Reports of Biochemistry & Molecular Biology
Vol.13, No.2, Jul 2024

Original article

www.RBMB.net

Assessment of Oxidative Stress Parameters in Iraqi Male Patients with Covid-19; A Case Control Study

Zainab Nazar Hasan Anber*[1], Basil Oied Saleh[2], Riyadh Hassan Majed[3]

Abstract

Background: SARS-CoV-2 infection can cause significant alterations in our lives. Oxidative stress (OS) has been proposed to play a major role in COVID-19 pathogenesis, and the determination of OS biomarkers provides insight into disease severity**.**

Methods: The study was conducted during the second wave of the pandemic in 2020. Fifty blood samples were collected from patients admitted to one of the COVID-19 isolation centers in Baghdad, Iraq. The samples were subdivided into 25 patients admitted to the intensive care unit (ICU) and 25 non-ICU patients, compared to 25 healthy controls. All participants were aged 35-52 years**.**

Results: The study showed that the mean (±SD) serum total oxidant status (TOS) and malondialdehyde (MDA) levels were significantly increased ($p< 0.001$) in the ICU group compared to the control and non-ICU groups. Conversely, the levels of serum total antioxidant capacity (TAC) and serum antioxidative enzymes superoxide dismutase (SOD), glutathione peroxidase (GPX), catalase, and glutathione (GSH) were significantly decreased ($p< 0.001$) in the ICU group compared to both the control and non-ICU groups. Serum zinc levels were significantly decreased ($p< 0.001$) in both ICU and non-ICU groups compared to the control group, while serum selenium (Se), copper (Cu), and vitamins C and E were significantly decreased ($p< 0.001$) in the ICU group compared to both the control and non-ICU groups**.**

***Conclusion*:** The presence of OS biomarkers in the sera of COVID-19 patients offers a potential new approach for the treatment of this disease.

Keywords: Antioxidants, COVID-19, Oxidative stress, Oxidants.

Introduction

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the main cause of COVID-19, appears as a critical health issue around the world, correlated with inflammation, cytokine production, cell death, and other pathophysiological processes, that were associated with oxidative stress (1). It was realized that an antioxidant absence with over manufacturing of reactive oxygen species (ROS) are critical for the viral proliferative diseases (2). Inflammation is recognized to be widely connected to OS (3). The OS is connected to the evolution of various diseases comprising cardiovascular, neurodegenerative, pulmonary diseases and cancer (4). Alongside inflammation, other pathophysiological mechanisms correlated with excess OS had been supported to illustrate COVID-19 pathogenesis (5). These biomarkers can be differentiated into oxidants such as MDA, and antioxidants such as enzymes like GSH, GPx, SOD, and catalase; free elements like Zn, Cu, and selenium; and vitamins, such as vitamin C and vitamin E, which function as antioxidants in lipid oxidative injuries (6, 7). Free radical reactions damage can be inhibited or delayed by antioxidants via neutralizing radicals through

1: Department of Pharmacy, Baghdad College of Medical Sciences, Baghdad, Iraq.
2: Department of Biochemistry, College of Medicine, University of Baghdad, Iraq.
3: Medico-legal Directorate, Ministry of Health, Baghdad, Iraq.
***Corresponding author: Zainab Nazar Hasan Anber; Tel: +96 47823287850; E-mail: zainab.nazar@bcms.edu.iq.**
Received: 29 Feb, 2024; Accepted: 5 Oct, 2024

[DOI: 10.61186/rbmb.13.2.167]

scavenging by metal- chelating, decomposition of peroxide, inhibiting of chain reactions and stimulation of antioxidant enzymes. Oxidative stress has a tremendous role in the pathophysiology of many diseases.

A great deal of interest has been focused on the role of OS in the pathogenesis of several human diseases. Oxidative stress can be manifested by increasing the levels of free radicals and/or decreasing antioxidant levels, especially in respiratory diseases such as COVID-19 (8). The objective of the current study was to estimate the OS markers levels in persons intended to have COVID-19 infection.

Materials and Methods

This case-control study was conducted at one of the COVID-19 isolation centers in Baghdad, Iraq. It was carried out during the second pandemic wave in March 2020. The study included 50 patients diagnosed with COVID-19, who were subdivided into 25 ICU patients and 25 non-ICU patients, compared to 25 healthy subjects. The age range for all patients and control groups was 35-52 years. Only patients with symptoms were included, while those with clinical manifestations of any other apparent diseases and those taking antioxidant supplements were excluded.

