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Prevalence of COVID-19 virus infection in asymptomatic volunteers in Baghdad city / Iraq during 2021

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***Abstract*---**Background: From February April 2020, the COVID-19 pandemic has swept through more than 200 countries and infected more than 100 million individuals globally, posing an unprecedented threat to human health. There are currently no specific antiviral treatments for COVID-19 and vaccination programs, whilst promising, remain in their infancy. A key to restricting the pandemic is the ability to minimize human-human transmission and predict the infection status of the population in the face of emerging SARS-CoV-2 variants. Success in this area is dependent on the rapid detection of COVID-19 positive individuals with current/previous SARS-CoV-2 infection status. The 3 categories of tests used to detect current or past viral infection are molecular, serologic, and antigen-detection assays**.** Aims: This study aimed to evaluate the performance of the fluorescence LFIA Finecare TM 2019-nCoV S-RBD test along with its reader (Model No.: FS-113) in Iraqi setting. Giving a look on the prevalence of Covid19 infection in Baghdad city is our second aim. Study design A retrospective study on 75 randomly selected serum samples collected between 10 and 15 April 2020 was performed. They were checked for Covid-19 specific IgG and IgM responses by

FinCare™ rapid semi-automated kit. Two aims were put in examinations. Participants were required to meet the following criteria: (i) Adult *Iraqi volunteers*; (ii) males aged between 12-65, (iii) Subject with any chronic disease was excluded from this study. Result: By Finecare™ 2019-nCoV RBD Antibody seems reliable for rapid diagnosis of Covid19 infections in Iqaqi patients. Test valid positive was 35 samples (46.7%), whereas IgG negative were 40 samples (53.3%). Valid IgM incidence was 20 (26.7 %) with negative IgM cases were 55 (73.3%). High prevalence of COVID-19 infection in Iraqi population probably due to high multiple transmission modes inducing early and recurrent infections. Conclusion: FinecareTM is a reliable assay and can be used as a tool to assess binding and antibody response after infection or vaccination, particularly in none or small laboratories.

***Keywords*---**SARS-CoV-2, COVID-19, fluorescence immunoassay, IgG COVID, IgM COVID, laboratory parameters COVID, lab test COVID.

Introduction

Soon after the emergence of the severe acute respiratory syndrome 2 (SARS-CoV-2) in December 2019 and its declaration as a pandemic by the World Health Organization (WHO), the need for accurate, sensitive, and rapid detection of SARS-CoV-2 for the control and prevention of the disease was urgent (1). Several commercial COVID-19 test kits were developed in response to this urgent need to detect either nucleic acid or antibodies (2). Although RT-PCR is used as a gold standard test, it requires specialized conditions, expensive equipment, and qualified personnel for sampling and testing. These limitations pose a challenge in a pandemic setting which requires rapid and reliable tests that can be used to screen populations (3). Thus, serological testing, particularly the conventional *lateral flow immunoassay* (*LFIA*), commonly known as a rapid test was introduced in clinical settings for diagnosing SARS-CoV-2 infection are done manually within 15 minutes, and the results are interrupted qualitatively by the naked eye. As a result, errors due to manual operation and inadequate visual sensitivity interpretation can occur. Wondfo Biotech developed Fluorescence-based LFIA to detect total IgG and IgM antibodies against SARS-CoV-2. Finecare™ 2019-nCoV Antibody Test is a fluorescence immunoassay that is done semiautomatically along with a small portable device (Model No.: FS-113). The combination of fluorescence and LFIA provides higher sensitivity, quantitative detection of antibodies. In addition, the test is easily affordable and accessible in small clinical laboratories, research settings, or point of care testing, including post-infection or vaccination. To the best of our knowledge, there is no data in the literature evaluating the rapid fluorescence in IRAQ. Hence, authors tried to examine the validity of this system on a group of asymptomatic volunteers.

Titers of IgG and IgM have been recorded at elevated levels in the serum of SARSCoV2 patients (4).Still , some patients suffer from the below-normal ratio of leukocytes while other infected persons sowed increased leukocyte and neutrophil ratios (5) indicating an immune derangements. Intravenously given

immunoglobulins to activate anti-infection responses in severe cases is to be practiced. Serological outcome of the disease involves detection of immunoglobulin G (IgG) as well as IgM in the patient specimens which were found to be induced by the spike protein of SARS-CoV-2 during the early first four days from the development of the disease. Additionally, the S and N proteins of this virus represent major detected immunogens in the patients infected with this virus (6). Effective anti-viral therapies for COVID-19, and the prevention of human-to-human transmission requires extensive testing, quarantine, and contact tracing (7).

