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## Original Research Article

## Evaluation of three commercial SARS-CoV-2 serology assays in a tertiary care hospital in the United Arab Emirates

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## ABSTRACT

**Background:** Serology assays have the potential to support RT-PCR in the diagnosis of SARS-CoV-2 infection. We studied three commercially available immunoassays for their diagnostic accuracy from blood specimens collected from 93 patients.

**Methods:** Blood samples from patients with confirmed COVID-19 infection were analysed using three different Immunoassays (Roche total antibody assay, Abbott IgG assay and Euroimmun IgG assay). Sensitivity, specificity, precision and time of seroconversion were evaluated.

**Results:** The sensitivity of Roche, Abbott and Euroimmun assays was 38.7%, 35.5% and 25.0% respectively for specimens collected <10 days and 84.4%, 84.4% and 70.0% respectively for specimens collected ≥10 days after the first positive RT-PCR. The specificity of all the three assays in this study was 100%. The timing of seroconversion occurred at day 1, 7 or 14.

**Conclusions:** The assays evaluated in this study have different sensitivities for detecting antibodies in SARS-CoV-2 infection. Sensitivity for detecting antibodies for all three assays was higher for specimens collected ≥10 days after first positive PCR compared with specimens collected <10 days. Time of seroconversion is variable and assay-dependent.

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## Introduction

Coronavirus disease 2019 (COVID-19) is caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). The COVID-19 pandemic which was initially reported in Wuhan, China in December 2019 has been since spreading worldwide [1]. At the time of writing this paper, The World Health Organization (WHO) has reported 21,294,845 cases and 761,779 deaths worldwide [2].

Currently, there are three types of laboratory tests available for the detection of SARS-CoV-2. Molecular tests detect the RNA of the virus while the antigen tests directly detect viral antigens [3–5]. Reverse transcriptase polymerase chain reaction (RT-PCR) is the gold standard test recommended for use by the WHO for the diagnosis of COVID-19 cases [6]. Serology tests, on the other hand, reflect the immune response to the virus by detecting the presence of antibodies in blood.

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Serology tests have generated substantial interest as a potential alternative to RT-PCR in the diagnosis of SARS-CoV-2 infection as they have faster turn-around time and they are cheaper and easier to perform in the laboratory in comparison to RT-PCR. According to the most recent publication from the Infectious Disease Society of North America (IDSA), serology assays can be used in selected diagnostic scenarios including providing evidence of COVID-19 infection in symptomatic patients with a high clinical suspicion and repeatedly negative PCR testing, confirmation of past infection and providing evidence of infection in paediatric patients with multisystem inflammatory syndrome [7].

Current evidence indicates that SARS-CoV-2 antibodies begin to develop approximately 6–10 days after infection with SARS-CoV-2 [8,9]. IgM appears to peak approximately 12–15 days after SARS-CoV-2 infection and persists in sufficient quantities for as long as 35 days, after which the quantity declines rapidly. IgG has been observed to peak approximately 17 days after SARS-CoV-2 infection and persist for at least 49 days [9,10].

Because of the pandemic situation and the increasing need for diagnostic testing, the Centres for Disease Control and Prevention (CDC), Food and Drug Administration (FDA) and other international organizations have supported the COVID-19 response, including

