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Evaluation of the Proportion of SARS-CoV-2 IgG Antibodies in those Who Received the Coronavirus Vaccine Compared to Those Who had Previous Infections

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Abstract

Of a total of 30 cases collected divided into three groups Including 10 (33.3 %) were participants who had received two doses vaccinated against the Coronavirus, the second groups10 (33.3 %) were people who have had a previous infection with Coronavirus and had received two doses vaccinated and the third groups10 (33.3 %) were the people who have only had a previous infection with the Coronavirus.

The results revealed high significance (p<0.05) in the specific IgG antibodies to anti- SARS-CoV-2 in the first group represented by individuals who took two dosages vaccinated against the Coronavirus When compared with the standard criteria used in the diagnostic kit used in the research (MAGLUMI 2019-nCoV IgG). This group included 10, which showed 10 (100%) the efficiency of the vaccines used against the Coronavirus has a significant IgG anti-SARS-CoV-2 concentration (positive) while there is no negative sample.

The present study revealed high significance (p<0.05) in the specific IgG antibodies to anti- SARS-CoV-2 in the second group represented by where those who have already been infected a previous with Coronavirus and had been received two doses of vaccination When compared to the diagnostic kit's conventional criteria, this group included 10, which showed 9 (90%) the efficiency of the vaccines used against the Coronavirus has a significant IgG anti-SARS-CoV-2 concentration (positive) while only 1 (10%) of the sample was negative, while the last group showed the percentage of acquired immunity specific IgG antibodies to anti- SARS-CoV-2 in people who have had a previous infection with the Coronavirus is a small percentage were 4 (40%)positive and 6 (60%) were negative.

The results of this research showed that high significance (p<0.05) in the specific IgG antibodies to anti- SARS-CoV-2 in the female in the first and second groups compared with a male which represented Individuals who took two doses vaccinated against the Coronavirus and people who have had a previous infection with Coronavirus and had been vaccinated respectively while in the third group the male showed high significance (p<0.05) compared to female which those who have already been infected a previous with Coronavirus.Finelly when compare between the vaccine which received in two doses.

Showed efficiency of the Pfizer vaccine (17.72) AU/mL compared to the Sinopharm vaccine (8.55 AU/)mL. Measurement was made in the first group only, which received vaccines in two doses and the rates were compared with the standard ratios of the kit used in the research.

Keywords: SARS-CoV-2 IgG, Vaccine, Pfizer Vaccine

Introduction

Wuhan City reported the first COVID-19 infections in December 2019. COVID-19—later termed SARS-CoV-2—caused the sickness. Chinese authorities detected retroactive cases of early December 2019 symptoms. Wuhan wholesale food market vendors, personnel, and customers were early patients. SARS-CoV-2 was discovered in Wuhan City market ambient samples in December 2019, suggesting the outbreak's genesis or early amplification. Market closed 2020-01-01. SARS-CoV-2 was found early January and its genetic sequence disclosed on January 11 and 12. The entire genetic sequence of the virus from the first human cases and the genomes of countless more viruses from patients in China and other countries after then reveal that SARS-CoV-2 originated in bat populations. Humans have not created the virus. SARSCoV-2's genetic features have not been proven lab-made by many specialists.

The virus's genome would be familiar if it were created. No. SARS-CoV-1 was closely related to bat coronaviruses. SARSCoV-1, SARS-CoV-2, and other coronaviruses may have developed in bat populations due to genetic similarities (WHO, 2020) [31].

SARS-CoV-2 produced 4.26 billion cases of COVID-19 on February 22, 2022. Disease-related fatalities exceeded 5.8 million. SARSCoV-2 may infect anybody and cause serious illness. On the other hand, people under the age of sixty, those residing in nursing homes or other long-term care facilities, and those who have had several health problems in the past are more prone to acquire a severe COVID-19 disease.

Large droplets that are produced by coughing and sneezing have the potential to disseminate COVID-19. It is believed that SARS-CoV-2 requires a period of 14 days to incubate after exposure. 4–5 days. SARS-CoV-2 often infects individuals in their middle years or later in life. While new cases are rising again, the worldwide health community has given 400 million immunisations, bringing hope and progress (Fu et al., 2020) [11].

