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INVESTIGATION OF THE EFFECT OF THE VIRUS ON THE HEPATIC ENZYMES OF PATIENTS INFECTED WITH SARS-COV-2

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INVESTIGATION OF THE EFFECT OF THE VIRUS ON THE HEPATIC ENZYMES OF PATIENTS INFECTED WITH SARS-COV-2

By Dalal Majid Abdullah AL-ALI

January 2023

We certify that we have read this thesis and that in our opinion it is fully adequate, in scope and in quality, as a thesis for the degree of Master of Science

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ABSTRACT

INVESTIGATION OF THE EFFECT OF THE VIRUS ON THE HEPATIC ENZYMES OF PATIENTS INFECTED WITH SARS-COV-2

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Master of Science in Chemistry

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January 2023

The SARS-CoV-2 or Coronavirus Disease 2019 (COVID-19) outbreak was brought on by severe acute respiratory syndrome. The goal of the present study is to investigate the effect of SARS-CoV-2 on the levels of hepatic enzymes and other biochemical markers. Patients with COVID-19 hospitalized in AL-Najaf city/Iraq between February 7 and May 7, 2022, had their clinical features and laboratory results collected. 80 individuals with COVID-19 had their serum collected, along with 40 samples used as controls. In the patient group, IgG and IgM titers were assessed. Liver enzymes ALT, AST, ALP, total bilirubin (TBIL), ferritin, C.R.P, D.Dimer, and IL-6 were analyzed in two groups. In the study, it was found that a highly significant difference between G.O.T, G.P.T, and ALP, and a significant difference between TSB and indirect TSB, but there is no significant difference between direct TSB in different groups. As for the other parameters (ferritin, CRP, D.Dimer, and IL-6) were highly significant differences between all of them in different groups. The results showed that the majority of SARS-CoV-2 patients have aberrant hepatic enzyme activity, which may be associated with virus replication in the liver or drug-induced liver harm caused by antiviral medication.

2022, 50 pages

Keywords: SARS-CoV-2, COVID-19, Liver, Enzymes, Corona patients, Biochemical markers

ÖZET

SARS-COV-2 ENFEKTE OLMUŞ HASTALARIN HEPATİK ENZİMLERİ ÜZERİNE VİRÜSÜN ETKİSİNİN ARAŞTIRILMASI

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Ocak 2023

SARS-CoV-2 veya Coronavirus Hastalığı 2019 (COVID-19) salgını, şiddetli akut solunum sendromu ile ortaya çıktı. Bu çalışmanın amacı, SARS-CoV-2'nin hepatik enzimler ve diğer biyokimyasal belirteçler üzerindeki etkisini araştırmaktır. 7 Şubat - 7 Mayıs 2022 tarihleri arasında AL-Necef şehrinde/Irak'ta hastaneye yatırılan COVID-19 hastalarının klinik özellikleri ve laboratuvar sonuçları toplandı. COVID-19'lu 80 kişinin serumları toplandı ve 40 numune kontrol olarak kullanıldı. Hasta grubunda IgG ve IgM titreleri değerlendirildi. Karaciğer enzimleri ALT, AST, ALP, total bilirubin (TBIL), ferritin, C.R.P, D.Dimer ve IL-6 iki grupta analiz edildi. Çalışmada farklı gruplarda G.O.T, G.P.T ve ALP arasında oldukça yüksek, TSB ile indirekt TSB arasında anlamlı fark olduğu ancak direkt TSB arasında anlamlı fark olmadığı bulundu. Diğer parametrelerde ise (ferritin, CRP, D.Dimer ve IL-6) farklı gruplarda hepsi arasında oldukça anlamlı fark vardı. Sonuçlar, SARS-CoV-2 hastalarının çoğunun karaciğerde virüs replikasyonu veya antiviral ilaçların neden olduğu ilaca bağlı karaciğer hasarı ile ilişkili olabilecek anormal hepatik enzim aktivitesine sahip olduğunu gösterdi.

2022, 50 sayfa

Anahtar Kelimeler: SARS-CoV-2, COVID-19, Karaciğer, Enzimler, Corona hastaları, Biyokimyasal belirteçler

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Dalal Majid Abdullah AL-ALI

Çankırı-2022

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LIST OF SYMBOLS

%	Percent
±	Plus-minus
°C	Degrees celsius
µg	Microgram
µL	Microliter
g	Gram
mg	Milligram
mL	Milliliters
ng	Nanogram
nm	Nanometer

LIST OF ABBREVIATIONS

ACE2	Angiotensin-converting enzyme 2
ACE2R	Angiotensin-converting enzyme 2 receptor
ALI	Acute lung injury
ALP	Alkaline phosphatase
ALT	Alanin transaminase
APR	Acute phase reactant
ARDS	Acute respiratory distress syndrom
AST	Aspartate transaminase
C.T	Computed tomography
CDC	A center for disease control and prevention
COVID-19	Coronavirus disease 2019
CRP	C-reactive protein
DNA	Deoxyribonucleic acid
GGT	Gamma-glutamyltrans peptidase
GOT	Glutamic-oxaloacetic transaminase
GPT	Glutamic-pyruvic transaminase
I.N.R	International normalized ratio
IgG	Immunoglubulin-G
IgM	Immunoglobulin-M
IL-6	Interlukein-6
IU\L	International unit
MERS	Middle east respiratory syndrom
PCR	Polymerase chain reaction
PT	Prothrombin time
RBD	A receptor-binding domain
RNA	Ribonucleic acid
SARS	Severe acute respiratory syndrom
TSB	Total serum bilirubin
ULN	Upper limit of normal
VLDL	Very-low-density lipoprotein
VTE	Venous thromboembolism

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1. INTRODUCTION

Coronaviruses were found to be the causative agent of the severe acute respiratory syndrome (SARS) outbreak in China in December 2019 (Zhu *et al.* 2019). The virus, initially called novel coronavirus 2019 (2019-nCoV), was later renamed SARS-COV-2. WHO has designated the 2019 coronavirus disease to be a pandemic (WHO Coronavirus Disease (COVID-19) 2020). In the family Coronaviridae and subfamily Orthocoronavirinae, enclosed single-stranded RNA viruses called coronaviruses are found. These are some of the biggest viruses (with a size of 27-34 kilobases). The virus is surrounded by a "halo" or "crown" in electron microscope images, which explains its name. Recently, coronaviruses have been responsible for a number of epidemics, including the 2003 pandemic brought on by Middle Eastern Respiratory Syndrome Coronavirus (MERS-CoV) and the 2012 pandemic brought on by Severe Acute Respiratory Syndrome Coronavirus (SARS-CoV) (Benvenuto *et al.* 2019).

Initially a zoonotic infection, the spread of SARS-CoV-2 is now extremely rapid, especially among close contacts, by coughing and sneezing. SARS-CoV-2 is resilient and, depending on the pathogen and the weather, can survive for 2 hours to 14 days (Doremalen *et al.* 2020).

The liver is the largest organ in the human body, making up about l/50 of a typical adult's total mass. As a result of its shape and histology, the liver may be broken down into five distinct tissue systems: Liver lobules and cells, hepatocytes, hepatic sinusoidal cells, bile ducts, and stroma (Gumucio *et al.* 1996). Also, it includes both parenchymatous and non-parenchymatous cell types. The liver has the largest network of reticulo-endothelial cells in the body, making it an important organ in the fight against infectious invaders (Ishibashi *et al.* 2009).

CoV-2 targets the lungs' alveolar cells, just like SARS does, and severely irritates them. Endocytosis mediated by ACE2 receptors (angiotensin converting enzyme 2) is how the virus is thought to enter cells. The receptor is widely distributed in the human body and

is probably the initial step in starting systemic illness, which can harm important organs like the liver. Liver damage brought on by SARS-CoV-2 can range in severity (Pirola *et al.* 2020).

Multiple studies show that people with COVID-19 have moderately raised liver enzymes, including alanine aminotransferase (ALT), aspartate aminotransferase (AST), and gamma-glutamyl transferase (GGT), and slightly elevated prothrombin time (PT) and total bilirubin (TB) (Liu *et al.* 2020).

Multiple cases of liver damage attributed to COVID-19 have been reported. Some research suggests that the antiviral drugs prescribed to patients with COVID-19 may increase the risk of liver damage. It is still unclear whether viruses or drugs have a larger role in causing liver damage in severe sickness. The liver damage observed in COVID-19 may also be attributable to innate immune response dysregulation. Therefore, some potential explanations for liver damage in COVID-19 patients are listed below: (i) pneumonia-related hypoxia and cytokine storm are examples of immune-mediated inflammation, (ii) death of liver cells due to viral replication and (iii) Drug-induced liver damage: People with severe illnesses should exercise caution when taking antiviral medications like lopinavir/ritonavir, chloroquine, remdesivir, tocilizumab, and uminefovir. Furthermore, it is unknown if SARS-CoV-2 infection worsens cholestasis in patients with cholestatic liver disease (Cai *et al.* 2020).

2. LITERATURE REVIEW

2.1 Corona Disease

2.1.1 Overview

Coronaviruses are a diverse family of viruses that can infect a wide variety of animals, including humans, and cause respiratory illnesses ranging in severity. Highly pathogenic coronaviruses of zoonotic origin, such as the Middle East respiratory syndrome coronavirus (MERS-CoV) and the severe acute respiratory syndrome coronavirus (SARS-CoV), emerged in humans and caused fatal respiratory illnesses in 2002 and 2012, respectively, suggesting that emerging coronaviruses in the 21st century are likely to have a significant impact. At the year's end, a novel coronavirus known as SARS-CoV-2 emerged and spread rapidly through Wuhan, China, causing an epidemic of a severe form of viral pneumonia. COVID-19, a recently discovered coronavirus, has rapidly disseminated throughout the world (Wu *et al.* 2019, Hui *et al.* 2019).

2.1.2 Corona virus and the history of disease

The order Nidovirales contains the family Coronaviridae, which contains all coronaviruses. The virus is also known as a coronavirus, after the crown-like spines that cover its outer surface. Most coronaviruses have a single-stranded RNA genome between 26 and 32kbs in size, despite how small they are (65-125 nm). The coronavirus family has four different branches: the alpha (a), beta (b), gamma (c), and delta (d). Acute respiratory distress syndrome (ARDS) and acute lung injury (ALI) are two symptoms of the severe acute respiratory syndrome coronavirus (SARS-CoV), influenza A virus H5N1, and influenza H1N1 virus 2009, and the Middle East respiratory syndrome coronavirus (MERS-CoV), which can lead to respiratory failure and death (ARDS). Before the SARS-CoV epidemic in Guangdong, China in 2002, it was believed that these viruses primarily affected animals (Zhong *et al.* 2003).

The Middle East Respiratory Syndrome Coronavirus (MERS-CoV) was a pathogenic coronavirus that swept through Middle Eastern countries ten years after SARS-CoV. (Wang *et al.* 2013). The 2003 pandemic in China's Guangdong Province was caused by the SARS virus. Once it was determined that SARS was caused by a beta-coronavirus, the virus was given the name SARS-CoV. (Pyrc *et al.* 2007). Pneumonia symptoms, such as extensive alveolar destruction, were reported by patients infected with the virus, and Acute Respiratory Distress Syndrome was the end result (ARDS). When severe acute respiratory syndrome (SARS) emerged in Guangdong, China, it rapidly spread around the world, affecting over 8,000 people and killing 776. Ten years later, in 2012, a different coronavirus was discovered to infect some people in Saudi Arabia. In honor of its initial geographic distribution, the coronavirus was dubbed "Middle East Respiratory Syndrome Coronavirus" (MERS-CoV). According to the World Health Organization, more than 2428 people contracted the MERS coronavirus and 838 of those people died (Rahman *et al.* 2019).

Although it shares a phylogenetic relationship with other human CoV, MERS-CoV is distinct from them due to its beta-coronavirus origins. The first sign of MERS-CoV infection is mild damage to the upper respiratory tract, followed by a severe respiratory illness. Patients with the MERS coronavirus, like those with the SARS coronavirus, develop pneumonia, ARDS, and renal failure (Memish *et al.* 2013). In late 2019, a new coronavirus outbreak swept across Wuhan, a developing industrial hub in China, killing over 800 people and infecting over 70,000 in the first fifty days (WHO 2020).

