Egyptian Journal of Medical Microbiology Volume 33 / No.2 / April 2024 83-90 Online ISSN: 2537-0979

ORIGINAL ARTICLE

Vaccines Defiance against SARS COV-2 Variants at Assiut Governorate

[1]Marwa Medhat M. Ahmed*, [2]Mohamed Ali M. Elfeky, [1]Safaa Ali Abo Elela [3]Rania Mohamed M. Bakry, [2]Amal A. Elkhawaga

[1]Assiut Chest Hospital
[2]Department of Medical Microbiology and Immunology, Faculty of Medicine Assiut University
[3]Clinical pathology department, South Egypt Cancer Institute Assiut University

Key words:
COVID-19, vaccination, IL-18, MCP-1

Corresponding Author:
Marwa Medhat M. Ahmed
Department of Microbiology
Faculty of Pharmacy Assiut University Egypt
Tel.: 01146510154
marmarsweet@hotmail.com

ABSTRACT

Background: *COVID-19 is a respiratory viral disease caused by the severe acute respiratory syndrome-Coronavirus type 2 (SARS-CoV-2). The World Health Organization (WHO) declared the SARS-CoV-2 infection as well as global health emergency. COVID-19 vaccines provide strong protection against serious illness, hospitalization, and death.* ***Objectives:*** *This study aimed to assess the correlation between the vaccine [type, doses] and the viral load, oxygen saturation, inflammatory markers, disease severity, and morbidity among Egyptian inpatients at Assiut chest hospital as well as assess the immune status relative to different types of cytokines, and level of anti-viral spike IgG.* ***Methodology:*** *The study involved eighty-nine subjects, including seventy-seven COVID-19 inpatients and twelve healthy controls. IL-18 and MCP-1, were evaluated using the Luminex® 100/200TM System, IgG antibodies against the SARS-CoV-2 spike protein's receptor binding domain (RBD) in human serum were detected using the ELISA technique.* ***Results:*** *increase in IgG concentration levels, oxygen saturation, clinical features (fever), and hospitalization time in vaccinated patients versus unvaccinated patients. The viral load and the levels of D-dimer, MCP-1, and IL-18 are related to vaccination status.* ***Conclusion:*** *According to the study, people who were not vaccinated got severe and life-threatening COVID-19 infections. The immunized population had a lower percentage of severe COVID-19 infections and overall mortality than the unvaccinated group. Two doses of vaccination were linked to a decreased mortality rate in hospitalized patients with moderate-to-severe COVID-19 infection.*

INTRODUCTION

Severe acute respiratory syndrome-Coronavirus type 2 (SARS-CoV-2) causes COVID-19 is a virus-related respiratory illness that was initially discovered in Wuhan, China. On January 30, 2020, the World Health Organization (WHO) classified the SARS-CoV-2 illness as a worldwide health emergency[1].

There are several COVID-19 vaccines validated for use by WHO. The first vaccination program started early December 2020[2].

The cytokine storm is caused by the unchecked and increased release of pro-inflammatory cytokines, which is a hallmark of COVID-19 syndrome.

It has been noted that the over-induction of proinflammatory cytokines contributes significantly to the development of the illness and eventually the death of SARS patients[3].

Pro-inflammatory cytokine interleukin IL-18 is a member of the IL-1 family, IL-18 plays a role in hemophagocytic lymphohistiocytosis, a life-threatening condition characterized by a cytokine storm that can be secondary to infections. A cytokine storm, which includes members of the interleukin IL-1 family, appears to be responsible for the damage in the final stages of the disease. Since IL-18 has been shown to be able to damage the lung tissue of infected animals, it is possible that it is involved in this hyperinflammation [4].

MCP-1 (monocyte chemoattractant protein-1), also known as chemokine (CC-motif) ligand 2 (CCL2). MCP-1 has been directly or indirectly linked to the etiology of many diseases, including cancer, neuroinflammatory diseases, cardiovascular diseases, and the new corona virus. It has been discovered that a biomarker associated with the severity of the illness in COVID-19 patients is a greater MCP-1 level[5].

The viral spike protein is the familiar spike that studs the surface of the coronavirus, giving it the appearance of a crown to electron microscopy[6]. The coronavirus spike protein is a multifunctional molecular machine that mediates coronavirus entry into host cells. Similar projecting proteins found in viruses are major

targets for the development of vaccines and therapeutic medications that aim to prevent the virus from infecting host cells[7].