**Table 1**  
Characteristics of SARS-CoV-2 serology assays used in this study.

	Roche Cobas antibody assay	Abbott Architect IgG assay	Euroimmun IgG assay	Diasorin LIAISON IgG assay
Type of assay Principle	Qualitative Electrochemiluminescent immunoassay (ECLIA)	Qualitative Chemiluminescent microparticle immunoassay (CMIA)	Qualitative Enzyme immunoassay (ELISA)	Quantitative Chemiluminescence immunoassay (CLIA)
Antigen target	Nucleocapsid (N) antigen	Undissolved epitope of nucleocapsid (N) antigen	S1 domain of viral spike protein	Recombinant S1 and S2 antigens
Result interpretation	Index < 1.0 = negative Index ≥ 1.0 = positive	<1.4 = negative ≥ 1.4 = positive	Ratio <0.8 = negative Ratio ≥ 0.8 to <1.1 = borderline Ratio ≥ 1.1 = positive	<12.0 AU/mL = negative ≥12.0–<15.0 = borderline ≥15.0 = positive
Manufacturer's sensitivity	65.5%, 88.1%, and 100% for specimens collected 0–6, 7–13, ≥ 14 days respectively post PCR	0%, 25%, 86.46%, and 100% for specimens collected <3, 3–7, 8–13, ≥14 days respectively post symptoms	43.7% and 94.4% for specimens collected <10 and >10 days respectively after symptoms	25%, 90.4%, and 97.4% for specimens collected ≤5, 5–15, >15 days respectively post PCR
Manufacturer's specificity	99.81%	99.6%	99.6%	98.5%

development of diagnostic assays and through issuing FDA emergency use authorization (EUA) for some of these assays. Due to the high number of different serologic assays available in the market that are based on different technologies and antigenic targets, there is a considerable uncertainty regarding the accuracy and the clinical performance of these tests.

We compared three of the commercially available immunoassay kits to assess their diagnostic accuracy for SARS-CoV-2 infection. We also included a fourth quantitative assay to measure antibody levels for the selection of convalescent plasma donors. Our objectives were to verify their reported performance and to compare sensitivities and specificities of different test methods. To our knowledge, this is the first study in the United Arab Emirates (UAE) that compares the performance of three different COVID-19 immunoassays.

## Material and methods

### Patients' specimens

Leftover blood specimens that were collected from patients admitted to Cleveland Clinic Abu Dhabi (CCAD) hospital with clinical manifestations suspicious for COVID-19 infection were utilized in this study. CCAD is a 364-bed tertiary care hospital in the United Arab Emirates (UAE). Clinical Information was obtained through reviewing the electronic medical records of patients. Testing was performed at the National Reference Laboratory (NRL). Patients' specimens were divided into three cohorts: the first cohort had 93 specimens to assess sensitivity, specificity and precision; the second cohort had multiple specimens collected from seven patients to evaluate seroconversion and the third cohort had 13 specimens to assess antibody levels from convalescent plasma donors.

### Evaluation of sensitivity, specificity and precision

For the evaluation of sensitivity and specificity of the serology assays, we tested leftover blood specimens from 93 patients (63 patients positive for COVID-19 by RT-PCR and 30 negative) using the Roche and Abbott assays and from 60 patients (30 positive for COVID-19 by RT-PCR and 30 negative) using the Euroimmun assay (first cohort of patients). The 30 COVID-19 negative patients included 10 who had a negative COVID-19 RT-PCR and 20 patients whose samples were previously collected before the COVID-19 pandemic. Some of the blood specimens were taken within 10 days after the first positive RT-PCR results and some specimens were collected after 10 days of the first positive RT-PCR results. We have also compared our results of 10 specimens tested using the Euroimmun assay with another laboratory that uses the same assay.

To assess the reproducibility of results, intra-run and inter-run precision studies were conducted for Roche, Abbott and Euroimmun and verified using CLSI EP5-A2 evaluation criteria. Negative and positive specimens and samples with a concentration near the cut-off point of the assay were processed in at least 10 replicates for intra-run precision while a minimum of 25 replicates in at least 3 days were completed for inter-run precision. Mean, standard deviation and coefficient of variation were calculated and compared with manufacturers' data.

To evaluate seroconversion, we tested seven specimens at days 1, 7 and 14 of the first positive RT-PCR, respectively collected from seven patients (second cohort). All samples were run on three platforms: Roche, Abbott and Euroimmun. Due to inadequate volume of samples, not all specimens were tested using all three assays. Finally, 13 specimens from 13 patients were used to assess antibody levels from convalescent plasma donors (third cohort). These patients were clinically symptomatic, had positive RT-PCR results and donated their plasma to be used for treatment of other COVID-19 patients. The timing of plasma donations varied between 14–46 days from the date of first positive RT-PCR test. These samples were run on Diasorin quantitative assay in addition to Roche and Abbott assays.