Venous blood samples were drawn into EDTA and citrate-containing tubes and centrifuged at 3000× g within 10 minutes. Serum and plasma samples were then kept at -80 °C until testing. A malondialdehyde colorimetric assay kit (Elabscience®, United States) was used for the spectrophotometric measurement of serum MDA concentration. Serum TOS was measured using the Erel method (9), (Rel Assay®, Diagnostics kits, Turkey), while TAC of plasma was determined using the Pouvoir AntiOxydant Total (PAOT®) method (Institut Européen des Antioxydants, Nancy, France) (10). Serum SOD and catalase were assayed using ELISA kits from Elabscience® (14780 Memorial Drive Suite 216, Houston, Texas), while erythrocyte GPx activity was determined using commercial RANSEL kits (Randox Laboratories, Crumlin, Northern Ireland, UK). Erythrocyte-reduced GSH levels were evaluated according to the Beutler et al. method (11).

Serum zinc and copper levels were determined using a flame atomic absorption spectrophotometer (AA-646 Shimadzu, Japan) (12).

Serum selenium levels were evaluated using a flameless atomic absorption spectrophotometer (Perkin-Elmer Model 503). Serum levels of Zn, Se, and Cu were calculated after implementing absorbances on an appropriate calibration curve for each element from standards.

Serum vitamin C and E levels were measured colorimetrically in accordance with the methods of Roe and Kuether (13) and Nield and Pearson, respectively (14).

Statistical Analysis

Statistical calculation of data was carried out using Statistical Package for the Social Sciences (SPSS), version 22. One-way analysis of variance (ANOVA) was used to test the significance of differences in means for all parameters, and the least significant difference (LSD) test was applied to evaluate significant differences between two groups. The correlation coefficient (r) was used to determine the relationship between two variables. A p-value < 0.05 was defined as significant.

Results

The demographic data showed that there was a non- significant difference in the (mean ±SD) values of the age and BMI between control, non- ICU and ICU groups (Table 1).

Table 1. Demographic characteristics of Covid-19 (ICU and non-ICU patients) and control groups.

Characteristic	Control (N=25)	Non-ICU (N=25)	ICU (N=25)	p-value
Age (years) ^{NS}	53.36±11.14	52.88 ± 8.1	54.6 ± 7.51	0.787 ^{NS}
BMI (kg/m^2) ^{NS}	26.99± 0.88	26.92±1.02	27.32±0.66	0.231 ^{NS}

All values were expressed as Mean (±SD). BMI: body mass index; ANOVA test revealed a non- significant difference among groups (NS)

[DOI: 10.61186/rbmb.13.2.167]

Also, all the Mean (±SD) values of oxidative stress markers and antioxidants between control, Non- ICU and ICU are shown in Table 2.

Table 2. Plasma levels of oxidative stress markers and antioxidants among healthy control, non-ICU and ICU Covid-19 patients.

Parameter	Control (N=25)	Non-ICU (N=25)	ICU (N=25)	p- value		
				Control & non-ICU	Control & ICU	Non- ICU& ICU
MDA	3.26±0.39	4.44±0.33	5.96±0.38	< 0.001**	< 0.001**	< 0.001**
TOS	2.54± 0.69	5.85±0.80	12.56±1.32	< 0.001**	< 0.001**	< 0.001**
TAC	118.96±7.39	83.68±6.72	58.52±7.92	< 0.001**	< 0.001**	< 0.001**
SOD	28.0±1.58	25.24±2.06	22.51±1.51	< 0.001**	< 0.001**	< 0.001**
Gpx	569.48±56.57	379.76±99.62	295.04±38.69	< 0.001**	< 0.001**	< 0.001**
Catalase	14.02±1.02	13.26±1.19	6.8±0.88	= 0.013 *	< 0.001**	< 0.001**
GSH	3.73±0.38	3.45±0.45	1.61±0.5	= 0.03 *	< 0.001**	< 0.001**
Zinc	88.48±7.13	75.00±4.83	70.08±5.76	< 0.001**	< 0.001**	= 0.005*
Se	80.60±5.85	76.4±6.29	69.68±3.86	= 0.008 *	< 0.001**	< 0.001**
Cu	135.04±2.92	131.88±3.41	127.76±4.01	=0.002 *	< 0.001**	< 0.001**
Vitamin C	0.87±0.15	0.60±0.71	0.42±0.085	< 0.001**	< 0.001**	< 0.001**
Vitamin E	0.83±0.10	0.59±0.78	0.4±0.06	< 0.001**	< 0.001**	< 0.001**

Data are expressed as mean (±SD). ** highly significant: P< 0.001, * significant: p< 0.05.

