Comparison between the Impact of Pfizer Vaccination and Covid-19 Infection on Human Creatine Kinase Activity and its Isoenzymes

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ABSTRACT
Background: One of the zoonotic illnesses, COVID-19, is spreading from person to person by a variety of methods, including coughing, sneezing, or speaking, which spreads tiny droplets from the infected person's lips or nose . Corona virus vaccination generally causes modest side effects that become more noticeable after the second dosage. Creatine kinase (CK) is a protein that catalyzes the transfer of a phosphate group from creatine phosphate to adenosine diphosphate.
Aim: This study aimed to compare between the impact of Pfizer vaccination and covid-19 infection on human creatine kinase activity and its isoenzymes.
Patients and methods: (150) blood samples; one hundred blood specimens were collected from Covid-19 patients who attended to Al- Fallujah Teaching Hospital and Al-Razzi private hospital and, fifty Blood samples were collected from volunteers who attended to the vaccination outlets to be vaccinated with the Pfizer vaccine in Al- Fallujah Teaching Hospital. The study was conducted through the period from September 2021 to December 2021.
Results: In the COVID-19 group of patients, there was a statistically significant difference ($P < 0.05$) in the mean levels of basal Total CK, basal CK-MB, and CK-MM in comparison with the vaccinated group. There was no statistically significant difference in mean of total CK after second dose between the two groups. There was a statistically significant decrease in mean of CK-mb and CK-mm in vaccinated group in comparison with COVID-19 group.
Conclusion: Covid-19 infection had more impact on CK-MB and CK-MM than the Pfizer vaccination. The activity of CK-MB and CK-MM after covid-19 infection were higher than activity after Pfizer vaccination and before vaccination
Keyword: mRNA vaccine, CK, CK-MB, CK-MM, Corona vaccination, Coronavirus, COVID-19.

INTRODUCTION

Coronaviruses are RNA viruses that actually cause illness in humans and animals. In 1962, human corona viruses (HCoVs) were identified. as causes of, acute upper, respiratory infection, (URI) [1]. The family Coronaviridae and order Nidovirales contain the largest group of viruses collectively known as the "corona virus" [2, 3]. They have a diameter of 125 nm and are polymorphic or spherical in shape [4]. It is a significant pathogen in both humans and vertebrates. They can harm the central nervous system, liver, gastrointestinal tract, and respiratory system of humans [5, 6] . Corona virus is a zoonotic infection. It spreads from person to person in a variety of ways, including small droplets from infected people's noses or mouths when they cough, sneeze, or talk. Real-time PCR allows for early detection [7]. Wuhan became the first pandemic epicenter [8, 10].

The corona virus family is divided into four classes, all of which have a positive-sense RNA genome with a single strand. The term "coronavirus" refers to the club-shaped protein spikes that can be seen on the surface when observed through transient electron microscope. The factor responsible for the corona virus pandemic belongs to the beta class [11]. The first signs of a covid-19 infection are fatigue, fever, difficulty breathing and a dry cough [12]. Corona virus is a systemic infection that affects hemostasis and the hematopoietic system significantly [13].

With the long-awaited arrival of COVID-19 vaccines, the side effects of vaccination are once again in the spotlight. While injection site pain, fever, malaise, and headache are common reactions, other, more serious side effects, such as vaccine-induced immune thrombocytopenia, have been reported [14].

The Pfizer-BioNTech corona virus vaccine, known as BNT162b2, is mRNA vaccine developed by BioNTech and Pfizer to prevent coronavirus disease, the causative agent of which is SARS-CoV-2. (SARS-CoV-2). Responsive genetic platforms have helped develop vaccines [15].

Gene-based vaccinations (which also include DNA and mRNA vaccines) include the genetic instructions for the vaccine recipient's cells to create an antigen. The surface spike protein, which the virus uses to connect and fuse with host cells, is the target antigen for coronavirus vaccines [16].

Pfizer corona virus vaccine (BNT162b2) is given intramuscularly as a series of two doses of 30 micrograms of the diluted vaccination solution (0.3 ml each), as follows: one dosage is given first, and the second dose is given 21 days later. The trial's time between the two dosages ranged from 19 to 45 days [17].

