Ministry of Higher Education and Scientific Research.

University of Babylon/ College of Science.

Department of Biology...Microbiology.

STUDY FOR IMMUNOGLOBULIN CONCENTRATIONS IN COVID 19 VACCINATED PERSON.

A research submitted to the College of Science, Department of Biology, as part of the requirements for obtaining a Bachelor's degree in Biology, Microbiology Branch.

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بسم الله الرحمن الرحيم

" وَلَقَدْ آتَيْنَا دَاوُودَ وَسُلَيْمَانَ عِلْمًا ۖ وَقَالَا الْحَمْدُ لِلَّهِ الَّذِي فَضَّلَنَا عَلَىٰ كَثِيرٍ
مِّنْ عِبَادِهِ الْمُؤْمِنِينَ* وَوَرِثَ سُلَيْمَانُ دَاوُودَ ۖ وَقَالَ يَا أَيُّهَا النَّاسُ
عُلِّمْنَا مَنطِقَ الطَّيْرِ وَأُوتِينَا مِن كُلِّ شَيْءٍ ۖ إِنَّ هَٰذَا لَهُوَ الْفَضْلُ الْمُبِينُ"

{ سورة النمل، الآيات 15، 16}

صدق الله العلي العظيم

الإهداء و الشكر :

الى صاحب السيرة العطرة، و الفكر المستنير؛

فلقد كان له الفضل الأوّل في بلوغي التعليم العالي

(والدي الحبيب)، أطال الله في عُمره.

الى من وضعتني على طريق الحياة، و جعلتني رابط الجأش،

وراعتني حتى صرت كبيرًا

(أمي الغالية)، طيّب الله ثراها.

الى إخوتي؛ من كان لهم بالغ الأثر في كثير من العقبات و الصعاب.

الى جميع أساتذتي الكرام؛ ممن لم يتوانوا في مد يد العون لي

أهدي اليكم بحثي.....

❖ *summary*

A study was conducted randomized on vaccinated people in Babylon governorate for both sexes, the number of vaccinated people were 695 people, including 561person taken Pfizer vaccine, 120 people taken Sinovac, and 14 people taken AstraZeneca, The study report shows that the highest rate of vaccinated people was Pfizer in the rate of (80.71%) and followed by the Sinovac vaccine in the rate of (17.36%), While less percentage was AstraZeneca vaccine in the rate of (2.01%). All vaccinated people in this study were distributed according to gender (males and females), were found the number of vaccinated females more than males, the highest number of vaccinated females with Pfizer vaccine (415), followed by the Sinovac vaccine (31), While less number of vaccinated females with the AstraZeneca vaccine (8).

Blood was collected from some vaccinated and unvaccinated ,then serum was separated and then stored in freezing until estimation total protein by Total protein kit and Estimation the immunoglobulin and complement component C3 and C4 by Mansin method (single radial immunodiffussion).

The results were appeared investigated the levels of C3, C4, IgG, IgM, and Total protein concentrations in the serum of the vaccinated people, and compared them with unvaccinated people. The study had shown elevated levels of C3, IgG, IgM, and Total protein concentrations serum of the vaccinated people compared with the control, while the concentration of C4 is low compared with the control .

The conclusions of this study found most common type of vaccine was taken was Pfizer followed with Sinopharm and less with AstraZeneca, and found vaccines Enhanced humoral immune response .

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CHAPTER ONE

INTRODUCTION

1.1: Introduction

Coronavirus is a zoonotic virus, an RNA virus in the family Coronaviridae of the order Nidovirales[1].It is a family of viruses that cause respiratory infections, which were first isolated in 1937 and designated coronaviruses, because they have a crown-like appearance under microscopy, in 1965[2].

The types of coronavirus known to date are as follows: the alpha corona -viruses HCoV-229E and HCoV-NL63; the beta coronaviruses HCoV-OC43 and HCoV-HKU1; SARS-CoV, which causes severe acute respiratory syndrome (SARS); MERS-CoV, which causes Middle East respiratory syndrome (MERS); and SARS-CoV-2, a

new coronavirus described in late 2019 after cases were reported in China[2], which causes the disease known as coronavirus disease 2019 (COVID-19) .

The different vaccine technologies like inactivation, attenuation, nucleic acid, viral vector, subunit, and viral particle based techniques are employed to develop a safe and highly efficient vaccine. The progress in vaccine development for SARS-CoV2 is much faster in the history of science. Even though there exist of lot of limitations, continuous efforts has put forward so as to develop highly competent and effective vaccine for many human and animal linked diseases due to its unlimited prospective. [3].

The aim of the research to survey Types of vaccine in immunization pupils in University of Babylon and study some humoral parameters in groups of them to achieve this aim, followed the steps

1- Information's from pupils were collected on type of vaccine and some Questioners.

2- Blood samples was collected randomly from immunized (test group) and un immunized (as control)

3- Serum was separated to determine total protein concentrations and C-Reactive protein

4- Estimation the Concentrations of immunoglobulin IgG and IgM by single radial immun diffusion

5- Estimations the concentrations of complement component C3 and C4 By single radial immun diffusion.

6- study the relation ships among immunological parameters by statistical analysis

CHAPTER TWO

LITERATURE REVIEW.

2-Literature Review:

2.1: Corona disease:

2.1.1: Historical review:

Severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2), a member of the subgenus Sarbecovirus, causes coronavirus disease 2019 (COVID-19), and the first cases occurred in late 2019 [4]. On 11 March 2020, the World Health Organization (WHO) declared the SARS-CoV-2 outbreak to be a pandemic. As of 28 May 2021, almost 3,520,000 deaths and 169,480,000 cases of SARS-CoV-2 infection have been reported worldwide [5]. An understanding of the immune responses to SARS-CoV-2 and the coronavirus vaccines is necessary because of its rapid spread.

In response to SARS-CoV-2 infection, humans produce specific antibodies, CD4+ T cells, which activate high-affinity antibodies produced by B cells, and CD8+ T cells, which destroy infected cells [6–8]

During a global pandemic, mRNA vaccines are the fastest available vaccines due to their short production time and low biological requirements [4]. These mRNA-based vaccines avoid the risk of integrating viral genetic material into the host cell's genome and are capable of producing pure viral protein. The technology of producing vaccines against COVID-19 in the form of lipid nanoparticles (LNP) enables the delivery of precise genetic information along with an adjuvant effect to antigen-presenting cells [4]. The SARS-CoV-2 vaccines are based on the virus's mRNA, specifically on the fragment encoding the spike (S) protein, which attaches the virion to the host cell's membrane [9]. Moreover, the S1 subunit of the S protein contains an immunologically relevant receptor-binding domain (RBD), which is a key antibody target [4]. According to clinical studies, subjects developed a strong dose-dependent antibody response to the S protein after the first and second inoculations [10]. Neutralizing antibodies were found in all subjects after the second inoculation, and the antibody titers were equal to or greater than the neutralizing antibody titers of COVID-19 patients [10].

The mRNA vaccine is a Comirnaty concentrate from Pfizer and BioNTech. One dose (0.3 mL) contained 30 micrograms of the COVID-19 mRNA vaccine. The active substance of the

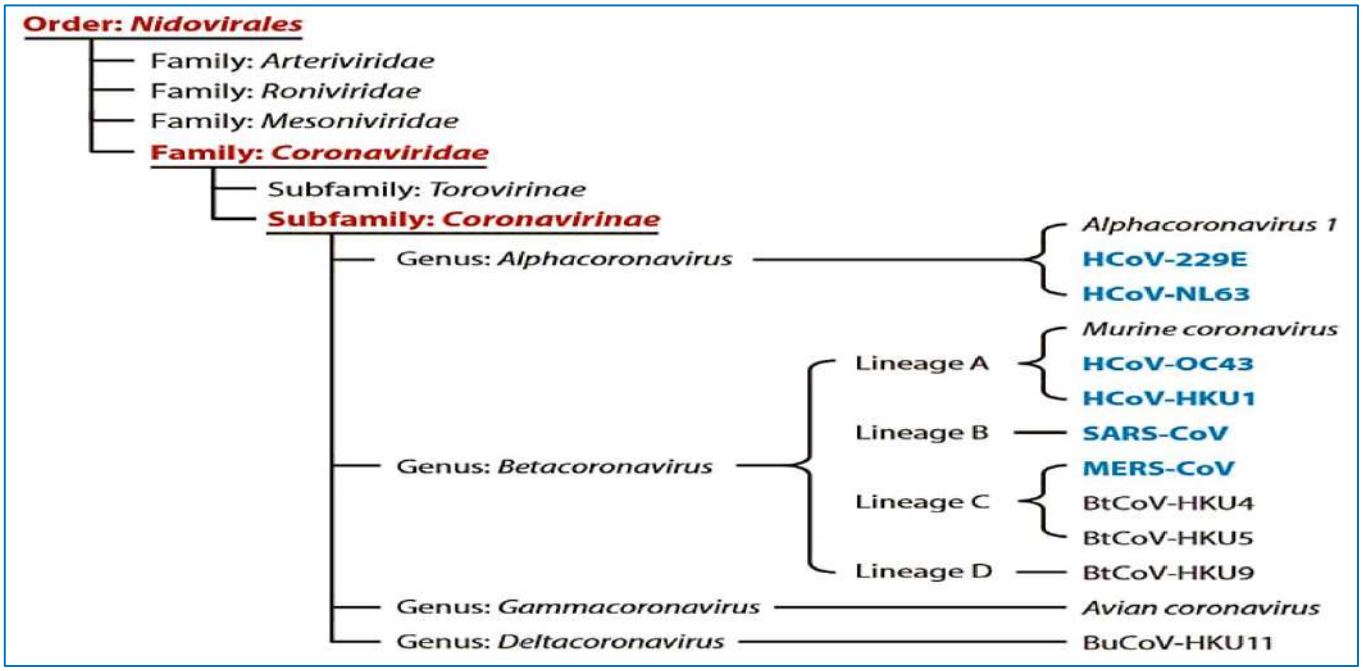
preparation is the mRNA that encodes the spike protein of the virus and acts as an antigen [11,12]. The vaccine also contains four types of fats in the form of lipid nanoparticles: (4-hydroxybutyl)azanediyl, bis(hexane-6,1-diyl), bis(hexyl-2-decanoate), (ALC-0315),2-((polyethylene glycol)-2000)-N, N-ditetradecylacetamide (ALC-0159), 1,2distearoyl-sn-glycero-3-phosphocholine (DSPC), cholesterol and other substances such as potassium chloride, potassium dihydrogen phosphate, sodium chloride, disodium phosphate dehydrate, saccharose, and water for injections [11]. Comirnaty is indicated for the active immunization of a person from the age of 16 years for the prevention of COVID19 disease caused by the SARS-CoV-2 virus. The product is administered intramuscularly into the deltoid muscle after dilution, and a second dose is administered after at least 21 days [11,12].

