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Studying the Risk Factors on COVID-19 Patients in Nineveh Governorate

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Abstract

The world witnessed a major crisis when the new Corona virus (Covid-19) appeared, and its effects are still continuing, as no gender, race, age, or color was excluded from infection, and given the disease represented by a pandemic that spread in most parts of the world in recent years and still poses a threat to Therefore, it is a fertile field for studies and to delve into it extensively. This research included a study of the effect of infection times, obesity, smoking and age on the levels of enzymes: lactoperoxidase (LP), lactate dehydrogenase (LDH), alanine aminotransferase (ALT), and aspartate aminotransferase (AST), as well as measuring interleukin-6 (IL-6). In Nineveh Governorate, (100) samples were collected from Covid patients (50) and a healthy group (50), whose ages ranged between (20-70) years and for both sexes. It was observed that there was a high significant increase (0.0001) in all the measured enzymes (LP, LDH, ALT and AST) as well as an increase in the level of IL-6 when comparing the total COVID-19 patients with the control group. The results showed that there was a significant increase in the levels of different enzymes (LP, LDH, ALT and AST), as well as IL-6 level in patients with COVID-19 aged 40 years and over, compared with the group of patients aged (20-39) years, as well as among patients who smoked compared to with non-smokers and control group. The results showed that there was a high significant increase in all variables measured in patients with obesity, more than patients with normal weights and control group. In addition, it was observed that there was a high significant increase in activity of LP when the number of infections increased in patients with Covid-19, and an increase in the activity of LDH, ALT and AST and the level of IL-6 in patients with Covid infected once compared with patients infected more than once and when compared with control group. The study concluded that there are clear effects of the levels of measured enzymes (LP, LDH, ALT, AST) and level of IL-6 in patients with COVID-19, and the factors of age, smoking, obesity, and the incidence of infection for one time had the greatest effect on the level of the measured variables, which reflect the increase in the severity of the disease in infected patients.

Introduction

Corona virus is a zoonotic virus that turned into isolated for the primary time in 1937 and identified. Corona virus belongs to the family Coronaviridae. It has crown-like protuberances at the outer floor of the virus, and on the premise of that it became referred to as the Corona

virus, and it's microscopic size, ranging in diameter among (65-125) nanometers, and it possesses a simple positive-sense single-stranded RNA as a pretty massive nuclear fabric whose size degrees between (26 -32) kb, and there are four genera of the coronavirus own family, which might be α, β, γ and δ viruses [1]. Bats and mice are a source of alpha and beta coronaviruses, while birds It is a source of delta and gamma coronaviruses [2].

Covid-19 it is dangerous highly contagious viral disease that caused a global pandemic is caused by a new type unknown strain of coronavirus, and it became known as severe acute respiratory syndrome coronavirus 2 (SARS-Cov-2) depending at the evolutionary courting with the Corona virus that It became the purpose of the outbreak of excessive acute breathing syndrome SARS in 2003, and Covid ailment turned into first observed at some stage in its outbreak in past December of the 12 months 2019 in Wuhan, Hubei Province, China, and then unfold all around the international, and this virus is one of the new lines of coronaviruses [3], and the incubation period ranges for the virus between 2 to 7 days and might extend to 14 days [4], and symptoms vary among not unusual inclusive of fever, dry cough, and shortness of breath similarly to secondary symptoms, inclusive of diarrhea and irritation of the intestines, might also attain a greater severe circumstance, together with CHF(Congestive heart failure) and death [5].

The virus binds via spike proteins to ACE2 (Angiotensin-converting enzyme 2) receptors all through its entry into the body [3]. After coming into the cell, it removes its capsid and edits the RNA genome to bind to the ribosome that allows you to translate it through having the enzyme RNA-based polymerase, and this lets in the genome to be copied. The virus produces new copies with the assist of the host cellular [6], the RNA genome is transcribed into along polypeptide containing all proteins linked together, because the virus has a protein peptidase which can separate proteins from every other in this translated chain, and this method of transcription, translation of proteins is a type of genetic and viral economic system that permits it to encode a huge range of genes in a as an alternative small number of nucleotides [1]. Many host-related factors were speculated to be a risk factors for the severity of disease [7] such as obesity, smoking, and age. Obesity is the maximum common underlying condition for COVID-19 patients Those under the age of, 64 years, and the danger of the COVID-19 virus can also make bigger to more youthful a long time. Accordingly, obesity ends in an growth in chance elements for these diseases and consequences in incredible results [8].