In present study, we checked for IgG and IgM antibodies levels based on fluorescent immunoassay serological assay kit FineCare™ validated for the detection of SARS-CoV-2 status. samples from blood donors collected in Baghdad, Iraq, between 10 and 15 April 2020 were assessed for current or previous SARS-CoV-2 infection.

Materials and Methods

Study design and blood collections

We evaluated the performance of FineCareTM 2019-nCOV Antibody test and its reader (Model No.: FS-113); from here on, abbreviated as FineCareTM. The serum samples were collected from volunteers between 10 and 15 April 2020. Ninety (90) subjects were included in this study. They were checked for SARS-CoV-2 IgG and IgM antibodies levels. Study participants were required to meet the following criteria included in this study: (i) Adult *Iraqi volunteers*; (ii) Males and Females aged between 12-65, (iii) Any subject with any chronic disease was excluded from our study (Accordingly, 15 individuals were excluded from this study).

Data statistical analysis

The data was analyzed using SPSS version 26 software and expressed as percent between IgG and IgM and *P*-value, demographic and clinical features of the study participants were summarized using descriptive statistics.

Results

Data depicted in the table-1 and figure-2 revealed that IgG valid positive samples were thirty-five (35) (46.7%) while negative samples were forty (40) (53.3%). In table-2 and figure 3 ,data revealed True IgM incidence reached twenty (20) (26.7 %) contrasted to negative IgM numbers which were Fifty-five (55) (73.3%). Data depicted in table -2 and figure -2 showed the number and percentages of IgG positive individuals significantly higher thant those IgM positive ones.

Statistical analysis

This study was designed by a completely randomized design (CRD). The correlation between IgG and IgM values in patient's serum sample groups was statistically analyzed by Pearson correlation coefficient. In addition, several statistical tests were used. Data were processed and analyzed using the

statistical program social science (SPSS 26) and results were expressed as percent with *P*-value (Table-1 and 2).

Discussion

This study validated the performance of FinecareTM 2019 nCoV RBD fluorescence immunoassay for detecting total antibodies against SARS-CoV2 S-RBD after infection. A panel of 75 samples collected from volunteer individuals. It is well-known that the production of antibodies against a particular virus is similar across patients (except for immunocompromised patients) during the acute and the chronic phases of infection.

The accurate and timely diagnosis of patients with asymptomatic as well as symptomatic SARS-CoV-2 infections remains crucial to limit current SARS-CoV-2 human-to-human transmission. The structural proteins of SARS-CoV-2 are highly immunogenic and lead to the generation of IgM and IgG antibodies (8) that are essential to the effective management of the pandemic. The present study showed a high incidence of IgG positive serum 35 (46.7%) representing the infected patients with SARS- Cov-19 viral infection at the acute or recovery stage. Concerning the IgM positive samples which reached 20% usually indicate the recent infection. In the total recruited 75 patients rate of Covid-19 infection was estimated to 73% when totaling both recent and old infections.

Detection of virus-specific antibody is effective for the diagnosis of SARS-CoV and middle east respiratory syndrome coronavirus (MERS-CoV) infection. Currently, according to the “Diagnosis and Treatment Protocol for Novel Coronavirus Pneumonia SARS-CoV-2 antibody assays are extensively carried out for COVID-19 diagnosis. Following its validation, the assay identified a single infected patient in the Baghdad /Iraq revealing its potential as a diagnostic tool for SARS-CoV-2. Importantly, the assay could easily be modified to increase capacity using stable SARS-CoV-2 cell lines and automated high throughput fluorescent analysis. We find a high prevalence of COVID-19 viral infection in Iraq probably due to multiple transmission modes inducing early and recurrent miscarriages (9).

Among the participants with antibodies all denied having any history of COVID-19-related symptoms. The most common symptom was fever, followed by myalgia. We found that antibody levels increased steadily with age (table-2 and figure-2). Similar observations were noticed in Hussein et al in their data analyzed in Duhok city in northern IRAQ. (10) There are several limitations in this study. First, this was a study on aggregate data and we lacked dynamic patient-level data, so this may have affected the results. Second, the sample size was small, which weakened the strength of our findings. Third, the study duration was only few weeks, so the results cannot fully explain the whole immune process in humans. More large-scale, long-term clinical studies focusing on patient-level data are needed to further confirm the conclusionsIn conclusion, this assay could be reliable for the quantitative detection of antibodies in the vaccinated population and recovered patients.