There are several hazards associated with vaccination, despite its long history in public health and preventative medicine. The COVID-19 vaccine frequently results in minor side effects, including swelling, soreness and redness at the injection site as well as more widespread symptoms of headache, fatigue, muscle pain, fever and chills. Side effects after the 2nd dose may be more severe than those after the 1st dose [18]. More serious side effects, such as myocarditis, have been observed with the mRNA vaccination Pfizer-BioNTech and Moderna [18].

Creatine phosphokinase is an enzyme that stimulates the conversion of creatine phosphate to adenosine diphosphate to form high-energy adenosine triphosphate (the main energy storage reservoir during muscle rest) [19]. The most common enzyme to diagnose and monitor muscle disease is creatine kinase, which is most abundant in the serum in response to muscle injury. CK isoenzymes are dimeric as subunit B or subunit M and the three cytoplasmic CPK isoenzymes are easily identified in human tissues [20, 21].

PATIENTS AND METHODS

The current study included 150 participant, one hundred of them were COVID-19 infected patients and fifty were free of the disease and had two dose of vaccine (pfizer). One hundred Blood specimens were collected from Covid-19 patients (mild and severe symptoms) who attended to Al- Fallujah Teaching Hospital and Al-Razzi private hospital and private laboratories in Al-Fallujah city (Al-Anbar province).

Fifty blood samples were collected from volunteers with age range (19-70 years old) who attended for vaccination outlets to be vaccinated with the Pfizer vaccine in Al- Fallujah Teaching Hospital and Al-Ramadi Teaching Hospital from September 2021 to December 2021.

The blood samples (5 ml) were taken from each volunteer included in this study, without using any anticoagulant (using gel tube). The serum was isolated from the human blood by centrifugation at 3000 rpm for ten minutes. The serum, was immediately placed at -20 °C in a deep freezer after separated in-multiple small tubes to be used for the desired measurements. Oliver method was used to evaluate the total Creatine phosphokinase activity of serum and its isoenzymes [22].

Ethical Approval: The study was approved by the Ethics Board of University of Baghdad and an informed written consent was taken from each participant in the study. This work has been carried out in accordance with The Code of Ethics of the World Medical Association (Declaration of Helsinki) for studies involving humans.

Statistical analysis

The data was interrupted using SPSS version 25.0 software, frequency, percentage, mean and stander deviation were used to describe data. Independent t-test was used to evaluate the difference in mean level of numeric data between 2 variables and paired t test to evaluate mean difference of mean to the same variable in two occasion. Histogram figures were used to show mean of studied biomarkers between the groups.

RESULTS

The current study included 150 participant, one hundred of them were COVID-19 infected patients and fifty were free of the disease and had two dose of vaccine, as presented in table (1). The study included participant from both sex, 62.7% were males and 36.3% were females. Regarding vaccinated group, 22% were females and 78% were males. While, Covid group (non-vaccinated) contained 45% females and 55% were

males (Figure 1). The mean of age was 34.44 ± 13.238, the lowest age was 12 years and highest age was 71 years. 16% of participant were aged ≤ 20 years, 25.3% were 31-40 years, 29.4% were 21-30 years, 18.7% were 41-50 years, 5.3% were 51-60 years and 5.3% were ≥ 61 years (Figure 2)

Table (1): group of participant and immunoglobulin result

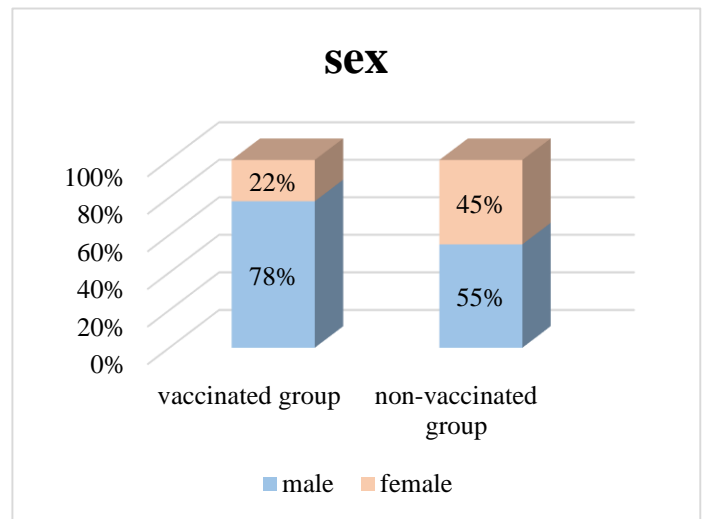|		N.	%
Group	Vaccine group	50	33.3%
	COVID group	100	66.7%
Ig G/ Ig M result	Negative	50	33.3%
	Positive	100	66.7%
	Total	150	100.0%