Smoking and electronic cigarette use increases the threat and severity of lung infections because of damage to the top airways and decreased pulmonary immune feature. Smokers are 1.91 instances morelikely to expand intense ailment with COVID-19 than non-smokers, because of the damaging consequences of smoking on pulmonary immune feature [9]. Age is a robust chance issue for the severity and results of the COVID-19 virus, as the proportion of humans with immunodeficiency is related to the age shape of that organization of society, as research conducted in China confirmed that maximum of those inflamed with the COVID-19 virus are aged age [10].

The studies objectives to examine the impact of danger elements consisting of the number of infections, obesity, smoking, and age on the activity of some enzymes and the level of IL-6 in patients in Nineveh Governorate, and comparing them with the control.

Materials and Methods

Sample collection

Collecting blood (serum) samples from patients with Covid-19 disease after they were clinically diagnosed, as well as the result of the (PCR) examination was positive and under the supervision of doctors specializing in respiratory diseases at Al-Shifa Hospital for Chest Diseases and Fever (Isolation Hospital) in the city of Mosul for the period between August 2022 and the end of the month January 2023, and after writing down the required information from the patients (age, gender, weight, height, smoking and number of infections), (100) samples were collected, including (50) samples of people with Covid-19 and (50) samples from healthy people considered as a control group. The ages ranged from (20-70) years for both sexes, in which the enzymes: LP,LDH, ALT and AST were measured, as well as IL-6.

LP activity was estimated using the colorimetric method used by the researcher Tayefi-Nnasrabadi et al. [11], as the enzyme oxidizes pyrogallol in the presence of hydrogen peroxide to turn it into purpurogallin. The activity of LDH was estimated using a kit from the Spanish company Spinreact (REF: 1001261) and depended on the spectroscopic method through the decrease in NADH concentration [12], and the effectiveness of the ALT & AST enzymes was estimated using kit from the English company Randox (Cat.No.HNI530), which is based on the colorimetric method for measuring enzyme activity [13]. The level of IL-6 was estimated using kit from Boditech Korean company (REF:FPRR020) (AFIAS) IL-6 based on the Flow injection analysis method, using the sandwich method for immunological detection. All the results were analyzed statistically using the statistical program (SPSS 25) and finding the difference between the values that appeared using the probability value (P) that occurs at $P \leq 0.05$ significant difference.

Results and Discussion

1. Comparison of patients with a control group

Table (1) indicated that there was a high significant increase in all measured enzymes (LP, LDH, ALT, AST) and IL-6 at a probability value less than (0.0001) in COVID patients.

The Covid-19 virus objectives the respiration machine, specifically the lung [14], because the LP enzyme performs a shielding function towards any viral assault [15], the significant increase in LP levels is attributed to the resistance shown by the body towards infection with the Covid-19 virus.

LDH is one of the biomarkers for the prognosis of sufferers with COVID-19, as an elevated LDH stage has been connected to patients with different viral infections earlier [16]. Severe infection can also cause tissue damage by way of cytokine and LDH secretion. Given the presence of LDH in lung tissue (LDH-3), sufferers with severe COVID-19 infection might be anticipated to excrete extra quantities of LDH into the circulate, as a severe form of interstitial pneumonia often progressing to acute respiratory distress syndrome, which it is a feature of COVID-19 sufferers [17].

Table 1: Levels of enzymes and interleukin-6 in total COVID-19 patients.