METHODOLOGY

Study Population and ethical aspects:

This study is a case-control study that was conducted at Assiut Chest Hospital. The study was conducted from March 2022 to November 2022 and was approved by the institutional review board (IRB approval number 17101909). All participants received a clear, written consent form indicating the purpose of the study and their freedom to participate or withdraw at any time. A total of 89 subjects were enrolled in the study, including 77 COVID-19 inpatients and 12 healthy controls.

Blood samples and patients' clinical data were collected after ministry of health approval at (1-3-2022).

Laboratory investigations: complete blood picture, measurement of ferritin, and D-dimer were measured using (Robonic Eco Plus).

Evaluation of IL-18 and MCP-1 using Luminex® 100/200™ System using commercially available kits cat. No. (LXSAHM) purchased from (R&D Systems, Inc.) according to the manufacturer's instructions.

Magnetic microparticles with color coding are pre-coated with antibodies specific to the analyte. Samples, standards, and microparticles were pipetted into wells, and the target analytes were bound by the immobilized antibodies. Each well receives a biotinylated antibody cocktail that is tailored to the target analytes after any unbound materials have been cleaned away. Streptavidin-phycoerythrin conjugate (Streptavidin-PE), which binds to the biotinylated antibody, was added to each well after a wash to eliminate any unbound biotinylated antibody. Unbound Streptavidin-PE was eliminated in a final wash, after which the microparticles were resuspended in buffer and scanned using the Luminex®100/200TM System. The superparamagnetic microparticles were drawn to and retained in a monolayer by a magnet within the analyzer. The beads were lit by two Light-emitting Diodes (LEDs) with different spectra. The amount of analyte bound is directly proportional to the magnitude of the PE-derived signal, which was determined by the second LED when one recognizes the analyte being detected. Each well was imaged with a CCD camera.

Detection of IgG antibodies against the SARS-CoV-2 spike protein's receptor binding domain (RBD) in human serum using an enzyme-linked immunosorbent assay (ELISA) technique for patients and controls using commercially available kits cat. No. (30181829) purchased from (TECAN IBL international GmbH) according to the manufacturer's instructions:

Solid-phase enzyme-linked immunosorbent assay (ELISA) based on the sandwich principle. The wells were coated with antigen. When certain antibodies from the sample bind to the antigen-coated wells, a secondary enzyme-conjugated antibody that is particular to human IgG was detected. The level of color created following the substrate reaction was directly correlated with the quantity of IgG-specific antibodies found. Sample results were obtained straight up from the standard curve.

Statistical analysis:

Version 26.0 of the Statistical Package for the Social Sciences (SPSS Inc., 233 South Wacker Drive, 11th Floor, Chicago, IL, USA) was used to analyze the data. Frequency and percentage were used to represent distributed categorical data, whereas mean, standard deviation (mean ± SD), and range were used to express distributed numerical data. The Kruskal-Walli's analysis of variance test, the Mann-Whitney U test, the Wilcoxon matched pairs test, the Chi-Square test, the Fisher-Exact or Monte Carlo test, the ANOVA with repeated measures test, and the Fisher-Exact test were used to analyze nonparametric data. These tests were used to determine whether or not two sample means were equal, to examine the relationship between two qualitative variables, and to determine the statistical significance of the difference between more than two parameters. When the p-value was less than 0.05, the data was considered significant; otherwise, they were not.

RESULTS

The current study included 77 Covid-19 patients and 12 healthy controls aged between 18 and 50 years old. 51.9% of the studied patients were males and 48.05% were females. According to the control group, 58.3% were males and 41.66% were females.

It was found that (54.5%) of COVID-19 patients had moderate disease severity, while (45.45%) had severe symptoms. More than half of the studied patients (55.8%) were not vaccinated, while 34 patients (44.15%) were vaccinated. Among the vaccinated patients, 12 patients (15.58%) were vaccinated with (BNT162b2) The Pfizer BioNTech COVID-19 vaccine, 7 patients (7.9%) with BIBP-CorV Sinopharm BIBP COVID-19 vaccine, 7 patients (7.9%) with chadox1 AstraZeneca vaccine, 7 patients (9.09%) with PiCoVacc Sinovac covid-19 vaccine and only 1 patient (1.29%) was vaccinated with Moderna Spikevax® Bivalent COVID-19 vaccine. 22 patients (28.57%) took the vaccination in two doses, and 12 patients (15.58%) consumed one dosage. There was a statistically significant difference among the studied patients according to the disease progression ($p<0.001$), type of vaccine ($p<0.001$) and the number of doses ($p<0.001$). The result is shown in table (1)

Table 1: Clinical characteristics of the studied patients:

