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Levels of Some Blood Clotting Factors in COVID-19 Patients: Comparative Study Before and After Pfizer- BioNTech Vaccinations

Lubna A.A. Al-Assaf*, Yasser A.H. Al-Issa
Department of Chemistry, College of Science, University of Baghdad, Baghdad, Iraq

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Abstract

One of the health issues that a coronavirus can induce is blood clotting. Coronavirus can be prevented in a number of ways. Vaccination is one of the critical methods for preventing illness or lessening its impact. This study seeks to estimate a few blood coagulation variables. 147 samples were collected from the Baghdad Governorate in the autumn of 2021. The samples were split into three groups: COVID-19 patients, healthy individuals before and after receiving the (Pfizer-BioNTech) vaccine, and healthy individuals only. Prothrombin Time (PT), Partial Thromboplastin Time (PTT), Protein C (PTN-C), Protein S (PTN-S), and International Normalized Ratio (INR) for 49 samples were measured and computed for each group. The results have shown that the PTN-C and PTN-S concentrations were significantly decreased in the COVID-19 patients compared to unvaccinated healthy individuals. While INR of COVID-19 patients showed a highly significant increase when compared with unvaccinated healthy individuals. At the same time, there was no significant difference for each PT and PTT between these studied groups. The parameters exhibit identical findings when COVID-19 patients are compared to those who have had vaccinations, with the exception of PT, which reveals a highly significant rise in COVID-19 patients when compared to those who have received vaccinations. Conclusion: COVID-19 causes blood clots and may be recognized by a decrease in PTN-C and PTN-S content.

Keywords: Protein C, Protein S, Prothrombin Time, Partial Thromboplastin Time, International normalized ratio, COVID-19, and Pfizer- BioNTech

مستويات بعض عوامل تخثر الدم لدى مرضى كوفيد-١٩ : دراسة مقارنة قبل وبعد التلقيحات من فايزر-بيونتيك

لبنى عامر عبد الوهاب*, ياسر عبدالحسين جعفر
قسم الكيمياء, كلية العلوم, جامعة بغداد, بغداد, العراق

الخلاصة

يمكن ان يتسبب فيروس كورونا في العديد من المشاكل الطبية ؛أحد هذه المشاكل هي تخثر الدم. هناك عدة طرق لتجنب الإصابة بفيروس كورونا اهمها الحصول على اللقاح. يعتبر اللقاح احدى طرق الوقاية من العدوى أو تقليل حدتها. تهدف هذه الدراسة إلى تقدير بعض عوامل التخثر في الدم. تم الحصول على 147

*Email: lubnaalassaf6@gmail.com

عينة في فصل الخريف لعام 2021 م من محافظة بغداد .وتم تقسيم العينات إلى ثلاث مجموعات : المرضى المصابين بكورونا ومجموعة افراد اصحاء قبل وبعد التطعيم بلقاح (فايزر– بيونتيك). تم تحليل وحساب تركيزالبروتين سي (PTN–C) ، والبروتين اس (PTN–S) ، ووقت البروثرومبين (PT) ، ووقت الثرومبوبلاستين الجزئي (PTT) والنسبة الطبيعية الدولية (INR) لـ 49 عينة لكل مجموعة. انخفضت تركيز كل من PTN–C و PTN–S بشكل ملحوظ في المرضى المصابين بفيروس كورونا مقارنة بالأفراد الأصحاء غير الملقحين. بينما أعطى INR لمرضى فيروس كورونا زيادة كبيرة للغاية مقارنة بالأفراد الأصحاء غير الملقحين. في الوقت نفسه ، لم يكن هناك فرق معنوي لكل PT و PTT بين هاتين المجموعتين. وعند إعادة المقارنة بين المرضى المصابين بالفيروس كورونا مع الذين حصلوا على اللقاح ، تظهر المعلمات نتائج مماثلة للمقارنه السابقة باستثناء PT الذي يظهر زيادة كبيرة للغاية في المرضى المصابين بفيروس كورونا مقارنة مع الملقحين. يمكن أن نستنتج أن مرض فيروس كورونا يتسبب في حدوث جلطات دموية ويمكن التعرف عليه من خلال انخفاض تركيز PTN–C و PTN–S.

1. Introduction

A virus is a tiny, biologically inert infectious entity that lives outside of the host cell and reproduces inside of living host cells [1]. Thousands of identical copies of the original virus are quickly produced by the host cell as a result of infection. It differs from other living species, whether they are eukaryotes or prokaryotes, for three primary reasons: (a) the nature of the environment in which it grows and multiplies, (b) the nature of its genome, and (c) the method of their multiplication [2].