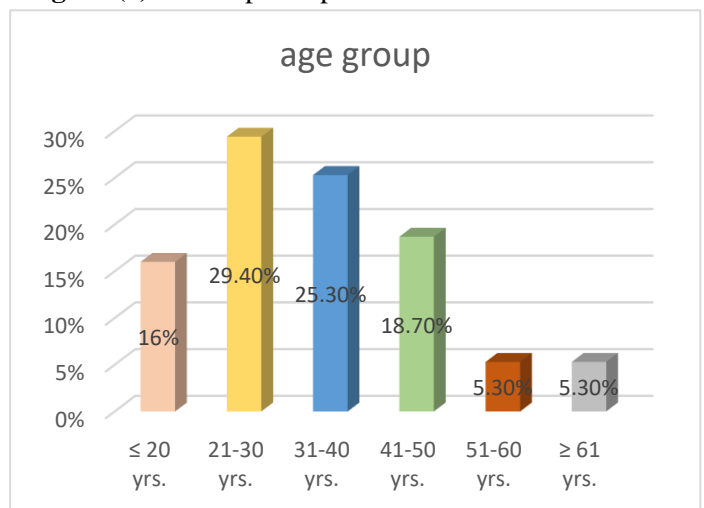**Figure (1):** sex of participant.

Figure (2): participant age group.

The total CK, CK-MB and CK-MM were evaluated in both group (using independent t-test), result revealed a statistical higher mean level of basal total CK, basal CK- MB and CK-MM in COVID-19 group of patients (53.38 ± 16.37, 27.06 ± 9.15 and 32.4 ± 14.8 respectively) as compared to vaccinated group (before taking vaccine) (34.54 ± 11.24, 7.44 ± 3.94 and

27.32 ± 10.65, p-value < 0.0001, < 0.0001 and < 0.03 successively).

In addition, there was no statistically significant difference in mean of total CK after second dose between the two groups, a statistically significant decrease in mean of CK-mb and CK-mm in vaccinated group as compared the COVID-19 group. While there was significant increase in CK, CK-MB and CK-MM of after 2nd dose when compared with before vaccine as presented in table (2) and figure (3).

Table (2): mean difference in total CK, CK-MB and CK-MM between the two groups   *p-value ≤ 0.05

	group	Mean ± Std. Deviation	p-value
Total CK(u/l)	Before Vaccine	34.54 ± 11.24	< 0.0001*
	COVID group	53.38± 16.37	
Basal CK- MB	Before Vaccine	7.44 ± 3.94	< 0.0001*
	COVID group	27.06 ± 9.15	
Basal CK-MM	Before Vaccine	27.32 ± 10.65	0.03*
	COVID group	32.4 ± 14.8	
Total CK	After 2nd dose	51.12 ± 12.43	0.39
	COVID group	53.38 ± 16.37	
CK-MB	After 2nd dose	12.52 ± 5.56	<0.0001*
	COVID group	27.06 ± 9.15	
CK-MM	After 2nd dose	38.9 ± 11.74	0.007*
	COVID group	32.40 ± 14.81	
Total CK	After 2nd dose	34.54 ± 11.24	< 0.0001*
	Before Vaccine	51.12 ± 12.43	
CK-MB	After 2nd dose	7.44 ± 3.94	< 0.0001*
	Before Vaccine	12.52 ± 5.56	
CK-MM	After 2nd dose	27.32 ± 10.65	< 0.0001*
	Before Vaccine	38.9 ± 11.74	