Variable	Parameter	COVID-19 patients (n=77)	Controls (n=12)	p-value
Disease progression	Control	0 (0%)	12 (100%)	<0.001
	Moderate	42 (54.5%)	0 (0%)	
	Severe	35 (45.45%)	0 (0%)	
Vaccination	Vaccinated	34 (44.15%)	6 (50%)	0.126
	Not-vaccinated	43 (55.8%)	6 (50%)	
Type of vaccine	Moderna	1 (1.29%)	0 (0%)	<0.001
	Sinovac	7 (9.09%)	1 (8.3%)	
	Astrazeneca	7 (9.09%)	2 (16.7%)	
	Sinopharm	7 (9.09%)	3 (25%)	
	Pfizer	12 (15.58%)	0 (0%)	
Number of doses	Single dose	12 (15.58%)	5 (41.7%)	<0.001
	Two doses	22 (28.57%)	7 (58.3%)	

Data expressed as number (N) (frequency %). p-value: the difference between the study parameters, p non-significant if >0.05, *P significant if <0.05, ** p highly significant if <0.001.

As shown in table (2): O_2 saturation recorded 95.29 ± 2.51 % among the vaccinated inpatients which was higher than the non-vaccinated inpatients (89.03 ± 4.06%). There was a highly statistically significant difference between vaccinated and non-vaccinated inpatients according to O_2 saturation (p<0.001).

D-dimer was higher in non-vaccinated inpatients (3.69 ± 1.29 ug/ml) than vaccinated inpatients (2.13 ± 1.03 ug/ml) with a statistically significant difference (p=0.02).

Ferritin level was higher in non-vaccinated inpatients (232.12 ± 172.3 ng/mL) than vaccinated inpatients (49.91 ± 37.84 ng/mL) with a statistically significant difference (p<0.001).

The mean IgG antibody concentration was higher in vaccinated inpatients (67.67 ± 31.27 U / ml) than non-vaccinated inpatients (60.11 ± 36.49 U / ml) with statistically significant difference (p=0.012).

IL18 level (713.56 ± 350.83 pg/ ml) among non-vaccinated inpatients which was higher than IL18 mean level of vaccinated inpatients (284.84 ± 201.98 pg/ ml) which was statistically significant difference (p=0.008).

MCP-1 level 275.69 ± 160.15 pg/ ml in non-vaccinated inpatients which was higher than the mean score of the vaccinated inpatients (193.55 ± 141.29 pg/ ml) without statistically significant difference (p=0.229).

According to the complete blood picture screening, lymphocyte count recorded 21.76 ± 8.2 $*10^3$/ul among vaccinated inpatients which was higher than non-vaccinated inpatients (16.06±9.94$*10^3$/ul) with a statistically significant difference (p=0.009).

RBC was higher in vaccinated inpatients 4.65 ± 0.69 $*10^6$/ul than non-vaccinated inpatients 3.63 ± 0.97$*10^6$/ul with a statistically significant difference (p<0.001).

WBC recorded 9.34±6.61 $*10^3$/ul among non-vaccinated inpatients, which was higher than the mean score of vaccinated inpatients 8.46±4.24$*10^3$/ul with no statistically significant difference (p=0.510).

PLT was found higher among vaccinated inpatients (298.1 ± 70.64$*10^3$/ul) than non-vaccinated inpatients (239.06 ± 112.99 $*10^3$/ul) with a statistically significant difference (p=0.011).

HGB was higher among vaccinated inpatients 12.76 ± 2.1 g/dL than in non-vaccinated inpatients 11.40 ± 1.75 g/dL with a statistically significant difference (p=0.002).

Table 2: Laboratory investigations among the studied patients:

Variable	Parameter	Non-vaccinated (n=43)	Vaccinated (n=34)	Control vaccinated (n=6)	Control non-vaccinated (n=6)	Normal range	p-value
o_2 saturation (%)	Mean ± SD	89.03 ± 4.06	95.29 ± 2.51	98.6 ± 1.53	99.6 ± 0.57	between 95 and 100	<0.001
	IQR (Min-Max)	75-98	89-99	97-100	99-100		
D-dimer (ug/ml)	Mean ± SD	3.69 ± 1.29	2.13 ± 1.03	1.15 ± 0.36	0.7 ± 0.4	0-0.50	0.02
	IQR (Min-Max)	0.4-11.5	0.3-7.9	0.8-1.52	0.3-1.2		
Ferritin (ng/mL)	Mean ± SD	232.12 ± 172.3	49.91 ± 37.84	32.96 ± 1.66	26.16 ± 8.5	Female:15-150; Male:30-400	<0.001
	IQR (Min-Max)	16.9-463.9	18.5-269.9	31.2-34.5	16.3-31.8		
IgG conc (U / ml)	Mean ± SD	60.11 ± 36.49	67.67 ± 31.27	103.31±12.07	39.78 ± 16.82	6.0 – 16.0	0.012
	IQR (Min-Max)	10.9– 117.4	7.8 – 120.9	89.88-113.26	25.1-58.1		
IL18 (pg/ ml)	Mean ± SD	713.56±350.83	284.84±201.98	669.35± 22.09	148.13±121.19		0.008
	IQR (Min-Max)	52.7 – 3227.2	67.1 – 715	654.15-694.707	64.713-287.157		
MCP-1 (pg/ ml)	Mean ± SD	275.69±160.15	193.55±141.29	325.09±5.507	174.35± 134.88		0.229
	IQR (Min-Max)	45.5 – 1623.5	36.3 – 560.4	320.95-331.34	53.64-319.94		
	Complete blood picture						
Lymphocyte ($*10^3$/ul)	Mean ± SD	16.06 ± 9.94	21.76±8.2	34.8 ± 2.86	29.13 ± 2.5	1-4	0.009
	IQR (Min-Max)	0.8-36.4	8.1 – 36.3	31.8-37.5	27.4-32		
RBC ($*10^6$/ul)	Mean ± SD	3.63 ± 0.97	4.65±0.69	5.27 ± 0.075	5.17 ± 0.57	(3.8-5.8)	<0.001
	IQR (Min-Max)	2 – 5.7	3.2-5.8	5.2-5.35	4.79-5.84		
WBC ($*10^3$/ul)	Mean ± SD	9.34 ± 6.61	8.46 ± 4.24	10.33 ± 0.802	11.6 ± 3.63	Female (4.5-6.5) Male (3.8-5.8)	0.510
	IQR (Min-Max)	1.3-45.7	1.6-21.4	9.5-11.1	9.5-15.8		
PLT ($*10^3$/ul)	Mean ± SD	239.06±112.99	298.1±70.64	348 ± 18.19	355.66±50.003	(150-400)	0.011
	IQR (Min-Max)	103-580	71-378	327-359	298-387		
HGB (g/dL)	Mean ± SD	11.40 ± 1.75	12.76±2.1	16.833 ± 0.89	17.2 ± 2.25	Female (14-18) Male (11-16)	0.002
	IQR (Min-Max)	8.3-15.3	9.5-17.7	15.8-17.4	14.6-18.6		

It was discovered that 62.79% of non-vaccinated patients had severe disease progression, while only 23.53% of the vaccinated patients had severe symptoms. 37.2% and 76.47% of the non-vaccinated and vaccinated patients had moderate disease progression respectively. There was a statistically significant difference between vaccinated and non-vaccinated patients according to disease progression (p<0.001) (fig.1).

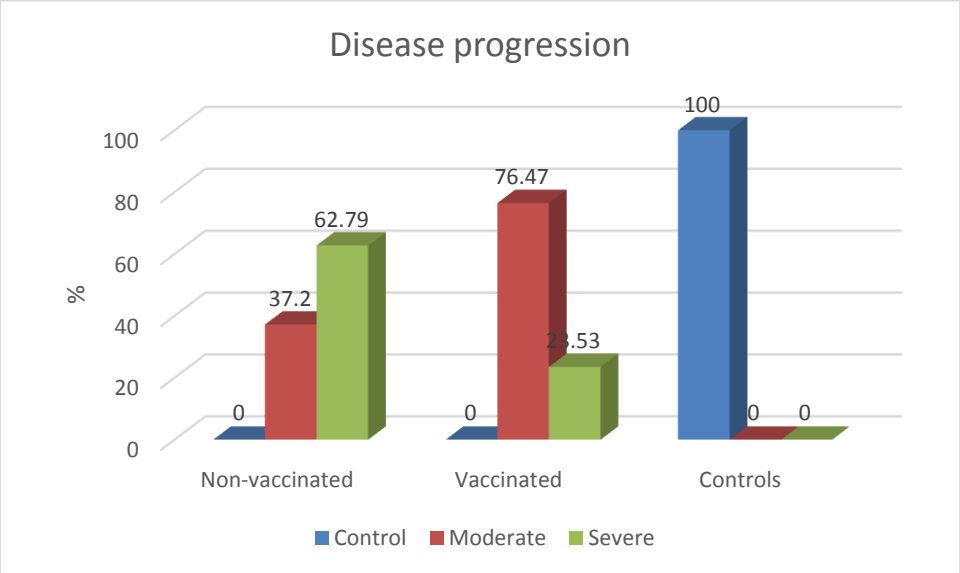

Fig. 1: Disease progression among vaccinated and non-vaccinated patients including, controls.

