microbiol Infect Dis. 40(5), 905-919.

[5] Noori M, Nejadghaderi SA, Sullman MJM, Carson-Chahhoud K, Ardalan M, Kolahi AA, Safiri S. [2012]: A Review on the Possible Pathophysiology of Potassium Abnormalities in COVID-19. Iran J Kidney Dis. 15(6), pp. 397-407.

[6] Timerga A, Kelta E, Kenenisa C, Zawdie B, Habte A, Haile K. [2020] :Serum electrolytes disorder and its associated factors among adults admitted with metabolic syndrome in Jimma Medical Center, South West Ethiopia: Facility based crossectional study. PLoS One. 15(11),e0241486.

[7] Electrolyte Disturbances in COVID-19 and Their Complications. Biomed Res Int. [2021]: doi: 10.1155/2021/6667047.

[8] LAB BOOK. [2019]: Mindary Medical. BS-800M Clinical Chemistry Analyzer. Clinical Chemistry . Shenzhen Mindary Bio-Medical Electronics Co., Ltd. https://healthcare-in-erope.com

[9] Rodriguez ; S. A randomised trial assess fluid and electrolyte balance responses following ingestion of different beverages in young and older men .[2023] Eur. J. Appl. Physiol. doi.org/10.1007ls00421-023-05241-0.

[10] Draz H, Hussein R , . Kamel M , Elmonem A,. Ibrahim B. [2019] :Study of the prevalence of sodium and potassium disturbances in the elderly.*ZUMJ.*, 25(6),825-834.

[11] Song HJJMD, Chia AZQ, Tan BKJ, Teo CB, Lim V, Chua HR, Samuel M, Kee A. [2023] Electrolyte imbalances as poor prognostic markers in COVID-19: a systemic review and meta-analysis. J Endocrinol Invest. 46(2), pp. 235-259.

[12] Lee JJ, Kilonzo K, Nistico A, Yeates K. [2014]: Management of hyponatremia. CMAJ. 186(8), E281-6..

[13] Yasari F, Akbarian M, Abedini A, Vasheghani M. [2022] : The role of electrolyte imbalances in predicting the severity of COVID-19 in the hospitalized patients: a cross-sectional study. Sci Rep. 12(1), pp.14732.

[14] Amin A, Moon R, Agiro A, Rosenthal N, Brown H, Legg R, Pottorf W. [2022] : In-hospital mortality, length of stay, and hospitalization cost of COVID-19 patients with and without hyperkalemia. Am J Med Sci. 364(4), pp 444-453.

[15] Zhang J, Huang X, Tao Z. [2022] : Correlation of clinical characteristics between patients with seasonal influenza and patients infected by the wild type or delta variant of SARS-CoV-2. Front Public Health. 18(10), pp. 981233.

[16] Bianchi S, Aucella F, De Nicola L, Genovesi S, Paoletti E, Regolisti G. [2019]]: Management of hyperkalemia in patients with kidney disease: a position paper endorsed by the Italian Society of Nephrology. J Nephrol. 32(4) pp. 499-516.

[17] Murugan C, Ramamoorthy S, Kuppuswamy G, Murugan RK, Sivalingam Y, Sundaramurthy A. [2021] COVID-19: A review of newly formed viral clades, pathophysiology, therapeutic strategies and current vaccination tasks. Int J Biol Macromol. 193(Pt B), pp.1165-1200.

[18] Sharmin Moutushi S, Akter T, Haq MA, Ahmad R, Sinha S, Adnan N, Haque M. [2023] : Electrolyte Imbalance Among Bangladeshi Patients With COVID-19. Cureus. 15(2), e35352.

[19] Pourfridoni M, Abbasnia SM, Shafaei F, Razaviyan J, Heidari-Soureshjani R. [2021]: Fluid and Electrolyte Disturbances in COVID-19 and Their Complications. Biomed Res Int. 14(2021), pp.6667047.

[20] Quade BN, Parker MD, Occhipinti R. The therapeutic importance of acid-base balance. [2020]: Biochem Pharmacol. 183(1), pp. 114278.

[21] Teshale AB, Tesema GA, Worku MG, Yeshaw Y, Tessema ZT. [2020]: Anemia and its associated factors among women of reproductive age in eastern Africa: A multilevel mixed-effects generalized linear model. PLoS One. 15(9), pp. e0238957.

[22] Jouda J, Abdul Kareem Jabbar E, Salih Abdulhadi F, Atiyah Kamil Y. [2022] : Assessment of some Physiological Biomarkers in COVID-19 Patients in Thi-Qar, Iraq. Arch Razi Inst. 77(3), pp. 1097-1104.

