Updated evaluation of diagnostic performance of RayBiotech COVID19 lateral flow serum IgM/IgG test kit with sample exclusion

RayBiotech Life, Inc.

April 24, 2020

Introduction

Study objective

Evaluation of the diagnostic performance of RayBiotech COVID19 lateral flow serum IgM/IgG test kit in subjects with Sars-COV-2 PCR test results, and disease-free subjects with clear free-of-exposure evidence to the Sars-COV-2 virus, or showing no symptoms of COVID19 in 3 weeks or more after sample collection.

Investigated product

Raybtiotech COVID19 lateral flow serum IgM/IgG test kit.

Study design

Blinded and/or controlled diagnostic test study with parallel reference test.

Study subjects

The study subjects including:

1) COVID19-affected patients with diagnosis based on Sars-cov-2 PCR test and clinical symptoms.
2) COVID19-free subjects confirmed by Sars-cov-2 PCR test at the timepoint of sample collection, or showing no COVID19 symptoms 3 or more weeks after sample collection (concurrent samples).
3) COVID19-free subjects of whom the serum samples were collected at least 3 months before the outbreak of COVID19 (retrospective samples, pre October 2019). These sample were previously acquired and in bio-sample reservoir in RayBiotech facilities.

The subjects in this study are from the USA and China.

Inclusion and exclusion criteria

Inclusion criteria

1) Sufficient serum of quality
2) Solid diagnosis of COVID19 (Sars-Cov-2 test, clinical symptoms, and free of Sars-Cov-2 exposure)

Exlusion Criteria

1) Insufficient serum of quality

Endpoint of the study

The endpoints of this study were defined as sensitivity, specificity, and accuracy of the product against the reference standard (Sars-COV-2 PCR test, free-of-exposure, and free-of-COVID19 symptom in 3 weeks or more after sample collection).

Reference standards

The reference standard was established as fulfillment of one of three criteria as following:

1) Sars-Cov-2 virus PCR test on nasopharyngeal swab specimens collected at the same timepoint or known timepoint close to the serum samples under investigation.

2) Free-of-exposure to Sars-Cov-2 virus. The outbreak of COVID19 caused by Sars-Cov-2, or novel coronavirus, were observed at the first time in late 2019. The subjects from whom the retrospective serum samples were collected at least 3 months before the outbreak of COVID19, had no reason to be exposed to the virus, and can be considered as disease-free subjects.

3) The subjects were free-of-symptoms in 3 weeks or more after sample collection. The COVID19 has a latent period of 4-21 days. A subject showing no COVID19 symptoms in 3 weeks or more after the sample collection, can be considered as disease-free safely at the timepoint of sample collection.

Primary endpoints

The sensitivity, specificity and accuracy of 'combined IgM and IgG' test against the reference standard. It is well known that IgM appears as the initial responding isotype in serum but fades rather quickly after initial onset, while the IgG isotype response occurs later in a stronger and more long term manner into and through recovery. Accordingly the 'combined' result, i.e., a subject was considered as positive when either of IgM and IgG was positive, and as negative when both IgM and IgG were negative, were defined as the primary outcome in the study. Overall both tests are recommended to be performed unless the timeline of infection is known prior to testing.

Secondary endpoints

There are two secondary endpoints in this study.

1) The sensitivity, specificity and accuracy of the IgM test only in subjects against the reference standard.

2) The sensitivity, specificity and accuracy of the IgG test only in subjects against the reference standard.