Also, the present study showed that there was a significant negative correlation in the ICU group between SOD and zinc level: r = -0.434, $p < 0.05$. There was also a significant negative correlation between GSH and Cu: r = -0.415, $p < 0.05$; MDA and TAC: r = -0.410, $p < 0.05$. In contrast, there was a significant positive correlation between catalase and GPX: r = 0.495, $p < 0.05$, and between MDA and both zinc and Se:r = 0.539, r = 0.522, $p < 0.05$, respectively. There was also a significant positive correlation between TOS and Cu: r = 0.570, $p < 0.05$. Additionally, there was a highly significant positive correlation between vitamin C and vitamin E: r = 0.710, $p < 0.001$. Considering the non-ICU group, there was a highly significant positive correlation between catalase and GPX levels: r = 0.738, $p < 0.001$. Furthermore, there was a significant positive correlation between MDA and TOS: r = 0.471, $p < 0.05$.

Discussion

There was no significant difference in both age and BMI among the studied groups (ICU, non-ICU, and control groups). In the current study, patients and control groups were similar concerning age and BMI. This similarity is helpful to inhibit any variation in age and BMI of the study. The increased levels of MDA in COVID-19 patients in comparison to the control group stated in the current research suggest the subsequent formation of MDA as byproducts due to the destruction of lipids by an increased production of free radicals.

The results of the current study also show that the serum level of TOS, as an OS biomarker, was higher in the ICU and non-ICU groups in comparison to the control group. Generally, an appropriate nutritional status, which is affected by drugs, dietary factors, and pollutants, is considered an important factor that supports the immune system during the COVID-19 crisis (15,16). In the present study, increasing TOS levels significantly impact the increase in oxidative stress status, which could be treated with antioxidant supplements and physical exercise (17). SARS-CoV-2 can generate ROS and deplete the antioxidant potential, which could greatly increase the severity of the disease (18).

[DOI: 10.61186/rbmb.13.2.167]

According to serum TAC levels, it was declined through increasing the disease severity. This will increase the rate of mortality in these patients. As TAC was used as a biochemical parameter to determine the antioxidant state in patients with viral infection (19). The present work also considers the levels of plasma SOD, catalase, erythrocytes GPx, erythrocyte reduced GSH activities in COVID-19 patients and controls. The levels of plasma SOD, GPx, catalase and erythrocytes GSH were noticed to be decreased in COVID-19 patients in comparison to control (20). In a comparable study performed by Dworzanski et al., a decline in the activity of SOD and GPx was observed (21). However, in contradiction to Strycharz-Dudziak study who reported higher levels of catalase in patients in comparison to control. This study also explained the formation of superoxide, H2O2 and OH free radicals via the Fenton reaction. As mentioned before, GSH/GPX and CAT considered as active antioxidants in these reactions (22). In a study reported by Qin et al.; it was observed that an infection with SARS-COV-2 could produce an activation of phagocytes to produce more ROS with a subsequent decrease in antioxidant levels (23). Current studies performed by Strycharz-Dudziak and Derouiche proposed that infection with SARS-COV-2, as with other RNA viruses, could produce an OS status together with the weakening in the antioxidant system (24). And that the conversion of free radicals from a highly reactive to a lower reactive state will demand more antioxidants to perform this process (25, 26). Dincer et al. reported all the reactions of GSH as an active antioxidant (27).

The levels of Zn, Se and Cu were significantly decrease in both the ICU and the non-ICU group in comparison to control group. In oxidative stress, Zn binds proteins to protect them from oxidation (28). SARS-CoV-2 can generate ROS and deplete the antioxidant potential, which could greatly increase the severity of the disease. The levels of Zn, Se, and Cu were significantly decreased in both the ICU and non-ICU groups in comparison to the control group. In oxidative stress, Zn binds to proteins to protect them from oxidation (28). Previously, it was proposed that zinc, selenium and copper play a significant role in both the innate and adaptive immune systems. The decreasing plasma zinc level in patients with Covid-19 in comparison to healthy control was similar to the results of recent studies conducted in Japan (29). Zinc was documented to play a role as antiviral, antioxidant and immunomodulatory agent during COVID-19 infection. Kardos et al. noted that zinc was an important antioxidant that could play a major role in ZnCu-SOD performance (30). Barazzoni et al. also observed a decrease in the plasma levels of zinc in COVID-19 patients and stated that the aggravation of this disease was accompanied by a decrease in the number and function of lymphocytes and the improvement of this state would interfere with the synthesis of proteins and replication of the virus (31). Similarly, the decrease in the plasma level of selenium observed in this study was also reported by Zhang et al. , who stated that the infection with RNA viruses will produce an insufficient amount of seleno-proteins, resulting in antioxidants that recall the infection more malicious nature (32).