Enzymes and IL-6	Control N=50		Covid patients N=50		P value
	mean	S.D	mean	S.D	
LP (U/ml)	51.38	13.88	109.79	15.13	0.0001***
LDH (U/L)	296.04	27.23	794.90	94.66	0.0001***
ALT (U/L)	7.37	0.87	35.58	3.47	0.0001***
AST (U/L)	5.64	0.73	29.49	2.95	0.0001***
IL-6 (Picogram/ml)	2.87	0.32	892.94	32.94	0.0001***

*** indicates that there is a significant difference at a probability level less than 0.0001 between the studied groups.

The high activity for ALT and AST are due to the fact that they are present in different quantities in the kidneys, heart and muscles, unlike the liver, which contains large amounts of ALT. Any injury to the liver cells can lead to an increase in the level of ALT enzyme, such as viral hepatitis and liver damage caused by toxins. While the AST enzyme is found in the highest concentration in heart compared to liver, skeletal muscles and kidneys [18]. The liver damage associated with COVID-19 can be explained by several mechanisms.

Interleukin (IL-6) is made by a variety of cells in the lung, including pulmonary type T lymphocytes, and lung fibroblasts. IL-6 is a multidirectional cytokine important in the regulation of immune and inflammatory responses. Its presence in high concentrations is an indication of pneumonia. This is supported by the often observed increase in IL-6 in acute respiratory distress syndrome and acute complications of lung transplantation [19]. It has been suggested recently that IL-6, which is likely derived from inflammatory monocytes, may be responsible for severe lung inflammation and impaired lung function in patients with severe COVID-19 who require more intensive care and treatment due to lung damage [4], patients with severe COVID-19 require more effective treatment. Elevation of interleukin-6 correlates with disease severity and course, as it increases with disease progression. Therefore, IL-6 may be a potential marker for disease surveillance in severe COVID-19 [19].

2: Obesity effect on COVID-19 Patients:

High significant increase in the levels of different enzymes (LP, LDH, ALT and AST), as well as a rise in the level of IL-6 in patients with obesity, more than patients with normal weights and control group, as noted in Table 2.

Table 2: Comparison of the control group with normal-weight and obese COVID-19 patients.

Enzymes and IL-6	Control N=50		Covid-19 patients with normal weight N=32		Covid-19 patients with Obesity N=18		P value
	mean	S.D	Mean	S.D	mean	S.D	
LP (U/ml)	51.38 a	13.88	95.83 b	19.10	124.16 c	17.81	0.0001***
LDH (U/L)	296.04 a	27.23	577.50 b	57.27	829.16 c	79.38	0.0001***
ALT (U/L)	7.37 a	0.87	18.79 b	2.49	35.00 c	4.50	0.0001***
AST (U/L)	5.64 a	0.73	19.45 b	18.52	31.00 c	25.40	0.0001***
IL-6 (Picogram/ml)	2.87 a	0.32	545.58 b	74.16	879.00 c	78.79	0.0001***

*** indicates that there is a significant difference at a probability level less than 0.0001 between the studied groups.

People with a body mass index (BMI) greater than 25 (kg/m2) are at increased risk of severe pneumonia associated with higher LDH levels. In-hospital deaths were higher in overweight patients and morbidly obese individuals [20], Studies have shown that people who suffer from obesity are more likely to be admitted to the hospital and intensive care unit or die, as it has been observed that there is a probability of hospitalization more than 6 times compared to adults of normal weight. In disease exacerbation in viral infections, obesity is associated with an increase in the frequency of respiratory tract infections [21], as the final products of the LP enzyme system, thiocyanate (SCN-) indicate that they are considered as parameters of cardiovascular and pulmonary infections and may be This is one of the reasons for the high LP enzyme, which is considered obesity as a risk factor for such infections [22], and as shown by all previous studies, which considered that obesity is a risk factor for Covid disease because it is one of the causes chronic diseases such as diseases of blood pressure, blood vessels and diabetes mellitus, and this is what the results showed in Table 2. We note that for those with Covid who have weight gain compared to Covid patients with normal weights when compared with the control group, and this confirms that obesity is a factor severity of COVID-19.

Obesity in for case of infection with the COVID-19 virus crystallizes in an increase in ACE2 in fat cells, which leads to the spread of targeting the rest of the body's organs, including the liver, and this is clear evidence of an increase in ALT and AST enzymes. Which leads to an increase in proteins associated with inflammation, including interleukin-6 (IL-6), and obesity causes a dysfunction in the immune system, such as increased insulin and leptin resistance, and weakness in T cells and macrophages. This negatively affects the ability of the immune system to resist foreign bodies, including the Covid virus. -19 [23].