The immune response to COVID-19 is poorly understood. There is insufficient knowledge about safe and immunologically effective vaccination strategies against SARS-CoV-2, and it is not known which vaccination strategies will be most effective [13]. Moreover, there is still insufficient information on the short- and long-term effects of these mRNA vaccinations. The aim of the research was to determine the humoral and cellular responses in vaccinated persons and in convalescents.

2.1.2: classification of coronavirus:

Coronaviruses members of Coronaviridae family, and are Orthocoronavirinae subfamily of the Nidovirales order. Among RNA viruses, CoVs have the largest genomes, with genome sizes ranging 26 - 32 kb. Depending on genetic and antigenic criteria, coronaviruses are classified into four genera: alphacoronavirus (a-CoV), betacoronavirus (B-CoV), gammacoronavirus (y-CoV), and deltacoronavirus (8-CoV)[14]. Bats and mice act as reservoirs for alpha and beta coronaviruses, while birds serve as reservoirs for gamma and delta coronaviruses[15].SARS-CoV-2 belongs to the subgenus Sarbecovirus of the genus Betacoronavirus, according to phylogenetic analysis as shown in Figure (1-1) [16].

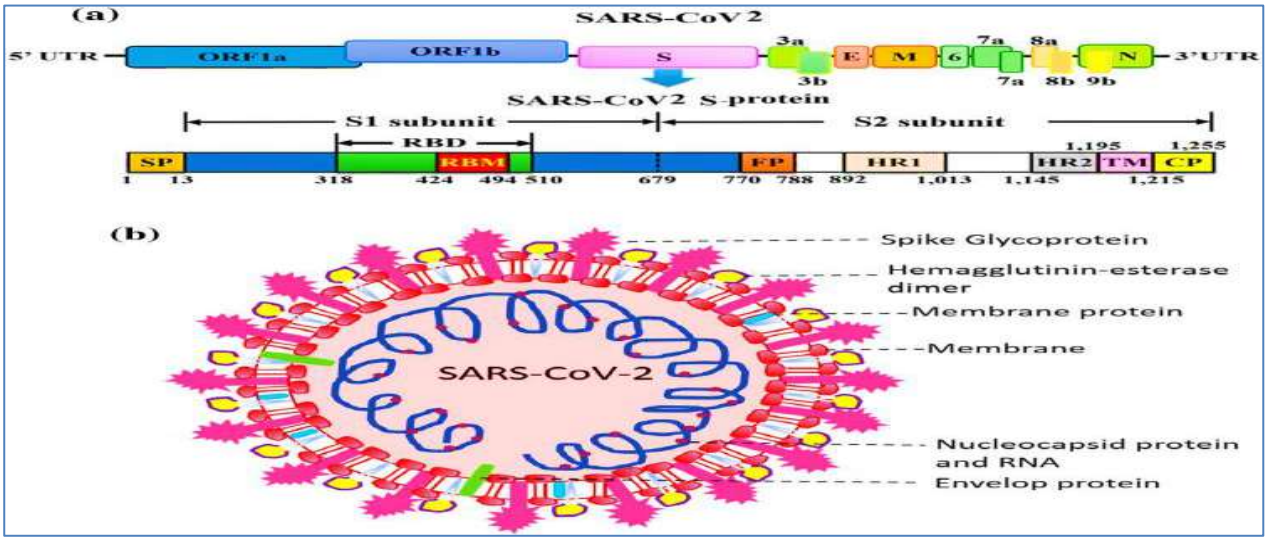
Figure(2.1): Classification of SARS-CoV-2.



2.1.3: Structure of coronavirus:

Coronaviruses are large particles viruses that are spherical in shape and have spikes that form a surface projection[17].The particles are about 125 nanometers in diameter. The envelope diameter is 85nm, and the spikes are 20 nm long; họwever, SARS-spikes Cov-2S ato larger, increasing its pathogenicity. The viral envelope, like any other membrane, was made up of a lipid bilayer and a variety of proteins with structural functions, including membrane (M), envelope (E), and spike (S)[17]in a ratio of E:S:M 1: 20:300[18].The particle's total number of spikes is about 74 [19].However, as shown in figure (1-2), SARS-CoV-2 has another short projection of a proteinous structure known as hemagglutinin esterase (HE)[20].

The spikes are folded into homotrimers and are divided into two sections, Sl is the spike's head structure, with receptor-binding domains (RBD) that The spikes are folded into homotrimers and are divided into two sections, contain the signal peptide, and s2 is the spike's stem, with heptad repeat regions (HR1 and HR2) and a putative fusion peptide (F). The transmembrane domain and endo-domain, are also present. All of these subunits aid in the activation of these subunits to facilitate fusion, which is essential for pathogenesis and maintaining envelope integrity [21].Finally, the nucleocapsid is made up of nucleic acid (positive-sense single-stranded RNA genome) folded on several copies of proteins (nucleocapsid, N). The organization is organized in a continuous bead-on-a- string pattern



[21].All of these structures are essential for the virus's defense when it is outside of host cells [22].

Figure (2.2): Structure of SARS-CoV-2(Cui et al.; 2019).

2.1.4:Coronavirus Transmission:

SARS-CoV-2, the virus, mainly spreads from person to person. People release respiratory fluids during exhalation (e.g., quiet breathing, speaking, singing, exercise, coughing, sneezing) in the form of droplets across a spectrum of sizes. These droplets carry virus and transmit infection. The largest droplets settle out of the air rapidly, within seconds to minutes. The smallest very fine droplets, and aerosol particles formed when these fine droplets rapidly dry, are small enough that they can remain suspended in the air for minutes to hours.

Risk of transmission is greatest within three to six feet of an infectious source where the concentration of these very fine droplets and particles is greatest. If you breathe them in or swallow them, the virus can get into your body. Some people who have the virus don't have symptoms, but they can still spread the virus.

While less likely, you can also get the virus from touching a surface or object the virus is on, then touching your mouth, nose, or possibly your eyes. Most viruses can live for several hours on a surface that they land on. A study shows that SARS-CoV-2 can last for several hours on various types of surfaces:

i. Copper (pennies, teakettles, cookware): 4 hours.
ii. Cardboard (shipping boxes): up to 24 hours.
iii. Plastic (milk containers, detergent bottles, subway and bus seats, elevator buttons):2 to 3 days.
iv. Stainless steel (refrigerators, pots and pans, sinks, some water bottles): 2 to 3 days.

That's why it's important to wash or sanitize your hands regularly and disinfect surfaces to get rid of the virus.

Some dogs and cats have tested positive for the virus. A few have shown signs of illness. There's no evidence that humans can catch this coronavirus from an animal, but it appears it can be passed from humans to animals.[23]

2.1.5: Symptoms of coronavirus :

People with COVID-19 have had a wide range of symptoms reported – ranging from mild symptoms to severe illness. Symptoms may appear 2-14 days after exposure to the virus. Anyone can have mild to severe symptoms. People with these symptoms may have COVID-19 including: Fever or chills, Cough, Shortness of breath or difficulty breathing, Fatigue, Muscle or body aches, Headache, New loss of taste or smell, Sore throat, Congestion or runny nose, Nausea or vomiting, and Diarrhea. This list does not include all possible symptoms . Older adults and people who have severe underlying medical conditions like heart or lung disease or diabetes seem to be at higher risk for developing more serious complications from COVID-19 illness [24].

2.2: Vaccines and types of vaccines used against COVID-19:

- The COVID-19 vaccine may help:

  - ✓ Prevents you from contracting COVID-19 or from becoming seriously ill or dying from COVID-19

- ✓ Preventing the spread of COVID-19 to others
- ✓ The number of vaccinated community members increases against COVID-19 - which slows the spread of the disease and contributes to herd immunity (so-called herd immunity(
- ✓ It prevents the virus that causes COVID-19 from spreading and replication, the two processes that allow it to create a mutation that may be better able to resist vaccines.

- **The COVID-19 vaccine can cause minor side effects after the first or second dose, including:** Pain, redness, or swelling where the vaccine was injected, fever, fatigue, headache, muscle pain, chills, Joint pain, Nausea and vomiting, feeling, unwell, and swollen lymph nodes

You may be monitored for 15 minutes after receiving the COVID-19 vaccine to be sure of any immediate reaction to your doctor. Most side effects go away within a few days. The side effects of the second dose may be more severe. Many people do not get side effects.

2.2.1: Pfizer-BioNTech COVID-19 Vaccine (known as COMIRNATY):

❖ General Information

a. Manufacturer: Pfizer, Inc., and BioNTech
b. Number of Shots: 2 shots, 21 days apart Moderately or severely immunocompromised people ages 5 years and older should get an additional primary shot at least 28 days after their second shot.
c. Booster Shot: Everyone ages 12 years and older is recommended to get a booster shot at least 5 months after completing their Pfizer-BioNTech primary series. Teens 12-17 years old can only get a Pfizer-BioNTech COVID-19 vaccine booster. For adults 18 years and older, a booster dose of either Pfizer-BioNTech or Moderna (mRNA COVID-19 vaccines) is preferred in most situations.
d. Type of Vaccine: mRNA
e. How Given: Shot in the muscle of the upper arm
f. Does NOT Contain: Eggs, preservatives, latex, metals
g. Name: BNT162b2
h. Brand name: COMIRNATY

❖ Ingredients in the original Pfizer-BioNTech COVID-19 vaccine for people ages 12 years and older

The original Pfizer-BioNTech COVID-19 vaccine for people ages 12 years and older contains the following ingredients:

Type of Ingredient:

A. Messenger ribonucleic acid (mRNA):

Nucleoside-modified mRNA encoding the viral spike (S) glycoprotein of SARS-CoV-2

Purpose : Provides instructions the body uses to build a harmless piece of a protein from the virus that causes COVID-19. This protein causes an immune response that helps protect the body from getting sick with COVID-19 in the future.

B. Lipids (fats):

- ✓ 2[(polyethylene glycol (PEG))-2000]-N,N-ditetradecylacetamide.
- ✓ 1,2-distearoyl-sn-glycero-3-phosphocholine.
- ✓ Cholesterol (plant derived).
- ✓ ((4-hydroxybutyl)azanediyl)bis(hexane-6,1-diyl)bis(2-hexyldecanoate).

