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Evaluation of a Serological Assay to Determine the Immunoglobulin Status in SARS-CoV-2 Infected Patients Reveals a Low Performance

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ABSTRACT

Introduction: Emergence of SARS-CoV-2 has promoted the development of several serological tests for antibody detection. The external evaluation of their performance is needed before use. This study aims at assessing the performance of a Commercial lateral flow device for IgM/IgG Antibody SARS-CoV-2 detection.
Methods: One-hundred twenty-two samples SARS-CoV-2 positive by RT-PCR were used. A serological test (Ichroma2 COVID-19 Ab kit) was used as gold standard. All these samples were tested > 14 day post symptoms onset. Among the 122 samples used after testing with gold standard: 41 were IgM / IgG positive, 43 IgM positive / IgG negative and 38 IgM negative / igG positive.
Results: The commercial test evaluated shows a sensitivity and specificity of 2.4% (95% confidence interval [CI]: 0.0%-5.7%) and 100% for IgM and 10.1% (95% confidence interval [CI]: 3.5%-16.8%) and 100% for IgG.
Conclusion: Our findings demonstrate the crucial importance to evaluate performance of an assay before implementing.
Keywords: [Serological methods, Immunochromatography, performance, SARS-CoV-2, COVID-19]

INTRODUCTION

The detection of SARS-CoV-2 RNA by real time reverse transcriptase-Polymerase chain reaction (RT-PCR) in respiratory tract samples is the gold standard method for screening and diagnosis [1-3] of infection state of individuals. False negative RT-PCR tests have been so far described. Moreover, due to the fact that not all SARS-CoV-2 infected persons will develop symptoms, it

is argued that true positive cases are probably missed and the prevalence will be considerably higher than reported.

The detection of antibodies is important to acquire knowledge about the seroprevalence of SARS-CoV-2, to have insight in the global dynamic of the current COVID-19's pandemy and to help in the estimation of unreported SARS-CoV-2 infections.

The emergence of new SARS-CoV-2 has promoted the development of several SARS-CoV-2 serological tests for antibody detection [4]. The assessing the performances of available tests is required before their introduction in a country. Thus, the objective of this study was to evaluate the performance of a Commercial lateral flow device for IgM/IgG Antibody SARS-CoV-2 detection.

MATERIALS & METHODS

Specimens collection

One hundred twenty two plasma samples collected between June 2020 and September 2020 from symptomatic individuals with PCR diagnosed infections with SARS-CoV-2 were used for this study. These samples were collected 14 days after the PCR detection. All these patients (62,5% of women, mean age was 41,6 years) were recruited to the "Unité Mixte de Recherche CIRMF-SSM" and before sampling; they have completed form to give their consent. For each patient, blood was been collected in an EDTA tube of 1,5mL then centrifuged at 3000 g during 10 minutes and immediately after sampling stored at 20°C.

Serological assay

IgM/IgG Antibody to Coronavirus (SARS-CoV-2) manufacturer by Zhuhai Livzon Diagnostics Inc is an immune colloidal lateral flow technique intended for the qualitative detection of IgG and IgM antibodies against the SARS-CoV-2 in human serum, plasma and whole blood samples. This test detects IgM and IgG SARS-CoV-2 antibodies separately. Samples were tested according to the manufacturer's instructions. Serological testing was performed in «Unité Mixte de Recherche VIH et Maladies infectieuses associées de l'Hôpital d'Instruction des Armées Omar Bongo Ondimba de Libreville (Gabon) ». Briefly, 10 µL of specimen was added onto the sample loading area followed by two drops of sample diluent. The results were read and interpreted 15 min after testing. In the presence of a control signal, any band, even weakly visible, located in the IgM and/or IgG position is considered positive. If the control line does not appear red, the test is invalid, and the test should be repeated with another cartridge.

Gold Standard

An *in vitro* diagnostic test system based on lateral flow sandwich detection immunofluorescence technology targeting anti-SARS-CoV-2 IgM and IgG antibodies (Ichroma2 COVID-19 Ab kits and Ichroma II Reader from Boditech Med Inc, South Korea) was used as gold standard. This test has a sensitivity of 95.8% and a specificity of 97% [5]. Among these 122 patients tested with gold standard: 41 were IgM / IgG positive, 43 IgM positive / IgG negative and 38 IgM negative / igG positive.

In total, for assess Livzon antibody test we used 84 IgM positive samples (41 IgM / IgG positive and 43 IgM positive / IgG negative), 38 IgM negative samples (IgM negative/IgG positive), 79 IgG positive samples (41 were IgM / IgG positive and 38 IgM negative / IgG positive) and 43 IgG negative samples (43 IgM positive / IgG negative) with Ichroma2 test.

STATISTICAL ANALYSIS

[Sensitivity (true positives/true positives + false negatives), specificity (true negatives / true negatives + false positives), positive predictive value (PPV, true positives / true positives + false positives) and negative predictive value (NPV, true negatives / true negatives) + false negatives) was calculated using MedCalc software for evaluate the performance of the test. The results were presented with IC.

RESULT

Of the 84 sera samples from IgM positive patients with Ichroma2, only 2 tested positive with Livzon test resulting a sensitivity of 2.4% [95% CI: 0.3-8.3]. All the 38 IgM negative serums with Ichroma2 were tested negative with the Livzon test generating a specificity of 100% [95% CI: 90.8-100]. The PPV and NPV for IgM were 100% and 31.7% [95% CI: 31-32.4] respectively for the IgM detection.