Materials and Methods
Patients Groups
A total of 30 serum samples were collected and divided into three groups, the first group represents the people vaccinated against the Coronavirus, the second group is the people who have had a previous infection with Coronavirus and have been vaccinated, and the third group is the people who have only had a previous infection with the Coronavirus. All samples ranged in age from (21-80) years they were collected from different places, including colleges from the University of Kufa and other regions Were collected during the period from December (2021) to March (2022).

Blood Sample Processing and Collection
A disposable 5 ml syringe was used to pierce a vein, drawing about 3–5 ml of blood, which was then collected in appropriate tubes without anticoagulant. To separate the serum, the samples were centrifuged at 3000 (rpm) for 10 minutes. The serum samples were then transferred into sterile test tubes using a micropipette with disposable, sterile tips, labeled with the patient's name and serial number, and then frozen at (-20°C) depending on the application in various assays.

Instruments and Equipment

Table 1: The main Instruments and Equipment used in the study

S. No	Instrument and Equipment's	Manufacturer'	Origen
1	Centrifuge	KOKUSAN	Japan
2	Centrifuge tube	Hettich	Germany
3	Disposable Syringes	Witeg	Malaysia
4	High Deep Freezer	GFL	Germany
5	Latex Gloves	Enana	Malaysia
6	Micro Pipettes 10-100 ml	Eppendorf	Germany
7	Microcentrifuge for Eppendorf Tube	Hettich	Germany
8	Refrigerator	Kiriazi	Egypt
9	Sterile Eppendorf Tube	Scharlau	Spain

Chemicals

Table 2: The chemical and biological materials used throughout the study and their sources (kits)

S. No	Items	Manufacturer's	Origen
1	MAGLUMI 2019-nCoV IgG(Kite)	Shenzhen New Industries Biomedical Engineering Co., Ltd. (SNIBE)	USA

Cassette	Components	Contents	100 tests
For SARS CoV-2 IgG antibodies	Magnetic Microbeads	Magnetic microbeads coated with 2019-nCoV recombinant antigen, PBS buffer and BSA, NaN_3 (<0.1%)	2.5 mL
	Calibrator Low	2019-nCoV IgG, PBS buffer and BSA, NaN_3 (<0.1%)	1.0 mL
	Calibrator High	2019-nCoV IgG, PBS buffer and BSA, NaN_3 (<0.1%)	1.0 mL
	Buffer	NaCl and BSA, NaN_3 (<0.1%).	23.5 mL
	ABEI Label	Anti-human IgG antibody labeled with ABEI, Tris-HCl buffer, Mouse IgG, Goat IgG, and BSA, NaN_3 (<0.1%)	23.5 mL
	Diluent	PBS buffer and BSA, NaN_3 (<0.1%)	23.5 mL
	Negative Control	PBS buffer, containing BSA, NaN_3 (<0.1%)	1.0 mL
	Positive Control	2019-nCoV IgG, PBS buffer, containing BSA and NaN_3 (<0.1%).	1.0 mL

All reagents are provided ready-to-use

The MAGLUMI 2019-nCoV IgM/IgG Test Kit Does Not Include Certain Components that are Necessary:

Component	Catalog number	Contents	Quantity/Volume
Reaction Module	630003	polypropylene	64/box

Starter Buffer	130299004M	Catalyst in 1.5% NaOH, 0.18% H2O2	230 mL×1
Wash Concentrate	130299005M	Tris-HCl buffer solution	714 mL×1
Light Check	130299006M	ABEI (II-(4-Aminobutyl)-N-ethylisoluminol), BSA	2mL×5

Test Methods
Reagent Preparation
1. Assembling the reagent kit to check for leaks, the sealing film and other components of the reagent kit were taken out of the box and examined.
2. an entrance to the reagent area Opened For roughly 2 seconds, the detector handle must be held to bring the RFID label close to the RFID reader; after the buzzer beeps, successful sensing has occurred.
3. The detector insert was kept straight downward along the detector blank pat.