For all enrolled patients, SARS-CoV-2 RT-PCR was performed on respiratory specimens using two assays: The U-TOP™ COVID-19 Detection Kit (Seasun biomaterials, Korea) which targets SARS-CoV-2 Open Reading Frame 1ab (ORF1ab) and Nucleocapsid (N) genes and Allplex™ 2019-nCoV Seegene Assay kit (Seegene, Korea) that detects three SARS-CoV-2 genes simultaneously (Envelope (E), RNA-dependent RNA polymerase (RdRp) and N genes).

### SARS-CoV-2 serology assays

Samples were analysed using three different Immunoassays kits (Roche total antibody assay performed on Elecsys machine, Abbott IgG assay performed on Abbott Architecti2000 and Euroimmun IgG assay performed on Euroimmun Analyzer). For the quantification of antibodies in plasma donors, we also included Diasorin quantitative IgG assay performed on the Diasorin LIAISON XL Analyzer. The description of each assay is shown in Table 1.

### Statistical analysis

We used SARS-CoV-2 RT-PCR as the gold standard to evaluate the performance of the four SARS-CoV-2 serology assays. EP evaluator software (Data Innovations, South Burlington, USA) was used to calculate the sensitivity, specificity and precision for each assay. We used the binomial distribution to calculate 95% confidence intervals around the point estimated provided by the EP evaluator software.

**Table 2**  
Diagnostic sensitivity of Roche, Abbott and Euroimmun assays.

Interval (days)	Positive	Negative	Total	Sensitivity % (95% Confidence interval)
<b>Roche Cobas antibody assay</b>				
<10 days	12	19	31	38.7 (21.85–57.81)
≥10 days	27	5	32	84.4 (67.21–94.72)
<b>Abbott Architect IgG assay</b>				
<10 days	11	20	31	35.5 (19.23–54.63)
≥10 days	27	5	32	84.4 (67.21–94.72)
<b>Euroimmun IgG assay</b>				
<10 days	5	15	20	25.0 (8.66–49.10)
≥10 days	7	3	10	70.0 (34.75–93.33)

**Table 3**  
Intra-run and inter-run precision studies of Abbott, Diasorin, Roche and Euroimmun assays.

	Manufacturer's mean	Verified mean	Manufacturer's CV (%)	Verified CV (%)	Manufacturer's SD	Verified SD
<b>Abbott protocol: 2 Levels of QC processed 20 replicates in one day and 25 replicates in 5 days</b>						
Abbott – Intra negative	0.39	0.061	5.9	3.7	0.002	0.002
Abbott – Intra positive	5.03	3.40	1.1	1.6	0.042	0.053
Abbott – Inter negative	0.39	0.061	5.9	4.7	0.002	0.000
Abbott – Inter positive	5.03	3.440	1.81	1.9	0.042	0.031
<b>Roche protocol: 2 Levels of QC processed 10 replicates in one day and 30 replicates in 3 days</b>						
Roche – Intra-run negative	0.089	0.00003	4.49	1.9	0.004	0.0016
Roche – Intra-run positive	28.9	19.2	6.22	0.8	1.8	0.240
Roche – Inter-run negative	0.089	0.085	4.49	2.4	0.004	0.002
Roche – Inter-run positive	28.9	28.3	6.22	1.0	1.8	0.028
<b>Euroimmun protocol: 3 Levels of patient pool samples run as QC materials processed 4 replicates in 1 day and 3 replicates in 5 days</b>						
Euroimmun – Intra-run negative	NA	0.28	16	5.3	NA	0.015
Euroimmun – Intra-run at cut-off	NA	1.33	5.4	3.3	NA	0.04
Euroimmun – Intra-run Positive	NA	7.09	4.4	2.4	NA	0.17
Euroimmun – Inter-run negative	NA	0.25	16.2	16.4	NA	0.04
Euroimmun – Inter-run at cut-off	NA	1.11	5.7	5.89	NA	0.06
Euroimmun – Inter-run positive	NA	6.87	5.5	6.6	NA	0.46

**Table 4**  
Seroconversion at day 1, 7 and 14 after the first positive SARS-CoV-2 RT-PCR using Roche, Abbott and Euroimmun serology assays.