Figure (3): The mean ± SD difference in total CK, CK-mb and CK-mm between the two groups.
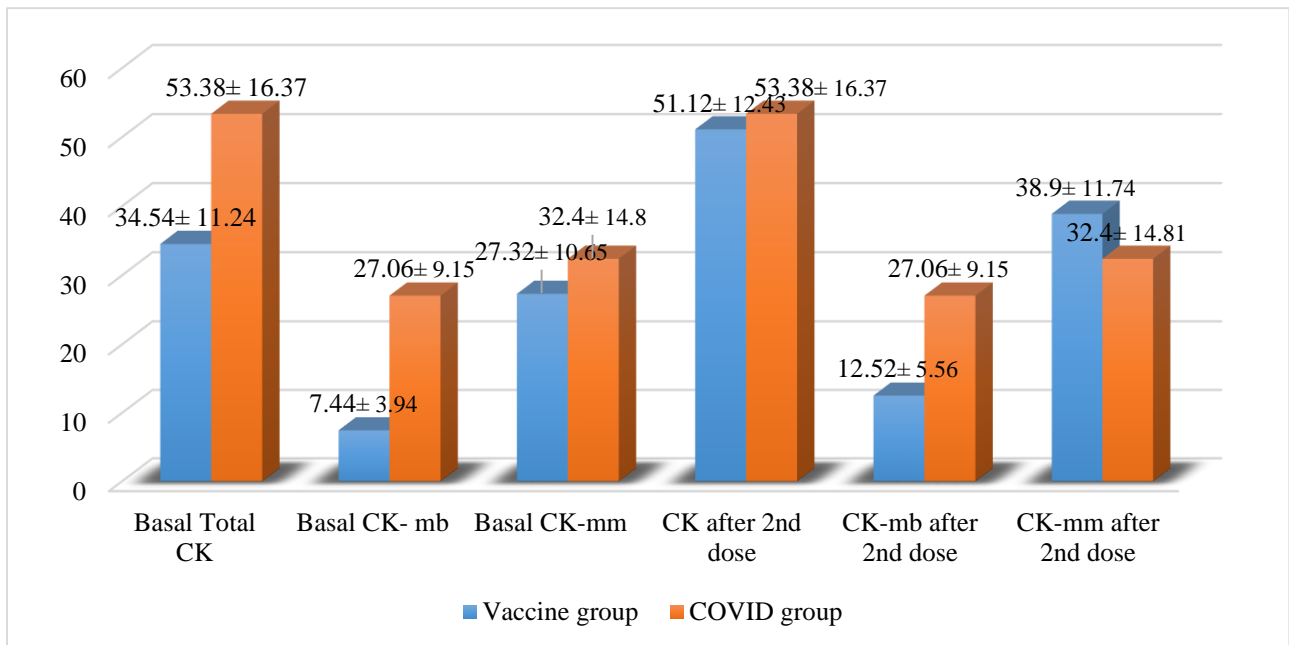
DISCUSSION

There was hope by the end of the year with the advent of COVID-19 vaccinations intended to end the pandemic. Corona virus sickness had a substantial impact on the economic, health and political systems in 2020 [23, 24].

The COVID-19 vaccine frequently results in minor side effects, including swelling, soreness and redness at the injection site as well as more widespread symptoms of headache, fatigue, muscle pain, fever and chills. Side effects after the 2nd doses may be more severe than those after the 1st dose. More serious side effects, such as myocarditis, have been observed with the mRNA vaccination Pfizer-BioNTech and Moderna [25].

In This study, result revealed a statistical higher mean level of basal total CK, basal CK- MB and CK-MM in COVID-19 group of patients (53.38± 16.37, 27.06 ± 9.15 and 32.4 ± 14.8 respectively) as compared to vaccinated group (before taking vaccine) (34.54 ± 11.24, 7.44 ± 3.94 and 27.32 ± 10.65, p-value < 0.0001,< 0.0001 and < 0.03 successively).

There was no statistically significant, difference in mean of total CK after second dose between the two groups, while there was a statistically significant decrease in mean of CK-mb and CK-mm in vaccinated group as compared to the COVID-19 group.