Purpose : Work together to help the mRNA enter cells.

C. Salts and sugar:

- ✓ Dibasic sodium phosphate dehydrate.
- ✓ Monobasic potassium phosphate.
- ✓ Potassium chloride (common food salt).
- ✓ Sodium chloride (basic table salt).
- ✓ Sucrose (basic table sugar).

Purpose: Work together to help keep the vaccine molecules stable while the vaccine is manufactured, frozen, shipped, and stored until it is ready to be given to a vaccine recipient.

The Pfizer-BioNTech vaccine is recommended for people ages 5 years and older but If you have had a severe allergic reaction or have a diagnosed allergy to any ingredient in the Pfizer-BioNTech COVID-19 vaccine (such as polyethylene glycol), you should not get this vaccine.

If you had a severe allergic reaction after getting a dose of the Pfizer-BioNTech COVID-19 vaccine, you should not get another dose of an mRNA vaccine.[25]

2.2.2: Sinopharm COVID-19 Vaccine (BBIBP-CorV):

The Sinopharm BIBP COVID-19 vaccine, also known as BBIBP-CorV,[26] the Sinopharm COVID-19 vaccine,[27] or BIBP vaccine,[27,28,29] is one of two inactivated virus COVID-19 vaccines developed by Sinopharm's Beijing Institute of Biological Products (sometimes written as Beijing Bio-Institute of Biological Products,[30] resulting in the two different acronyms BBIBP and BIBP for the same vaccine). It completed Phase III trials in Argentina, Bahrain, Egypt, Morocco, Pakistan, Peru, and the United Arab Emirates (UAE) with over 60,000 participants[31]. BBIBP-CorV shares similar technology with CoronaVac and Covaxin, other inactivated virus vaccines for COVID-19[32]. Its product name is SARS-CoV-2 Vaccine (Vero Cell),[33,34,35] not to be confused with the similar product name of CoronaVac[36,37]. Peer-reviewed results published in JAMA of Phase III trials in United Arab Emirates and Bahrain showed that the vaccine 78.1% effective against symptomatic cases and 100% against severe cases (21 cases in vaccinated group vs. 95 cases in placebo group)[38]. In December 2020, the UAE previously announced interim results showing 86% efficacy[39].

While mRNA vaccines like the Pfizer–BioNTech COVID-19 vaccine and Moderna COVID-19 vaccine showed higher efficacy of over 90%, those present distribution challenges for some nations as they require deep-freeze facilities and trucks. The BIBP vaccine could be transported and stored at normal refrigerated temperatures.[40]

The vaccine is being used in vaccination campaigns by certain countries in Asia,[41,42,43] Africa,[44,45,46]. South America,[47,48,49] and Europe.[50,51,52] Sinopharm expects to produce one billion doses of the vaccine in 2021[53]. By May, Sinopharm had supplied 200 million doses[54].

On (7 May 2021), the World Health Organization approved the BIBP vaccine for use in COVAX[55,56]. Sinopharm has signed purchase agreements for 170 million doses from COVAX[57].

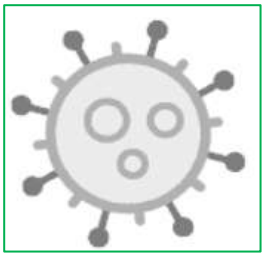
The similarly-named Sinopharm WIBP COVID-19 vaccine is also an inactivated virus vaccine.

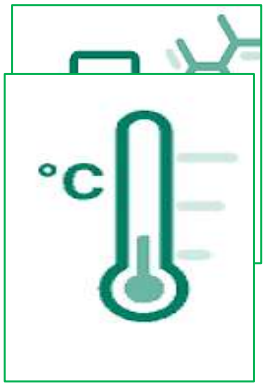
Similar to Sinovac, Sinopharm vaccine is a vaccine that uses inactivated COVID-19 virus to teach your immune system to make antibodies against COVID-19, creating an immune response to COVID-19.

When introduced to your body, your immune system will make antibodies against the coronavirus, attaching to viral proteins. As the vaccine uses a dead or inactivated virus, the vaccine can be safely injected into the arm without causing the individual to develop COVID-19.

Once the vaccine is in your system, some of the inactivated viruses will be absorbed by a type of immune cell called the anti-gen presenting cell. This type of cell will then display some of the fragments of inactivated virus on its surface, then known as a helper T cell. If the fragment fits onto the surface protein of the cell, it then can be activated and gets other immune cells to also respond to the vaccine.

When you are vaccinated with Sinopharm, your body's immune system can respond to an infection of live coronaviruses by producing antibodies to block the virus.





❖ General Information

- Inactive COVID-19 virus It uses inactivated virus to fight against COVID-19,

Sinopharm uses a traditional and widely known method of inactivated virus to protect against COVID-19.

- The virus is inactivated through chemical treatment,

Beta-propiolactone is the chemical that binds to the SARS-CoV-2 virus' genetic material and inactivates it, preventing it from replicating and causing COVID-19.

- It is stored at normal fridge temperatures,

Like the traditional flu vaccination, Sinopharm can be stored at normal refrigerator temperatures.

2.2.3: AstraZeneca vaccine:

The Oxford–AstraZeneca COVID-19 vaccine, codenamed AZD1222[58], and sold under the brand names Covishield[59] and Vaxzevria[60,61] among others, is a viral vector vaccine for prevention of COVID-19. Developed in the United Kingdom by the Oxford University and British-Swedish company AstraZeneca [62,63,64]. Using as a vector the modified chimpanzee adenovirus ChAdOx1[65]. The vaccine is given by intramuscular injection. Studies carried out in 2020 showed that the efficacy of the vaccine is 76.0% at preventing symptomatic COVID-19 beginning at 22 days following the first dose, and 81.3% after the second dose[66]. A study in Scotland found that, for symptomatic COVID-19 infection after the second dose, the vaccine is 81% effective against the Alpha variant (lineage B.1.1.7), and 61% against the Delta variant (lineage B.1.617.2)[67].

The vaccine is stable at refrigerator temperatures and has a good safety profile, with side effects including injection-site pain, headache, and nausea, all generally resolving within a few days[68,69]. More rarely, anaphylaxis may occur; the UK Medicines and Healthcare products Regulatory Agency (MHRA) has 268 reports out of some 21.2 million vaccinations as of 14 April 2021[69]. In very rare cases (around 1 in 100,000 people) the vaccine has been associated with an increased risk of blood clots when in combination with low levels of blood platelets (Embolic and thrombotic events after COVID-19 vaccination)[70,71,60]. According to the European Medicines Agency as of 4 April 2021, a total of 222 cases of extremely rare blood clots had been recorded among 34 million people who had been vaccinated in the European Economic Area (a percentage of 0.0007%)[72].

- Medical uses

The Oxford–AstraZeneca COVID-19 vaccine is used to provide protection against infection by the SARS-CoV-2 virus in order to prevent COVID-19 in adults aged 18 years and older[60]. The medicine is administered by two 0.5 ml (0.017 US fl oz) doses given by intramuscular injection into the deltoid muscle (upper arm). The initial course consists of two doses with an interval of 4 to 12 weeks between doses. The World Health Organization (WHO) recommends an interval of 8 to 12 weeks between doses for optimal efficacy[73].

There is no evidence that a third booster dose dose is needed to prevent severe disease in healthy adults[73,74].

- **Pharmacology:**

The Oxford–AstraZeneca COVID-19 vaccine is a viral vector vaccine containing a modified, replication-deficient chimpanzee adenovirus ChAdOx1[65]. Containing the full-length codon-optimized coding sequence of SARS-CoV-2 spike protein along with a tissue plasminogen activator (tPA) leader sequence[75,76]. The adenovirus is called replication-deficient because some of its essential genes required for replication were deleted and replaced by a gene coding for the spike protein. However, the HEK 293 cells used for vaccine manufacturing, express several adenoviral genes, including the ones required for the vector to replicate[77,78,79]. Following vaccination, the adenovirus vector enters the cells and releases its genes, in the form of DNA, which are transported to the cell nucleus; thereafter the cell's machinery does the transcription from DNA into mRNA and the translation into proteins.[80] The approach to use adenovirus as a vector to deliver spike protein is similar to the approach used by the Johnson & Johnson COVID-19 vaccine and the Russian Sputnik V COVID-19 vaccine[81,82].

The protein of interest is the spike protein, an external protein that enables the SARS-type coronavirus to enter cells through the ACE2 receptor[72]. Producing it following vaccination will prompt the immune system to attack the coronavirus through antibodies and T-cells if it later infects the body[28].

2.3: The role of non-specialized immunity:

2.3.1:Immunoglobulins :

2.3.1.1: Introduction to Immunoglobulins:

Immunoglobulins, also known as antibodies, are glycoprotein molecules produced by plasma cells (white blood cells). They act as a critical part of the immune response by specifically recognizing and binding to particular antigens, such as bacteria or viruses, and aiding in their destruction. The antibody immune response is highly complex and exceedingly specific. The various immunoglobulin classes and subclasses (isotypes) differ in their biological features, structure, target specificity and distribution. Hence, the assessment of the immunoglobulin isotype can provide useful insight into complex humoral immune

response. Assessment and knowledge of immunoglobulin structure and classes is also important for selection and preparation of antibodies as tools for immunoassays and other detection applications.[83]

- **Soluble vs. membrane-bound immunoglobulins**

Immunoglobulins occur in two main forms: soluble antibodies and membrane-bound antibodies. (The latter contain a hydrophobic transmembrane region.) Alternative splicing regulates the production of secreted antibodies and surface bound B-cell receptors in B cells.

Membrane-bound immunoglobulins are associated non-covalently with two accessory peptides, forming the B-cell antigen receptor complex. The first antigen receptors expressed by B cells are IgM and IgD. The receptor is a prototype of the antibody that the B cell is prepared to produce. The B cell receptor (BCR) can only bind antigens. It is the heterodimer of Ig alpha and Ig beta that enables the cell to transduce the signal and respond to the presence of antigens on the cell surface. The signal generated causes the growth and proliferation of the B cell and antibody production inside the plasma cell [83].

2.3.1.2:Structure of immunoglobulins:

Antibody (or immunoglobulin) molecules are glycoproteins composed of one or more units, each containing four polypeptide chains: two identical heavy chains (H) and two identical light chains (L). The amino terminal ends of the polypeptide chains show considerable variation in amino acid composition and are referred to as the variable (V) regions to distinguish them from the relatively constant (C) regions. Each L chain consists of one variable domain, VL, and one constant domain, CL. The H chains consist of a variable domain, VH, and three constant domains CH1, CH2 and CH3. Each heavy_chain has about twice the number of amino acids and molecular weight (~50,000) as each light chain (~25,000), resulting in a total immunoglobulin monomer molecular weight of approximately 150,000[84].