Almost everyone agrees that this virus is a member of the coronavirus b family. Wuhan coronavirus (or 2019 novel coronavirus (2019 nCov)) is the name given by Chinese researchers to the new virus. The International Committee on the Taxonomy of Viruses (ICTV) designated the virus as SARS-CoV-2 and the disease as COVID-19 (Cui *et al.* 2019). The 2003 outbreak of SRAS-CoV infected 8098 people and killed 9 percent of them across 26 countries; the 2019 outbreak of new Corona virus has infected 120,000 people and killed just 2.9 percent of them across 109 nations as of this writing. A genetic recombination event at the S protein in the RBD region of SARS-CoV-2 may be responsible for the virus's faster rate of spread than that of SRAS-CoV (Figure 2.1).

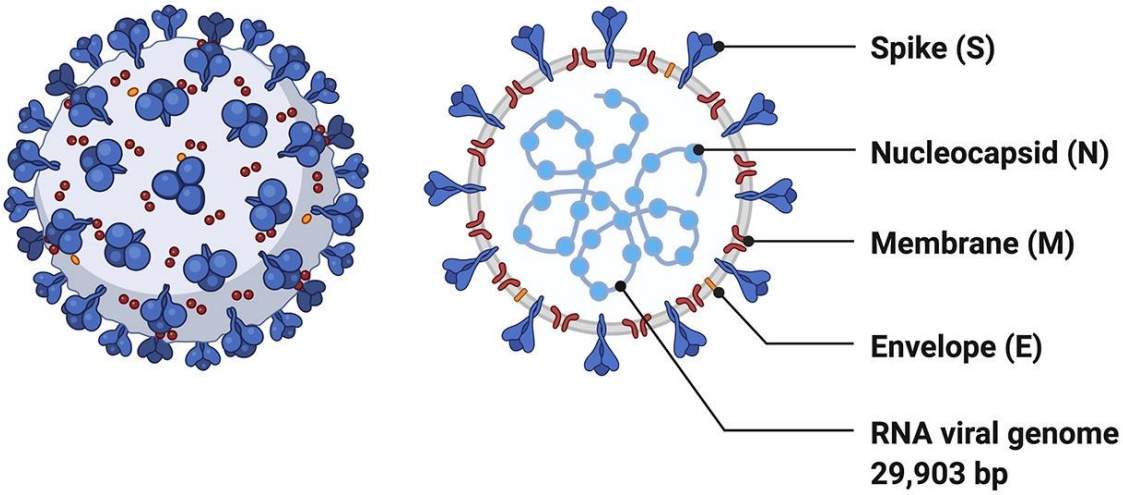

Figure 2.1 Corona virus structure (Amirian *et al.* 2020)

2.1.3 Symptoms of COVID -19

Flu-like symptoms are common, including high body temperature, hacking cough, muscle aches, fatigue, sore throat, and difficulty breathing. The group of infected persons older than 60 is a higher likelihood of developing a severe COVID-19 state and exhibiting comorbidity in the context of chronic conditions (Santos-Sánchez and Salas-Coronado 2020). Fevers, coughing, shortness of breath, and extreme fatigue are all possible side effects of acute respiratory infections (Tang *et al.* 2020). The most common cause of death in people infected with COVID-19 is acute respiratory distress syndrome (ARDS), brought on by diffuse lung damage (Nardo *et al.* 2021).

In addition to having negative effects on breathing, Infection with SARS-CoV-2 may impact the cardiovascular, gastrointestinal, hepatic, renal, neurological, olfactory, gustatory, ophthalmic, cutaneous, and hematological systems, among others (Figure 2.2) (Lai *et al.* 2020).

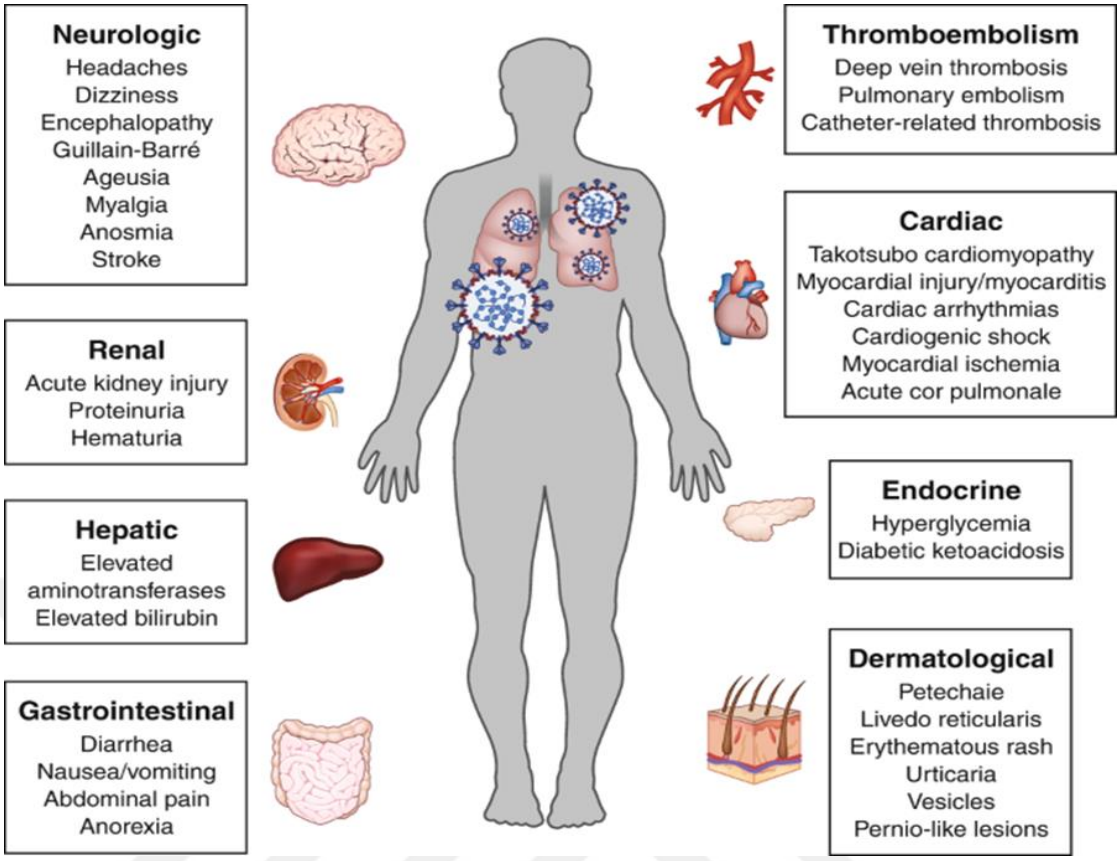

Figure 2.2 SARS-impact CoV-2's on several bodily functions (Gupta 2020)

2.1.4 Viral transmission and clinical features

When an infected person coughs or sneezes near another person, the respiratory droplets spread the COVID-19 virus. In this case, the host's eyes, nose, and mouth become infected after being exposed to infected respiratory droplets (Li *et al.* 2020).

Bedding, kitchenware, medical equipment (including stethoscopes), and linens are just some of the objects that might be contaminated when an infected person touches them. Documented procedures that result in aerosols capable of spreading COVID-19 through the air include endotracheal intubation, bronchoscopy, open suctioning, nebulization with oxygen, bronchodilators, or steroids, bag and mask ventilation prior to intubation, tracheostomy, and cardiopulmonary resuscitation (Burke *et al.* 2020).

According to earlier studies, 19.6% to 73% of SARS patients had gastrointestinal symptoms. SARS-CoV was present in the enterocytes of the small intestine, where it was actively replicating. During the SARS epidemic, there is concern for fecal-oral transmission of SARS-CoV because RNA from the virus has been detected in patient feces (Leung *et al.* 2003). In general, it takes 5-6 days for symptoms to appear after exposure to the COVID-19 virus, but it may take up to 14 days. The "presymptomatic" period is when the virus may spread from sick persons to healthy people in the community.

2.1.5 Diagnosis imaging and molecular tests (RT-PCR)

Nasopharyngeal and oropharyngeal swabs can be used to collect samples from the upper respiratory tract, while expectorated sputum and bronchoalveolar lavage can be used to collect samples from the lower respiratory tract. The samples are delivered to the lab where reverse transcription is used to ramp up the viral genetic material after being kept at 4°C. This involves converting the previously present viral RNA into a double-stranded DNA molecule with the use of a reverse transcription polymerase chain reaction (RT-PCR) or real-time RT-PCR (Chan *et al.*2015).

Finally, the amplified DNA reveals which parts of the genetic code for SARS-CoV-2 are stable. Repeat testing is indicated in cases where the COVID19 test is positive in order to confirm the positive result and to ensure viral clearance. Oropharyngeal swab positivity was found in approximately 53.3% of COVID-19 confirmed patients, and RT-PCR positivity was found in approximately 71% of patients using sputum samples, indicating that these tests are not particularly sensitive (Fang *et al.*2020, Zhang *et al.*2020). After 2 to 8 days, the RT-PCR results typically indicate positive (Huang *et al.*2020).

Serology:

It has not been possible to develop a viable antibody test as of yet. The CDC is now investigating application of a diagnostic tool created by the National Institutes of Health's Center for Vaccine Research. There appear to be a high degree of the utmost detail (above 99%) and sensitivity (96%) (Cascella *et al.* 2020).

Laboratory analyses of blood:

Reports of lymphopenia (normal white blood cell count) and normal white blood cell count (anemia) are both quite common and are often taken as an indicator of a poor prognosis. C reactive protein, lactate dehydrogenase, aspartate aminotransferase, alanine aminotransferase and creatine kinases (CK MB and CK MM) all rise in response to inflammation (Cascella *et al.* 2020).

Increased ratio of neutrophils to lymphocytes and concentration of D-dimer have been seen in certain cases (Yang *et al.* 2020).

Increases in prothrombin time and international normalized ratio indicate coagulation abnormalities in extreme situations (I.N.R).

The X-ray of the chest:

During the preliminary stages of a disease, an X-ray of the chest may not reveal any obvious alterations. Multifocal alveolar opacities are seen on both sides as the illness worsens; they may be connected to pleural effusion (Cascella *et al.* 2020).

CT:

The most efficient approach for high-resolution CT (HRCT) is useful for the early diagnosis of COVID-19 pneumonia because of its high sensitivity. The most frequent

symptoms are patchy peripheral dispersion and multifocal bilateral “ground-glass” patches linked to consolidation, with increased involvement of the lower lobes (Cascella *et al.* 2020).

2.1 The Liver

The liver is a vital organ that plays many important roles in the body, including immunity, digestion, and detoxification, metabolism, and vitamin storage, to name a few. About 2% of a person’s total weight comes from fat. Approximately 75% of the blood flow to the liver comes from the portal vein, while the remaining 25% comes from the hepatic artery are the two main blood vessels that supply the liver with oxygenated blood (Kalra *et al.* 2022).

2.2.1 Organ systems involved

The liver aids digestion and metabolism by communicating with the endocrine system and the digestive system. The liver regulates cholesterol homeostasis and stores fat-soluble vitamins. It keeps copper and iron. It contributes to protein synthesis and clotting factor in hematology. Hemoglubin is converted in the liver from unconjugated bilirubin to conjugated bilirubin. The hormones involved in sperm and egg development couldn’t function without it, and neither could the carrier proteins they help make. The immune system’s Kupffer cells and pit cells were first found in the liver (Kalra *et al.* 2022).

2.2.2 Function

Bile production:

The secretion of bile salts and acids facilitates fat absorption and digestion and assists in the excretion of substances that the kidneys are unable to eliminate. The hepatocytes

secrete bile, which is primarily composed of water, electrolytes, bile salts, bile acids, cholesterol, bile pigment, bilirubin, and phospholipids.

Storing and/or using vitamins that are fat-soluble:

Absorption of fat-soluble vitamins occurs mostly in the small intestine, and they are then carried to the liver by chylomicrons or very low-density lipoprotein. The liver is where fat-soluble vitamins such as vitamins A, D3, and E are stored or processed.

Drug metabolism:

The liver's First and Second Stages processes aid due to the radical change of xenobiotics from their lipophilic form to their hydrophilic form. Hepatocytes' smooth endoplasmic reticulum is often the site of these reactions.