Anti-SARS-CoV-2 method comparison										Patient information		
Patient #	Day 1 Roche	Abbott	Euroimmun	Day 7 Roche	Abbott	Euroimmun	Day 14 Roche	Abbott	Euroimmun	Clinical notes	Sex	Age
1	Negative	Negative	Negative	Positive	Positive	Negative	Positive	Positive	Positive	Cerebral infarction	M	47
2	Negative	–	–	Positive	Positive	Positive	Positive	Positive	Positive	Pneumonia	M	59
3	Negative	Negative	Negative	Positive	Positive	Positive	Positive	Positive	Positive	Fever	M	34
4	Negative	–	–	–	–	–	Negative	Negative	Positive	Respiratory failure	M	62
5	Negative	Negative	Negative	–	–	–	Negative	Negative	Negative	Kidney transplant	M	20
6	Positive	–	–	Positive	–	–	Positive	Positive	Positive	Respiratory failure	M	62
7	Positive	Positive	Positive	Positive	Positive	Positive	Positive	Positive	Positive	Pneumonia	M	49

## Results

### Sensitivity, specificity and precision

Diagnostic sensitivity was calculated using RT-PCR as the gold standard. The sensitivity of Roche assay was 38.7% for specimens collected <10 days and 84.4% for specimens collected ≥10 days after the first positive RT-PCR test (Table 2). Abbott assay, on the other hand, had a sensitivity of 35.5% for specimens collected <10 days and 84.4% for specimens collected ≥10 days after the first positive RT-PCR test. The sensitivities for Euroimmun were 25.0% and 70.0% for specimens collected <10 days and ≥10 days, respectively. Specificity was 100% for all the three assays that were used in this study (95% Confidence Interval: 88.43–100%). We have also compared our Euroimmun assay results with another local laboratory performing the same assay and all results matched. For precision, all assays performed similarly with CV values <10% (Table 3).

### IgG seroconversion against SARS-CoV-2 in COVID-19 patients

Specimens collected from seven patients during hospitalization were used to evaluate the kinetics of IgG seroconversion (Table 4).

There was an overall agreement between the Abbot, Roche and Euroimmun SARS-COV-2 assays except for patient #1 at day seven and patient #4 at day 14. Seroconversion did not occur in two patients (patients # 4 and 5) at all time points except for patient #4 who converted at day 14 using Euroimmun assay only.

### Selection of plasma donors after recovery using SARS-CoV-2 IgG

The Diasorin quantitative results were in agreement with both Roche and Abbott results as qualitative tests for only 8 out of 13 positive samples for patients # 1 through 8 (Table 5). Five patients that tested positive using both Abbott and Roche assays, were found to be negative using Diasorin assay (patients # 9–13).

## Discussion

In this study, we evaluated the diagnostic sensitivity and specificity of Roche, Abbott and Euroimmun commercial assays. The sensitivity of these assays varied depending on time from first positive RT-PCR results and increased with longer periods. Sensitivity was higher in the ≥10-day group compared to <10-day group

**Table 5**

Comparison of SARS-CoV-2 serology test results for 13 plasma donors using Diasorin, Abbott and Roche assays. Diasorin is a quantitative assay while Abbott and Roche are qualitative assays.