Kardos et al. noted that selenium deficiency increases mutations in the viral genome, transforming the virus into a more virulent (30). Current studies also suggest that there is a correlation between selenium level and COVID-19 consequences. In China, it has been reported that people with low selenium level are at high risk for death than people with

[DOI: 10.61186/rbmb.13.2.167]

a higher level (33). Serum Cu level has also been observed in this study in comparison to other studies. It has been recorded that Cu is essential for the activity of antioxidant enzymes such as Cu Zn-SOD in COVID-19 patients. (34). Kardos et al. noted that Cu decrease is associated with an increase in OS and an increase frequency of infections with weakening of the human immune system (30). Zabetakis et al. stated that COVID-19 infection was associated with a rise in inflammatory cytokines accompanied by Cu deficiency (35). The effect of COVID 19 on plasma levels of vitamins C and E are also considered by the current study. Vitamins C and E were significantly decreased in COVID 19 patients in comparison to healthy control. These results are identical to those of Chiscano-Camón et al., who recorded a considerable decrease of vitamin C in COVID-19 patients (36). These results are also supported by Chris et al., who investigated the scavenging effect of antioxidants in COVID-19 infection (37). Shakoor et al. proposed methods for improving the level of these antioxidant vitamins in these patients (38). The decreased serum level of the antioxidant vitamins is accompanied by a great rise of ROS produced by phagocytes in COVID-19 patients (39). An increase in ROS will activate the redox transcription factors and thus lead to the release of more pro-inflammatory factors (40). Vitamins C and E were determined as great scavengers for these ROS (41). The negative correlation observed in this study between Zn and SOD is in contrast to that reported by Maradi R et al., who observed a significant correlation between them reporting the strong effect of Zn on SOD (42). Along with increasing of reactive oxygen species (ROS) and its peroxides that are represented as the second messengers, activate redox sensitive transcription factors and stimulate the release of different proinflammatory factors. Oxidative stress markers play a significant role in the pathogenesis of COVID-19. Thus, determination of the level of these markers could play a role in the management and treatment of COVID-19 infection.

Acknowledgments

We thank all staffs and technicians at Baghdad College of Medical Sciences to their valuable support throughout the research project.

Financial support

Authors declare that they had no financial support.

Ethical Information and conflict of interest

The entire work had permitted by the Ethical Committees of local authorities. All participants provided an inscribed informed consent, and the research had conducted in line with the ethical morals identified in the 1975 treaty of Helsinki. The authors declare no potential conflicts of interest related to the present research.

References

1. Delgado-Roche L, Mesta F. Oxidative Stress as Key Player in Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV) Infection. Arch Med Res. 2020;51(5):384-387.
2. Khomich OA, Kochetkov SN, Bartosch B, Ivanov AV. Redox Biology of Respiratory Viral Infections. Viruses. 2018;10(8):392.
3. Steven S, Frenis K, Oelze M, Kalinovic S, Kuntic M, Bayo Jimenez MT, et al. Vascular Inflammation and Oxidative Stress:Major Triggers for Cardiovascular Disease. Oxid Med Cell Longev. 2019;2019:7092151.
4. Ito F, Sono Y, Ito T. Measurement and Clinical Significance of Lipid Peroxidation as a Biomarker of Oxidative Stress:Oxidative Stress in Diabetes, Atherosclerosis, and Chronic Inflammation. Antioxidants (Basel). 2019;8(3):72.