2.3.1.3:Classes of immunoglobulins:

The five primary classes of immunoglobulins are IgG, IgM, IgA, IgD and IgE. These are distinguished by the type of heavy chain found in the molecule. IgG molecules have heavy

chains known as gamma-chains; IgMs have mu-chains; IgAs have alpha-chains; IgEs have epsilon-chains; and IgDs have delta-chains.

Differences in heavy chain polypeptides allow these immunoglobulins to function in different types of immune responses and at particular stages of the immune response. The polypeptide protein sequences responsible for these differences are found primarily in the Fc fragment. While there are five different types of heavy chains, there are only two main types of light chains: kappa (κ) and lambda (λ).

Antibody classes differ in valency as a result of different numbers of Y-like units (monomers) that join to form the complete protein. For example, in humans, functioning IgM antibodies have five Y-shaped units (pentamer) containing a total of 10 light chains, 10 heavy chains and 10 antigen-binding [84].

2.3.1.4:Complement C3/C4:

Complement (C3/C4) are proteins that are part of the immune system. Measuring complement involved a simple blood test that measures the levels of C3 and C4 in the blood.

complement levels is typically done in autoimmune diseases that affect the levels of complement. For example, in patients with lupus the complement levels can be low when the disease is active.[85]

C3 and C4 Levels

Complement are small proteins that are produced by the liver. The complement system is part of the immune system. It's normal job is to enhance (or *complement*) the ability of the immune system to clear foreign invaders.

When the immune system becomes activated (with autoimmune diseases) the complement system may become involved in the attack. This can result in a reduction in the levels of complement as it is consumed in the autoimmune attack.

A complement test may be used to monitor people with an autoimmune disorder. It is done to see if treatment for their condition is working. When the complement system is turned on during inflammation, levels of complement proteins may go down. For example, people with active lupus erythematosus may have lower-than-normal levels of the complement proteins C3 and C4.[85]. The test may also be done for the following conditions such as Fungal

infections, Gram negative septicemia, Parasitic infections, such as malaria, Paroxysmal nocturnal hemoglobinuria (PNH),and Shock.

2.3.2: Total protein:

The total protein test measures the total amount of two classes of proteins found in the fluid portion of your blood. These are albumin and globulin.

Proteins are important parts of all cells and tissues.

- Albumin helps prevent fluid from leaking out of blood vessels. It also carries chemicals in your blood.
- Globulins are an important part of your immune system.[86]

2.3.2.1:Total protein range:

The normal range for total protein is between 6 and 8.3 grams per deciliter (g/dL). This range may vary slightly among laboratories. These ranges are also due to other factors such as age, gender, population, test method. The total protein measurement may increase during pregnancy. If total protein is abnormal, additional tests must be performed to identify which specific protein is low or high before a diagnosis can be made. Elevated total protein may indicate: inflammation or infections, such as viral hepatitis B or C, or HIV bone marrow disorders, such as multiple myeloma or Waldenstrom's disease.

Low total protein may indicate:

Bleeding, liver disorder, kidney disorder such as a nephrotic disorder or glomerulonephritis, malnutrition, malabsorption conditions such as celiac disease or inflammatory bowel disease, extensive burns, agammaglobulinemia, which is an inherited condition in which your blood doesn't have enough of a type of globulin, affecting the strength of the immune system, inflammatory conditions and delayed post-surgery recovery [87].

2.3.3: C-Reactive Protein:

C-reactive protein (CRP) is an annular (ring-shaped) pentameric protein found in blood plasma, whose circulating concentrations rise in response to inflammation. It is an acute-

phase protein of hepatic origin that increases following interleukin-6 secretion by macrophages and T cells. Its physiological role is to bind to lysophosphatidylcholine expressed on the surface of dead or dying cells (and some types of bacteria) in order to activate the complement system via Clq[88]. CRP is synthesized by the liver[89] in response to factors released by macrophages and fat cells (adipocytes)[90]. It is a member of the pentraxin family of proteins[89]. It is not related to C-peptide (insulin) or protein C (blood coagulation). C-reactive protein was the first pattern recognition receptor (PRR) to be identified[91], CRP binds to the phosphocholine expressed on the surface of dead or dying cells and some bacteria. This activates the complement system, promoting phagocytosis by macrophages, which clears necrotic and apoptotic cells and bacteria [92]

2.3.3.1: Role of CRP test in COVID-19:

Based on worldwide data and recommendations from experts, CRP test has become a requisite for people with COVID-19 admitted in hospitals. What is most significant is that elevated levels of CRP may help in early detection of cases that can progress into severe COVID-19.

Though coronavirus is a respiratory virus that replicates in the nose, throat, and lungs, moderate or severe disease can cause hyper inflammation in the body. This dysregulated immune response can be lethal. Hence, keeping a track of inflammatory markers like CRP becomes crucial.

Normally, CRP level in blood is less than 5 mg/L. According to a study that looked at the clinical characteristics of people with COVID-19, a significantly elevated CRP levels (average 20 to 50 mg/L) were seen in COVID-19 cases. People who had severe COVID-19 had a far elevated CRP level as compared to the people with mild disease. CRP elevations were observed in up to 86% in severe COVID-19 cases. Another study reported that while those who had severe symptoms had on average CRP levels of 39.4 mg/L, those with mild symptoms had an average CRP levels of 18.8 mg/L. Other evidence has also shown that CRP is increased at the initial stages in the moderate or severe group than those in the mild group.

The authors also observed that the chances of developing severe symptoms is increased by 5% for every one-unit rise in CRP levels in people with COVID-19.

High CRP levels in COVID-19 cases can indicate the need for hospitalization and advanced treatment modalities. In a study, people who died from COVID-19 had about 10 fold higher levels of CRP than those who recovered.[93]

CHAPTER THREE

MATERIALS AND METHODS

3-*Materials and methods*

3.1: Materials:

3.1.1: Instrument and Equipment:

The equipment used in the current study were listed in the table (3-1) below .

Table (3.1) : Instruments and Equipment in this study

Table (3.1) : Materials

Instrument and Equipment	Origin
Gloves	Malaysia
Cotton	Iraq
Alcohol	Iraq
Tournica	China
Syringe	China
Gel tube	Jordan
Eppendorf tubes	China
pastol pipet	China
Centrifuges	Germany
ELISA reader +Washer	USA _ Bioteck

3.1.2: Serological kits test:

Table (3.2) : Serological kits test

Kits	origin	company
Easy RID	Italy	Liofilchem
Colorimetric total protein kit	France	Biolabo

3.2:Methods:

3.2.1: Sample collection :

Blood samples were collected from Babylon University students, aged between 20-30 years, of both genders (males and females), for the period from {14-11 to 14-12} for the year 2021-2022.

3.2.2: Separation of the serum:

Separation of the serum A blood sample of 5 ml was withdrawn from the humeral vein using a special one-time syringe. The drawn blood was placed in tubes containing a gel that helps to separate the serum. Then the blood was separated using a centrifuge at a power of 4500 for 5 minutes, then the serum was separated using Pasteur pipette and the sera were kept in Eppendorf tubes and kept by freezing at a temperature of 5 ° C...

3.2.3: Nonspecific humeral immunity:

3.2.3.1: Total protein test :

This test was done to detect total protein by depending of kit procedure.

3.2.3.1.1: Test principle:

Colorimetric method described according to manufacture company The peptide bound of protein react with Cu+ in alkaline solution to form a coloured complex which absorbance, proportional to be concentration of total protein in the specimen is measured at 550 nm. The biuret reagent contain sodium potassium tartrate to complex cupric ions and maintain their solubility in alkalin solution.

3.2.3.1.2: Testing procedure:

1- The stand reagents and specimen was putted at room temperature.

Pipette into well identified test tube	Reagent blank	Standard	Assay
Reagent R1	1ml	1ml	1ml
Standard		20 ul	
Specimen			20 ul
Demineralized Water	20 ul		

2- All was mixed well and waited for 10 minutes at room temperature.

3- Absorbance was recorded at 550 nm(530-570)against reagent blank

3.2.3.1.3 : Calculation:

Result = $\frac{Abs(Assay)}{Abs(Standar)} \times Standar\ concentration$

3.2.3.2: Estimation of Immunoglobulins and Complements Levels (IgG,IgM,C3,C4):

ESAY RID(Radial immune diffusion) are plates with 12 wells for quantitative determination in radial immunodiffusion of human plasma proteins in serum and plasma to estimate of antibodies IgG,IgM ,lgA and complements C3,C4 were used in the study.

3.2.3.2.1: Princeple of method :

When antigen diffuses from a well into agar containing suitably diluted antiserum, initially it is present in a relatively high concentration and forms soluble complexes; as the antigen diffuses further the concentration continuously falls until the point is reached at which the reactants are nearer optimal proportions and a ring of precipitate is formed. The higher the concentration of antigen, the greater the diameter of this ring. By incorporating, say, three standards of known antigen concentration in the plate, a calibration curve can be obtained and used to determine the amount of antigen in the unknown samples tested.

3.2.3.2.2: Test procedure:

1- ESAY RID was removed from the envelope, open the plate and leave to stand for about 5 minutes at room temperature so that any condenser water in the well can evaporate.

2- The well was filled with 5 µl of undiluted patient sample.

3- The plate was closed with the lid, after the sample has diffused into the gel for about 20,leaved to stand ,overturned into the envelope at room temperature for 48 hours.

3.2.3.2.3: Interpretation of result:

After 48h, measure the diameter of the precipitin ring using a suitable measuring device (mm) .The protein concentration for the precipitin ring diameters can be read using the standard table.

CHAPTER FOUR

RESULTS AND DISCUSSION .

Study Population:

A study was conducted randomized on vaccinated people in Babylon governorate for both sexes, the number of vaccinated people were 695 people, including 561person taken Pfizer vaccine, 120 people taken Sinovac, and 14 people taken AstraZeneca, The study report shows that the highest rate of vaccinated people was Pfizer in the rate of (80.71%) and followed by the Sinovac vaccine in the rate of (17.36%), While less percentage was AstraZeneca vaccine in the rate of (2.01%) as shown in figure (4.1).