3. Smoking effect on COVID- 19 patients

A significant increase in the levels of different enzymes (LP, LDH, ALT and AST), as well as a rise in the level of IL-6 in patients with smokers, compared with non-smokers and the control group. As Table (3) shows, the increase in the levels of the aforementioned enzymes, in addition to interleukin-6, is strikingly higher in the smokers group, which may be due to the high oxidative stress in smokers, which leads to an increase in infections resulting from infection.

Table 3: Comparison of the control group with smoking and non- smoking COVID-19 patients.

Enzymes and IL-6	Control N=50		Covid-19 patients Non -smoking N=30		Covid-19 patients Smoking N=20		P value
	mean	S.D	mean	S.D	mean	S.D	
LP (U/ml)	51.38 a	13.88	100.00 b	18.55	115.53 c	18.60	0.0001***
LDH (U/L)	296.04 a	27.23	703.33 b	42.07	742.07 c	41.65	0.0001***
ALT (U/L)	7.37 a	0.87	26.89 b	4.60	29.30 c	5.24	0.0001***
AST (U/L)	5.64 a	0.73	25.47 b	2.65	28.23 c	4.82	0.0001***
IL-6 (Picogram/ml)	2.87 a	0.32	712.29 b	42.65	822.76 c	89.50	0.0001***

*** The difference in the letters (a, b, c) horizontally indicates that there is a significant difference at a probability level less than or equal to 0.0001 between the studied groups.

Tobacco smoke is a complicated aggregate of more than 5,000 chemical substances, carcinogens, and pollutants. Smoking is a chance component for the improvement and exacerbation of multiple respiration sicknesses, which includes infections. In specific, the liver is a clear out to eliminate these pollution, and smoking contributes to the deposition of fat at the liver, which quickens its poor performance and failure. It is likewise an impartial threat component for community-received pneumonia due to disruption of breathing epithelial repair and reduced clearance of micro organism from the airways [24].

Strong proof shows that numerous mechanisms may boom the hazard of breathing tract contamination in people who smoke. Smoking weakens the immune gadget and almost doubles the danger of contracting tuberculosis (each latent and active). Specifically, smoking affects the macrophage and mobile reaction and as a result the capability to comprise infection. Similarly, the chance of contracting pneumococcus and streptococcus is set 3-5 times better in people who smoke. Smokers also are five times much more likely to trap influenza than non - smokers [25].

Smoking leads to changes in airway biology [26], as it has been shown that cigarette smoke increases IL-8 production and decreases IL-8. -10, from airway epithelial cells which is

one of the sites of human LP enzyme when stimulated experimentally with human rhinoviruses and this can lead to altered immune cell [27].

Moreover, cigarette smoke is also known to decrease serum IgM levels and raise IgM levels in the lungs [28]. Smoking has been shown to increase the expression of intercellular adhesion molecule-1 (ICAM-1), a known receptor of human rhinoviruses [29]. This explains the high LP enzyme that boosts the immune system.

Most studies indicated that the mechanism of entry of the Covid-19 virus into the host cell is through ACE2 receptors, which smoking is considered a risk factor for COVID-19 disease, as smoking increases ACE2 levels, and this gives a greater opportunity for the virus to enter the host cell [30] In addition to the above, smoking is considered the most important cause of poor lung efficiency, which leads to high levels of LDH and lack of oxygen, which leads to a weak immune system, increased inflammation, and high levels of interleukin-6, which portends greater risks when contracting Covid-19 [31].

4. Age effect on COVID-19 patients

Increase in activity of enzymes (LP, LDH, ALT and AST), as well as a rise in the level of IL-6 in patients with COVID-19 aged 40 years and over, compared with the group of patients aged (20-39) years.