[DOI: 10.61186/rbmb.13.2.167]

5. Cecchini R, Cecchini AL. SARS-CoV-2 infection pathogenesis is related to oxidative stress as a response to aggression. Med Hypotheses. 2020;143:110102.
6. Pincemail J, Cavalier E, Charlier C, Cheramy-Bien JP, Brevers E, Courtois A, et al. Oxidative Stress Status in COVID-19 Patients Hospitalized in Intensive Care Unit for Severe Pneumonia. A Pilot Study. Antioxidants (Basel). 2021;10(2):257.
7. M VJ, Koopmans M, van Doremalen N, van Riel D, de Wit E. A Novel Coronavirus Emerging in China - Key Questions for Impact Assessment. N Engl J Med. 2020;382(8):692-694.
8. Ntyonga-Pono MP. COVID-19 infection and oxidative stress:an under-explored approach for prevention and treatment? Pan Afr Med J. 2020;35(Suppl 2):12.
9. Erel O. A new automated colorimetric method for measuring total oxidant status. Clin Biochem. 2005;38(12):1103-11.
10. Joël P, Mouna-Messaouda K, Jean-Paul CB, Jean-Olivier D, Smail M. Electrochemical Methodology for Evaluating Skin Oxidative Stress Status (SOSS). Diseases. 2019;7(2):40.
11. Beutler E, Duron O, Kelly BM. The improved method for the determination of blood glutathione. J Lab Clin Med. 1963;61:882-8.
12. Meret S, Henkin KI Clin Chem. 1971:17:369. Cited by:Gowenlock HA, McMurray RJ, McLauchlan MD (1988):Varly's Practical Clinical Biochemistry. 6th Ed. Heinemann Medical Books. London.
13. Lykkesfeldt J. Determination of ascorbic acid and dehydroascorbic acid in biological samples by high-performance liquid chromatography using subtraction methods:reliable reduction with tris[2-carboxyethyl]phosphine hydrochloride. Anal Biochem. 2000;282(1):89-93.
14. Neeld JB Jr, Pearson WN. Macro- and micromethods for the determination of serum vitamin A using trifluoroacetic acid. J Nutr. 1963;79:454-62.
15. Muhammad Y, Kani YA, Iliya S, Muhammad JB, Binji A, El-Fulaty Ahmad A, et al. Deficiency of antioxidants and increased oxidative stress in COVID-19 patients: A cross-sectional comparative study in Jigawa, Northwestern Nigeria. SAGE Open Med. 2021;9:2050312121991246.
16. Iddir M, Brito A, Dingeo G, Fernandez Del Campo SS, Samouda H, La Frano MR, Bohn T. Strengthening the Immune System and Reducing Inflammation and Oxidative Stress through Diet and Nutrition:Considerations during the COVID-19 Crisis. Nutrients. 2020;12(6):1562.
17. Delgado-Roche L, Mesta F. Oxidative Stress as Key Player in Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV) Infection. Arch Med Res. 2020;51(5):384-387.
18. Baqi HM, Farag HAM, El Bilbeisi AHH, Askandar AH, El Afifi AM. Oxidative Stress and Its Association with COVID-19: A Narrative Review. Kurdistan J Appl Res. 2020;5 (3):97-105.
19. Rahimi B, Vesal A, Edalatifard M. Coronavirus and Its effect on the respiratory system:Is there any association between pneumonia and immune cells. J Family Med Prim Care. 2020;9(9):4729-4735.
20. Martín Giménez VM, Inserra F, Tajer CD, Mariani J, Ferder L, Reiter RJ, Manucha W. Lungs as target of COVID-19 infection:Protective common molecular mechanisms of vitamin D and melatonin as a new potential synergistic treatment. Life Sci. 2020;254:117808.
21. Dworzański J, Strycharz-Dudziak M, Kliszczewska E, Kiełczykowska M, Dworzańska A, Drop B, Polz-Dacewicz M. Glutathione peroxidase (GPx) and superoxide dismutase (SOD) activity in patients with diabetes mellitus type 2 infected with Epstein-Barr virus. PLoS One. 2020;15(3):e0230374
22. Strycharz-Dudziak M, Kiełczykowska M, Drop B, Świątek Ł, Kliszczewska E, Musik I, Polz-Dacewicz M. Total Antioxidant Status (TAS), Superoxide Dismutase (SOD), and Glutathione Peroxidase (GPx) in Oropharyngeal Cancer Associated with EBV Infection. Oxid Med Cell Longev. 2019;2019:5832410.
23. Qin M, Cao Z, Wen J, Yu Q, Liu C, Wang F, et al. An antioxidant enzyme therapeutic for COVID-19. Adv Mater. 2020;32(43):e2004901.