The metabolism of drugs can be aided by other organs including the kidney and stomach. Metabolism of a drug can be affected by a wide variety of factors, including but not limited to: age, gender, medication-drug interactions, diabetes, pregnancy, liver/kidney disease, inflammation, and genetics (Almazroo *et al.* 2017).

Bilirubin metabolism:

To properly break down heme, the liver plays a crucial role. A variety of tissues and organs are capable of hemolysis, including the liver, spleen, and bone marrow. When heme is broken down into biliverdin, unconjugated bilirubin is the end result. Albumin carries unconjugated bilirubin from the bloodstream to the liver. The UGT system uses uridine diphosphate glucuronyltransferase to conjugate the unconjugated bilirubin. After being released into the bile or in trace quantities after dissolving in the circulation, the newly conjugated bilirubin is filtered by the kidneys for disposal. Because the intestinal wall cannot absorb it, most conjugated bilirubin is excreted along with bile in the stool. Unconjugated bilirubin, or urobilinogen, is produced when bacteria in the stomach

break down a portion of the bilirubin and absorbed it through the enterohepatic circulation (Figure 2.3) (O'Brien *et al.* 2015, Stec *et al.* 2016).

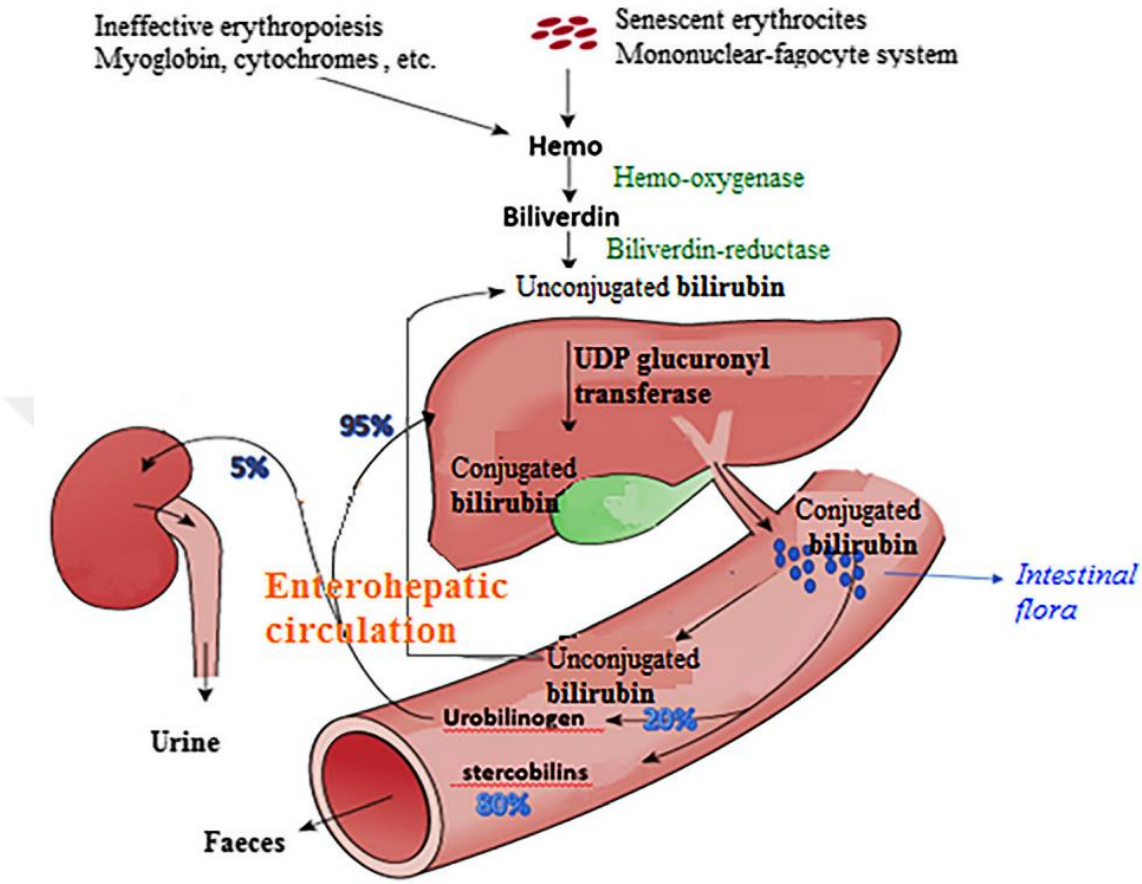

Figure 2.3 The process of bilirubin metabolism (Ruiz *et al.* 2016)

Other functions:

The liver, which is where T4 is converted to T3, plays a part in the activity of thyroid hormones. Most plasma proteins are synthesized in the liver, which plays an essential role in regulating blood pressure, including albumin, binding globulins, protein C, and protein S. Blood coagulation factors, both intrinsic and extrinsic, with the exception of factor VIII, are synthesized and secreted by the liver, which is a significant liver function.

2.2.3 Related testing

Clinical professionals frequently request a panel of liver function tests (LFTs) in order to help them assess the liver function of a patient; these are some of the things they look for:

The enzyme aspartate transaminase (AST).

An enzyme called alanine transaminase (ALT).

Bilirubin.

Enzyme that breaks down alkaline phosphates.

Gamma-glutamyltranspeptidase (GGT)

The panel only describes the extent to which cell damage is occurring in the liver in order to assist develop a picture of what is happening there. Injuries to the liver trigger the release of specific enzymes into the blood; their levels are a more accurate reflection of the existence of injury.

ALT and AST are both vital enzymes in gluconeogenesis; however, AST is found in many different tissues whereas ALT is found only in the liver. Alkaline phosphatase (ALP) is less specific because it can be seen in the biliary tree and bone, but when combined with the other tests on the panel, it can provide proof of hepatocellular injury. Elevated ALP, in particular, denotes bile tract lining injury (Hoekstra *et al.* 2013).

Besides factor VIII, all clotting factors are produced in the liver. The PT test evaluates the extrinsic pathway's coagulation proteins. A high PT may indicate liver injury, vitamin K insufficiency, or ongoing warfarin therapy. Because that the process by

which these components are carboxylated in the liver requires vitamin K (Hoekstra *et al.* 2013).

2.2.4 Liver injury

Increased levels of either ALT or AST that were less than twice the upper limit of normal were considered to be indicative of mild liver injury (ULN), moderate if levels were high 2-5, severe if levels were elevated 5-15 (Kwo *et al.* 2017).

2.3 Laboratory Assessment

2.3.1 Ferritin

Soluble ferritin has a molecular weight of 450 kilodaltons. The highest numbers of macrophages are found in the liver, spleen, and bone marrow, although it is found in every cell of the body. It offers safe and convenient intracellular storage of bioavailable iron. It shields cells against the toxicity and production of free radicals caused by iron (Harrison *et al.* 1996).

Extreme hyperferritinemia, which feeds the cytokine storm, is a condition in which ferritin plays a crucial modulatory role in immunological dysregulation via its direct immuno-suppressive and pro-inflammatory effects. It has been postulated that the severity of the disease is reliant on the cytokine storm syndrome given that COVID-19 catastrophic results are frequently accompanied by this condition. (Huang *et al.* 2020).

Based on this, data have quickly been discussed in favor of the theory that ferritin levels may play a significant role in establishing the degree of harm caused by COVID-19. Ferritin levels within the normal range are anywhere from 12 to 300 ng/mL in males and 2 to 150 ng/mL in females (Ferri 2019).

2.3.2 D.Dimer

D-dimer refers to a readily quantifiable terminal fibrin degradation byproduct and may be used to assess venous thrombosis and widespread intravascular coagulopathy. D-dimer levels may also increase in pregnancy, cancer, inflammation, and after surgery (Thachil *et al.* 2010). As of December 2019, the COVID-19 (coronavirus disease 2019) pandemic has been ongoing in Wuhan, China; it has infected over 120.77 million people worldwide and claimed 2,672,099 lives. An increased risk of developing venous thromboembolism (VTE), also known as hypercoagulability, which might result in thrombo-inflammation in serious situations, is predominantly linked to COVID-related mortality (Thachil *et al.* 2010).

Therefore, coagulation biomarkers may be used to help with patient selection, treatment planning, and prognosis monitoring. They can also be used to convey information about a disease's lethality and degree of severity. D-dimer, a byproduct of fibrin breakdown, is a mechanism that aids in the thrombo-inflammatory process seen in COVID-19. High levels of D-dimer (up to 46.4% prevalence) have been linked in multiple studies to increased COVID-19 severity and negative outcomes. There is a 20-fold increase in mortality risk in patients whose D-dimer levels are greater than 1000 ng/ml compared to those whose levels are less than 1000 ng/ml (Zhou *et al.* 2020). Therefore, D-dimer could be used as a VTE screening test for people with COVID-19. D-dimer elevation warrants a change in anticoagulant dosing, but patients would benefit more from a therapeutic shift than a preventative one. That's why it's important to check COVID patients' D-dimer levels soon after they check in.

2.3.3 CRP

As an acute-phase protein, C-reactive protein (CRP) may be utilized as a precursor to other inflammatory markers. A gene on chromosome 1 codes for a protein produced in the interaction between IL-1 and IL-6 and the liver; normal blood levels are below 10 mg/L. Within 6-8 hours of infection or inflammation, CRP levels spike rapidly, peaking at 350-400 mg/L after 48 hours (Kolb-Bachofen *et al.* 1991).

In the first publication on COVID-19, the importance of monitoring C-reactive protein (CRP) levels was shown. A correlation was found between a measure of the levels of the protein C-reactive and the presence of lung lesions. The size of lung lesions was shown to be positively correlated with CRP levels. Since COVID-19 disease has no known cure, biomarker research has becoming increasingly important (Pepys *et al.* 1981). When looking for biomarkers, the most common ones were CRP and D-dimer. CRP was discovered in pneumococcal pneumonia patients' blood and is one of acute-phase reactant (APR) markers (Palosuo *et al.* 1986).

2.3.4 IL-6

The cytokine IL-6 performs a broad range of biological roles. It serves as a mediator for the acute phase response regulation and immunoglobulin class substitution. It serves as a body's indicator of inflammation as well. IL-6 is a useful research marker for studying the progression of bacteremia (Remick *et al.* 2005).

It has been hypothesized that IL-6 also makes people more likely to develop type 2 diabetes mellitus ...and systemic juvenile rheumatoid arthritis (Tanaka and Kishimoto 2012). Resting normal B-cells, T-lymphocytes, and cells from myeloid and hepatic cell lines all carry IL-6 receptors (Kubistova *et al.* 2012). Journal articles detailing a description of the immune system of people who test positive for COVID-19 show that interleukin (IL)-6 Humoral immune system activation is a major mediator of multiple organ failure, including respiratory failure and shock. This dysregulation of host immune responses may be an important targets for therapies due to the possibility that cytokine release syndrome (CRS) can form a pathologic basis for the progression of disease in patients with severe COVID-19 infection (Xu *et al.* 2020).

2.3.5 ALT

ALT is a special to hepatocellular damage enzyme that is largely found in liver (with smaller quantities in heart, renal, and muscular tissue). ALT stimulates hepatocyte

glutamate and pyruvate synthesis, two building blocks for the production of ATP (Aulbach *et al.* 2017).

2.3.6 AST

Like Alanine transaminase, Aspartate transaminase is an enzyme that is found in the liver but is also found in other locations where it is more prevalent than ALT. The main sites include the brain, heart muscle, renal tissue, and skeletal muscle. AST promotes the metabolism of amino acids (Sparling *et al.* 2016).

2.3.7 ALP

Concentrations of ALP are highest in the hepatobiliary system, followed by the placenta, bone, and to a lesser degree, intestinal tissue. The enhanced osteoblastic activity linked to bone formation is the main reason why ALP is often greater in children and teenagers (Lowe *et al.* 2021).

2.3.8 Bilirubin

In serum, bilirubin, the byproduct of heme breakdown, is first bound to albumin. It is conjugated in the liver and then secreted in the bile. Direct hyperbilirubinemia and indirect hyperbilirubinemia are further classifications for elevated bilirubin levels. Cholestasis, Rotor syndrome, and Dubin-Johnson syndrome are all liver excretion disorders that may cause hyperbilirubinemia in a direct fashion. Hemolysis and intrinsic liver injury are two potential causes of indirect hyperbilirubinemia (Gopal *et al.* 2000). Liver damage caused by COVID-19, and some insight into additional variables that may contribute to that damage (Chen *et al.* 2020, Tian *et al.* 2020).