[23] Hunt LC, Demontis F. [2022] :Age-Related Increase in Lactate Dehydrogenase Activity in Skeletal Muscle Reduces Life Span in Drosophila. J Gerontol A Biol Sci Med Sci. 77(2), pp. 259-267.

[24] Frank AC, Raue R, Fuhrmann DC, Sirait-Fischer E, Reuse C, Weigert A, Lütjohann D, Hiller K, Syed SN, Brüne B.[2021] : Lactate dehydrogenase B regulates macrophage metabolism in the tumor microenvironment. 11(15), pp.7570-7588.

[25] Sochocka M, Karska J, Pszczołowska M, Ochnik M, Fułek M, Fułek K, Kurpas D, Chojdak-Łukasiewicz J, Rosner-Tenerowicz A, Leszek J. [2023] : Cognitive Decline in Early and Premature Menopause. Int J Mol Sci 24(7), pp.6566.

[26] Salive ME, Cornoni-Huntley J, Guralnik JM, Phillips CL, Wallace RB, Ostfeld AM, Cohen HJ. [1992] : Anemia and hemoglobin levels in older persons: relationship with age, gender, and health status. J Am Geriatr Soc. 40(5), pp. 489-96.

[27] Abbaspour N, Hurrell R, Kelishadi R. [2014] : Review on iron and its importance for human health. J Res Med Sci.19(2), pp. 164-74.

[28] Larson LM, Braat S, Hasan MI, Mwangi MN, Estepa F, Hossain SJ, Clucas D, Biggs BA, Phiri KS, Hamadani JD, Pasricha SR. [2021] : Preanalytic and analytic factors affecting the measurement of haemoglobin concentration: impact on global estimates of anaemia prevalence. BMJ Glob Health. 6(7), e005756.

[29] Marengo-Rowe AJ. [2006] :Structure-function relations of human hemoglobins. Proc (Bayl Univ Med Cent). 19(3), pp.239-45.

[30] Yuan X, Huang W, Ye B, Chen C, Huang R, Wu F, Wei Q, Zhang W, Hu J. [2020] : Changes of hematological and immunological parameters in COVID-19 patients. Int J Hematol. 112(4), pp. 553-559.

[31] Lechuga GC, Souza-Silva F, Sacramento CQ, Trugilho MRO, Valente RH, Napoleão-Pêgo P, Dias SSG, Fintelman-Rodrigues N, Temerozo JR, Carels N, Alves CR, Pereira MCS, Provance DW Jr, Souza TML, De-Simone SG. [2021] : SARS-CoV-2 Proteins Bind to Hemoglobin and Its Metabolites. Int J Mol Sci. 22(16), pp. 9035.

[32] An Y, Jang J, Lee S, Moon S, Park SK. [2019] : Sex-specific Associations Between Serum Hemoglobin Levels and the Risk of Cause-specific Death in Korea Using the National Health Insurance Service-National Health Screening Cohort (NHIS HEALS). J Prev Med Public Health. 52(6), pp. 393-404.

[33] Eshale AB, Tesema GA, Worku MG, Yeshaw Y, Tessema ZT. [2020] :Anemia and its associated factors among women of reproductive age in eastern Africa: A multilevel mixed-effects generalized linear model. PLoS One. 15(9), e0238957.

[34] Jasim RZ . [2017]: Positive and negative aspects of copaxone (Glutiramer acetate) action on TC , TG , HbA1c and iron levels in the sera of Iraqi women with multiple sclerosis. Baghdad Sci. J. 14(1), pp.157-166.

[35] Horstman AM, Dillon EL, Urban RJ, Sheffield-Moore M. [2012] : The role of androgens and estrogens on healthy aging and longevity. J Gerontol A Biol Sci Med Sci. 67(11), pp. 1140-52.

[36] Farshbafnadi M, Kamali Zonouzi S, Sabahi M, Dolatshahi M, Aarabi MH. [2021] : Aging & COVID-19 susceptibility, disease severity, and clinical outcomes: The role of entangled risk factors. Exp Gerontol. 15(154) , pp.111507.

[37] Shirbhate E, Pandey J, Patel VK, Kamal M, Jawaid T, Gorain B, Kesharwani P, Rajak H. [2021] : Understanding the role of ACE-2 receptor in pathogenesis of COVID-19 disease: a potential approach for therapeutic intervention. Pharmacol Rep. 73(6), pp.1539-1550.

[38] Alam S, Bhuiyan FR, Emon TH, Hasan M. [2021]: Prospects of nutritional interventions in the care of COVID-19 patients. Heliyon. 7(2), e06285.