Table 1. IgG antibodies according to the age ranges with their descriptive statistics

			Positive		Negative		*P*-Value
Age category	No.	%	No.	%	No.	%	
<20 Years old	7	9.3	4	57.1	3	42.9	>0.05
20-31 Years old	31	41.3	15	48.4	16	51.6	>0.05
32-40 Years old	25	33.3	10	40	15	60	>0.05
>40 Years old	12	16	6	50	6	50	>0.05
Total	75	100	35	46.7	40	53.3	>0.05

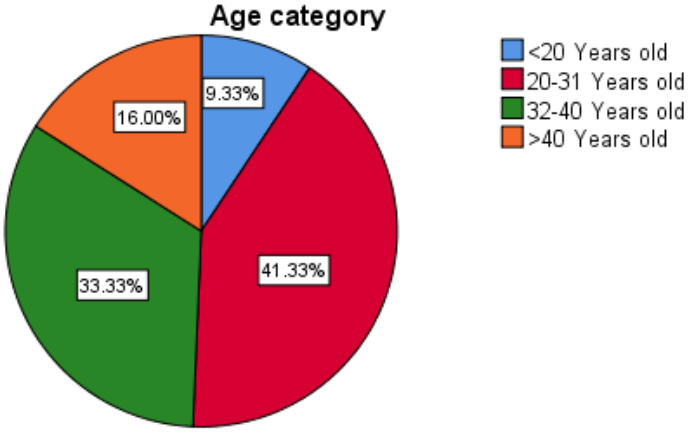

Figure 1. Descriptive statistics of Age ranges; Significant differences were noticed in the 20-31 years old range (percentage reached approximately 41.3%).

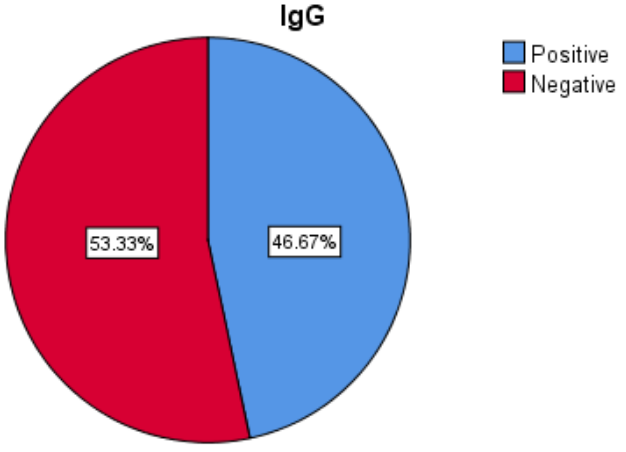

Figure 2. Percent of IgG antibodies in the total 75 patients. No significant differences between positive and negative results according to *P*-value.

Table 2. IgM antibodies according to the age ranges with their descriptive statistics

			Positive		Negative		*P*-Value
Age category	No.	%	No.	%	No.	%	
<20 Years old	7	9.3	2	28.6	5	71.4	>0.05
20-31 Years old	31	41.3	8	25.8	23	74.2	<0.05*
32-40 Years old	25	33.3	5	20	20	80	<0.05*
>40 Years old	12	16	5	41.7	7	58.3	>0.05
Total	75	100	20	26.7	55	73.3	<0.05*

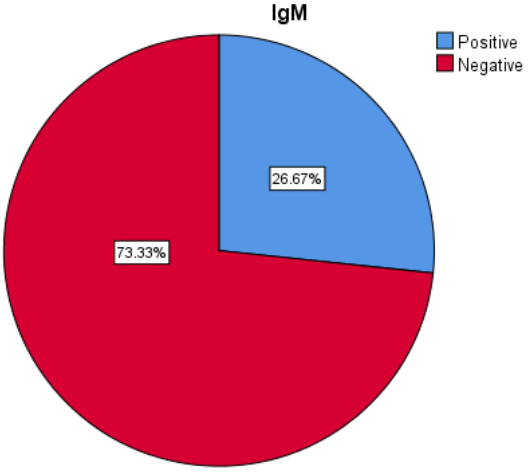

Figure 3. Percent of IgM antibodies in the total 75 patients. Significant differences between positive and negative results according to *P*-value.

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