New information indicating that more than half of COVID-19 patients demonstrated varying degrees of liver disease sheds some light on the impact of COVID-19 on other organs (Chau *et al.* 2004). Levels of alanine aminotransferase (ALT) and aspartate

aminotransferase (AST) rose by 14–53% (Wang *et al.* 2020). A new study suggests that the SARS-CoV-2 may interact with angiotensin-converting enzyme 2 (ACE2) in cholangiocytes, causing cholangiocyte dysfunction and triggering a systemic inflammatory response that damages the liver (Chai *et al.* 2019). In addition, microvesicular steatosis and moderate lobular and portal activity were found in the liver biopsy samples of a deceased COVID-19 patient, suggesting that SARS-CoV-2 may have been the underlying cause of this liver damage (Xu *et al.* 2020).

However, there is a dearth of data that comprehensively evaluates the spectrum of COVID-19-related liver enzymes and clinical signs of liver failure. Lung injury is linked to the most serious clinical symptoms. It's not only the kidneys and liver and heart and pancreas that might exhibit strange clinical signs. The angiotensin II (ACE2R) receptor is a popular entryway for viruses into cells, which is found in many tissues. When the liver isn't working properly, the bile ducts are a possible target for the SARS-CoV-2 infection and liver since ACE2R is found all throughout the body (Zhou *et al.* 2020). In COVID-19, abnormal liver biochemistry is multifactorial and related to formation of microthrombi in the hepatic parenchyma, immunological liver dysfunction brought on by the inflammatory response (cytokine storm), and direct virus-induced cytotoxicity. Numerous medications used to alleviate COVID-19-related symptoms are liver-damaging (Remdesivir, Tocilizumab, etc.). Chronic liver disease, hemodynamic instability, also Hypoxia is all risk factors that might diminish hepatic function (Fix *et al.* 2020). Scientists may have discovered a connection between COVID-19 illness severity and abnormal liver function tests (Wang *et al.* 2020). Patients with SARS-CoV-2 infections who also suffer from hepatic dysfunction have worse clinical outcomes and a higher risk of dying from their infection (Hajifathalian K *et al.*2019). A similar retrospective study found that after being discharged, one patient was readmitted with elevated blood ALT and AST levels and a positive Reverse transcription-PCR for SARS-CoV-2 (Cao *et al.* 2020). Recent research by Hundt *et al.* published in Hepatology on July 29, 2020, confirmed that a significant percentage of COVID-19 patients with hospital admissions had abnormal liver tests (AST 66.9%, ALT 41.6%) and at the peak of their hospital stay (AST 83.4%, ALT 61.6%) (Hundt *et al.* 2020).

This study aimed to learn more about how COVID-19 patients' liver enzyme levels and other indicators fluctuate over time by comparing them to those in control groups. MiniVIDIS SYSTEM IgM and IgG titers were used to identify COVID -19 patients and compare them to controls in this investigation. C-reactive protein, interleukin-6, and ferritin levels were also analyzed for their potential impact on illness severity. After everything else was done, researchers at Columbia University measured D. Dimer levels in people with COVID -19 and compared them to those in healthy people.

3. MATERIALS AND METHODS

3.1 Instruments

The primary tools and equipment employed in this investigation are detailed in Table 3.1, Table 3.2 and Table 3.3.

Table 3.1 Laboratory equipments and apparatus

No.	Materials	Company	Country
1	MiniVIDIS SYSTEM	Biomerieux	France
2	Afias ten	Boditech.co	Korea
3	Spectrophotometer	APEL	Japan
4	Centrifuge	Sigma	Germany
5	Freezer	Froilabo	France
6	Micropipettes (100-1000) µL, Micropipettes (5-50) µL	Slamed	Japan
7	Water bath	Klostermann	Germany
8	Refrigerator	Hitachi	Japan
9	Printer	Canon	Japan
10	Stop watch	-----	China
11	Cooling boxes/ICE pack	Abbott	USA
12	Vortex	IKA	Germany

Table 3.2 Tools and materials used in the study

No.	Materials	Origin
1.	Cotton	China
2.	Eppendorf	Denmark
3.	Gloves	China
4.	Tourniquet	China
5.	Syringe	China
6.	Glass gel tube	China
7.	Tips (blue 1mL,yellow 100M)	China
8.	Used laboratory glassware	Afco-despo/ Jordan
9.	Alcohol(Ethanol 68.6%&methanol 3.7%)	UAE

Table 3.3 Chemical used in the study

No.	Kits	Company	Origin
1	GOT kit	BIOLABO	France
2	GPT kit	BIOLABO	France
3	Alkaline phosphatase kit	BIOLABO	France
4	TSB & direct and indirect bilibubin kit	BIOLABO	France
5	C.R.P kit	Boditech.co	korea
6	Ferritine kit	Biomerieux	France
7	D.Dimer kit	Biomerieux	France
8	IL-6 kit	Boditech.co	korea
9	COVID-19 IgM kit	Biomerieux	France
10	COVID-19 IgG kit	Biomerieux	France

3.2 Subjects

Patients:

Eighty COVID-19 patients between the ages of 18 and 88 took part in the present investigation. Patients infected with COVID-19 were quarantined in one of three official centers in Najaf, Iraq: Al-Hakim Hospital, Al-Shifa Center, or Al-Amal Specialized Hospital for Infectious Diseases between the time frame of the study (February 2022 to May 2022). All cases of SARS-CoV-2 were confirmed by acids. COVID-19 was detected by rRT-PCR, a positive reactive protein test, and the classic signs and symptoms of the illness (fever, fatigue, shortness of breath, hacking cough, diminished smell, and taste).

In addition, Chest X-rays and CT scans were performed to check for lung abnormalities in each patient. More than 70% of COVID-19 cases verified by Using Reverse Transcription-Polymerase Chain Reaction have had CT scan findings published. Patients were assessed through a complete medical history to explore the presence of any systemic diseases that might have an impact on the studied criteria.

Controls:

As a control group, forty individuals aged 20-40 have participated in the present study. Subjects were chosen to be free from any systemic or inflammatory disorders.

3.3 Methods

3.3.1 Sampling

Somewhere approximately 5 mL took blood samples through a venous puncture. After letting the blood clot for 5-10 minutes at room temperature, the sample spent 10 minutes being centrifuged at 3000 rpm in a sodium citrate tube and 3 ml in a gel tube without anticoagulant. Serum was collected by centrifugation, and then split between six Eppendorf tubes marked with patient IDs and names, and stored at -20°C until use.

3.3.2 Determination of COVID-19 IgG and COVID-19 IgM by Mini VIDIS SYS

The kit was removed from storage at +2°C or +8°C, and the necessary reagents were extracted. The kit was returned to +2°C or +8°C after carefully resealing the SPR pouch. For every sample, control, and standard, one strip and one SPR were taken out of the apparatus. The 9COG code is for IgG testing, whereas the 9COM code is for IgM testing. Both copies of the "S" calibrator and the "C" control underwent testing. A vortex-type mixer was used to combine the calibrator, controls, and samples. In this experiment, 100 µl aliquots as the standard, control, and sample volumes were used. Strip readers and SPR devices were installed in the apparatus. The test was fully automated by the equipment. In around 27 minutes, the experiments were completed. The SPR probes and strips were taken out of the analyzer after the test was done.

3.3.3 Determination of ferritin by Mini VIDIS SYS

For each sample, control, and calibrator, one “FER” strip and one “FER” SPR strip were taken from the set. The "FER" symbol on the analyzer identifies this particular check. Two duplicates of the titrator “S1” and control “C1” were evaluated. A vortex-type mixer was used to combine the titrator, controls, and samples. Both the calibrator and controls, as well as the sample, were diluted to a volume of 100 µl. To prepare the instrument, “FER” reagent strips and SPRs were placed in their respective holes. The color labels of the check code were scanned. Results were available within 30 minutes.

3.3.4 Determination of D.dimer by mini VIDIS SYS

For each sample, control, and calibrator, the kit was emptied of one "DEX2" strip and one "DEX2" SPR. VIDAS preparation/loading tray or work surface was used to set down the "DEX2" strip and "DEX2" SPR. The "DEX2" indicator on the machine denotes this particular examination. The calibrator, labeled S1, passed both sets of tests. C1 and C2 represent the two knobs used to adjust the settings. A vortex-type mixer was used to combine the calibrator, controls, and samples. All three volumes, calibrator, control, and sample, were 200 µl. DEX2 SPRs and DEX2 strips were installed in their designated slots. The STRs were compared to the assay codes printed on the colored labels of the SPRs. All of the assay procedures were carried out mechanically by the instrument after the operator followed the instructions in the handbook. The wait for results was just 20 minutes.

3.3.5 Determination of C.R.P and IL-6 by AFIAS-10

Firstly, the “General Mode” in the instrument for AFIAS tests was selected. 100 µL of the sample (serum) was taken with through the cartridge’s sample well after being drawn into the pipette. After installing the cartridge in its holder, a tip was inserted into the cartridge’s tip hole. The last step was to tap on the screen. “START” icon was

clicked. After 3 minutes for CRP and 12 minutes for IL-6, the results were displayed on the screen.

3.3.6 Determination of GOT and GPT

Tonhazy, White, and Umbreit developed a colorimetric method, which Reitman and Frankel used to measure serum activity. Reaction scheme is as follows Equation 3.1 and Equation 3.2:

$$L-Alanine+2-Oxoglutarate \overset{ALT}{\Longleftrightarrow} Pyruvate + L-Glutamate \quad (3.1)$$

$$L-Asparate + 2-Oxoglutarate \overset{AST}{\Longleftrightarrow} Oxaloacetate + L-Glutamate \quad (3.2)$$

Then, 2,4 Dinitrophenylhydrazones are created when Pyruvate or Oxalate interacts with 2,4 DNPH, and their absorbance at 505 nm in an alkaline solution correlates with the amount of AST or ALT activity in the reactional mixture.

3.3.7 Determination of ALP

ALP activity was measured colorimetrically using the following Equation 3.3 scheme:

$$Phenylephosphate \xrightarrow{Alkaline\ phosphatase} Phenol + Phosphate \quad (3.3)$$

This free phenol is obtained through substrate hydrolysis. This free phenol reacts with 4-amino-antipyrine in the presence of alkaline potassium ferricyanide to produce a red-colored complex whose absorbance at 510 nm is directly proportional to the ALP activity in the material. The addition of sodium arsenate to the reagent stops any further enzyme activity and prevents the dilution of color seen in traditional methods.

3.3.8 Determination of direct and total serum bilirubin

The azobilirubin compound is formed when bilirubin and diazotized sulfanilic acid react. This chemical is colored in very acidic or basic environments. Concept of Malloy-Evelyn with a few tweaks (Walters *at al.* 1970). In an aqueous solution, only DB can be seen interacting. Isolating TB requires severing the bond between unconjugated bilirubin and albumin. Dimethyl sulfoxide (DMSO) was added at this stage. At 550 nm, the absorbance of the azobilirubin that is subsequently created is proportional to the bilirubin concentration (530-580).

Indirect serum bilirubin (mg\L) = total serum bilirubin – direct serum bilirubin

3.4 Statistical Analysis

The correlation between corona virus infection and elevated liver enzyme levels was analyzed using the Statistical Analysis System- SAS (2012) software. A meaningful comparison between means was performed using statistical Analysis Using the Least Significant Difference (LSD) (Analysis of Variance-ANOVA). The chi-squared test was performed to find statistically significant differences between the two percentages (probability levels of 0.05 and 0.01). Moreover, the correlation coefficient was estimated between variables in this study.

4. RESULTS AND DISCUSSION

There were 120 participants in the trials covered by this study, who were divided into groups based on age and gender. Group I is the control group, and Group II consists of COVID-19 infected patients as a whole. As part of the study's parameters, as well as the other parameters ferritin, IgG, IL-6, C-reactive protein, D-dimer, and IgM titers, liver profile tests were performed in the blood serum samples of all cases.

4.1 Comparison Between Patients and Control Groups in Age

In this study, eighty samples came from patients while forty came from the control group. The results showed that the mean age in group I and group II was (33.42 ± 1.47 and 48.52 ± 2.06) respectively, the results of which are displayed in Table 4.1 and Figure 4.1. Differences between the study's control group and its patient group were found to be statistically and clinically significant.