Among the studied non-vaccinated inpatients, 23.25% died and 76.7% survived. All vaccinated inpatients survived. There was a statistically significant difference among the studied vaccinated and non-vaccinated inpatients according to morbidity (p=0.002) (fig.2).

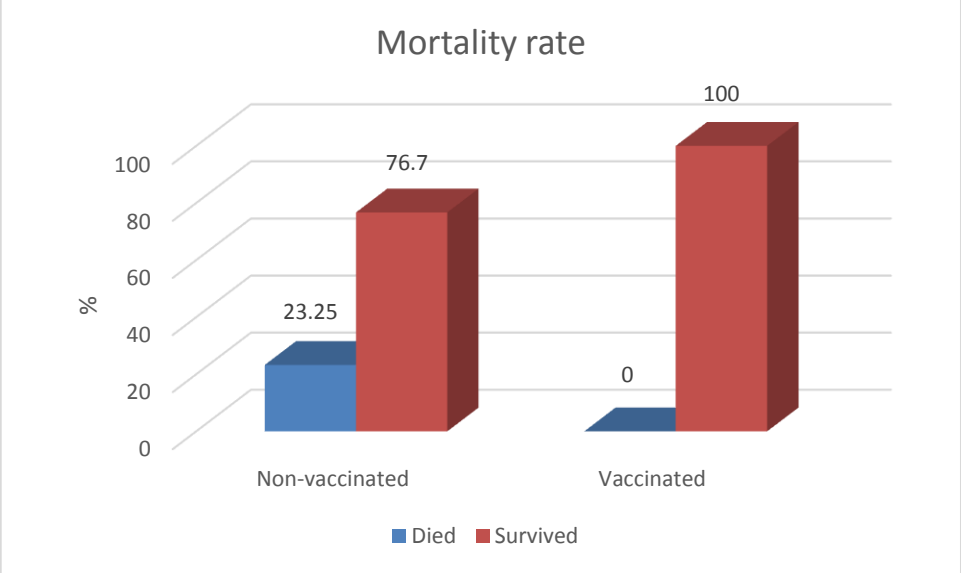

Fig. 2: Mortality rate among the vaccinated and non-vaccinated inpatients.

It has been demonstrated that there was a negative correlation between IgG and MCP-1 levels among COVID-19 inpatients (r = -0.318) with a statistically significant difference (p = 0.003). Another positive correlation discovered between MCP-1 and IL18 levels among the studied COVID-19 inpatients (r = 0.554), with a statistically significant difference (p<0.001).

There was a positive correlation between lymphocyte and IgG levels among the studied COVID-19 inpatients (r = 0.323) with a statistically significant difference (p = 0.003). Another negative correlation discovered between lymphocyte and IL18 levels among COVID-19 inpatients (r = -0.397) with a statistically significant difference (p<0.001). Moreover, there was a negative association between lymphocyte and MCP-1 levels in COVID-19 inpatients (r = -0.239) with a statistically significant difference (p = 0.029) (table3).

Table 3: Hospitalization time in relation to the level of anti- viral spike IgG and cytokines, viral load and cytokines among the studied inpatients

Variable		IL18	MCP-1	Lymphocyte	Hospitalization time (days)
IgG conc	r-value	-.482	-.318	.323	.084
	p-value	.000**	.003**	.003**	.451
IL18	r-value	1	.554	-.397	.023
	p-value		.000**	.000**	.836
MCP-1	r-value		1	-.239	.066
	p-value			.029*	.554
Lymphocyte	r-value			1	.137
	p-value				.217

**. Correlation is significant at the 0.01 level (2-tailed). *. Correlation is significant at the 0.05 level (2-tailed).

DISCUSSION

The present investigations reported a significant correlation between disease severity and oxygen saturation level, indicating that as disease severity increased, oxygen saturation level decreased[8].

There was a statistically significant difference between vaccinated and non-vaccinated inpatients according to O_2 saturation, which means that vaccination decreases the need for oxygen ventilation.

This study provided evidence of a substantial increase in protection against symptomatic COVID-19 disease after a booster dose of the BNT162b2 or mRNA-1273 vaccine. With a BNT162b2 booster, very high levels of defense against hospitalization or mortality were seen. Regardless of the vaccination used

in the initial course, the effectiveness of the second dose was extremely similar [9].