Amphibians, birds, and mammals are all susceptible to the Coronaviridae family of viruses, which are encased in a single positive-strand RNA genome [3]. They are members of the sizable viral group known as Nidovirales. (a) Arteriviridae, (b) Coronaviridae, (c) Mesoniviridae and (d) Roniviridae each represent one of four families. Two more subfamilies, Coronavirinae and Torovirinae, are added to the second family (Coronaviridae) [4]. Large genomic RNA (26000-32000) bases and spike-shaped glycoprotein mutations that are extruded from the surface of the viral envelope are two structural characteristics of coronaviruses. The term "coronaviruses" comes from spikes, which give the viruses a characteristic ultra-structural feature that mimics the solar corona [5]. Compared to other viruses, COVID-19 has a relatively high mutation capacity, which makes it difficult to design specialized diagnostic tools, therapies, and vaccinations [6].

Coronavirus can be avoided in a number of methods, one of which is vaccination [7]. Its biological formulation offers effective acquired defence against a certain infectious disease [8]. It often comprises pure products made from spiked proteins or mRNA from viruses that have been attenuated, inactivated, or dead [9]. It causes the immune system of the body to recognize a threat, get rid of it, and then recognize and get rid of any germs it may come into contact with in the future [10].

Blood produces cohesive blood groupings called blood clots during the intricate process of blood coagulation, which stops bleeding [11]. Coagulation, protein activation, adhesion and platelet aggregation, fibrin deposition and maturation, as well as bleeding from the injured artery being stopped and eventually repaired, are all parts of this process [12].

Various substances are required for the proper functioning of the coagulation cascade known as cofactors such as calcium and phospholipids, Vitamin K [13]. PTN-C can inhibit the activities of factors Va (Labile factor) and VIIIa (Antihacmophilic factor), while PTN-S is cofactor for Activated Protein C (APC, inactive when bound to C4b-binding protein) [14].

Prothrombin Time (PT) and Partial Thromboplastin Time (PTT) measure the following coagulation factors: I (fibrinogen), II (prothrombin), V (Labile factor), and X (Stuart–Prower factor). PT is more selective for VII (proconvertin) in comparison to PTT which is selective for VIII (anti-haemophilic factor), XI (plasma thromboplastin antecedent), and XII (Hageman factor) [15].

This study intends to examine how COVID-19 and its vaccination affect levels of certain blood clotting factors in individuals who have the virus and in those who received the vaccine both before and after.

2. Materials and Methods

In this study, samples were obtained over the course of four months, beginning in September. In the first week of hospital admission, 49 samples from both genders of COVID-19 patients (first time infected) with a mean age of (50 ± 12) years were chosen from Al-Specialized Shifa's Crisis Centre, Private Nursing Hospital, and Noor Al-Kawthar laboratory, depending on them. The second samples were collected after receiving the second dosage of the Pfizer vaccine, which was administered 56 days after the first dose and one week following the second dose. With excluded the people who infected with COVID-19 before vaccination. All samples were collected according to the controls and instructions of the Iraqi Ministry of Health (IMoH), after obtaining the approval of patients and individuals. The plasma PTN-C and PTN-S concentrations were measured for all groups by sandwich ELISA kit (MyBioSource Company / Vancouver, British Columbia, Canada) [16, 17] and the plasma PT and PTT were measured by manual method for (Spinreact Company / Girona, Spain) [18] while INR was calculated by using the following equation:

$$INR = \left(\frac{PT\ (sec)}{PT\ reference\ (sec)}\right)^{ISI}$$

INR = International normalized ratio
PT = Prothrombin time
PT reference = Established as the geometric mean of the prothrombin times (PT) of a reference sample group [18]

3. Statistical Analysis

The statistical analysis was conducted using IBM SPSS version 25 statistical software. The difference between COVID-19 patients and healthy individuals before and after vaccination was analysed using the independent t-test. A value of $p<0.05$ was considered statistically significant and $p<0.01$ was considered a highly significant.

4. Results

Statistical analysis of PTN-C, PTN-S concentration, PT, PTT and INR of two groups COVID-19 and healthy peoples before vaccination was performed. The data indicated two things: first, there was a very significant drop in PTN-C and PTN-S concentration, whereas COVID-19 patients' INR exhibited a highly significant rise when compared to healthy, uninfected people. According to Table (1), PT and PTT do not significantly differ across the groups.