Table 4.1 In terms of age, a comparison was made between the patients and the controls

Group	No	Mean ± SE of Age (year)
Patients	80	48.52 ± 2.06
Control	40	33.42 ± 1.47
T-test	--	6.181 **
P-value	--	0.0001
** (P≤0.01).		

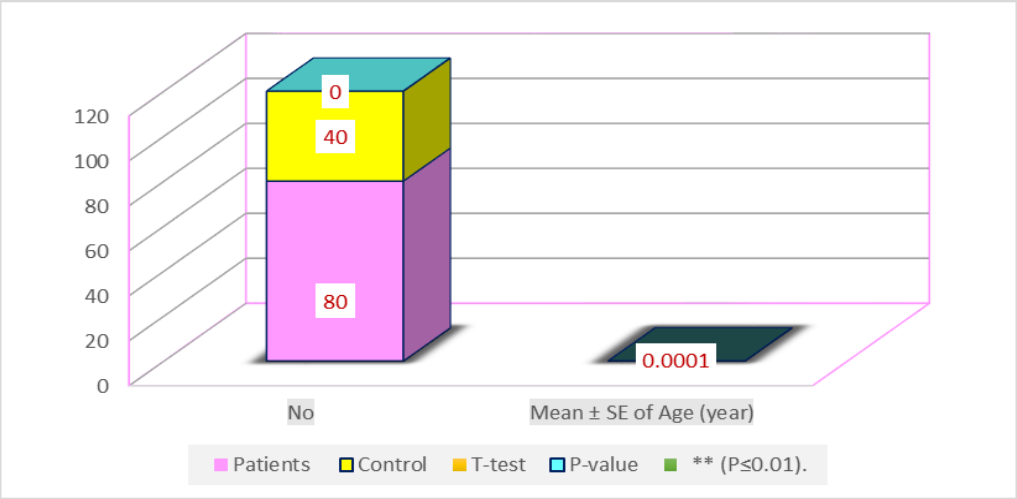

Figure 4.1 The age difference between patients and controls

4.2 Comparison Between Patients and Control Groups in Sex

The present study included, the patient group was 80 people, while the control group was 40 people. In the patient group, there were 32 females (40%) and 48 males (60%). On the other hand, in the control group, N = 26 (65.00%), while N = 14 (35.00%), as shown in Table 4.2. The results showed that between the sexes, there were substantial differences between the two groups statistically. Equally, no differences can be observed between males and females of different sexes within any of the groups. This study agrees with many studies reported in the literature. In Basra (Iraq), a study showed that regarding sex distributions were 161 (57.5%) males and 119 (42.5%) females (Al-khaqani *et al.* 2022). In Erbil city, in an investigation, it was found that males had a greater incidence of COVID-19 than females (52.7% for males and 47.3% for females) (Hwaiz *et al.* 2021). In China, Tolia *et al.* (2020) mentioned that the majority of diagnosed COVID-19 cases were male individuals.

According to a recent survey, 58 percent of the 1,099 COVID-19 in 552 hospital patients across 30 Chinese cities were men. These findings indicate that COVID-19 may exhibit a gender pattern, with men being more susceptible to being affected. Due to Chinese men's higher smoking rates than women, this gendered inclination may exist

(In 2018, there were 288 million male smokers and 12.6 million female smokers) (Guan *et al.* 2020).

Nonetheless, in a second research including critically ill patients, it was found that females were 67% increased risk of contracting COVID-19 compared to males (Zhang *et al.* 2020).

Table 4.2 Patient and control group sex distribution in a sample study

Group	No	Male No. (%)	Female No. (%)	P-value
Patients	80	48 (60.00%)	32 (40.00%)	0.0097 **
Control	40	26 (65.00%)	14 (35.00%)	0.0061 **
P-value	--	0.788 NS	0.788 NS	---
** (P≤0.01), NS: Non-Significant.				

4.3 Comparison Between Difference Groups in Ferritin, CRP, D-Dimer and IL-6

According to the data, the average levels of ferritin, C.R. P, D-Dimer, and IL-6 in the patient group was (760.59 ±35.46, 87.79 ±7.08, 660.38 ± 32.98, and 31.65 ±3.40). On the other hand, the mean of ferritin, C.R. P, D. Dimer, and IL-6 in the representative sample of the healthy population was (124.86 ±11.54, 6.67 ±0.40, 303.37 ±12.26, 1.46 ±0.13). Table 4.3 and Figure 4.2 show that the study found statistically significant differences across all parameters when comparing the groups.

For ferritin:

This study agrees with the study in Bahrain which showed revealed that D-dimer was the second most accurate predictor, after serum ferritin and CRP (Farid *et al.* 2021). One research including 20 COVID-19 patients in Mexico indicated that those with the most severe cases of the disease had higher than average blood ferritin levels. In the COVID-19 cohort, the concentration was 1006.16 ng/ml (Zhou *et al.* 2020). Patients who died from COVID-19 had higher ferritin levels upon hospital admission and

throughout their hospital stay, according to another study from China. High correlations were found between serum ferritin levels and the severity of COVID-19 disease (Liu *et al.* 2020). COVID-19 patients under home observation in Egypt had elevated ferritin levels (Yameny *et al.* 2021).

For IL-6:

According to a Chinese study, patients with COVID-19 who developed ARDS had significantly higher IL-6 levels (median 7.39 pg/mL, IQR 5.63-10.89 vs median 6.29 pg/mL, IQR 5.36-7.83; P =.03). And there was a correlation between high levels of IL-6 and premature death. COVID-19 fatalities were also associated with significantly higher levels of IL-6 (11.4 8.5 pg/mL vs. 6.8 3.6 pg/mL, P .001) (Ruan *et al.* 2020).

In patients with persistently high IL-6 levels, which may be crucial in the emergence and maintenance of critical COVID-19 stages, the IL-6 level was assessed on many occasions, However, these studies only included a small sample size (ranging from 21 to 100 patients), but only a small number of patients (21-100) were included in these studies demonstrating that increased levels of IL-6 and other cytokines were connected with the severity of this condition. Both groups of researchers have confirmed this trend (Chen *et al.* 2020; Tao *et al.* 2020). Increased levels of IL-6 and other pro-inflammatory cytokines have been observed in those with severe COVID-19 in Italy, according to a number of studies (Conti *et al.* 2020).

For CRP:

The results here corroborate those of the Spanish research. Patients who died in the hospital with COVID-19 strain had substantially increased C-reactive protein levels, evidenced by a single-center retrospective study of 540 patients (CRP) (Bernal-Monterde *et al.* 2020). High C-reactive protein levels have been linked to predicting the probability of COVID-19 aggravation in moderately ill adult hospital patients (Wang *et al.* 2020). In India Chronic Obstructive Venous Inflammatory Disease Sufferers who

passed away in hospitals in 19 had CRP levels that were roughly ten times higher than those who survived (Ali 2020).

This research contradicts a study conducted in India in which 10 out of 25 individuals (about 40%) exhibited just minor symptoms and a normal CRP. Six out of the twenty-five individuals (24%) showed mild symptoms and abnormally high C-reactive protein levels. C-reactive protein levels were elevated in 36% of patients (9/25) (Arjunan *et al.* 2021).

For D.dimer:

This study agrees with many studies in china that reported increasing of D-dimer levels compared with control in 94 cases (Han *et al.* 2020), Wu et al, reported increasing in D-dimer levels in ARDS cases (Wu *et al.* 2020), a study by Gao *et al.* on 43 cases (28 mild and 15 severe cases) showed a high level of D-dimer for extreme conditions (Gao *et al.* 2020), showed elevation of D-dimer level in 17% of cases and the study was conducted on 30 cases (Liu *et al.* 2020), also in china a study 0f 140 cases showed elevation of D-dimer in severe cases (Zhang *et al.* 2020). In Egypt, As the study's findings indicated, there were 84 (36.4%) patients with abnormal D-dimer from 231 Outpatients with mild symptoms and patients under home observation were included in this study, which has a significant p-value (Yameny *et al.* 2021).

Research conducted in Jakarta, Indonesia revealed that 194 Sufferers of COVID-19 participated in the research. The median D-dimer concentration was 2240 ng/mL, in the range of 890 - 5640 ng/mL. Sufferers of Severe COVID-19 disease had three times the median D-dimer levels of those with moderate COVID-19 disease (median 1870mg/L vs 630mg/L).

Table 4.3 Comparison between difference groups in ferritin, CRP, D-dimer and IL-6

Group	Mean ± SE			
	Ferritin (ng\mL)	CRP (mg\L)	D-dimer (ng\mL)	IL-6 (pg\mL)
Patients	760.59 ±35.46	87.79 ±7.08	660.38 ±32.98	31.65 ±3.40
Control	124.86 ±11.54	6.67 ±0.40	303.37 ±12.26	1.46 ±0.13
T-test	100.84 **	19.88 **	94.13 **	9.54 **
P-value	0.0001	0.0001	0.0001	0.0001
** (P≤0.01).				

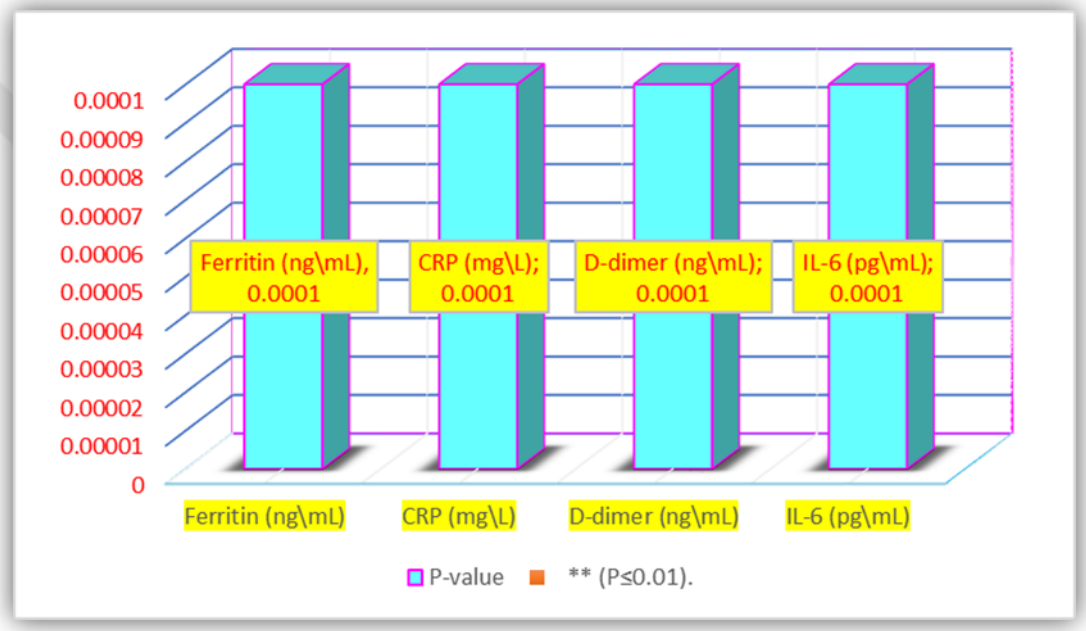

Figure 4.2 Comparison between difference groups in ferritin, CRP, D-dimer and IL-6

4.4 Comparison Between Difference Groups in Liver Enzyme

The results of the current study showed that the average G.O.T, G.P.T, ALP, Direct TSB, indirect TSB, and total serum bilirubin in the patient group was (46.40 ±2.34, 42.30 ±2.37, 119.03 ±5.63, 0.150 ±0.01, 0.293 ±0.03, 0.438 ±0.04). On the other hand, the mean of these parameters in the healthy control group was (20.12 ±0.96, 23.77 ±1.46, 73.19 ±1.79, 0.117 ±0.01, 0.207 ±0.01, 0.325 ±0.01).

According to the results, there is a statistically significant distinction between G.O.T, G.P.T and ALP, while this study shows a significant difference between TSB and indirect TSB, but we find no discernible distinction between direct TSB in different groups, as shown in Table 4.4, Figure 4.3.