Individuals of all ages are at risk of contracting COVID-19 and risk of severe consequences. However, the risk of severe COVID-19 infection is increased in elderly patients over 60 years of age and those with underlying medical conditions or risks (Obesity, cardiovascular disease, smoking, and cancer) [32]. With age, the ability of the liver to filter a lot of drugs and cholesterol decreases, and thus leads to cirrhosis of the liver, which leads to high activity of ALT and AST. Also, with age, oxidative stress increases in the elderly, which increases inflammation and a rise in cytokine storm, including interleukin. -6 (IL-6) [33].

Table 4: Comparison of COVID-19 patients between ages groups.

Enzymes and IL-6	20-39 years N=24		40 years and over N=26		P value
	mean	S.D	mean	S.D	
LP (U/ml)	94.20	13.34	104.14	11.01	0.021*
LDH (U/L)	629.70	46.64	755.92	67.12	0.037*
ALT (U/L)	23.25	2.83	29.50	1.31	0.045*
AST (U/L)	20.20	1.82	29.53	2.72	0.035*
IL-6 (Picogram/ml)	519.30	59.29	850.14	83.53	0.0014*

* indicates that there is a significant difference at the level of probability less than or equal to 0.05 between the studied groups.

With the continued spread of the disease, an increase in cases was observed among people aged 65 years and over, and the number of males was higher [14]. According to the data of the Chinese Center, for Disease Control and Prevention (China, CDC) [34], which was published in February 2020, the percentage of children infected with Covid-19 since the

beginning of the disease outbreak was small, as children under the age of 10 years, as well as the age group between (11-19) years, their percentage was 1% of all cases [35]. Since this age group represents 20% of the total population, this indicates a low prevalence rate of COVID-19 among children.

The reason for the lack of infection can be attributed to the lack of corona examination on children, as well as the lack of symptoms associated with the virus, and the young age groups have less incidence of chronic diseases compared to the elderly, which negatively affect the immune system, which leads to exacerbation of infection and the occurrence of acute pneumonia, which is accompanied by high levels of LDH [14]. This confirms what the results indicated in Table 4, as we notice a high increase in the level of measured enzymes in addition to interleukin-6 in the older age group of people with Covid-19, who are more at risk of severe infection due to their weak immune system and are also exposed to chronic diseases that double their risk factor. compared to the same patients with younger ages.

5. The effect number of infections on COVID -19 patients

A high significant increase in different levels of the enzyme (LP) when the number of infections increased in patients, and an increase in the activity of LDH, ALT and AST enzymes and the level of IL-6 in patients who were infected once compared with patients who were infected more than once, as well as when compare them with the control group.

Table 5: Comparison of the control group with COVID-19 patients with one infection and patients with more than one infection.

Enzymes and IL-6	Control N=50		COVID-19 patients with one infection N=33		COVID-19 patients with more than one infection N=17		P value
	mean	S.D	Mean	S.D	mean	S.D	
LP (U/ml)	51.38 a	13.88	99.22 b	17.06	109.33 c	21.43	0.0001***
LDH (U/L)	296.04 a	27.23	776.86 c	33.13	531.91 b	44.99	0.0001***
ALT (U/L)	7.37 a	0.87	29.77 c	2.74	17.62 b	1.41	0.0001***
AST (U/L)	5.64 a	0.73	27.42 c	2.34	16.83 b	1.50	0.0001***
IL-6 (Picogram/ml)	2.87 a	0.32	825.90 c	21.93	607.16 b	34.57	0.0001***

*** The difference in the letters (a, b, c) horizontally indicates that there is a significant difference at a probability level less than or equal to 0.0001 between the studied groups.

Recovery from infection with Covid-19 does not mean not being infected a second or third time. The first infction with Covid -19 provides protection against reinfection with an effectivness rate that may reach 80% during the same period, while the presence of IgG immunoglobulin antibodies is evidence of the immune memory for a period of time. It extends to eight months [36].

These considerations are whether the second infection is with the same virus as the first infection, because the immune system is aware of the protein of the virus that it dealt with previously, but the infection may be more severe if there is a mutation in SARS-COV-2, as the antibodies cannot recognize it, which makes it worse. It negatively affects immune memory, and repeated infections with Covid-19 are usually less severe than the first infection as the body has shown some immune response to the disease [37], and this explains the rise in enzyme levels (LDH, ALT, AST) and IL-6 in the first infection compared to the second and third infection. This indicates that the levels of inflammation in the second infection are lower, which leads to a lower rise of IL-6, and since the severity of the disease is less severe than the first infection, as we notice LDH levels are lower than its levels in the first infection, as well The severity of the disease is less stressful on the liver, which makes the levels of ALT and AST enzymes less high than the levels in the first infection.