The present study is in an agreement with **Alrubaye and Choudhary** [4] who showed that a 35-year old lady who had a fever, chills, a cough, and myalgia was taken to the hospital. She had not recently undergone trauma, engaged in any hard exercise, or had her urine color changed when she was diagnosed with COVID-19. She didn't have a long medical history. She had no known drug allergies and wasn't taking any prescription or over-the-counter drugs. She did not use tobacco or alcohol. Creatine kinase levels in the patient rose and reached a peak of 71 000 U/L. In another study, a 88-yeares old woman entered the hospital's emergency room with sudden onset of discomfort and weakness in both of his thighs, she was diagnosed with coronavirus. Her blood tests were notable for the significant increase in creatine phosphokinase (CPK), which was 13581 units per liter (U/L) [26]. In another study, a cough, breathing difficulty and diarrhea were present for two days in a 65-yeares old African American man. The patient had a positive SARS-CoV-2 test, and his creatine kinase level was 7854 U/L (normal range, 20–300 U/L) [27]. CK was increased to 4625 U/L with concurrent transaminitis in African-American man, 58 years old. The patient complained of headaches, breathing difficulty, and dry cough [28].

The present study is in an agreement with **Woodworth et al.** [29] who revealed that patients did not take any medication on a regular basis. They denied engaging in strenuous exercise, heavy lifting, or experiencing post-vaccination physical trauma. The patients supported social marijuana use but denied using any other drugs, alcohol, or tobacco. Creatine kinase (CK) levels were greater than 22000 U/L (nr 20e190 U/L).

In another study, Two days after getting the 3rd dosage of Pfizer vaccination in her left upper arm, a 76-year-old Japanese woman with myalgia attended emergency room. Seven and eight months had passed since the patient's 1st and 2nd doses of Pfizer-BioNTech, corona viruses vaccination were administered in her left upper arm. She experienced no negative side effects following the prior COVID-19 vaccinations. Laboratory findings showed that her concentration of creatine kinase (CK) was 9816 U/L, CK-MB was 106 U/L, Day 1 CK 9816, Day 2 CK 5416, Day 4 CK 1630, Day 8 CK 164 and Day 10 CK 101. The CK concentration gradually decreased to normal [30]

CONCLUSION

Covid-19 infection had more impact on CK-MB and CK-MM than Pfizer vaccination. The activity of CK-MB and CK-MM after covid-19 infection were higher than activity after Pfizer vaccination and before vaccination.

REFERENCES

1. **Abbasi J (2020):** COVID-19 and mRNA vaccines—first large test for a new approach. JAMA., 324 (12): 1125-1127.
2. **Al-Suhail R, Ali L (2021):** Statistical Analysis of COVID-19 Pandemic Across the Provinces of Iraq. https://doi.org/10.24996/ijs.2021.62.3.12
3. **Albert E, Aurigemma G, Saucedo J et al. (2021):** Myocarditis following COVID-19 vaccination. Radiology case reports, 16 (8): 2142-2145.
4. **Alrubaye R, Choudhary H (2020):** Severe rhabdomyolysis in a 35-year-old woman with COVID-19 due to SARS-CoV-2 infection: a case report. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7440748/
5. **Bohlmeyer T, Wu A, Perryman M (1994):** Evaluation of laboratory tests as a guide to diagnosis and therapy of myositis. Rheumatic Disease Clinics of North America, 20 (4): 845-856.
6. **Bozkurt B, Kamat I, Hotez P (2021):** Myocarditis with COVID-19 mRNA vaccines. Circulation, 144 (6): 471-484.
7. **Callaway E (2020):** Time to use the p-word? Coronavirus enters dangerous new phase. https://pesquisa.bvsalud.org/global-literature-on-novel-coronavirus-2019-ncov/resource/pt/covidwho-2009
8. **Chamling B, Vehof V, Drakos S et al. (2021):** Occurrence of acute infarct-like myocarditis following COVID-19 vaccination: just an accidental co-incidence or rather vaccination-associated autoimmune myocarditis?. Clinical Research in Cardiology, 110 (11): 1850-1854.
9. **Chen, Y, Guo D (2016):** Molecular mechanisms of coronavirus RNA capping and methylation. Virologica Sinica, 31 (1): 3-11.

10. **Cucinotta D, Vanelli M (2020):** WHO declares COVID-19 a pandemic. Acta Bio Medica: Atenei Parmensis, 91 (1): 157.

11. **Cui J, Li F, Shi Z (2019):** Origin and evolution of pathogenic coronaviruses. Nature reviews microbiology, 17 (3): 181-192.