Multiple studies find agreement with this one: In Erbil city of Iraq, an analysis of 74 people infected with SARS-CoV-2 found that 25 patients (34.7%) had high ALT, 28 patients (40%) had elevated AST, 12 patients (20.3%) had raised ALP, and 39 patients (52.7%) had abnormal TBili. There were abnormalities in the liver function tests of nearly three-quarters of patients (74.3%) due to SARS-CoV-2 were men who had never been diagnosed with liver illness before (Hwaiz *et al.* 2021). In Jordan, 22.6 percent of the study's sample population had elevated liver enzymes (Fataftah *et al.* 2022). In Romania, patients with COVID-19 experience respiratory insufficiency, which impairs oxygen delivery, negatively affects hepatic cells and causes hepatic damage (Baroiu *et al.* 2021).

In U.S.A, in 43 (3%), or 74% of the 1555 patients, the ALT elevation became severe (above 20x ULN) throughout hospitalization. Increase in ALT and ALP readings was connected to being transferred to the ICU (Sobotka *et al.* 2021). Hepatic patients (230/274, 83.9%) were more likely to have increased ALT than those without liver damage (220/334, 65.9%), while 42.2% of 657 people with raised ALT and 4.9% of those with elevated TBIL had hepatic impairment (Wang *et al.* 2020).

More than three times the average high (>3x ULN) AST levels were related with a higher potential for passing away and China has a serious need for mechanical ventilation. High levels of ALP and TBIL were also significantly correlated with these two unfavorable outcomes (Huang *et al.* 2020).

Out of 281 individuals studied in France, 102 had liver damage. The most prevalent alteration in GGT levels among COVID-19 patients was a rise of 24.3%, followed by increases in AST values of 12.8% and ALT values of 24.3% (Chaibi *et al.* 2021).

Conforming to the findings in the previous study, a large multicentric cohort from hospitals in 11 Latin American countries found that 45.2% of 1611 COVID-19 patients had abnormal LFTs. They had a higher rate of death (18.7 vs 12.2 per cent, p 0.0001) and were more likely to be admitted to the intensive care unit (Mendizabal *et al.* 2021).

This study disagrees with the study that was made by Chaibi et al (2021). In this study conducted in France, in a sample of 281 patients, the most prevalent liver abnormality was in a high level of GGT (25.3%), while ALP was not always significantly elevated.

Table 4.4 Comparison between difference groups in liver enzyme

Group	Mean ± SE					
	GOT (U\L)	GPT (U\L)	ALP (U\L)	TSB (U\L)	Direct TSB(U\L)	Indirect TSB(U\L)
Patients	46.40 ±2.34	42.30 ±2.37	119.03 ±5.63	0.438 ±0.04	0.150 ±0.01	0.293 ±0.03
Control	20.12 ±0.96	23.77 ±1.46	73.19 ±1.79	0.325 ±0.01	0.117 ±0.01	0.207 ±0.01
T-test	6.731 **	6.960 **	16.001 **	0.11 *	0.039 NS	0.081 *
P-value	0.0001	0.0001	0.0001	0.0428	0.103	0.0352
* (P≤0.05), ** (P≤0.01).						

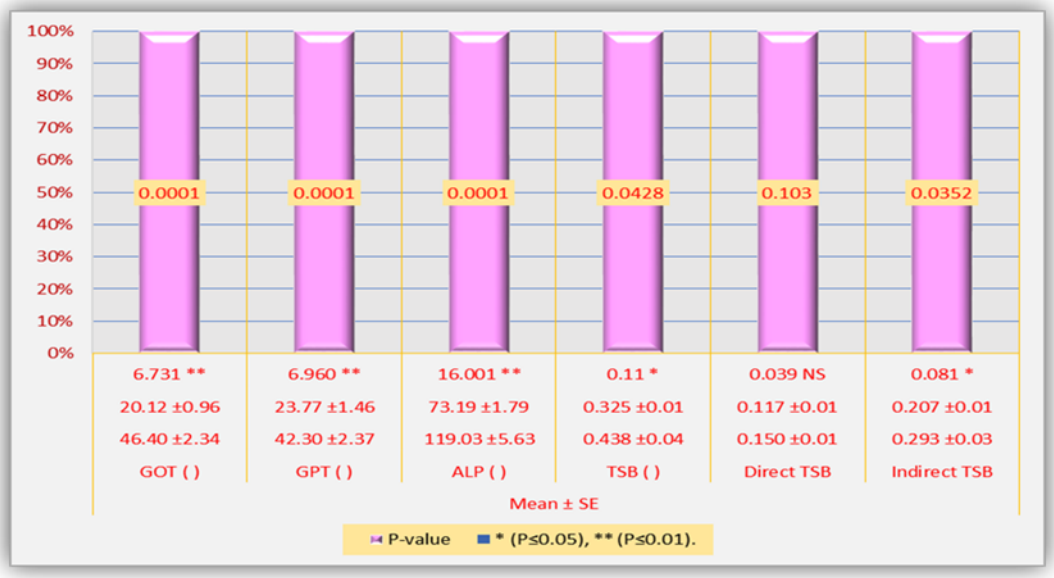

Figure 4.3 Comparison between difference groups in liver enzyme

4.5 Correlation Coefficient Between IgG and Other Parameters

The results showed higher statistical significance with some parameters, which are CRP, D-dimer, GOT, GPT, ALP, TSB, and direct TSB. In statistical tests conducted on the test subjects, the p-value was lower than 0.01 ($P \leq 0.01$). There was a statistically significant relationship between IL-6, indirect TSB, and IgM ($P \leq 0.05$). There was only one parameter that showed no significant difference, which was ferritin. As shown in Table 4.5.

Table 4.5 Correlation coefficient between IgG and others parameters

Parameters	Correlation coefficient-r with IgG	
	Correlation coefficient-r	P-value
Ferritin	0.19 NS	0.0898
CRP	0.30 **	0.0063
D-dimer	0.32 **	0.0033
IL-6	0.24 *	0.0327
GOT	0.46 **	0.0001
GPT	0.38 **	0.0006
ALP	0.40 **	0.0002
TSB	0.29 **	0.0078
Direct TSB	0.33 **	0.0027
Indirect TSB	0.24 *	0.0322
IgM	0.27 *	0.0210
* ($P \leq 0.05$), ** ($P \leq 0.01$).		

4.6 Correlation Coefficient Between IgM and Other Parameters

The results showed higher statistical significance with some parameters, which are ferritin, D-dimer, IL-6, GOT, GPT, ALP, TSB, direct TSB, and indirect TSB. There was statistical evidence of significance at the $P \leq 0.01$ level. There is a significant difference in CRP and IgG. The p-value was less than 0.05 ($P \leq 0.05$), see Table 4.6 for details.

Table 4.6 Correlation coefficient between IgM and others parameters

Parameters	Correlation coefficient-r with IgM	
	Correlation coefficient-r	P-value
Ferritin	0.44 **	0.0001
CRP	0.24 *	0.0281
D-dimer	0.32 **	0.0039
IL-6	0.63 **	0.0001
GOT	0.81 **	0.0001
GPT	0.72 **	0.0001
ALP	0.68 **	0.0001
TSB	0.64 **	0.0001
Direct TSB	0.44 **	0.0001
Indirect TSB	0.65 **	0.0001
IgG	0.27 *	0.0210
* (P≤0.05), ** (P≤0.01).		

4.7 Effect of Sex in Parameters of Patients

The present study revealed that the mean of ferritin, C.R. P, D-Dimer and IL-6 G.O.T, G.P.T, ALP, total serum bilirubin, Direct TSB, indirect TSB, IgG and IgM in the patient's group for males were (852.94 ±37.67, 99.29 ±9.78, 712.33 ±49.63, 32.02 ±4.33, 48.20 ±2.73,42.60 ±2.86, 125.35 ±7.17, 0.470 ±0.06, 0.160 ±0.02, 0.314 ±0.04, 11.46 ±1.57, 4.26 ±0.42). On the other hand, the mean of these parameters for females were (622.07 ±61.28, 70.53 ±9.26, 582.46 ±31.66, 31.12 ±5.56, 43.69 ±4.21, 41.86 ±4.14, 109.54 ±8.95, 0.390 ±0.04, 0.134 ±0.02, 0.262 ±0.03, 12.93 ±2.15, 3.78 ±0.47). Table 4.7 displays that while there is a highly significant difference in ferritin, there is a difference in CRP and D-Dimer, but no difference in IL-6, G.O.T, G.P.T, ALP, total serum bilirubin, Direct TSB, indirect TSB, IgG, or IgM between groups. Many other studies from different countries agree with this one: There were more females than males tested for the D.Dimer parameter in Egypt (n=119, 51.5% vs. n=112, 48.5%). (Yameny *et al.* 2021). C-reactive protein levels were found to be significantly higher in male patients compared to female patients in Turkey (Yazar *et al.* 2021). A majority of SARS-CoV-2 patients in Iraq are male; however, approximately 74.3% of those with abnormal liver function tests were male (Hwaiz *et al.* 2021).

Table 4.7 Effect of sex in parameters of patients

Parameters	Mean ± SE		P-value
	Male	Female	
Ferritin (ng\mL)	852.94 ±37.67	622.07 ±61.28	0.0004 **
CRP (mg\L)	99.29 ±9.78	70.53 ±9.26	0.0336 *
D-dimer (ng\mL)	712.33 ±49.63	582.46 ±31.66	0.0391 *
IL-6 (pg\mL)	32.02 ±4.33	31.12 ±5.56	0.888 NS
GOT(U\L)	48.20 ±2.73	43.69 ±4.21	0.301 NS
GPT(U\L)	42.60 ±2.86	41.86 ±4.14	0.868 NS
ALP(U\L)	125.35 ±7.17	109.54 ±8.95	0.126 NS
TSB(U\L)	0.470 ±0.06	0.390 ±0.04	0.287 NS
Direct TSB(U\L)	0.160 ±0.02	0.134 ±0.02	0.335 NS
Indirect TSB(U\L)	0.314 ±0.04	0.262 ±0.03	0.351 NS
IgG	11.46 ±1.57	12.93 ±2.15	0.532 NS
IgM	4.26 ±0.42	3.78 ±0.47	0.432 NS
* (P≤0.05), ** (P≤0.01), NS: Non-Significant.			

4.8 Effect of Age Groups in Parameters of Patients

People under the age of 30 made up one of the study's three age groups which showed that the mean of all parameters (ferritin, C.R. P, D. Dimer and IL-6, G.O.T, G.P.T, ALP, total serum bilirubin, Direct TSB, indirect TSB, IgG and IgM) was (511.52 ±84.70, 68.20 ±13.69, 484.01 ±49.50, 16.34 ±5.823, 33.37 ±2.85, 30.70 ±1.34, 104.92 ±7.42, 0.300 ±0.02, 0.100 ±0.01, 0.200 0.01, 6.66 ±2.53, 2.35 ±0.19).

The second group was the people's age (30-50) years which show the mean of all parameters as following (762.69 ±47.51, 66.78 ±9.45, 616.95 ±29.32, 21.66 ±2.72, 40.86 ±2.46, 36.12 ±3.25, 95.77 ±7.04, 0.343 ±0.03, 0.126 ±0.02, 0.230 ±0.03, 7.65 ±1.18, 3.54 ±0.32). The third group was people more than 50 years which show the mean of all parameters as following (865.53 ±51.56, 114.19 ±11.46, 773.21 ±62.47, 46.79 ±6.14, 56.73 ±4.16, 52.58 ±4.01, 145.01 ±9.29, 0.580 ±0.07, 0.191 ±0.02, 0.388 ±0.05, 18.13 ±2.12, 5.24 ±0.60).

According to the results, there is a statistically significant distinction between all parameters accepts direct TSB which showed a significant difference in different groups, as shown in Table 4.8.