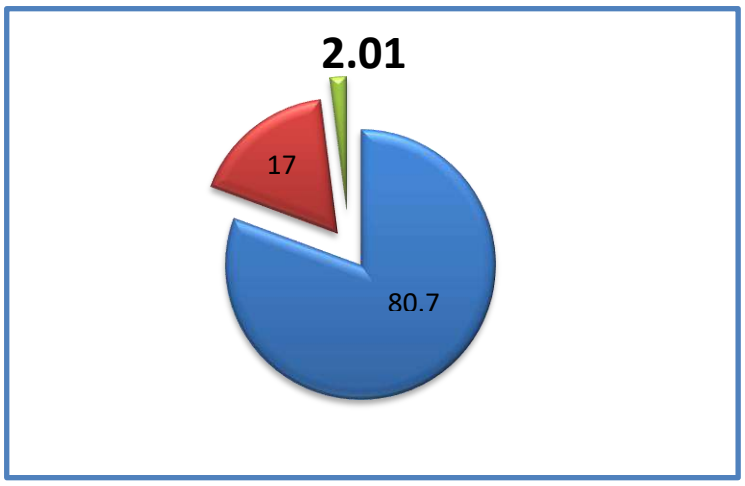


Figure (4.1): Percentage of vaccination groups 1-Pfizer vaccine, 2-Sinovac , 3- AstraZeneca.

The highest rate of vaccinated people was Pfizer in the rate of (80.71%) this result agrees with (Halim et al., 2021) who said based on the research, the Pfizer vaccine has the highest percentage efficacy of 95 %, showing that the vaccine is effective, 78% for sinovac vaccine and 70.4% for AstraZeneca vaccine respectively.

A total of (695) people (males and females) were distributed into two groups according to Symptom and without symptoms associated with Pfizer vaccine, Sinovac, and AstraZeneca where the symptoms were fever and pain. The results showed that the highest rate of symptoms was in the AstraZeneca vaccines, where the percentage of symptoms of AstraZeneca(98.85%), followed by the percentage of symptoms of Pfizer vaccine(75.22%), and the percentage of symptoms of Sinovac (65%) as shown in figure (4.2).

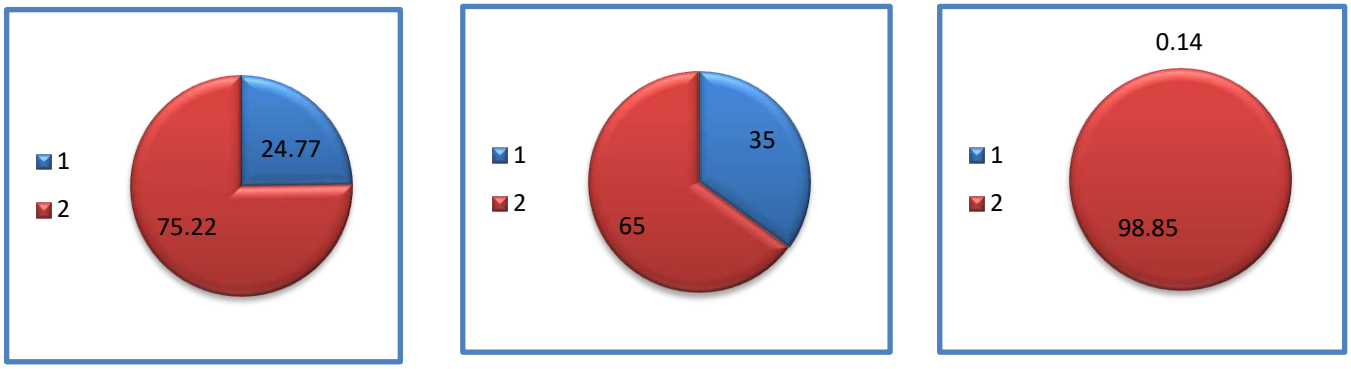


Figure (4.2): Percentage Symptom and without symptom associated with a-Pfizer vaccine , b-Sinovac c- AstraZeneca 1- without symptom 2- symptom such as fever and pain.

This agree with (Meo *et al*., 2021) they reported the COVID-19 vaccines can cause mild adverse effects after the first or second doses, including pain, redness or swelling at the site of vaccine shot, fever, fatigue, headache, muscle pain, nausea, vomiting, itching, chills, and joint pain.

Also, agree with (Malayala, *et al.*, 2021) they reported in the trials related to the vaccine in the United States for the prevention of COVID-19 infection since December 2020, appeared Mild to moderate intensity side effects like low-grade fever, myalgia, chills, and malaise after being taken where the "Moderna vaccine".

All vaccinated people in this study were distributed according to gender (males and females), were found the number of vaccinated females more than males, the highest number of vaccinated females with Pfizer vaccine (415), followed by the Sinovac vaccine (31), While less number of vaccinated females with the AstraZeneca vaccine (8) shown in figure(4.3).

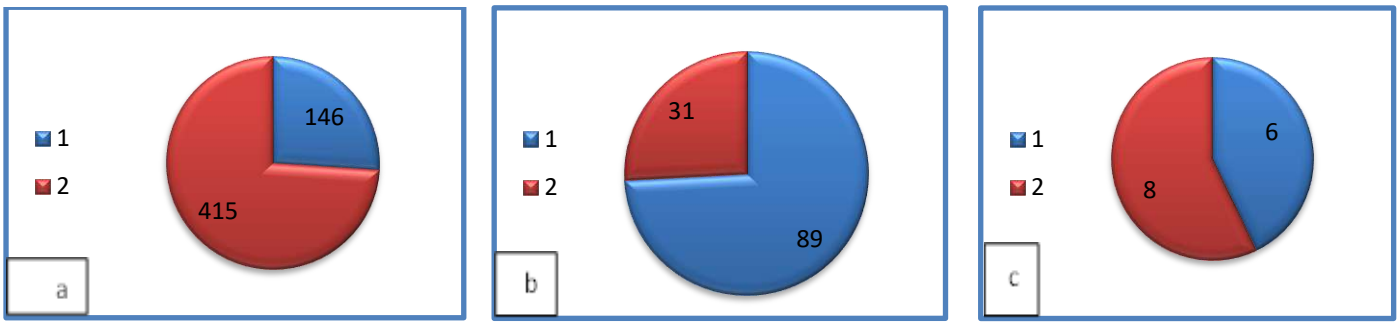


Figure (4.3): Number of a-Pfizer vaccine, b-Sinovac, c- AstraZeneca, groups according gender 1-male 2 –female.

The study included the recognition of the Concentrations of C3, C4, IgG, IgM, and Total protein concentrations, by using the special kit.

In this study, we investigated the levels of C3, C4, IgG, IgM, and Total protein concentrations in the serum of the vaccinated people, and compared them with non-vaccinated people.

The study has shown elevated levels of C3, IgG, IgM, and Total protein concentrations serum of the vaccinated people compared with the control, while the concentration of C4 is low compared with the control as shown in tables (1,2,3,4,5).

The mean of C3 concentration in serum of vaccinated people of Pfizer vaccine was256.2250±78.56689 pg/ml, Sinovac vaccine was 291.3333±43.16901pg/ml and AstraZeneca vaccine was 328.9000±29.01293pg/ml while control was 282.028±46.18720 pg/ml, there significantly as show in table (4.1).

Table (4.1): Concentrations of C3 among immunization groups

groups	Mean ±Sd
Control	282.028±46.18720
F	256.2250±78.56689
S	291.3333±43.16901
A	328.9000±29.01293

LSD ≥0.05 sig=0.27

Complement protein C3 is a central molecule in the complement system, and its activation is required for all of the system's important functions, including phagocytosis, local inflammatory responses against pathogens, and instructing the adaptive immune system to select appropriate antigens for humoral responses (Sahu, and Lambris, 2002).

Kurtovic and Beeson (2021) they said we hypothesize that complement may also have a protective role and could function to enhance virus neutralization by antibodies, promote virus phagocytosis by immune cells, and lysis of virus. These functions might be exploited in the development of effective therapeutics and vaccines against SARS-CoV-2.

In one study, researchers looked at the link between complement and mortality in COVID-19 disease in humans and discovered that higher C3 levels in the blood were linked to a lower risk of death compared to those with lower levels (Fanga *et al.*, 2020).

The mean of C4 concentration in serum of vaccinated people of Pfizer vaccine was 77.5500±21.40537pg/ml, Sinovac vaccine was 74.9667±15.95755 pg/ml and AstraZeneca vaccine was 73.5000±27.36110 pg/ml while control was 89.0060±14.18771 pg/ml there significantly as show in table (4.2).

Table (4.2): Concentrations of C4 among immunization groups

groups	Mean ±Sd
Control	89.0060±14.18771
F	77.5500±21.40537
S	74.9667±15.95755
A	73.5000±27.36110

LSD ≥0.05 sig=0.34

The complement system is vital for anti-microbial defense. In the classical pathway, pathogen-bound antibody recruits the C1 complex that initiates a cleavage cascade involving C2, C3, C4, and C5 and triggering microbial clearance, human genetics suggest that C4 has important immune. The importance of C4 in immunity from viral mechanisms where is that the C4-dependent antiviral mechanism is independent of downstream complement components (Bottermann *et al.,* 2019).

The study has shown elevated levels of Immunoglobulin G (IgG), and Immunoglobulin M (IgM) in serum of vaccinated people to all vaccines (Pfizer vaccine, Sinovac vaccine, and AstraZeneca vaccine), and showed there a significance of (IgG), and (IgM) in serum of vaccinated people compared with control, as shown in the tables (4.3).

The mean of IgG concentration in serum of vaccinated people of Pfizer vaccine was 3115.0000±620.38648pg/ml, Sinovac vaccine was 3115.0000±620.38648 pg/ml and AstraZeneca vaccine was 3582.9667±459.37998 pg/ml while control was 2836.4000±943.85630 pg/ml, there significantly as show in table (4.3).

Table (4.3): Concentrations of IgG among immunization groups

groups	Mean ±Sd
Control	2836.4000±943.85630
F	3115.0000±620.38648
S	3582.9667±459.37998
A	3485.8333±307.69131

LSD ≥0.05 sig=0.20

Immunoglobulin G (IgG) is an antibody that interacts with a range of antigens and is essential for protection against invading pathogens. It is one of the most abundant proteins in human serum, IgG playing a central role in systemic antiviral immunity. Initial results suggested that IgM antibodies to SARS-Cov-2 appeared earlier than IgG antibodies, and that testing both IgM and IgG antibodies might enhance SARS-Cov-2 infection detection (Elslande, *et al.*, 2020)

The study has shown the concentration of IgG more than compared with control, this agree with (Wisnewski, *et al.*, 2021) they reported that IgG levels rose exponentially and plateaued 21 days after the initial vaccine dose. After the second vaccine dose IgG levels increased further, reaching a maximum approximately 7–10 days later, and remained elevated (average of 58% peak levels) during the additional >100 days follow up period.