Conclusions

The results showed that the Covid-19 virus, which causes acute respiratory distress, and the target organ is the lung in the first place, but it has serious repercussions on the rest of the body's organs. All age groups, but the elderly are more affected by the disease compared to the young group, because they are more susceptible to chronic diseases with age and less resistant to the risk of infection. The results also showed the danger of smoking and what it causes of weakening the efficiency of the lungs and lack of oxygen, that infection with Covid-19 for the first time and survival from it This does not make you isolated from getting infected with it a second and third time, but on the contrary, repeated injuries may be more severe.

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Conflicts of Interest: None.

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دراسة عوامل الخطورة على مرضى كوفيد-19 في محافظة نينوى

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الخلاصة:

شهد العالم ازمة كبيرة عند ظهور فايروس كورونا المستجد (كوفيد-19) ومازالت اثاره مستمرة، اذ لم يستثني لا جنسا، او عرقا، او عمرا، او لونا من الاصابة، ونظرا لما مثله المرض من جائحة انتشرت في معظم ارجاء العالم في السنوات الاخير ومازالت تمثل خطرا على البشرية لذا فهي مجال خصب للدراسات والتعمق فيها بشكل موسع. اذ تضمن هذا البحث دراسة تأثير عدد مرات الاصابة والسمنة والتدخين والعمر على مستويات الانزيمات كل من لاكتوبيروكسديز (LP) ولاكتيت ديهيدروجينيز (LDH) والانين امينوترانسفيريز (ALT) واسبارتيت امينوترانسفيريز (AST) فضلا عن قياس انترليوكين-6 (IL-6)، اجريت الدراسة في محافظة نينوى على (100) عينة من مرضى كوفيد (50) ومجموعة اصحاء (50) واعمارهم ما بين (20-70) سنة ولكلا الجنسين. لوحظ ان هناك ارتفاع معنوي عالي في جميع الانزيمات المقاسة (LP، LDH، ALT وAST) فضلا ارتفاعه ايضا لمستوى IL-6 عند مقارنة المرضى كوفيد-19 الكلي مع مجموعة السيطرة. اظهرت النتائج ان هناك ارتفاع معنوي بمستويات مختلفة لانزيمات (LP، LDH،ALT وAST) فضلا ارتفاعه ايضا لمستوى IL-6 لدى مرضى الكوفيد للاعمار 40 سنة فما فوق مقارنة مع مجموعة المرضى ذوي الاعمار (20-39) سنة وكذلك لدى مرضى الكوفيد المدخنين مقارنة مع غير المدخنين ومجموعة السيطرة. اظهرت النتائج ان هناك ارتفاع معنوي عالي لجميع المتغيرات المقاسة لدى مرضى الكوفيد ذوي السمنة أكثر من مرضى ذوي الاوزان الطبيعية ومجموعة السيطرة. فضلا عن ذلك لوحظ بحصول ارتفاع معنوي عالي بفعالية إنزيم (LP) عند زيادة عدد الاصابات لدى مرضى كوفيد-19، وارتفاع في فعالية انزيمات ALT،LDH وAST ومستوى IL-6 لدى مرضى الكوفيد المصابين لمرة واحدة مقارنة مع مرضى الكوفيد المصابين لاكثر من مرة فضلا عند مقارنتهم مع مجموعة السيطرة. استنتجت الدراسة ان هناك تأثيرات واضحة لمستويات الانزيمات المقاسة (LP، LDH، ALT وAST) ومستوى IL-6 وارتفاعها بمستوى عالي لدى مرضى كوفيد-19 وكانت لعوامل العمر والتدخين والسمنة وحدوث الاصابة لمرة واحدة التأثير الاكبر على مستوى المتغيرات المقاسة والتي تعكس زياد حدة المرض لدى المصابين.