[DOI: 10.61186/rbmb.13.2.167]

24. Derouiche S. Oxidative stress associated with SARS-Cov-2 (COVID-19) increases the severity of the lung disease—a systematic review. J Infect Dis Epidemol. 2020;6(3):1–6.
25. Dorjgochoo T, Gao Yt, Chow Wh, Shu xo, Yang G, Cai Q, et al. Major metabolite of F2-isoprostane in urine may be a more sensitive biomarker of oxidative stress than isoprostane itself. Am J Clin Nutr. 2012;96:405-14.
26. Mesaros C, Arora JS, Wholer A, Vachani A, Blair IA. 8-Oxo-2'-deoxyguanosine as a biomarker of tobacco-smoking-induced oxidative stress. Free Radic Biol Med. 2012;53(3):610-7.
27. Dinçer Y, Alademir Z, Ilkova H, Akçay T. Susceptibility of glutatione and glutathione-related antioxidant activity to hydrogen peroxide in patients with type 2 diabetes:effect of glycemic control. Clin Biochem. 2002;35(4):297-301.
28. Taheri M, Bahrami A, Habibi P, Nouri F. A Review on the Serum Electrolytes and Trace Elements Role in the Pathophysiology of COVID-19. Biol Trace Elem Res. 2021;199(7):2475-2481.
29. Yasui Y, Yasui H, Suzuki K, Saitou T, Yamamoto Y, Ishizaka T, et al. Analysis of the predictive factors for a critical illness of COVID-19 during treatment － relationship between serum zinc level and critical illness of COVID-19. Int J Infect Dis. 2020;100:230-236.
30. Kardos J, Héja L, Simon Á, Jablonkai I, Kovács R, Jemnitz K. Copper signalling:causes and consequences. Cell Commun Signal. 2018;16(1):71.
31. Barazzoni R, Bischoff SC, Breda J, Wickramasinghe K, Krznaric Z, Nitzan D, et al. ESPEN expert statements and practical guidance for nutritional management of individuals with SARS-CoV-2 infection. Clin Nutr. 2020;39(6):1631-1638.
32. Zhang J, Taylor EW, Bennett K, Saad R, Rayman MP. Association between regional selenium status and reported outcome of COVID-19 cases in China. Am J Clin Nutr. 2020;111(6):1297-1299.
33. Zhang J, Taylor EW, Bennett K, Saad R, Rayman MP. Association between regional selenium status and reported outcome of COVID-19 cases in China. Am J Clin Nutr. 2020;111(6):1297-1299.
34. Fooladi S, Matin S, Mahmoodpoor A. Copper as a potential adjunct therapy for critically ill COVID-19 patients. Clin Nutr ESPEN. 2020;40:90-91.
35. Zabetakis I, Lordan R, Norton C, Tsoupras A. COVID-19:The Inflammation Link and the Role of Nutrition in Potential Mitigation. Nutrients. 2020;12(5):1466.
36. Chiscano-Camón L, Ruiz-Rodriguez JC, Ruiz-Sanmartin A, Roca O, Ferrer R. Vitamin C levels in patients with SARS-CoV-2-associated acute respiratory distress syndrome. Crit Care. 2020;24(1):522.
37. Ntyonga-Pono MP. COVID-19 infection and oxidative stress:an under-explored approach for prevention and treatment? Pan Afr Med J. 2020;35(Suppl 2):12.
38. Shakoor H, Feehan J, Al Dhaheri AS, Ali HI, Platat C, Ismail LC, et al. Immune-boosting role of vitamins D, C, E, zinc, selenium and omega-3 fatty acids:Could they help against COVID-19? Maturitas. 2021;143:1-9.
39. Delgado-Roche L, Mesta F. Oxidative Stress as Key Player in Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV) Infection. Arch Med Res. 2020;51(5):384-387.
40. Nazar Hasan Anber Z, Oead Mohammed Saleh B, Waheab Al-Obidy M. Hepatocellular Damage and Severity of COVID-19 Infection in Iraqi Patients:A Biochemical Study. Rep Biochem Mol Biol. 2022;11(3):524-531.
41. Jan H, Usman H, Zainab R. COVID-19:a brief overview on the role of vitamins specifically vitamin C as immune modulators and in prevention and treatment of SARS-Cov-2 infections. Biomed J Sci Tech Res 2020;28(3):21580-6.
42. Maradi R, Joshi V, Balamurugan V, Susan Thomas D, Goud M. Importance of Microminerals for Maintaining Antioxidant Function After COVID-19-induced Oxidative Stress. Rep Biochem Mol Biol. 2022;11(3):479-486.

[DOI: 10.61186/rbmb.13.2.167]