The mean of IgM concentration in serum of vaccinated people of Pfizer vaccine was 439.0500±57.44635 pg/ml, Sinovac vaccine was 459.1333±120.95488 pg/ml and AstraZeneca vaccine was 525.4667±112.06770 pg/ml while control was 402.3500±172.40591 pg/ml, there significantly as show in table (4.4).

Table (4.4): Concentrations of IgM among immunization groups

groups	Mean ±Sd
Control	402.3500±172.40591
F	439.0500±57.44635
S	459.1333±120.95488
A	525.4667±112.06770

LSD ≥0.05 sig=0.22

These data revealed some potential differences in the nature of the humoral response induced by the three vaccines, these results may be agree with (Parry *et al.*, 2021) who reported antibody titers by Roche ELISA were 691-fold higher in vaccinated donors with a previous infection compared to vaccinated people without a previous infection.

Correlation between IgG and IgM, Correlations between C3 and C4 in serum of vaccinated people:

Correlation between IgG and IgM was positive correlation and Show in figure (5), while the correlation between C3 and C4 was negative correlation Show in figure (6).

Immunoglobulin M (IgM) and immunoglobulin G (IgG) II antibody tests are clinically important because they enable the assessment of humoral immunity after infection and vaccination (Kitagawa, *et al.*, 2021).

Table (4.5): Correlations between IgG and IgM

		IgG	IgM
IgG	Pearson Correlation	1	.711**
	Sig. (2-tailed)		.010
IgM	Pearson Correlation	.711**	1
	Sig. (2-tailed)	.010	

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

IgM and IgG antibody levels vary with time after infection in COVID-19, which is expected because antibodies are produced during the lag period after infection, early IgM antibodies are later replaced by IgG antibodies, and antibody levels in blood generally decrease with time after infection resolution (Shah, *et al.*, 2021).

This result agree with (Shah, *et al.*, 2021) who said detection of either IgG or IgM antibodies was better than IgG or IgM alone for assessing.

Table (4.6): Correlations between C3 and C4

		C3	C4
C3	Pearson Correlation	1	-.298-
	Sig. (2-tailed)		.281
C4	Pearson Correlation	-.298-	1
	Sig. (2-tailed)	.281	

The complement **system** has been observed to be activated in coronavirus illness 19 (COVID-19). Immunoassays are commonly used in clinical practice to determine and monitor complement activation using the complement proteins C3 and C4. C3 and C4 testing in COVID-19 patients may provide useful information about the balance of 'physiological' vs. 'abnormal' complement activation and overall clinical risk, C3 is often decreased through

consumption during infections whereas a combined reduction in C3 and C4 is observed in immune complex diseases (Zinellu and Mangoni 2021).

Table (4.7): Total protein concentrations

groups	Mean ±Sd
Control	5.7780±.16574
F	6.1660±.16562
S	5.5880±.18913
A	5.3980±.15482

LSD ≥0.05 sig=0.000

conclusions

- most common type of vaccine was taken was Pfizer followed with Sinopharm and less with AstraZeneca ,and found vaccines
- Vaccine Enhanced humoral immune response

✓

Recommendation

-Study the other immunoglobulin isotypes IgE and IgA

- study some cytokine invaccinated

References

1- Brasil, Ministério da Saúde. Protocolo de manejo clínico para o novo-corona vírus

(2019-nCoV). [cited 2020 Feb 12]. Available from: https://portalar quivos2. saude.gov.br/images/pdf/2020/fevereiro/11/protocolo-manejo-coronavirus.pdf

2- Brasil. Ministério da Saúde. Coronavírus: o que você precisa saber e como

prevenir o contágio. [cited 2020 Feb 18]. Available from: https://saude.gov

br/saude-de-a-z/coronavirus

3- Megha K.B.; Seema A; Nayar Mohanan (2021)Vaccine and vaccination as a part of human life: In view of COVID-19.Biotichnology journal.

4- Li, D.D.; Li, Q.H. SARS-CoV-2: Vaccines in the pandemic era. Mil. Med. Res. 2021, 8, 1. [CrossRef]

5 - WHO Coronavirus Disease (COVID-19) Dashboard. Available online: https://covid19.who.int/ (accessed on 28 May 2021).

6- Grifoni, A.; Weiskopf, D.; Ramirez, S.I.; Mateus, J.; Dan, J.M.; Moderbacher, C.R.; Rawlings, S.A.; Sutherland, A.; Premkumar, L.; Jadi, R.S.; et al. Targets of T Cell Responses to SARS-CoV-2 Coronavirus in Humans with COVID-19 Disease and Unexposed

Individuals. Cell 2020, 181, 1489–1501.e1415. [CrossRef]

7- Sette, A.; Crotty, S. Adaptive immunity to SARS-CoV-2 and COVID-19. Cell 2021, 184, 861–880. [CrossRef]

8-Bonifacius, A.; Tischer-Zimmermann, S.; Dragon, A.C.; Gussarow, D.; Vogel, A.; Krettek, U.; Godecke, N.; Yilmaz, M.; Kraft ,

A.R.M.; Hoeper, M.M.; et al. COVID-19 immune signatures reveal stable antiviral T cell function despite declining humoral

responses. Immunity 2021, 54, 340–354.e346. [CrossRef]

9-Ashour, H.M.; Elkhatib, W.F.; Rahman, M.M.; Elshabrawy, H.A. Insights into the Recent (2019 Novel Coronavirus (SARS-CoV-2

in Light of Past Human Coronavirus Outbreaks. Pathogens 2020, 9, 186. [CrossRef]

10-Pfizer. Pfizer and BioNTech Announce Vaccine Candidate against COVID-19 Achieved Success in First Interim Analysis from Phase 3

Study; Pfizer: New York, NY, USA, 2020.

11-Summary of Product Characteristics. Comirnaty Concentrate for Dispersion for -Injection. COVID-19 mRNA Vaccine. Avail

able online: https://www.ema.europa.eu/en/documents/product-information/comirnaty-epar-product-information_en.pdf

.(accessed on 13 March 2021)

12-Information for Healthcare Professionals on Pfizer/BioNTech COVID-19 Vaccine. :Updated 31 March 2021. Available online

https://www.gov.uk/government/publications/regulatory-approval-of-pfizer-biontech--vaccine-for-covid-19/information

for-healthcare-professionals-on-pfizerbiontech-covid-19-vaccine (accessed on 12 April .(2021

13-Wang, J.; Peng, Y.; Xu, H.; Cui, Z.; Williams, R.O., 3rd. The COVID-19 Vaccine Race: Challenges and Opportunities in Vaccine

Formulation. AAPS PharmSciTech 2020, 21, 225. [CrossRef]

14- Woo P.C., et al. (2012) Discovery of seven novel Mammalian and avian coronaviruses in the genus coronaviruses as the gene betacoronavirus and avian coronaviruses as the gene source of deltacoronavirus supports bat source of alphacoronavirus and gammacoronavirus and deltacoronavirus. J Virol. 86(7):3995-4008.

15- Li Q., Guan X., Wu P., Wang X., and Zhou L. (2020) Early transmission dynamicsin Wuhan, China, of novel coronavirus-infected pneumonia. N Engl J Med. 382:1199-1207.

16- Lorusso A., Calistri P.., Petrini A.. Savini G., and Decaro N. (2020) Novel coronavirus .(SARS-CoV-2) epidemic:a 26 veterinary perspective. Vet Ital56:5-10

Lu R., Zhao X., Li J., Niu P., and Yang B. (2020) Genomic characterisation and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding. Lancet. 395: 565-574.

17- Singer B.S., and Blinov S.K. (2014) The epidemiological characteristics of Ebola Virus Disease. Am J BioMed 2: 1095-1109

18- Susan P. (2020) Family Coronaviridae. Viruses, Academic Press, United States.

19-Yousif N.G., Al-Amran F.G., Hadi N., Lee J., and Adrienne J. (2013) Expression of IL-32 modulates NF-KB and p38 MAP kinase pathways in human esophageal cancer. Cytokine 61: 223-227

20-Masters P.S. (2006) The molecular biology of coronaviruses. Adv Virus Res. ;66:193-292.McIntosh K. (1974) Coronaviruses: Comparative Review, Curr Topics Microbiol Immunol. Berlin, Heidelberg: Springer. 63: 85-129.

21-Cui J., Li F., and Shi Z.L. (2019) Origin and evolution of pathogenic coronaviruses. Nat Rev Microbiol 17: 181- 192.

22-Decaro N., Tidona C., and Darai G. (2011) Betacoronavirus. The Springer Index of Viruses, Springer, New York, United States.

23-MedRxiv: “Viral infection and transmission in a large, well-traced outbreak caused by the SARS-CoV-2 Delta variant”.

24-Centers for Disease Control and Prevention. CDC twenty four seven. Saving Lives, Protecting People

25-Centers for Disease Control and Prevention. CDC twenty four seven. Saving Lives, Protecting People

26-Xia S, Zhang Y, Wang Y, Wang H, Yang Y, Gao GF, et al. (January 2021). "Safety and immunogenicity of an inactivated SARS-CoV-2 vaccine, BBIBP-CorV: a randomised, double-blind, placebo-controlled, phase 1/2 trial". The Lancet. Infectious Diseases. 21 (1): 39–51. doi:10.1016/s1473-3099(20)30831-8. PMC 7561304. PMID 33069281.

27- " The Sinopharm COVID-19 vaccine: What you need to know". World Health Organization. 10 May 2021. Retrieved 5 July 2021.

28-Nguyen S (5 June 2021). "Coronavirus: Vietnam approves Sinopharm's vaccine, but will people take it?". South China Morning Post. Retrieved 5 July 2021.

29-Lahiri T, Li J (16 June 2021). "What we now know about the efficacy of China's Covid-19 vaccines". Quartz. Retrieved 5 July 2021.

30-"WHO lists additional COVID-19 vaccine for emergency use and issues interim policy recommendations". World Health Organization. 7 May 2021. Retrieved 3 July 2021.

31-Reuters Staff (19 November 2020). "China Sinopharm's coronavirus vaccine taken by about a million people in emergency use". Reuters. Retrieved 9 December 2020.

32-Corum J, Zimmer C (26 April 2021). "How the Sinopharm Vaccine Works". The New York Times. ISSN 0362-4331. Retrieved 29 April 2021.