4. The detector information has been successfully displayed in the program interface.
5. When the kit is loaded successfully, the magnetic microbeads are automatically resuspended, ensuring they are completely and uniformly resuspended before usage.

Sample Testing
1. 160 µl serum samples were gently transferred into tubes.
2. Sample tubes were loaded for testing.
3. Samples were arranged in the software's sample area and the <Start> button was pressed to test patient want information.
4. Samples from the same patient must be put in various cassettes due to clearing equipment design.

A chemiluminescence immunoassay analyzer will assess MAGLUMIITM series fullyautomated IgG SARS-CoV-2 antibodies, and each patient will get a report.

Result Calculation
The analyzer uses a calibration curve produced by a two-point calibration master curve technique to automatically determine the numerical output in each sample. The outcomes are given in absorbance units (AU/mL). The end user is given two results options: "Reactive" and "Non-Reactive." To the end user, no numerical AU/mL numbers are provided. Please see the fully automated chemiluminescence immunoassay analyzer's MAGLUMI series for more information. Operating instructions for further details.

Result Interpretation
1. Non-reactive results are those with a value of less than 1.00 AU/mL (1.00 AU/mL).
2. Reactive: A result is deemed reactive if it is greater than or equal to 1.00 AU/mL (1.00 AU/mL).

Analyte	Results	Interpretation	Description*
SARS-CoV-2 IgG	<1.00 AU/mL	SARS-CoV-2 IgG Non-Reactive	IgG antibodies against SARS-CoV-2 are not detected
	≥1.00 AU/mL	SARS-CoV-2 IgG Reactive	IgG antibodies against SARS-CoV-2 are detected

Analytical Statistics
The Statistical Package for the Social Sciences (SPSS) version 20 was used to conduct the statistical analysis for the study that was presented. The ANOVA test was used to identify age variation. A P value of less than 0.05 was used to demonstrate statistical significance.

Result and Dissection
Clinical Samples Distribution
Of a total of 30 cases collected divided into three groups Including 10 (33.3%) were participants who had received two doses vaccinated against the Coronavirus, the second groups10 (33.3%) were those who have already been infected previous with Coronavirus and had received two doses vaccinated and the third groups 10 (33.3%) were the people who have only had a previous infection with the Coronavirus (Fig 1).
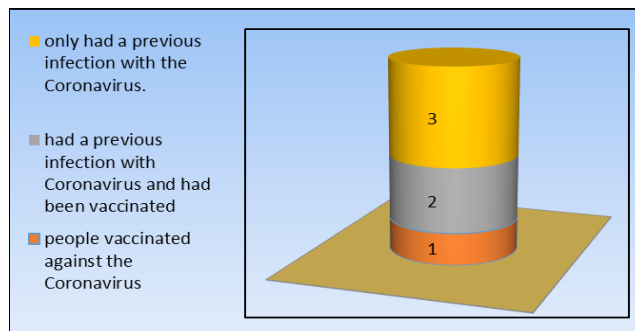

Fig 1: Samples Distribution

Distribution of Sample According to Sex
The study groups were divided into six the highest number 20 (66.6%) were male and 10 (33.3%) were female as in (Fig 2).
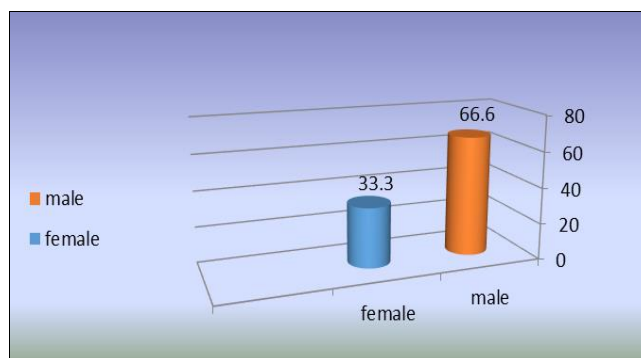