Patients with COVID-19 may develop more serious conditions after being admitted to the hospital, such as respiratory failure and even death. Because of their previous immunization, individuals who have received the SARS-CoV-2 vaccine are likely to develop memory antibodies and cellular responses as a reaction to the illness. These immune responses may slow the progression of the disease and even avoid a possible fatal organ failure.[10, 11]

our laboratory findings demonstrated that The increase in ferritin level is associated with the worsening of COVID-19[12]. The cytokine storm and the exaggerated host immune response (i.e., ferritin) participate in the development of ARDS, which is the most important reason for mortality if progresses to respiratory failure[13].

Non-survivors had higher D-dimer levels than survivors do[14], indicating that elevated D-dimer levels represent an independent risk factor for death in COVID-19 patients[15]. Therefore, patients were categorized based on their D-dimer level, and the findings indicated that irrespective of the clinical characteristic, the group with greater D-dimer level had a higher MCP-1 level than the group with lower D-dimer level.

Unlike the unvaccinated patients, All serum samples from vaccinated people (34) were positive when tested with RBD IgG ELISA, which is matching with the general belief that antibodies appear at least 15 days after vaccination[16].

In SARS caused by SARS-CoV-2, IL-18 levels were considerably elevated compared with those in healthy subjects[17]. In addition, IL-18 concentrations were significantly higher in non-survivors compared with survivors. High IL-18 levels were associated with a risk of heart damage and death[18].

MCP-1 is involved in the pathophysiology of illnesses where there is an invasion of monocytes. According to our research, critically sick patients had greater serum levels of MCP-1 than severe patients did compared to patients with severe disease, those with critical disease had distinctly higher white blood cell counts, procalcitonin levels, and D-dimer levels, and lower hemoglobin levels and lymphocyte counts[19].

One biomarker linked to the severity of COVID-19 illness is MCP-1. Additionally, the group with higher D-dimer levels had higher MCP-1 levels than the group with lower levels, suggesting that in COVID-19 patients, MCP-1 may be linked to an increased risk of death.

lymphocytopenia (64.5%) and leukocytopenia (29.4%) were more common. They were all consistent with a respiratory virus infection overall. In order to diagnose newly discovered coronavirus infections, the lymphocytopenia may be utilized as a reference index. Studies have shown that the levels of inflammatory cytokines may be related to the severity of the disease,[20, 21] which is expected to be an indicator of the severity of the disease.

Individuals admitted to the ICU with COVID-19 were more likely to have lymphocytopenia at the time of hospital admission compared to patients who did not get admitted to the ICU[22].

Patients with worse outcomes or more severe illnesses had lower platelet counts, while non-survivors had even lower platelet counts[23].

Severe COVID-19 individuals had higher ferritin and lower hemoglobin and RBC levels than moderate cases[24].

All the vaccinated inpatients survived. There was a statistically significant difference among the studied vaccinated and non-vaccinated inpatients according to morbidity, and disease progression.

It has been shown that A negative correlation was seen between the levels of MCP-1 and IgG[25]; another positive correlation was found between MCP-1 and IL18 levels[26]; there was a positive correlation between lymphocytes and IgG levels[27]; and another negative correlation was found between lymphocytes and IL18 and MCP-1 levels in COVID-19 inpatients[28].

CONCLUSION

The current study suggests an increase in IgG concentration levels, increase in oxygen saturation, decreases in clinical features (fever) and an average duration of hospital stay in vaccinated patients versus unvaccinated patients. The viral load of SARS-CoV-2 is associated with the vaccination status as well as inflammatory markers such as D-dimer and IL-18.

However, based on these early findings, we may conclude that individuals who choose not to receive the vaccination experience severe and life-threatening COVID-19 infection. The percentage of those with severe COVID-19 infection, the need for ventilatory support, and overall mortality has decreased when compared to the unvaccinated population. Two vaccine doses were associated with a decreased mortality rate in hospitalized patients those with moderate-to-severe COVID.

Declarations:

Consent for publication: Not applicable

Availability of data and material: Data are available upon request.

Competing interests: The author(s) declare no potential conflicts of interest with respect to the research, authorship and/or publication of this article.

This manuscript has not been previously published and is not under consideration in another journal.

Funding: Authors did not receive any grants from funding agencies.

Acknowledgement: We would like to thank Prof. Mohamed Ali Mohamed Elfeky, Prof. Rania Mohamed Mohamed Bakery, Dr. Safaa Ali Abo-Elela, Dr. Amal Ahmed Mahmoud Elkhawaga, for their kind supervision, continuous encouragement, direct guidance and valuable support.