12. **Fehr A, Perlman S (2015):** Coronaviruses: an overview of their replication and pathogenesis. https://cir.nii.ac.jp/crid/1573950398846102272

13. **Greinacher A, Thiele T, Warkentin T et al. (2021):** Thrombotic thrombocytopenia after ChAdOx1 nCov-19 vaccination. New England Journal of Medicine, 384 (22): 2092-2101.

14. **Habas K, Nganwuchu C, Shahzad F et al. (2020):** Resolution of coronavirus disease 2019 (COVID-19). Expert review of anti-infective therapy, 18 (12): 1201-1211.

15. **Kahn J, McIntosh K (2005):** History and recent advances in coronavirus discovery. The Pediatric infectious disease journal, 24 (11): S223-S227.

16. **Khosla S, Nylen E, Khosla R (2020):** Rhabdomyolysis in patients hospitalized with COVID-19 infection: five case series. Journal of investigative medicine high impact case reports.DOI: https://doi.org/10.1177/2324709620984

17. **Kimura M, Niwa J, Doyu M (2022):** Recurring Weakness in Rhabdomyolysis Following Pfizer–BioNTech Coronavirus Disease 2019 mRNA Vaccination. Vaccines, 10 (6): 935.

18. **Mahmood Z, Fadhil H, Ad A(2021):** Estimation of Hematological Parameters of Disease Severity in Iraqi Patients with COVID-19. Iraqi Journal of Science, 62 (10): 3487-3496 DOI: https://doi.org/10.24996/ijs.2021.62.10.8

19. **Nassar M, Chung H, Dhayaparan Y et al. (2021):** COVID-19 vaccine induced rhabdomyolysis: case report with literature review. Diabetes & Metabolic Syndrome, 15 (4): 102170.

20. **Neuman B, Adair B, Yoshioka, C et al. (2006):** Supramolecular architecture of severe acute respiratory syndrome coronavirus revealed by electron cryomicroscopy. Journal of virology, 80 (16): 7918-7928.

21. **Oliver I (1955):** A spectrophotometric method for the determination of creative phosphokinase and myokinase. https://pubmed.ncbi.nlm.nih.gov/13260184

22. **World Health Organization (2020):** Coronavirus disease 2019 in children—United States, february 12–april 2, 2020. Morbidity and Mortality Weekly Report, 69 (14): 422

23. **World Health Organization (2020):** mRNA vaccines against COVID-19: Pfizer-BioNTech COVID-19 vaccine BNT162b2: prepared by the Strategic Advisory Group of Experts (SAGE) on immunization working group on COVID-19 vaccines, 22 December 2020 (No. WHO/2019-nCoV/vaccines/SAGE_evaluation/BNT162b2/2020.1). World Health Organization.

24. **Song Z, Xu Y, Bao L et al. (2019):** From SARS to MERS, thrusting coronaviruses into the spotlight. viruses, 11 (1): 59.

25. **Suwanwongse K, Shabarek N (2020):** Rhabdomyolysis as a presentation of 2019 novel coronavirus disease.Cureus, 12(4): 7561

26. **Sweeney H (1994):** The importance of the creatine kinase reaction: the concept of metabolic capacitance. Medicine and science in sports and exercise, 26 (1): 30-36.

27. **Tetsuka S, Tominaga,K, Ohta E et al. (2013):** Paraneoplastic cerebellar degeneration associated with an onconeural antibody against creatine kinase, brain-type. Journal of the Neurological Sciences, 335 (1-2): 48-57.

28. **Wang L, Xiang Y (2020):** Spike glycoprotein-mediated entry of SARS coronaviruses.Viruses, 12 (11): 1289.

29. **Wang L (2006):** F, Shi Z, Zhang S, Field H, Daszak P, Eaton BT. Review of bats and SARS. Emerg Infect Dis., 12 (12): 1834-1840.

30. **Woodworth K, Moulia, D, Collins J et al. (2021):** The advisory committee on immunization practices' interim recommendation for use of Pfizer-BioNTechCOVID-19 vaccine in children aged 5–11 years—United States, November 2021. Morbidity and Mortality Weekly Report, 70 (45): 1579.

31. **Zhou, P, Yang X, Wang X et al. (2020):** A pneumonia outbreak associated with a new coronavirus of probable bat origin. nature, 579 (7798): 270-273.