Fig 2: Distribution of sample according to sex

Evaluation of the First Group's Adaptive Immune Response to SARS-CoV-2.
The results revealed high significance (p<0.05) in the specific IgG antibodies to anti- SARS-CoV-2 in the first group represented by individuals who took two dosages vaccinated against the Coronavirus Within a period of time not exceeding six months from the date of the second doseWhen compared with the standard criteria used in the diagnostic kit used in the research (MAGLUMI 2019-nCoV IgG). This group included 10, which showed 10 (100%) the efficiency of the vaccines used against the Coronavirus contains a significant amount of IgG anti-SARS-CoV-2 (positive) while there is no negative sample. Table (1). This results in agreement with Victoria Hall., et al (2022) [13] who found Exceptional short-term defense against SARS-CoV-2 infection was associated with receiving two doses of the vaccine; however, this protection began to decline significantly after six months.

Table 1: Titer of IgG anti SARS-CoV-2 in people received two doses vaccinated against the Coronavirus

Mean of SARS-CoV-2 IgG NO(10)	SARS-CoV-2 IgG Non-Reactive	SARS-CoV-2 IgG Reactive	P Value
15.89 AU/mL ***	<1.00 AU/mL	≥1.00 AU/mL	(p<0.05)

*Non-Reactive: (IgG antibodies against SARS-CoV-2 are not detected)
*Reactive: (IgG antibodies against SARS-CoV-2 are detected)

Measurement of the Adaptive Immune Response to SARS-CoV-2 in the Second Group

The present study revealed high significance (p<0.05) in the specific IgG antibodies to anti- SARS-CoV-2 in the second group represented by where people who have had a previous infection with Coronavirus and had been received two doses of vaccination Within a period of time not exceeding six months from the date of the second dose.When compared to the diagnostic kit's conventional criteria, the investigation found (MAGLUMI 2019-nCoV IgG). This group included 10, which showed 9 (90%) the efficiency of the vaccines used against the Coronavirus with a high concentration of IgG anti-SARS-CoV-2 (positive) while only 1(10%) of the sample was negative. Table (2). This result is similar to Joseph *et al*., (2021) who discovered that some people had previously been infected with SARS-CoV-2 Development of BNT162b2 (Pfizer-BioNTech) mRNA vaccine-induced antibody responses after a single dose that were comparable to antibody responses following a two-dose vaccination.

Table 2: Titer of IgG anti-SARS-CoV-2 in people who have had a previous infection with Coronavirus and had been vaccinated

Mean of SARS-CoV-2 IgG NO(9)	SARS-CoV-2 IgG Non-Reactive	SARS-CoV-2 IgG Reactive	P Value
15.5 AU/mL**	<1.00 AU/mL	≥1.00 AU/mL	(p<0.05)

*Non-Reactive: (IgG antibodies against SARS-CoV-2 are not detected)
*Reactive: (IgG antibodies against SARS-CoV-2 are detected)

Measurement of the Adaptive Immune Response to SARS-CoV-2 in the Thrid Group

The current study showed the percentage of acquired immunity specific IgG antibodies to anti- SARS-CoV-2 in people who have had a previous infection with the Coronavirus is a small percentage were 4 (40%) positive while 6 (60%) were negative When compared to the diagnostic kit's conventional criteria, the investigation found (MAGLUMI 2019-nCoV IgG). Table (3). This result in concept to Nabin., *et al* (2020) who discovered previously noted that those who had coronavirus disease 2019 (COVID-19), Nevertheless, had an extremely low cumulative incidence of COVID-19 during 5.5 months of follow-up and was caused by the SARS-CoV-2 coronavirus (SARS-CoV-2).