Study workflow

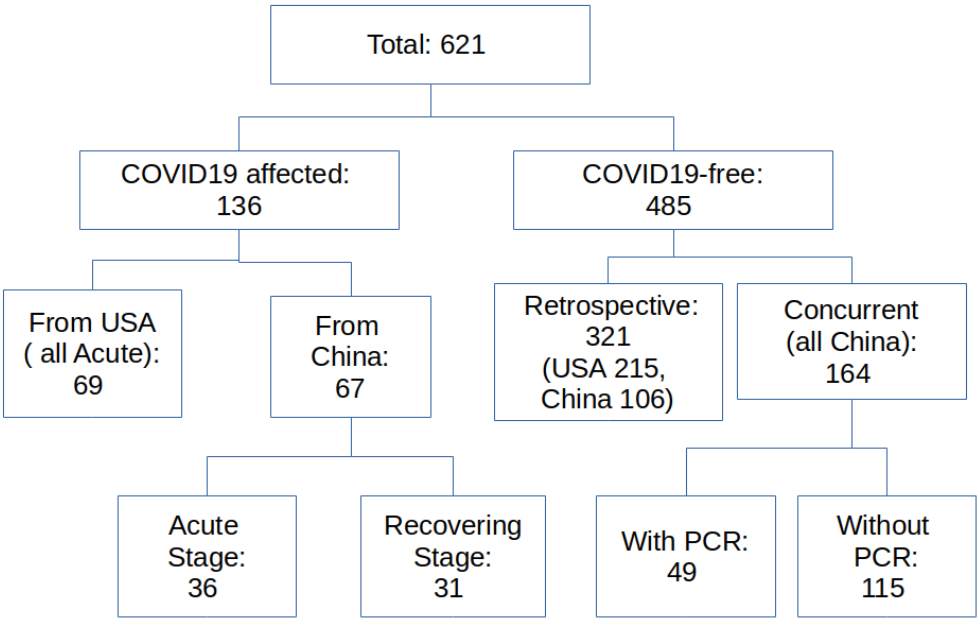
Statistical analysis

Study population

621 subjects were enrolled into this study, including 136 COVID19 cases (21.9%) confirmed by Sars-Cov-2 PCR test. 69 among the COVID19 cases came from USA, while the other 67 were enrolled in China. 31 COVID19-affected serum samples were collected during recovering stage from the China patients.

321 among 485 COVID19-free samples were retrospective, i.e., retrieved from biological samples reservoir in facilities of RayBiotech, which were collected at least 3 moths before the outbreak of COVID19. The other 164 COVID19-free samples were collected from China concurrently, w/wo Sars-Cov-2 PCR test results(49 and 115, respectively).

66 COVID19-free subjects were retrospectively tested that were confirmed for other virus infections (RSV, HBV, HCV, etc) or auto-immunity disease (ANA+).



Analysis in overall study subjects

Overall 136 COVID19 cases and 485 COVID19-free subjects were enrolled into this study. The IgM in serum were tested in all of the 621 subjects using the product under investigation, while IgG in serum being tested in 608 subjects. Accordingly the 'combined IgM/IgG' were evaluated in only 608 subjects.

Primary endpoints in overall study population

The sensitivity of IgM and IgG tests combined against reference standard in overall study population is 90.44% (123/136, 95%CI: 84.21-94.81%).

The specificity of IgM and IgG tests combined against reference standard in overall study population is 97.25% (459/472, 95%CI: 95.34-98.53%).

The accuracy of IgM and IgG tests combined against reference standard in overall study population is 95.72% (582/608, 95%CI: 93.8-97.19%), with Kappa value of 0.8769.

Performance of IgM and IgG tests combined in overall study population

combined	PCR/other COVID19	PCR/other COVID19-free	Total
Raybio Test +	123	13	136
Raybio Test -	13	459	472
Total	136	472	608

Secondary endpoints in overall study population

Performance of IgM only in overall study population

The sensitivity of IgM against reference standard in overall study population is 61.03% (83/136, 95%CI: 52.3-69.27%).

The specificity of IgM against reference standard in overall study population is 97.32% (472/485, 95%CI: 95.46-98.57%).

The accuracy of IgM against reference standard in overall study population is 89.37% (555/621, 95%CI: 86.68-91.68%), with Kappa value of 0.6525.

Performance of IgM in overall study population

IgM	PCR/other COVID19	PCR/other COVID19-free	Total
Raybio Test +	83	13	96
Raybio Test -	53	472	525
Total	136	485	621

Performance of IgG only in overall study population

The sensitivity of IgG against reference standard in overall study population is 63.24% (86/136, 95%CI: 54.55-71.33%).

The specificity of IgG against reference standard in overall study population is 100% (472/472, 95%CI: 99.22-100%).

The accuracy of IgG against reference standard in overall study population is 91.78% (558/608, 95%CI: 89.3-93.83%), with Kappa value of 0.7276.

Performance of IgG in overall study population

IgG	PCR/other COVID19	PCR/other COVID19-free	Total
Raybio Test +	86	0	86
Raybio Test -	50	472	522
Total	136	472	608