REFERENCES

1. Ortiz-Prado E, Simbaña-Rivera K, Gómez-Barreno L, Rubio-Neira M, Guaman LP, Kyriakidis NC, Muslin C, Jaramillo AMG, Barba-Ostria C, Cevallos-Robalino DJD. m, disease, i., Clinical, molecular, and epidemiological characterization of the SARS-CoV-2 virus and the Coronavirus Disease 2019 (COVID-19), a comprehensive literature review.J Diagnostic microbiology and infectious disease. 2020, *98* (1), 115094.
2. Gee J, Marquez P, Su J, Calvert GM, Liu R, Myers T, Nair N, Martin S, Clark T, Markowitz LJM. report, m. w., First month of COVID-19 vaccine safety monitoring—United States, December 14, 2020–January 13,J centers for disease control and prevention. 2021. 2021, *70* (8), 283.
3. Karimabad, M. N, Hassanshahi, G, Kounis, N. G, Mplani, V, Roditis, P, Gogos, C, Lagadinou, M, Assimakopoulos, S. F, Dousdampanis, P, Koniari, I. J. V., The Chemokines CXC, CC and C in the Pathogenesis of COVID-19 Disease and as Surrogates of Vaccine-Induced Innate and Adaptive Protective Responses.J MDPI. 2022, *10* (8), 1299.
4. Abbate, A, Toldo, S, Marchetti, C, Kron, J, Van Tassell, B. W, Dinarello, C. A. J. C. r., Interleukin-1 and the inflammasome as therapeutic targets in cardiovascular disease.J AHA. 2020, *126* (9), 1260-1280.
5. Singh, S, Anshita, D, Ravichandiran, V. J. I. i., MCP-1: Function, regulation, and involvement in disease.J science direct. 2021, *101*, 107598.
6. Robson, B. J. C. i. b, medicine, Computers and viral diseases. Preliminary bioinformatics studies on the design of a synthetic vaccine and a preventative peptidomimetic antagonist against the SARS-CoV-2 (2019-nCoV, COVID-19) coronavirus. J science direct. 2020, *119*, 103670.
7. Martinez-Flores, D, Zepeda-Cervantes, J, Cruz-Resendiz, A, Aguirre-Sampieri, S, Sampieri, A, Vaca, L. J. F. i. i., SARS-CoV-2 vaccines based on the spike glycoprotein and implications of new viral variants. J frontiers. 2021, *12*, 701501.
8. Qadir FI, Kakamad FH, Abdullah IY, Abdulla B A, Mohammed SH, Salih RQ, Ali RK, Salh AMJA.o. M. Surgery, The relationship between CT severity infections and oxygen saturation in patients infected with COVID-19, a cohort study. J wolters klwuer. 2022, *76*, 103439.
9. Arbel R, Hammerman A, Sergienko R, Friger M, Peretz A, Netzer D, Yaron SJNEJ.o M. BNT162b2 vaccine booster and mortality due to Covid-19. J PubMed. 2021, *385* (26), 2413-2420.
10. Cromer D, Juno JA, Khoury D, Reynaldi A, Wheatley AK, Kent SJ, Davenport MPJNRI. Prospects for durable immune control of SARS-CoV-2 and prevention of reinfection.J nature reviews immunology. 2021, *21* (6), 395-404.
11. Sadarangani M, Marchant A, Kollmann TRJNRI/ Immunological mechanisms of vaccine-induced protection against COVID-19 in humans. J nature reviews immunology. 2021, *21* (8), 475-484.
12. Zhou F, Yu T, Du R, Fan G, Liu Y, Liu Z, Xiang J, Wang Y, Song B, Gu XJT. l., Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. J the lancet. 2020, *395* (10229), 1054-1062.
13. Wu C, Chen X, Cai Y, Zhou X, Xu S, Huang H, Zhang L, Zhou X, Du C, Zhang YJJ. i. m., Risk factors associated with acute respiratory distress syndrome and death in patients with coronavirus disease 2019 pneumonia in Wuhan, China. J JAMA internal medicine. 2020, *180* (7), 934-943.
14. Zhong L, Chuan J, Gong B, Shuai P, Zhou Y, Zhang Y, Jiang Z, Zhang D, Liu X, Ma SJSCL S., Detection of serum IgM and IgG for COVID-19 diagnosis. J Sci China Life Sci. 2020, *63* (5), 777.
15. Wu D, Lu J, Liu Y, Zhang Z, Luo LJIJ. o. I. D., Positive effects of COVID-19 control measures on influenza prevention. J International Journal Of infectious Diseases. 2020, *95*, 345-346.
16. Keyhanian K, Umeton RP, Mohit B, Davoudi V, Hajighasemi F, Ghasemi MJJ. o. n., SARS-CoV-2 and nervous system: From pathogenesis to clinical manifestation. J Journal Of Neuroimmunology 2021, *350*, 577436.
17. Marino L, Criniti A, Guida S, Bucci T, Ballesio L, Suppa M, Galardo G, Vacca A, Santulli M, Angeloni AJC, medicine, e., Interleukin 18 and IL-18 BP response to Sars-CoV-2 virus infection. J Clinicl And Experimental medicine 2023, *23* (4), 1243-1250.
18. Mehta P, Samanta RJ, Wick K, Coll RC, Mawhinney T, McAleavey PG, Boyle AJ, Conlon J, Shankar-Hari M, Rogers AJ. t., Elevated ferritin, mediated by IL-18 is associated with systemic inflammation and mortality in acute respiratory distress syndrome (ARDS). thorax; 2023.