Table 3: Titer of IgG anti-SARS-CoV-2 in people who have had a previous infection with Coronavirus

Mean of SARS-CoV-2 IgG NO(4)	SARS-CoV-2 IgG Non-Reactive	SARS-CoV-2 IgG Reactive	P Value
9.5 AU/mL **	<1.00 AU/mL	≥1.00 AU/mL	(p<0.05)

*Non-Reactive: (IgG antibodies against SARS-CoV-2 are not detected)
*Reactive: (IgG antibodies against SARS-CoV-2 are detected)

Comparison of the Acquired Immune Response IgG Anti-SARS-CoV-2 According to Sex

The result in the Table 4 showed that high significance (p < 0.05) in the specific IgG antibodies to anti- SARS-CoV-2 in the female in the first and second groups compared with a male which represented people who received two doses vaccinated against the Coronavirus and those who have already been infected previous with Coronavirus and had been vaccinated respectively while in the third group the male showed high significance (p<0.05) compared to female which people who have had a previous infection with Coronavirus,this result agreement with Qing., *et al* (2020) Researchers discovered that hospitalization rates were greater for males than for females in all reported nations, ranging from 55% to 62%, after analyzing 19 databases and 45 publications. Additionally, men were even more likely to enter the ICU, ranging from 65% to 74%, the mortality rates of men were greater than those of women, ranging from 59% to 69%. Men presented, according to early reports from China, in most cases (but not always).

Table 4: Titer of IgG anti-SARS-CoV-2 according to sex

groups	NO (+)	Mean of SARS-CoV-2 IgG male	NO (+)	Mean of SARS-CoV-2 IgG female	SARS-CoV-2 IgG Non-Reactive	SARS-CoV-2 IgG Reactive	P Value
Group(1) people received two doses vaccinated against the Coronavirus.	7	11 AU/mL	3	17.9 AU/mL	<1.00 AU/mL	≥1.00 AU/mL	(p<0.05)
Group(2) people who have had a previous infection with Coronavirus and had been vaccinated	6	14.9 AU/mL	3	16.6 AU/mL	<1.00 AU/mL	≥1.00 AU/mL	(p<0.05)
Group(3) people who have had a previous infection with Coronavirus.	3	10.7	1	5.8	1.00 AU/mL	≥1.00 AU/mL	(p<0.05)

*Non-Reactive: (IgG antibodies against SARS-CoV-2 are not detected)
*Reactive: (IgG antibodies against SARS-CoV-2 are detected)

Measurement of Acquired Immunity IgG Anti-SARS-CoV-2 in Participants According to the Type of Vaccine

The results of the current study indicated in (Table 4) the efficiency of the Pfizer vaccine (17.72) AU/mL compared to the Sinopharm vaccine (8.55 AU/)mL. Measurement was made in the first group only, which received vaccines in two doses and the rates were compared with the standard ratios of the kit used in the research (MAGLUMI 2019-nCoV IgG). These results are similar to a study conducted by Rajasekaran, (2021) that found Ninety-three percent of the 141 subjects who received the Pfizer–BioNTech COVID–19 vaccination had positive IgG titers. In contrast, 147 people received the Sinopharm vaccination, and 85.7% of them had positive IgG titers.

Table 5: Titer of IgG anti-SARS-CoV-2 according to Type of Vaccine

Type of Vaccine	Mean of SARS-CoV-2 IgG	SARS-CoV-2 IgG Non-Reactive	SARS-CoV-2 IgG Reactive	P Value
Pfizer BioNTech (BNT162b2)	17.72 AU/mL**	<1.00 AU/mL	≥1.00 AU/mL	(p<0.05)
Sinopharm vaccine	8.55 AU/mL*			

*Non-Reactive: (IgG antibodies against SARS-CoV-2 are not detected)
*Reactive: (IgG antibodies against SARS-CoV-2 are detected)

Conclusions

Using a (MAGLUMI 2019-nCoV IgG) fully-automated chemiluminescence immunoassay analyzer. That is a useful and good technique in measuring acquired immunity specific IgG anti-SARS-CoV-2, 2- The results revealed high significance (p<0.05) in the specific IgG antibodies to anti-SARS-CoV-2 in the first group represented by people who received two doses vaccinated against the Coronavirus Compared with the second group vaccinated and infected and the third group infected only, The previous infection with the Coronavirus gives a small percentage of acquired immunity (40%), The women's acquired immunity was higher in both groups first and second than in males.

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