Among the 79 samples from IgG positive patients with Ichroma2, 8 were tested positive with the Livzon test resulting in a sensitivity of 10.1% [95% CI: 4.5-19] (Table 1). All the 43 IgG negative sera were tested negative generating a specificity of 100% [95% CI: 91.2-100]. The VPP and VPN for IgG detection were 100% and 37.7% [95% CI: 36.0-39.5] respectively.

Table 1. Performance of the Livzon IgM/IgG Antibody to Coronavirus (SARS-CoV-2) detection

	Ichroma results			
Livzon test results	**Positive (IgM) N (%)**	**Negative (IgM) N (%)**	**Positive (IgG) N (%)**	**Negative (IgG) N (%)**
Positive, N (%)	2 (97.6)	0 (0.0)	8 (10.1)	0 (0.0)
Negative, N (%)	82 (2.4)	38 (100)	71 (89.9)	43 (100)

DISCUSSION

The emergence of SARS-CoV-2 infection has conducted to the development of many serological assays for SARS-CoV-2, assessment of their analytical performance by using clinical specimens is of critical importance [6-8]. In our study, we have evaluated the performance of a serological test manufactured by Zhuhai Livzon Diagnostics Inc, the IgM/IgG Antibody to Coronavirus (SARS-CoV-2) test. Our findings reveal a low sensitivity for both IgM and IgG antibody, 2.4% and 10.1% respectively and a high specificity for the two antibodies (100%). There is great heterogeneity in terms of sensitivity and specificity in published studies on serological testing in general. However, concerning the Livzon IgM/IgG Antibody to Coronavirus (SARS-CoV-2) test, two countries, Argentina and Denmark, have already reported an abnormal low sensitivity [9-10]. Low sensitivity of a test may falsely conduct to assuming that a person has not infected or has never been infected. It will compromise the accuracy of seroepidemiological studies in SARS-CoV-2 infection and underestimate the spread of the virus among the population.

Some studies have evaluated the performance of serological tests using RT PCR as gold standard rather than other serological test or the time from symptoms onset inducing a risk of misestimating sensitivity and may decrease the real number of patients infected by SARS-CoV-2 due to false negative results [4,11]. However in our study, in addition to PCR result, a serological test was used as gold standard.

CONCLUSION

Overall, our data demonstrate a poor performance of the IgM/IgG Antibody to Coronavirus (SARS-CoV-2) manufactured by Zhuhai Livzon Diagnostics Inc test. These findings reinforce the need to evaluate carefully a test before it uses on a national scale. Utilisation of high quality of serological test will be an important component for epidemiological study based on the detection of SARS-CoV-2 antibodies.

Declaration by Authors

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REFERENCES

1. Beatriz Araujo Oliveira, Lea Campos de Oliveira, Ester Cerdeira Sabino, et al. SARS-CoV-2 and the COVID-19 disease: a

mini review on diagnostic methods. Journal of the Sao Paulo Institute of Tropical Medicine. 2020; 62: 1-8
2. Wang M, Wu Q, Xu W, Qiao B, Wang J, Zheng H, et al. Clinical diagnosis of 8274 samples with 2019-novel coronavirus in Wuhan. medRxiv. 2020; In Press.
3. Singh A, Shaikh A, Singh R, Singh AK. COVID-19: from bench to bed side. Diabetes Metab Syndr. 2020 ; 14 : 277-81.
4. Nicol T, Lefeuvre C, Serri O, et al. Assessment of SARS-CoV-2 serological tests for the diagnosis of COVID-19 through the evaluation of three immunoassays: Two automated immunoassays (Euroimmun and Abbott) and one rapid lateral flow immunoassay (NG Biotech). J Clin Virol. 2020; 129: 104511. doi: 10.1016/j.jcv.2020.104511
5. Pinto Borges L, Fagundes Martins A, Santos de Melo M, et al., Seroprevalence of SARS-CoV-2 IgM and IgG antibodies in an asymptomatic population in Sergipe, Brazil. Rev Panam Salud Publica.2020; 44 (108).
6. Jääskeläinen A, Kuivanen S, Kekäläinen E et al. Performance of six SARS-CoV-2 immunoassays in comparison with microneutralisation. J Clin Virol. 2020; 129 : 104512. doi: 10.1016/j.jcv.2020.104512.
7. A. Petherick, Developing antibody tests for SARS-CoV-2. Lancet.2020 ; 395: 1101-1102. doi: https://doi.org/10.1016/S0140-6736(20)30788-1
8. Tuaillona E, Bolloré K, Pisoni A, et al. Detection of SARS-CoV-2 antibodies using commercial assays and seroconversion patterns in hospitalized patients. Journal of Infection. 2020; 81: 39-45. doi: 10.1016/j.jinf.2020.05.077.
9. Digital Military Medecine. Argentina: COVID-19 Tests Bought from China Present Problems. https://dialogo-americas.com/articles/argentina-covid-19-tests-bought-from-china-present-problems/, 2020 (accessed 25 June 2020)
10. Reuters: Denmark to send back inaccurate antibody tests from China's Livzon. https://www.reuters.com/article/us-health-coronavirus-denmark-kits-idUSKBN22W2TC , 2020 (accessed 25 June 2020)
11. Mayara L-B, Gamuchirai T, Tavazivasyed K-A, et al. Diagnostic accuracy of serological tests for covid-19: systematic review and meta-analysis. BMJ. 2020; 370 (2616). doi: 10.1136/bmj.m2516

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