33-"WHO lists additional COVID-19 vaccine for emergency use and issues interim policy recommendations". World Health Organization. 7 May 2021. Retrieved 13 June 2021. The Sinopharm product is an inactivated vaccine called SARS-CoV-2 Vaccine (Vero Cell).

34-Chen W, Al Kaabi N (18 July 2020). "A Phase III clinical trial for inactivated novel coronavirus pneumonia (COVID-19) vaccine (Vero cells)". Chinese Clinical Trial Registry. Retrieved 13 June 2021.

35- Yang Y. "A Study to Evaluate The Efficacy, Safety and Immunogenicity of Inactivated SARS-CoV-2 Vaccines (Vero Cell) in Healthy Population Aged 18 Years Old and Above". Archived from the original on 14 September 2020. Retrieved 13 June 2021.

36-"Sinovac's Coronavac™, SARS-CoV-2 Vaccine (Vero Cell), Inactivated, Announces Approval for Phase I/II Clinical Trial in Adolescents and Children" (Press release). Beijing: Bloomberg. Business Wire. 23 September 2020. Retrieved 13 June 2021.

37-"A Multi-center, Randomized, Double-blind, Placebo-controlled Phase II/III Clinical Trial to Evaluate the Safety and Immunogenicity of a SARS-CoV-2 Inactivated (Vero Cell) Vaccine in the Elderly 60–80 Years of Age, Coronovac ENCOV19 Study". registry.healthresearch.ph. Philippine Health Research Registry. Retrieved 13 June 2021.

38-Al Kaabi N, Zhang Y, Xia S, Yang Y, Al Qahtani MM, Abdulrazzaq N, et al. (26 May 2021). "Effect of 2 Inactivated SARS-CoV-2 Vaccines on Symptomatic COVID-19 Infection in Adults: A Randomized Clinical Trial". JAMA. 326 (1): 35–45. doi:10.1001/jama.2021.8565. PMC 8156175. PMID 34037666.

39-UAE: Ministry of Health announces 86 per cent vaccine efficacy". gulfnews.com. Retrieved 13 December 2020.

40- China State-Backed Covid Vaccine Has 86% Efficacy, UAE Says". Bloomberg.com. 9 December 2020. Retrieved 9 December 2020.

41- Liu R (31 December 2020). "China gives its first COVID-19 vaccine approval to Sinopharm". Reuters. Retrieved 31 December 2020.

42-Turak N (18 January 2021). "The UAE is on track to have half its population vaccinated by the end of March". CNBC. Retrieved 21 January 2021.

43-PM Imran kicks off Pakistan's Covid-19 vaccination drive". Dawn.com. 2 February 2021. Retrieved 3 February 2021.

44-Reuters Staff (24 January 2021). "Sisi says Egypt to begin COVID-19 vaccinations on Sunday". Reuters. Retrieved 24 January 2021.

45-Dumpis T (27 January 2021). "Morocco Receives Half a Million Doses of Chinese Sinopharm Vaccine". Morocco World News. Retrieved 28 January 2021.

46-Zimbabwe starts administering China's Sinopharm vaccines". thestar.com. 18 February 2021. Retrieved 20 February 2021.

47-Argentina autoriza la vacuna china Sinopharm para mayores de 60 años". El Comercio. Retrieved 26 March 2021.

48-Aquino M (10 February 2021). "'The best shield': Peru launches inoculation drive with Sinopharm vaccine". Reuters. Retrieved 10 February 2021.

49-Bolivia begins inoculation with Sinopharm jabs | The Star". www.thestar.com.my. Retrieved 28 February 2021.

50-Serbia Becomes First European Nation To Use China's Sinopharm Vaccine". RadioFreeEurope/RadioLiberty. Retrieved 21 January 2021

51-Hungary first EU nation to use China's Sinopharm vaccine against COVID". euronews. 24 February 2021. Retrieved 26 February 2021

52-Belarus begins COVID-19 vaccinations with Chinese shots". eng.belta.by. 15 March 2021. Retrieved 16 March 2021.

53-Which companies will likely produce the most COVID-19 vaccine in 2021?". Pharmaceutical Processing World. 5 February 2021. Retrieved 28 February 2021.

54-WHO approves Sinopharm vaccine in potential boost to COVAX pipeline". Reuters. 7 May 2021. Retrieved 14 May 2021.

55-WHO lists additional COVID-19 vaccine for emergency use and issues interim policy recommendations" (Press release). World Health Organization (WHO). 7 May 2021. Retrieved 7 May 2021.

56-Taylor A (7 May 2021). "WHO grants emergency use authorization for Chinese-made Sinopharm coronavirus vaccine". The Washington Post. Retrieved 7 May 2021.

57-Chinese drugmakers agree to supply more than half a billion vaccines to COVAX". Reuters. 12 July 2021. Retrieved 13 July 2021.

58-"AstraZeneca COVID-19 Vaccine (AZD1222)" (PDF). AstraZeneca. 27 January 2021. Archived (PDF) from the original on 27 January 2021. Retrieved 7 March 2021.

59-"Covishield and Covaxin: What we know about India's Covid-19 vaccines". BBC News. 4 March 2021. Archived from the original on 7 March 2021. Retrieved 8 March 2021.

60-"Vaxzevria (previously COVID-19 Vaccine AstraZeneca) EPAR". European Medicines Agency (EMA). Archived from the original on 21 April 2021. Retrieved 27 March 2021. Text was copied from this source which is © European Medicines Agency. Reproduction is authorized provided the source is acknowledged.

61-AstraZeneca vaccine renamed 'Vaxzevria'". The Brussels Times. 30 March 2021. Archived from the original on 31 March 2021. Retrieved 6 April 2021.

62-"Investigating a Vaccine Against COVID-19". ClinicalTrials.gov (Registry). United States National Library of Medicine. 26 May 2020. NCT04400838. Archived from the original on 11 October 2020. Retrieved 14 July 2020 .

63-"A Phase 2/3 study to determine the efficacy, safety and immunogenicity of the candidate Coronavirus Disease (COVID-19) vaccine ChAdOx1 nCoV-19". EU Clinical Trials Register (Registry). European Union. 21 April 2020. EudraCT 2020-001228-32. Archived from the original on 5 October 2020. Retrieved 3 August 2020.

64-O'Reilly P (May 2020). "A Phase III study to investigate a vaccine against COVID-19". ISRCTN (Registry). doi:10.1186/ISRCTN89951424. ISRCTN89951424.

65,0

65- Voysey M, Clemens SA, Madhi SA, Weckx LY, Folegatti PM, Aley PK, Angus B, Baillie VL, and et.al. (January 2021). "Safety and efficacy of the ChAdOx1 nCoV-19 vaccine (AZD1222) against SARS-CoV-2: an interim analysis of four randomised controlled trials in Brazil, South Africa, and the UK". Lancet. 397 (10269): 99–111. doi:10.1016/S0140-6736(20)32661-1. ISSN 0140-6736. PMC 7723445. PMID 33306989.

66-Voysey M, Costa Clemens SA, Madhi SA, Weckx LY, Folegatti PM, Aley PK, et al. (February 2021). "Single-dose administration and the influence of the timing of the booster dose on immunogenicity and efficacy of ChAdOx1 nCoV-19 (AZD1222) vaccine: a pooled analysis of four randomised trials". Lancet. 397 (10277): 881–891. doi:10.1016/S0140-6736(21)00432-3. PMC 7894131. PMID 33617777.

67-Sheikh A, McMenamin J, Taylor B, Robertson C (14 June 2021). "SARS-CoV-2 Delta VOC in Scotland: demographics, risk of hospital admission, and vaccine effectiveness". The Lancet. 397 (10293). Table S4. doi:10.1016/S0140-6736(21)01358-1. ISSN 0140-6736. PMC 8201647. PMID 34139198 .

68-Belluz J (23 November 2020). "Why the AstraZeneca-Oxford Covid-19 vaccine is different". Vox. Archived from the original on 29 January 2021. Retrieved 26 November 2020.

69-"Coronavirus Vaccine : Summary of Yellow Card reporting" (PDF). Archived (PDF) from the original on 16 March 2021. It is known from the clinical trials that the more common side effects for both vaccines can occur at a rate of more than one in 10 doses (for example, local reactions or symptoms resembling transient flu-like symptoms(

70-"AstraZeneca's COVID-19 vaccine: benefits and risks in context". European Medicines Agency (EMA) (Press release). 23 April 2021. Archived from the original on 23 April 2021. Retrieved 23 April 2021.

71-"Annex 1: Summary of Product Characteristics" (PDF). European Medicines Agency (EMA). Archived (PDF) from the original on 28 March 2021. Retrieved 29 March 2021.

72-"AstraZeneca's COVID-19 vaccine: EMA finds possible link to very rare cases of unusual blood clots with low platelets". European Medicines Agency (EMA) (Press release). 7 April 2021. Archived from the original on 20 May 2021. Retrieved 9 April 2021. Text was copied from this source which is © European Medicines Agency. Reproduction is authorized provided the source is acknowledged.

73-Interim recommendations for use of the ChAdOx1-S [recombinant] vaccine against COVID-19 (AstraZeneca COVID-19 vaccine AZD1222, SII Covishield, SK Bioscience) (Guidance). World Health Organization. 21 April 2021. WHO/2019-nCoV/vaccines/SAGE_recommendation/AZD1222/2021.2. Archived from the original on 8 March 2021. Retrieved 12 February 2021.

74-Interim statement on COVID-19 vaccine booster doses" (Press release). World Health Organization. 10 August 2021. Retrieved 26 August 2021.

75-Arashkia A, Jalilvand S, Mohajel N, Afchangi A, Azadmanesh K, Salehi-Vaziri M, et al. (October 2020). "Severe acute respiratory syndrome-coronavirus-2 spike (S) protein based vaccine candidates: State of the art and future prospects". Reviews in Medical Virology. 31 (3): e2183. doi:10.1002/rmv.2183. PMC 7646037. PMID 33594794.

76-Watanabe Y, Mendonça L, Allen ER, Howe A, Lee M, Allen JD, et al. (January 2021). "Native-like SARS-CoV-2 spike glycoprotein expressed by ChAdOx1 nCoV-19/AZD1222 vaccine". bioRxiv: 2021.01.15.426463. doi:10.1101/2021.01.15.426463. PMC 7836103. PMID 33501433.