19. Hashimoto S, Yoshizaki K, Uno K, Kitajima H, Arai T, Tamura Y, Morishita H, Matsuoka H, Han Y, Minamoto SJF i. m., Prompt reduction in CRP, IL-6, IFN-γ, IP-10, and MCP-1 and a relatively low basal ratio of ferritin/CRP is possibly associated with the efficacy of tocilizumab Monotherapy in severely to critically ill patients with COVID-19.J Frontiers in Medicine 2021, *8*, 734838.
20. Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, Zhang L, Fan G, Xu J, Gu XJTl. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. J The Lancet. 2020, *395* (10223), 497-506.
21. Chen L, Liu H, Liu W, Liu J, Liu K, Shang J, Deng Y, Wei SJZ. Analysis of clinical features of 29 patients with 2019 novel coronavirus pneumonia. J Chinese Journal of tuberculosis and respiratory diseases 2020, *43*, E005-E005.
22. Zhao Q, Meng M, Kumar R, Wu Y, Huang J, Deng Y, Weng Z, Yang LJI j. o. i. d., Lymphopenia is associated with severe coronavirus disease 2019 (COVID-19) infections: A systemic review and meta-analysis.J International journal of infectious diseases. 2020, *96*, 131-135.
23. Zong X, Gu Y, Yu H, Li Z, Wang YJLM, Thrombocytopenia is associated with COVID-19 severity and outcome: an updated meta-analysis of 5637 patients with multiple outcomes.J Laboratory medicine. 2021, *52* (1), 10-15.
24. Richardson S, Hirsch JS, Narasimhan M, Crawford JM, McGinn T, Davidson KW, Barnaby DP, Becker LB, Chelico JD, Cohen SLJJ, Presenting characteristics, comorbidities, and outcomes among 5700 patients hospitalized with COVID-19 in the New York City area. J JAMA. 2020, *323* (20), 2052-2059.
25. Yang F, Jing J.-J, Fu S.-Y, Su X.-Z, Zhong Y.-L, Chen D.-S, Wu X.-Z, Zou Y.-Q JMN. Spinal MCP-1 Contributes to Central Post-stroke Pain by Inducing Central Sensitization in Rats. J Molecular Neurobiology. 2023, *60* (4), 2086-2098.
26. Kazmi N, Wallen GR, Yang L, Alkhatib J, Schwandt ML, Feng D, Gao B, Diazgranados N, Ramchandani VA, Barb JJJF i. P., An exploratory study of pro-inflammatory cytokines in individuals with alcohol use disorder: MCP-1 and IL-8 associated with alcohol consumption, sleep quality, anxiety, depression, and liver biomarkers. J Frontiers in psychiatry. 2022, *13*, 931280.
27. Achiron A, Mandel M, Gurevich M, Dreyer-Alster S, Magalashvili D, Sonis P, Dolev M, Menascu S, Harari G, Flechter SJJ o. n., Immune response to the third COVID-19 vaccine dose is related to lymphocyte count in multiple sclerosis patients treated with fingolimod.J Journal of Neurology. 2022, *269* (5), 2286-2292.
28. Huang W, Li M, Luo G, Wu X, Su B, Zhao L, Zhang S, Chen X, Jia M, Zhu JJTJ. o. I. The inflammatory factors associated with disease severity to predict COVID-19 progression. J The Journal of immunology. 2021, *206* (7), 1597-1608.