Table 4.8 Patient parameter analysis by age range

Parameters	Mean ± SE			P-value
	<30	30-50	>50	
Ferritin (ng\mL)	511.52 ±84.70	762.69 ±47.51	865.53 ±51.56	0.0003 **
CRP (mg\L)	68.20 ±13.69	66.78 ±9.45	114.19 ±11.46	0.0027 **
D-dimer (ng\mL)	484.01 ±49.50	616.95 ±29.32	773.21 ±62.47	0.0023 **
IL-6 (pg\mL)	16.34 ±5.823	21.66 ±2.72	46.79 ±6.14	0.0002 **
GOT(U\L)	33.37 ±2.85	40.86 ±2.46	56.73 ±4.16	0.0001 **
GPT(U\L)	30.70 ±1.34	36.12 ±3.25	52.58 ±4.01	0.0003 **
ALP(U\L)	104.92 ±7.42	95.77 ±7.04	145.01 ±9.29	0.0076 **
TSB(U\L)	0.300 ±0.02	0.343 ±0.03	0.580 ±0.07	0.0043 **
Direct TSB(U\L)	0.100 ±0.01	0.126 ±0.02	0.191 ±0.02	0.021 *
Indirect TSB(U\L)	0.200 0.01	0.230 ±0.03	0.388 ±0.05	0.011 **
IgG	6.66 ±2.53	7.65 ±1.18	18.13 ±2.12	0.0001 **
IgM	2.35 ±0.19	3.54 ±0.32	5.24 ±0.60	0.0013 **
* (P≤0.05), ** (P≤0.01), NS: Non-Significant.				

4.9 Sensitivity and Specificity in Patients and Control Groups

This study showed that the sensitivity in the patients' group was 0.39, while in the healthy control group was 0.47. On the other side the specificity in patients' group was 0.65 while in control group was 0.55 (as showed in Table 4.9).

Table 4.9 Sensitivity and specificity in patients and control groups

Group	**Sensitivity**	**Specificity**
Patients	0.39	0.65
Control	0.47	0.55

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5. CONCLUSIONS AND RECOMMENDATION

We used a cross-control study design to assess the levels of liver enzymes and a few biochemical markers in COVID-19 patients. Age and gender distributions were found to be significantly different from the control group in the present study. Blood levels of GOT, GPT, ALP, TSB, and indirect TSB from patients with COVID-19 were all significantly different from those from the healthy control group, with the exception of direct TSB. The study also found statistically significant mean ferritin levels. The average value of the patients' D. Dimer levels is higher than the reference value range, as measured by comparison to a control group. Both inflammatory indicators were significantly different between healthy people and COVID-19 patients (CRP and IL-6).

Based on the results of the present study, these suggestions are offered as possible solutions.

- The number of samples and the number of areas where samples are taken should be increased.
- Correlation analysis should be performed with other diseases such as pancreatic dysfunction and diabetes.
- More Advanced Biochemical testing studies for COVID-19 Disease such as gamma-glutamyl transpeptidase (GGT) enzyme should be conducted.
- Body mass index (BMI) and hemoglobin A1C (HbA1C) levels are two additional parameters that should be analyzed for correlation.
- There is a need to employ more refined immunological markers like interleukin (IL)-1, IL-8, IL-7, IL-2, tumor necrosis factor (TNF), and procalcitonin.

REFERENCES

Ali, N. 2020. Elevated level of C-reactive protein may be an early marker to predict risk for severity of COVID-19. Journal of medical virology, 92(11): 2409–2411.

Al-khaqani, F., Sabeeh, Y. A., Hashim, A. R. and Farid, H. A. 2022. The Occurrence and the Characteristics of Liver Injury, and its Impactamong Hospitalized COVID-19 Patients in Basrah City-Iraq. Indian Journal of Forensic Medicine and Toxicology, 16(3): 166-172.

Almazroo, O. A., Miah, M. K. and Venkataramanan, R. 2017. Drug metabolism in the liver. Clinics in liver disease, 21(1): 1-20.

Amirian, E. S. and Levy, J. K. 2020. Current knowledge about the antivirals remdesivir (GS-5734) and GS-441524 as therapeutic options for coronaviruses. One health, 9: 100128.

Arjunan, M., Anoop, U. R., Verma, K. and Manimarane, O. 2021. C Reactive Protein based early intervention for home quarantined COVID-19 patients to avoid complications and hospitalization. medRxiv, 2: 13-15.

Aulbach, A. D. and Amuzie, C. J. 2017. Biomarkers in nonclinical drug development. In A Comprehensive guide to toxicology in nonclinical drug development, pp. 447-471, Academic Press.

Baroiu, L., Dumitru, C., Iancu, A., Leșe, A. C., Drăgănescu, M., Baroiu, N. and Anghel, L. 2021. COVID-19 impact on the liver. World Journal of Clinical Cases, 9(16): 3814.

Benvenuto, D., Giovanetti, M., Ciccozzi, A., Spoto, S., Angeletti, S. and Ciccozzi, M. 2020. The 2019-new coronavirus epidemic: evidence for virus evolution. Journal of medical virology, 92(4): 455-459.

Bernal-Monterde, V., Casas-Deza, D., Letona-Giménez, L., de la Llama-Celis, N., Calmarza, P., Sierra-Gabarda, O. and M. Arbones-Mainar, J. 2020. SARS-CoV-2 infection induces a dual response in liver function tests: association with mortality during hospitalization. Biomedicines, 8(9): 328.

Burke, R. M., Midgley, C. M., Dratch, A., Fenstersheib, M., Haupt, T., Holshue, M. and Rolfes, M. A. 2020. Active monitoring of persons exposed to patients with

confirmed COVID-19—United States, January–February 2020. Morbidity and Mortality Weekly Report, 69(9): 245.

Cai, Q., Huang, D., Ou, P., Yu, H., Zhu, Z., Xia, Z. and Chen, J. 2020. COVID-19 in a designated infectious diseases hospital outside Hubei Province, China. Allergy, 75(7): 1742-1752.

Cao, H., Ruan, L., Liu, J. and Liao, W. 2020. The clinical characteristic of eight patients of COVID-19 with positive RT-PCR test after discharge. Journal of medical virology, 92(10): 2159-2164.

Cascella, M., Rajnik, M., Aleem, A., Dulebohn, S. C. and Di Napoli, R. 2022. Features, evaluation, and treatment of coronavirus (COVID-19). Statpearls [internet].

Chai, X. 2019. Hu l, Zhang y, Han W. Lu Z, Ke A, *et al.* Specific ACE2 expression in cholangiocytes may cause liver damage after.

Chaibi, S., Boussier, J., El Hajj, W., Abitbol, Y., Taieb, S., Horaist, C. and Nahon, S. 2021. Liver function test abnormalities are associated with a poorer prognosis in COVID-19 patients: results of a French cohort. Clinics and research in hepatology and gastroenterology, 45(5): 101556.

Chan, J. F. W., Choi, G. K. Y., Tsang, A. K. L., Tee, K. M., Lam, H. Y., Yip, C. C. Y. and Yuen, K. Y. 2015. Development and evaluation of novel real-time reverse transcription-PCR assays with locked nucleic acid probes targeting leader sequences of human-pathogenic coronaviruses. Journal of clinical microbiology, 53(8): 2722-2726.

Chau, T. N., Lee, K. C., Yao, H., Tsang, T. Y., Chow, T. C., Yeung, Y. C. and Lai, C. L. 2004. SARS-associated viral hepatitis caused by a novel coronavirus: report of three cases. Hepatology, 39(2): 302-310.

Chen, G., Wu, D. I., Guo, W., Cao, Y., Huang, D., Wang, H. and Ning, Q. 2020. Clinical and immunological features of severe and moderate coronavirus disease 2019. The Journal of clinical investigation, 130(5): 2620-2629.

Chen, N., Zhou, M., Dong, X., Qu, J., Gong, F., Han, Y. and Zhang, L. 2020. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. The lancet, 395(10223): 507-513.

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

Fang, Y., Zhang, H., Xie, J., Lin, M., Ying, L., Pang, P. and Ji, W. 2020. Sensitivity of chest CT for COVID-19: comparison to RT-PCR. Radiology, 4: 27-29.

Farid, E., Sridharan, K., Alsegai, O. A., Khawaja, S. A., Mansoor, E. J., Teraifi, N. A. and Salman, J. A. 2021. Utility of inflammatory biomarkers in patients with COVID-19 infections: Bahrain experience. Biomarkers in Medicine, 15(8): 541-549.

Fataftah, J. Z., Tayyem, R., Qandeel, H., Baydoun, H. A., Al Manasra, A. R. A., Tahboub, A. and Al-Mousa, A. 2022. The effect of COVID 19 on liver parenchyma detected and measured by CT scan hounsfield units. Journal of Research in Medical Sciences: The Official Journal of Isfahan University of Medical Sciences, 2: 7.

Fix, O. K., Hameed, B., Fontana, R. J., Kwok, R. M., McGuire, B. M., Mulligan, D. C. and Chung, R. T. 2020. Clinical best practice advice for hepatology and liver transplant providers during the COVID-19 pandemic: AASLD expert panel consensus statement. Hepatology, 72(1): 287-304.

Gao, Y., Li, T., Han, M., Li, X., Wu, D., Xu, Y. and Wang, L. 2020. Diagnostic utility of clinical laboratory data determinations for patients with the severe COVID-19. Journal of medical virology, 92(7): 791-796.

Gong, J., Dong, H., Xia, Q. S., Huang, Z. Y., Wang, D. K., Zhao, Y. and Lu, F. E. 2020. Correlation analysis between disease severity and inflammation-related parameters in patients with COVID-19: a retrospective study. BMC infectious diseases, 20(1): 1-7.

Gopal, D. V. and Rosen, H. R. 2000. Abnormal findings on liver function tests: Interpreting results to narrow the diagnosis and establish a prognosis. Postgraduate medicine, 107(2): 100-114.

Guan, W. J., Ni, Z. Y., Hu, Y., Liang, W. H., Ou, C. Q., He, J. X. and Zhong, N. S. 2020. Clinical characteristics of coronavirus disease 2019 in China. New England journal of medicine, 382(18): 1708-1720.

Gumucio, J. J. 1996. Structural and functional organization of the liver. Liver and biliary disease, 2: 3-19.

Gupta, A., Madhavan, M. V., Sehgal, K. and Nair, N. Mahajan Sh., Sehrawat TS, *et al.*(2020). Extrapulmonary manifestations of COVID-19. Nature Medicine, 26(7): 1017-32.

Hajifathalian, K., Krisko, T., Mehta, A., Kumar, S., Schwartz, R., Fortune, B. and Cohen, D. 2020. Gastrointestinal and hepatic manifestations of 2019 novel coronavirus disease in a large cohort of infected patients from New York: clinical implications. Gastroenterology, 159(3): 1137-1140.

Han, H., Yang, L., Liu, R., Liu, F., Wu, K. L., Li, J. and Zhu, C. L. 2020. Prominent changes in blood coagulation of patients with SARS-CoV-2 infection. Clinical Chemistry and Laboratory Medicine (CCLM), 58(7): 1116-1120.

Harrison, P. M. and Arosio, P. 1996. The ferritins: molecular properties, iron storage function and cellular regulation. Biochimica et biophysica acta (BBA)-bioenergetics, 1275(3): 161-203.

Hoekstra, L. T., de Graaf, W., Nibourg, G. A., Heger, M., Bennink, R. J., Stieger, B. and van Gulik, T. M. 2013. Physiological and biochemical basis of clinical liver function tests: a review. Annals of surgery, 257(1): 27-36.

Huang, C., Wang, Y., Li, X., Ren, L., Zhao, J., Hu, Y. and Cao, B. 2020. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. The lancet, 395(10223): 497-506.

Huang, H., Chen, S., Li, H., Zhou, X. L., Dai, Y., Wu, J. and Ge, M. 2020. The association between markers of liver injury and clinical outcomes in patients with COVID-19 in Wuhan. Alimentary pharmacology and therapeutics, 52(6): 1051-1059.

Huang, P., Liu, T., Huang, L., Liu, H., Lei, M., Xu, W. and Liu, B. 2020. Use of chest CT in combination with negative RT-PCR assay for the 2019 novel coronavirus but high clinical suspicion. Radiology, 3: 23-25.

Hui, D. S., Azhar, E. I., Madani, T. A., Ntoumi, F., Kock, R., Dar, O. and Petersen, E. 2020. The continuing 2019-nCoV epidemic threat of novel coronaviruses to global health—The latest 2019 novel coronavirus outbreak in Wuhan, China. International journal of infectious diseases, 91: 264-266.

Hundt, M. A., Deng, Y., Ciarleglio, M. M., Nathanson, M. H. and Lim, J. K. 2020. Abnormal liver tests in COVID-19: a retrospective observational cohort study of 1,827 patients in a major US hospital network. Hepatology, 72(4): 1169-1176.