77-He TC, Zhou S, da Costa LT, Yu J, Kinzler KW, Vogelstein B (March 1998). "A simplified system for generating recombinant adenoviruses". Proc Natl Acad Sci U S A. 95 (5): 2509–

14. Bibcode:1998PNAS...95.2509H. doi:10.1073/pnas.95.5.2509. PMC 19394. PMID 94829 16.

78-Thomas P, Smart TG (2005). "HEK293 cell line: a vehicle for the expression of recombinant proteins". J Pharmacol Toxicol Methods. 51 (3): 187–200. doi:10.1016/j.vascn.2004.08.014. PMID 15862464

79-Kovesdi I, Hedley SJ (August 2010). "Adenoviral producer cells". Viruses. 2 (8): 1681–703. doi:10.3390/v2081681. PMC 3185730. PMID 21994701.

80- Dicks MD, et al. (2012). "A novel chimpanzee adenovirus vector with low human seroprevalence: improved systems for vector derivation and comparative immunogenicity". PLOS ONE. 7 (7): e40385. Bibcode:2012PLoSO...740385D. doi:10.1371/journal.pone.0040385. PMC 3396660 . PMID 22808149.

81-"Drug Levels and Effects". COVID-19 vaccines. Bethesda (MD): National Library of Medicine (US). 2006. PMID 33355732. Retrieved 15 October 2021.

82-Mishra SK, Tripathi T (2021). "One year update on the COVID-19 pandemic: Where are we now?". Acta Tropica. Elsevier BV. 214: 105778. doi:10.1016/j.actatropica.2020.105778. ISSN 0001-706X. PMC 7695590. PMID 33253656.

83-https://www.thermofisher.com/iq/en/home/life-science/antibodies/antibodies-learning-center/antibodies-resource-library/antibody-methods/introduction-immunoglobulins.html

84-Alberts, B., et al. (1983). Molecular Biology of the Cell. Garland Publishing, Inc., New York, NY.

Harlow, E. and Lane, D. (1988). Antibodies: A Laboratory Manual. Cold Spring Harbor Laboratory, Cold Spring Harbor, NY.

Sites, D.P., et al. (1976). Basic & Clinical Immunology. Lange Medical Publication, Los Altos, CA.

85-Bean KV, Massey HD, Gupta G. Mediators of inflammation: complement. In: McPherson RA, Pincus MR, eds. Henry's Clinical Diagnosis and Management by Laboratory Methods. 24th ed. Philadelphia, PA: Elsevier; 2022:chap 48.

86-Pincus MR, Abraham NZ. Interpreting laboratory results. In: McPherson RA, Pincus MR, eds. Henry's Clinical Diagnosis and Management by Laboratory Methods. 23rd ed. St Louis, MO: Elsevier; 2017:chap 8.Review Date: 04/29/2019

87-https://www.healthline.com/health/total-protein#results

88-Thompson D, Pepys MB, Wood SP (February 1999). "The physiological structure of human C-reactive protein and its complex with phosphocholine". Structure. 7 (2): 169–77. doi:10.1016/S0969-2126(99)80023-9. PMID 10368284.

89-Pepys MB, Hirschfield GM (June 2003). "C-reactive protein: a critical update". The Journal of Clinical Investigation. 111 (12): 1805–12. doi:10.1172/JCI18921. PMC 161431. PMID 12813013.

90- Lau DC, Dhillon B, Yan H, Szmitko PE, Verma S (May 2005). "Adipokines: molecular links between obesity and atheroslcerosis". American Journal of Physiology. Heart and Circulatory Physiology. 288 (5): H2031–41. doi:10.1152/ajpheart.01058.2004. PMID 15653761

91-Mantovani A, Garlanda C, Doni A, Bottazzi B (January 2008). "Pentraxins in innate immunity: from C-reactive protein to the long pentraxin PTX3". Journal of Clinical Immunology. 28 (1): 1–13. doi:10.1007/s10875-007-9126-7. PMID 17828584. S2CID 20300531.

92- Enocsson H, Karlsson J, Li HY, Wu Y, Kushner I, Wetterö J, Sjöwall C (December 2021). "The Complex Role of C-Reactive Protein in Systemic Lupus Erythematosus". Journal of Clinical Medicine. 10 (24). doi:10.3390/jcm10245837. PMC 8708507. PMID 34945133.

93-https://www.metropolisindia.com/blog/prevention-healthcare/crp-test-in-covid-19-all-you-need-to-know/

94-Halim, M.; , Halim, A. and Tjhin, Y. (2021). COVID-19 Vaccination Efficacy and Safety Literature Review. J Clin Med Res ISSN: 2582-4333.

95-Meo, S. A.; Bukhari, I. A.; Akram, J. and Klonoff, D. C. (2021). COVID-19 vaccines: comparison of biological, pharmacological characteristics and adverse effects of Pfizer/BioNTech and Moderna Vaccines. Eur Rev Med Pharmacol Sci 25(3):1663-1669.

96-Malayala, S. V.; Mohan, G.; Vasireddy, D. and Atluri, P. (2021). Purpuric Rash and Thrombocytopenia After the mRNA-1273 (Moderna) COVID-19 Vaccine. Cureus 13(3.(

97-Sahu, A. and Lambris, J. D. (2002). Structure and biology of complement protein C3, a connecting link between innate and acquired immunity. Immunological Reviews.

98-Kurtovic, L. and Beeson, J. G. (2021). Complement Factors in COVID-19 Therapeutics and Vaccines. Trends in Immunology, 42(2), 94-103.

99-Fanga, S.; Wang, H.; Lu, L.; Jia, Y. and Xia, Z. (2020). Decreased complement C3 levels are associated with poor prognosis in patients with COVID-19: A retrospective cohort study. International Immunopharmacology, Volume 89, Part A

100-Bottermann, M.; Foss, S.; Caddy, S. L.; Clift, D.; Laurens, M.; Tienen, V.; Vaysburd, M.and et al., (2019). Complement C4 Prevents Viral Infection through Capsid Inactivation. Cell Host & Microbe 25, 617–629.

101-Elslande, J.V.; Houben, E.; Depypere, M.; Brackenier, A.; Desmet, S.; André, E.; Ranst, M. V.; Lagrou, K. and Vermeersch, P. (2020). Diagnostic performance of seven rapid IgG/IgM antibody tests and the Euroimmun IgA/IgG ELISA in COVID-19 patients. Clinical Microbiology and Infection, 26(8): 1082-1087.

102-V. Wisnewski, A. V.; Luna, J. C. and Redlich, C. A. (2021). Human IgG and IgA responses to COVID-19 mRNA vaccines. journal.pone.0249499

103-Parry, H. M.; Bruton, R.; Tut, G.; Ali, M.; Stephens, C.; Faustini, S. and et al., (2021). Single Vaccination with BNT162b2 or ChAdOx1 in Older People Induces Equivalent Antibody Generation but Enhanced Cellular Responses after ChAdOx1. The Lancet, SSRN.15 Pages.

104-Kitagawa, Y.; Imai, K.; Matsuoka, M.; Fukada, A.; Kubota, K. and et al., (2021). EvaluationofthecorrelationbetweentheaccessSARS-CoV-2IgM and IgG II antibody tests with the SARS-CoV-2 surrogatevirus neutralization test. J Med Virol. 2022;94:335–341.

105-Shah, J.; Liu, S.; Potula, H.; Bhargava, P.; Cruz, I.; Force, D.; Bazerbashi, A. and Ramasamy, R. (2021). IgG and IgM antibody formation to spike and nucleocapsid proteins in COVID-19 characterized by multiplex immunoblot assays. BMC Infectious Diseases 21:325.

106-Zinellu, A. and Mangoni, A. A. (2021).Serum Complement C3 and C4 and COVID-19 Severity and Mortality: A Systematic Review and Meta-Analysis With Meta-Regression. Front. Immunol 12:696085.

❖ **الخلاصة :**

أجريت دراسة عشوائية على المتلقين اللقاح في محافظة بابل لكلا الجنسين ،حيث بلغ عدد الملقحين 695 شخصاً .منهم 561 تلقى لقاح فايزر ، و 120 شخصاً سينوفاك ، و 14 أسترازينيكا ، ووضحت النتائج أن أعلى نسبة تطعيم كان الملقحين فايزر بنسبة (80.71٪) يليه لقاح سينوفاك بنسبة (17.36٪) بينما اقل نسبة لقاح استرازينيكا بنسبة (2.01٪). تم توزيع جميع الملقحين في هذه الدراسة حسب الجنس (ذكور وإناث) ، ووجد أن عدد الإناث الملقحات أكثر من الذكور ، وكان أعلى عدد للإناث الملقحات بلقاح فايزر (415) ، يليه لقاح أسترازينيكا (8) ، بينما قل عدد الاناث الملقحات بلقاح سينوفاك (31).

تم جمع الدم من بعض الملقحين وغير الملقحين ، ثم فصل المصل ثم تخزينه في التجميد حتى لتقدير البروتين الكلي بواسطة مجموعة البروتين الكلي وتقدير الغلوبيولين المناعي والمكون التكميلي C3 و C4 بطريقة مانسين (مناعة شعاعية مفردة).

ظهرت النتائج بفحص مستويات C3، C4، IgG، IgM ، وتركيز البروتين الكلي في مصل الأشخاص الملقحين ، ومقارنتها مع غير الملقحين . أظهرت الدراسة ارتفاع

مستويات تركيز C3 و IgG و IgM وإجمالي البروتينات في مصل الأشخاص الملقحين مقارنة مع المجموعة الغير ملقحة ، في حين أن تركيز C4 منخفض مقارنة بالمجموعة الغير ملقحه.

وجدت نتائج هذه الدراسة أن أكثر أنواع اللقاح شيوعًا هو Pfizer متبوعًا بـ Sinopharmوأقل مع AstraZeneca ، ووجدت اللقاحات استجابة مناعية خلطية معززة.

وزارة التعليم العالي والبحث العلمي.

جامعة بابل / كلية العلوم.

قسم علوم الحياة ... فرع الأحياء المجهرية.

دراسة لتراكيز الغلوبيولينات المناعية في الاشخاص المصابين بفيروس كوفيد 19.

بحث مقدم لكلية العلوم/ جامعة بابل

كجزء من متطلبات الحصول على درجة البكالوريوس في علوم الحياة فرع الأحياء المجهرية.

تم اعداده من قبل كل من :

سماء أسعد حميد. **حسن فلاح محمد.** **فاطمة خالد علي.**

شذا رويض شذر.

بإشراف :

أ.د. فريال جميل. **أ.د. ازهار عمران.** **م.م انمار مهدي .**

2022 A.D. **1443 A.H.**