Table 1: Comparisons between the studied coagulation factors of COVID-19 patients' group and healthy subject group before vaccination

Parameters	Groups	N	Mean	±SD	*P-value*
Conc. PTN-C (µg/ml)	COVID-19 Patients	31	1.20	0.27	0.000**
	Before Vaccination	31	2.39	0.85	
Conc. PTN_S (µg/ml)	COVID-19 Patients	31	1.03	0.30	0.001**
	Before Vaccination	31	1.70	0.32	
PT (sec)	COVID-19 Patients	49	14.81	2.24	0.637
	Before Vaccination	49	14.59	2.40	
PTT (sec)	COVID-19 Patients	49	31.15	6.84	0.205
	Before Vaccination	49	29.34	7.25	
INR	COVID-19 Patients	49	1.20	0.24	0.000**
	Before Vaccination	49	1.01	0.20	

**** Significant at p<0.05 for 2-tailed.***
***** Significant at p<0.01 for 2-tailed***

Table (2) shows the comparison between COVID-19 patients' group and healthy individual group after vaccination. The results revealed presence of high significant decrease in the concentration of PTN-C and PTN-S contrary to the findings of PT and INR a high significant increase, while PTT shows no significant difference.

Table 2: Statistical analysis of some coagulation factors of COVID-19 patients' group and the after-vaccination group

Parameters	Groups	N	Mean	±SD	*P-value*
Conc. PTN-C (µg/ml)	COVID-19 Patients	31	1.20	0.27	0.000**
	After Vaccination	31	2.45	1.11	
Conc. PTN_S (µg/ml)	COVID-19 Patients	31	1.03	0.30	0.006**
	After Vaccination	31	1.23	0.22	
PT (sec)	COVID-19 Patients	49	14.81	2.24	0.000**
	After Vaccination	49	12.24	2.72	
PTT (sec)	COVID-19 Patients	49	31.15	6.84	0.113
	After Vaccination	49	28.89	7.17	
INR	COVID-19 Patients	49	1.20	0.24	0.000**
	After Vaccination	49	0.79	0.25	

**** Significant at p<0.05 for 2-tailed.***
***** Significant at p<0.01 for 2-tailed***

This study demonstrated that, for all of the measures PTN-C, PTN-S, PTT, and INR, the comparison between the group of COVID-19 patients and the vaccinated group yielded the same findings, with the exception of the PT, which showed a highly significant drop.

5. Discussion

Due of the pandemic's devastating mental and economic effects, inoculation against COVID-19 is crucial. Because of the relatively new mRNA technology utilized in the BioNTech Pfizer vaccine, there has been substantial public debate. However, several nations employ the vaccination since it has been shown to be successful in preventing COVID-19 [19].

According to the PTN-C and PTN-S data mentioned above, individuals with COVID-19 had thrombosis following well monitored significant fibrin accumulation in the lungs and circulation [20]. Contrarily, PTN-C with cofactor PTN-S inactivates factors VIII (anti-haemophilia factor) and V (labile factor) [21] . Factor V participates in the process of converting prothrombin to thrombin when it functions as a co-factor. Although it is involved in activating factor X, factor VIII also functions as a co-factor [22]. Anticoagulants, a lack of coagulation factors, and the use of fibrinolysis as a laboratory technique to predict bleeding are all associated with prolonged PT and PTT [23].

In the present investigation, along with six other studies [24-29], it was discovered that there are no consistent, significant differences between coronavirus patients and healthy individuals for each of the parameters, PT and PTT (p>0.05). Failure of PT and PTT to detect COVID-19 early was likely driven by many reasons (including anticoagulants) [30]. Another explanation for this failure is that the intensity of the infection with the coronavirus might vary depending on a number of parameters, including age, chronic conditions, and co-morbidities with COVID-19. This reinforces the likelihood of varying results depending on other circumstances [31].The high value of INR indicates the presence of coagulopathy is highly significantly with severe disease and increased mortality in patients with COVID-19 [32]. It has been suggested that elevation of INR can be used as rapid and inexpensive diagnosis for COVID-19 infection [15].

6. Conclusions

The study's conclusions include that COVID-19 infection causes blood clots and may be recognized by a decline in PTN-C and PTN-S concentrations, which block factors V (Labile factor) and VIII (anti-hemophilic factor). This study supports the idea that a high INR may be utilized to make a precise and rapid diagnosis of COVID-19 infection. When comparing the effects of various variables on COVID-19 patients to healthy individuals, these data also offer further information and proof that PT and PTT have no discernible impact on the clotting process in COVID-19 patients.

Ethical Clearance

The Research Ethical Committee at scientific research by ethical approval of environmental, health, higher education, and scientific research ministries in Iraq.

Conflict of Interest

The authors declare that they have no conflict of interest.

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