Hwaiz, R., Merza, M., Hamad, B., HamaSalih, S., Mohammed, M. and Hama, H. 2021. Evaluation of hepatic enzymes activities in COVID-19 patients. International Immunopharmacology, 97: 107701.

Ishibashi, H., Nakamura, M., Komori, A., Migita, K. and Shimoda, S. 2009. Liver architecture, cell function, and disease. In Seminars in immunopathology. Springer-Verlag, 31(3): 399-409.

Kalra, A., Yetiskul, E. and Wehrle, C.J. 2022. Web site. https://www.ncbi.nlm.nih.gov/books/NBK535438 Date of access: 08.03.2022.

Kolb-Bachofen, V. 1991. A review on the biological properties of C-reactive protein. Immunobiology, 183(1-2): 133-145.

Kubistova, A., Horacek, J. and Novak, T. (2012). Increased interleukin-6 and tumor necrosis factor alpha in first episode schizophrenia patients versus healthy controls. Psychiatria danubina, 24(1): 153-156.

Kwo, P.Y., Cohen, S. M. and Lim, J. K. 2017. ACG clinical guideline: evaluation of abnormal liver chemistries. Official journal of the American College of Gastroenterology| ACG, 112(1): 18-35.

Lai, C.C., Ko, W. C., Lee, P. I., Jean, S. S. and Hsueh, P. R. 2020. Extra-respiratory manifestations of COVID-19. International journal of antimicrobial agents, 56(2): 106024.

Leung, W. K., To, K. F., Chan, P. K., Chan, H. L., Wu, A. K., Lee, N. and Sung, J. J. (2003). Enteric involvement of severe acute respiratory syndrome-associated coronavirus infection. Gastroenterology, 125(4): 1011-1017.

Li, Q., Guan, X., Wu, P., Wang, X., Zhou, L., Tong, Y. and Feng, Z. 2020. Early transmission dynamics in Wuhan, China, of novel coronavirus–infected pneumonia. New England journal of medicine., 3(2): 33-35.

Liu, M., He, P., Liu, H. G., Wang, X. J., Li, F. J., Chen, S. and Li, C. H. 2020. Clinical characteristics of 30 medical workers infected with new coronavirus pneumonia. Zhonghua jie he he hu xi za zhi= Zhonghua jiehe he huxi zazhi= Chinese journal of tuberculosis and respiratory diseases, 43: E016-E016.

Liu, T., Zhang, J., Yang, Y., Ma, H., Li, Z., Zhang, J. and Yi, J. 2020. The potential role of IL-6 in monitoring severe case of coronavirus disease 2019. MedRxiv, 4(1): 91.

Lowe, D., Sanvictores, T. and John, S. 2021. Alkaline phosphatase.[updated 2021 Aug 11]. Stat pearls [internet]. Treasure Island (FL): Stat Pearls Publishing.

Memish, Z. A., Zumla, A. I., Al-Hakeem, R. F., Al-Rabeeah, A. A. and Stephens, G. M. 2013. Family cluster of Middle East respiratory syndrome coronavirus infections. New England Journal of Medicine, 368(26): 2487-2494.

Mendizabal, M., Pinero, F., Ridruejo, E. anders, M., Silveyra, M. D., Torre, A. and Silva, M. O. 2021. Prospective Latin American cohort evaluating outcomes of patients with COVID-19 and abnormal liver tests on admission. Annals of hepatology, 21, 100298.

Nardo, A. D., Schneeweiss-Gleixner, M., Bakail, M., Dixon, E. D., Lax, S. F. and Trauner, M. 2021. Pathophysiological mechanisms of liver injury in COVID-19. Liver International, 41(1): 20-32.

O'Brien, L., Hosick, P. A., John, K., Stec, D. E. and Hinds Jr, T. D. 2015. Biliverdin reductase isozymes in metabolism. Trends in Endocrinology and Metabolism, 26(4): 212-220.

Palosuo, T., Husman, T., Koistinen, J. and Aho, K. 1986. C-reactive protein in population samples. Acta Medica Scandinavica, 220(2): 175-179.

Pepys, M. B. 1981. C-reactive protein fifty years on. The Lancet, 317(8221): 653-657.

Pirola, C. J. and Sookoian, S. 2020. COVID-19 and ACE2 in the liver and gastrointestinal tract: putative biological explanations of sexual dimorphism. Gastroenterology, 159(4): 1620.

Pyrc, K., Berkhout, B. and Van Der Hoek, L. (2007). Identification of new human coronaviruses. Expert review of anti-infective therapy, 5(2): 245-253.

Rahman, A. and Sarkar, A. 2019. Risk factors for fatal middle east respiratory syndrome coronavirus infections in Saudi Arabia: analysis of the WHO Line List, 2013–2018. American journal of public health, 109(9): 1288-1293.

Ruan, Q., Yang, K., Wang, W., Jiang, L. and Song, J. 2020. Clinical predictors of mortality due to COVID-19 based on an analysis of data of 150 patients from Wuhan, China. Intensive care medicine, 46(5): 846-848.

Ruiz, A. R. G., Crespo, J., Martínez, R. M. L., Iruzubieta, P., Mercadal, G. C., Garcés, M. L. and Ruiz, M. M. 2021. Measurement and clinical usefulness of bilirubin in liver disease. Advances in Laboratory Medicine/Avances en Medicina de Laboratorio, 2(3): 352-361.

Santos-Sánchez, N. F. and Salas-Coronado, R. 2020. Origin, structural characteristics, prevention measures, diagnosis and potential drugs to prevent and COVID-19. Medwave, 20(8): e8037.

Schofield, J., Leelarathna, L. and Thabit, H. 2020. COVID-19: Impact of and on Diabetes. Diabetes Therapy, 11(7): 1429-1435.

Sobotka, L. A., Esteban, J., Volk, M. L., Elmunzer, B. J. and Rockey, D. C. 2022. Acute liver injury in patients hospitalized with COVID-19. Digestive diseases and sciences, 67(8): 4204-4214.

Sparling, D. W. 2016. Ecotoxicology essentials: environmental contaminants and their biological effects on animals and plants. Academic Press.

Stec, D. E., John, K., Trabbic, C. J., Luniwal, A., Hankins, M. W., Baum, J. and Hinds Jr, T. D. 2016. Bilirubin binding to PPARα inhibits lipid accumulation. PloS one, 11(4): e0153427.

Tanaka, T. and Kishimoto, T. 2012. Targeting interleukin-6: all the way to treat autoimmune and inflammatory diseases. International journal of biological sciences, 8(9): 1227.

Tang, D., Comish, P. and Kang, R. 2020. The hallmarks of COVID-19 disease. PLoS pathogens, 16(5): e1008536.

Thachil, J., Fitzmaurice, D. A. and Toh, C. H. (2010). Appropriate use of D-dimer in hospital patients. The American journal of medicine, 123(1): 17-19.

Tian, S., Hu, N., Lou, J., Chen, K., Kang, X., Xiang, Z. and Zhang, J. 2020. Characteristics of COVID-19 infection in Beijing. Journal of infection, 80(4): 401-406.

Tolia, V. M., Chan, T. C. and Castillo, E. M. 2020. Preliminary results of initial testing for coronavirus (COVID-19) in the emergency department. Western Journal of Emergency Medicine, 21(3): 503.

Walters, M. I., & Gerarde, H. W. 1970. An ultramicromethod for the determination of conjugated and total bilirubin in serum or plasma. Microchemical Journal, 15(2), 231-243.

Wang, D., Hu, B., Hu, C., Zhu, F., Liu, X., Zhang, J. and Peng, Z. 2020. Clinical characteristics of 138 hospitalized patients with 2019 novel coronavirus–infected pneumonia in Wuhan, China. Jama, 323(11): 1061-1069.

Wang, G., Wu, C., Zhang, Q., Wu, F., Yu, B., Lv, J. and Zhong, Y. 2020. C-reactive protein level may predict the risk of COVID-19 aggravation. In Open forum infectious diseases, 7(5): 153.

Wang, M., Yan, W., Qi, W., Wu, D., Zhu, L., Li, W. and Ning, Q. 2020. Clinical characteristics and risk factors of liver injury in COVID-19: a retrospective cohort study from Wuhan, China. Hepatology international, 14(5): 723-732.

Wang, N., Shi, X., Jiang, L., Zhang, S., Wang, D., Tong, P. and Wang, X. 2013. Structure of MERS-CoV spike receptor-binding domain complexed with human receptor DPP4. Cell research, 23(8): 986-993.

WHO, S. A. 2020. WHO coronavirus disease (COVID-19) dashboard. World Health Organization.

World Health Organization. 2020. Laboratory testing for coronavirus disease 2019 (COVID-19) in suspected human cases: interim guidance, 2 March 2020 (No. WHO/COVID-19/laboratory/2020.4). World Health Organization.

Wu, C., Chen, X., Cai, Y., Zhou, X., Xu, S., Huang, H. and Song, Y. 2020. Risk factors associated with acute respiratory distress syndrome and death in patients with coronavirus disease 2019 pneumonia in Wuhan, China. JAMA internal medicine, 180(7): 934-943.

Wu, J. T., Leung, K. and Leung, G. M. 2020. Nowcasting and forecasting the potential domestic and international spread of the 2019-nCoV outbreak originating in Wuhan, China: a modelling study. The Lancet, 395(10225): 689-697.

Xu, Z., Shi, L., Wang, Y., Zhang, J., Huang, L., Zhang, C. and Wang, F. S. 2020. Pathological findings of COVID-19 associated with acute respiratory distress syndrome. The Lancet respiratory medicine, 8(4): 420-422.

Yameny, A. A. 2021. D-dimer levels in COVID-19 out-hospitalized patients in Egypt. Journal of medical and life science, 3(1): 19-24.

Yang, A. P., Liu, J. P., Tao, W. Q. and Li, H. M. 2020. The diagnostic and predictive role of NLR, d-NLR and PLR in COVID-19 patients. International immunopharmacology, 84: 106504.

Yazar, H., Kayacan, Y. and Zdin M. 2019. Investigation of C-reactive Protein and D-dimer Findings in Patients with COVID-19. Bezmialem Science, 9(1): 4-8.

Zhang, J. J., Dong, X., Cao, Y. Y., Yuan, Y. D., Yang, Y. B., Yan, Y. Q. and Gao, Y. D. 2020. Clinical characteristics of 140 patients infected with SARS-CoV-2 in Wuhan, China. Allergy, 75(7): 1730-1741.

Zhang, J. J., Dong, X., Cao, Y. Y., Yuan, Y. D., Yang, Y. B., Yan, Y. Q. and Gao, Y. D. 2020. Clinical characteristics of 140 patients infected with SARS-CoV-2 in Wuhan, China. Allergy, 75(7): 1730-1741.

Zhang, J., Xie, B. and Hashimoto, K. 2020. Current status of potential therapeutic candidates for the COVID-19 crisis. Brain, behavior, and immunity, 87, 59-73.

Zhang, W., Du, R. H., Li, B., Zheng, X. S., Yang, X. L., Hu, B. and Zhou, P. 2020. Molecular and serological investigation of 2019-nCoV infected patients: implication of multiple shedding routes. Emerging microbes and infections, 9(1): 386-389.

Zhong, N. S., Zheng, B. J., Li, Y. M., Poon, L. L. M., Xie, Z. H., Chan, K. H. and Guan, Y. (2003). Epidemiology and cause of severe acute respiratory syndrome (SARS) in Guangdong, People's Republic of China, in February, 2003. The Lancet, 362(9393): 1353-1358.

Zhou, B., She, J., Wang, Y. and Ma, X. 2020. Utility of ferritin, procalcitonin, and C-reactive protein in severe patients with 2019 novel coronavirus disease.

Zhou, F., Yu, T., Du, R., Fan, G., Liu, Y., Liu, Z. and Cao, B. 2020. Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. The lancet, 395(10229): 1054-1062.

Zhu, N., Zhang, D., Wang, W., Li, X., Yang, B., Song, J. and Tan, W. 2020. A novel coronavirus from patients with pneumonia in China, 2019. New England journal of medicine, 2: 19-21.

APPENDICES

APPENDIX 1. Etik Kurul Raporu (Arapça)

APPENDIX 2. Etik Kurul Raporu (Türkçe tercüme edilmiş)

APPENDIX 1. Etik Kurul Raporu (Arap

APPENDIX 2. Etik Kurul Raporu (Türkçe tercüme edilmiş)

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