Clinical Validation Report of

Tigsun COVID-19 Saliva Antigen Rapid Test

For in vitro diagnostic use only. For professional use only.

1. Introduction

Since December 2019, continuous surveillance on influenza and relevant diseases had been carried out, and several patients with viral pneumonia had been found and diagnosed with viral pneumonia/pulmonary infection. Relevant viruses were typed for detection. On January 7, 2020, the laboratory detected a novel coronavirus. The "2019 novel coronavirus (2019-nCoV)" was identified in the viral pneumonia cases and named by the World Health Organization (WHO) on January 12, 2020. The 2019-nCoV infected cases typically have symptoms like fever, fatigue, dry cough as the main respiratory syndrome, and gradually develop dyspnea. Severe patients presented with acute respiratory distress syndrome, septic shock, refractory metabolic acidosis, and coagulation dysfunction. Some patients had mild onset symptoms, but no fever. Most patients had good prognosis, while a few patients were critically ill and even died.

Within days of the full SARS-CoV-2 genome being made available, PCR assay protocols were posted on the World Health Organization (WHO) website from partner laboratories. Diagnostic test manufacturers have responded rapidly to the needs of countries, and over 700 products have been released onto the market to detect SARS-CoV-2 specific nucleic acids, antigens (proteins) and human antibodies. In settings where RT-PCR is unavailable or turnaround times for results are slow (e.g., several days to weeks), rapid antigen detecting tests may facilitate earlier diagnosis and required actions.

2. Purpose of Research

Tigsun COVID-19 Saliva Antigen Rapid Test is an lateral flow immunochromatographic assay intended for the rapid and qualitative detection of antigen from severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in

saliva samples from individuals suspected of COVID-19 by their healthcare provider within the first seven days of symptom onset. To validate its clinical effectiveness, according to the requirements of the Technical Guidance for Clinical Trials of In Vitro Diagnostic Reagents and Key Points for Technical Review of the Registration of 2019 -nCoV Antigen/Antibody Detection Reagents (Interim), a comparative study was carried out between the Beijing Tigsun reagent and the available clinical diagnostic results, and the relevant test data were obtained and statistically analyzed.

3. Clinical Trial Design

3.1. Trail Objectives

Following the *Provisions for In Vitro Diagnostic Reagent Registration*, *Technical Guidance for Clinical Trials of In Vitro Diagnostic Reagents* and *Key Points for Technical Review of the Registration of 2019- nCoV Antigen/Antibody Detection Reagents (Interim)*, comparative analysis was performed for the data and results, to evaluate the clinical performance of the test reagent. Test results were analyzed, and statistical results were obtained. Finally, the clinical report was completed.

The enrolled specimens were from patients suspected with 2019-nCoV pneumonia. The test results of the test kit were compared with 2019-nCOV PCR reference product results, so as to evaluate the clinical performance of the kit to be tested.

3.2. Administrative Information

1. Instruction supplied with the reagents must be followed exactly unless explicitly stated.
2. The assays during the period of the evaluation should be double checked.
3. The results for both Novel Coronavirus (2019-nCoV) Antigen and any reference assay methods must be properly identified.

3.3. Materials

3.3.1. Target reagent:

Tigsun COVID-19 Saliva Antigen Rapid Test produced based on technological process and the outputted Standard Operating Procedure.

3.3.2. Reference reagent:

2019-nCoV PCR test

3.3.3. Specimens to be tested

1) Specimen types

Nasopharyngeal swab and saliva should be used.

2) Specimen quantity

Specimens above should be collected from known PCR positive and known PCR negative patients' samples.

3.4. Statistical Methods

The products launched on the market shall be subject to comparative study and evaluation Kappa inspection: each sample shall be tested with the product tested and the reference product respectively, and then the consistency in statistical results of these two inspection methods shall be compared through Kappa inspection. The data shall be subject to Kappa inspection and analysis and the Kappa coefficient shall be calculated. Favorable consistency can be proven if Kappa is greater than 80%. The consistency in test results of the product tested and the reference product is evaluated as per the evaluation standards.

The coincidence rate shall be calculated by comparing with the reference product whose marketing is approved.

1) Coincidence rate of negative: the sample whose test results are negative for both the product tested and the reference product.

2) Coincidence rate of positive: the sample whose test results are positive for both the product tested and the reference product.

3) Total coincidence rate: the sample whose test results are the same for the product tested and the reference product.

Test System (Antigen)	Reference System (PCR)		Total
	Positive (+)	Negative (-)	
Positive (+)	(A)	(B)	(A+B)
Negative (-)	(C)	(D)	(C+D)
Total	(A+C)	(B+D)	(A+B+C+D)

In general, the formula calculating the coincidence rate of positive/negative is:

Coincidence rate of positive =A/(A+C) *100%

Coincidence rate of negative =D/(B+D) *100%

Total coincidence rate=(A+D)/(A+B+C+D) *100%

95%CI is calculated by approximate normal distribution method or Wilson score interval.

Kappa consistency analysis shall be adopted for statistical analysis of similar reference kits. If conducting Kappa consistency analysis for the base data above, high consistency can be judged if the Kappa coefficient is >0.8, and both systems

are considered as equivalent. The results of the product tested are statistical materials and can be analyzed as per the table below:

$$\text{Kappa}=\frac{2(ad-bc)}{(a+b)(b+d)+(a+c)(c+d)}$$

4. Clinical Study Results and Analysis

4.1 Sample Inclusion

In this study, 465 cases were included, of which 128 cases were confirmed by 2019-nCOV PCR result and 337 cases were negative by 2019-nCOV PCR result. Swab and saliva specimens collected according to proper procedure.

4.2 Analysis of Results

A total of 465 samples had nucleic acid test results in this clinical trial. The nucleic acid test results were compared and analyzed as shown in the following table:

Comparative Analysis of Saliva Antigen Test with PCR Result

		PCR Assay		Total
		Positive	Negative	
Antigen Test	Positive	120	1	121
	Negative	8	336	344
	Total	128	337	465
		Result	95% LCI	95% UCI
PPA-Sensitivity		93.75%	88.15%	96.80%
NPA-Specificity		99.70%	98.34%	99.95%
OPA-Accuracy		98.06%	96.36%	98.98%

Kappa Analysis:

P_0 is 0.98 and P_e is 0.61. This give Kappa value equals to 0.95, which is greater than 0.8.

5. Conclusion

The clinical performance of the Tigsun COVID-19 Antigen Rapid Test researched and developed by Beijing Tigsun Diagnostics Co., Ltd. was investigated.

In this study, a total of 465 cases were included for statistical analysis. The number of confirmed cases and COVID-19 negative cases was 128 and 337, respectively. The consistency of the test reagent results with the reference product results was analyzed.

By analyzing the test results, the consistency percentage of negative/positive and the total consistency percentage are high. Results showed a good consistency between Tigsun COVID-19 Saliva Antigen Rapid Test and PCR results. In summary, this clinical study showed that Tigsun COVID-19 Saliva Antigen Rapid Test researched and developed by Beijing Tigsun Diagnostics Co., Ltd. is of great value in clinical application for meeting clinical test requirements with good performance.

Version: 1417-1.6

Manufacturer

Beijing Tigsun Diagnostics Co.,Ltd.

Add: No. 16 Guba Road, District #1, Chengguan Street, Fangshan District, Beijing, China,102400

Tel : +86-10-89350910 400-0666-093 (Toll Free)

Fax: +86-10-89350980

E-mail: world.sales@tigsun.com

Website: www.tigsun.com/en

Linkedin: www.linkedin.com/company/tigsun follow us to get latest updates