Patient #	Diasorin assay (Arbitrary units (AU/mL))	Abbott assay (Index)	Roche assay (Signal/cut off)
1	82.7 (Positive)	5.86 (Positive)	83.46 (Positive)
2	25.6 (Positive)	3.17 (Positive)	6.05 (Positive)
3	79.3 (Positive)	8.96 (Positive)	86.44 (Positive)
4	355 (Positive)	7.42 (Positive)	38.58 (Positive)
5	>400 (Positive)	8.27 (Positive)	35.14 (Positive)
6	24.6 (Positive)	1.99 (Positive)	7.84 (Positive)
7	217 (Positive)	6.36 (Positive)	141.4 (Positive)
8	20.9 (Positive)	4.61 (Positive)	13.38 (Positive)
9	5.6 (Negative)	35.4 (Positive)	50.16 (Positive)
10	6.75 (Negative)	44.4 (Positive)	47.94 (Positive)
11	8.44 (Negative)	122 (Positive)	57.36 (Positive)
12	7.87 (Negative)	386 (Positive)	41.56 (Positive)
13	8.86 (Negative)	167 (Positive)	81.21 (Positive)

for all three assays. Sensitivity for specimens collected after day 10 was similar for both Roche and Abbott assays. The results of the Abbott assay are consistent with previous results reported by Chew et al. The authors reported sensitivity of 8.6% at  $\leq 6$  days, 43.6% at 7–13 days, 84% at 14–20 days and 84.4% at  $\geq 21$  days [11]. Better sensitivity has been reported for both Roche and Abbott assays 20 days after onset of symptoms: 97.2% for Roche assay and 92.7% for Abbott assay [12]. The specificity of the three assays measured using specimens negative for RT-PCR and specimens collected before the COVID-19 pandemic was 100%. Perkmann et al. reported specificities of 99.2% and 99.7% for Abbott and Roche respectively for specimens collected  $\geq 14$  days of the onset of symptoms [13].

We have also evaluated seroconversion by testing specimens from seven patients at days 1, 7 and 14. The timing of seroconversion was assay-dependent and occurred at day 1, 7 or 14. There was an overall agreement between the Abbott, Roche and Euroimmun SARS-CoV-2 assays except for two results. The different results in these assays can be explained by variations in the epitopes and the measured antibody (total versus IgG) in these assays.

For the evaluation of antibody levels in convalescent plasma donors, we tested 13 patients using Diasorin quantitative assay in addition to Roche and Abbott assays. The Diasorin quantitative results were in agreement with both Roche and Abbott results for only 8 out of 13 samples. All the 8 patients had fever, longer hospital stay and multiple positive RT-PCR results. The 5 discrepant results were for patients who presented with mild upper respiratory symptoms and single positive RT-PCR. The amount of antibodies produced in COVID-19 infections is variable [14]. Wu et al. reported that the majority of patients with COVID-19 develop high neutralizing antibody titers while 30% develop low titers and 5.7% had antibody levels below the threshold of the assay [14].

This study has few limitations. The use of leftover samples limited our study to a relatively small number of patients to evaluate the performance characteristics of different COVID-19 serology assays. The interrupted supply of serology kits especially in the beginning of the COVID-19 pandemic and the insufficient volume of specimens used for testing by four assays prevented us from including more patients. More studies are required to confirm the results of this study. Because sera containing antibodies against other respiratory viruses were not available, cross-reactivity with these antibodies was not evaluated for the four COVID-19 serology assays. Although the seroconversion in the majority of COVID-19 patients in this study occurred at day 14, no specimens were tested after this time.

## Conclusion

The assays evaluated in this study have different sensitivities for SARS-CoV-2 infection. Time of seroconversion is variable and assay-dependent. Not all plasma donors with previous history of COVID-19 tested positive for antibodies by all assays. More independent validation studies are required to study cross-reactivity of those assays.

## Ethical approval

This study was approved by the Cleveland Clinic Abu Dhabi Research Ethics Committee (REC Number: A-2020-063).

## Funding source

None.

## Conflict of interest

None.

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