Novel Coronavirus Spike Glycoprotein Detection Kit

(Ligand-receptor Competitive Chromatography)

Pre-Clinical Study Report

Name of in vitro diagnostic reagents used in the test: Novel Coronavirus Spike Glycoprotein Detection Kit (Ligand-receptor Competitive Chromatography)

Specifications: 25 Tests/Box

Start and end time of the test: April 23th, 2020- April 30th, 2020

Clinical location: Center for Disease Control and Prevention

Applicant: New Gene (Hangzhou) Bioengineering Co., Ltd.

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Summary

The Novel Coronavirus Spike Glycoprotein Detection Kit (Ligand-receptor Competitive Chromatography) developed by New Gene (Hangzhou) Bioengineering Co., Ltd. can quickly and qualitatively detect the spike glycoprotein of novel coronavirus (SARS-COV-2) in human sputum/stool samples. It can be used as a supplementary test for COVID-19 diagnosis.

According to the clinical trial plan, the Novel Coronavirus Spike Glycoprotein Detection Kit (Ligand-receptor Competitive Chromatography) or test reagent , is to test sputum/stool samples from healthy subjects and confirmed COVID-19 patients. Test results are compared with another commercial SARS-COV-2 nucleic acid detection kit with CFDA approval, which is defined as the gold standard . The sensitivity, specificity, and total agreement rate are used to evaluate the feasibility of the test reagent in clinical applications.

Method: A collection of clinical samples are examined by the test reagent and the gold standard in parallel, to calculate the clinical sensitivity, clinical specificity, and total agreement rate of the test reagent.

Standard of criteria for a qualified test reagent: C[illegible] 0%, and total agreement [illegible] 85%.

Results: Compared to the gold standard, the clinical sensitivity of test reagent reached 90.0%, the clinical specificity reached 95.0%, and the total coincidence rate reached 93.3%.

Conclusion: The performance of test reagent has a high agreement rate with the gold standard, proving its good feasibility in diagnosing suspected COVID-19 cases.

Acronyms

Test reagent: The Novel Coronavirus Spike Glycoprotein Detection Kit (Ligand-receptor Competitive Chromatography) developed by New Gene (Hangzhou) Bioengineering Co., Ltd.

SARS-COV-2: Novel Corona Virus 2019

Main contents

Introduction

The novel coronavirus SARS-COV-2 is the causative pathogen for the global pandemic of COVID-19. It is contagious in humans, either symptomatically or asymptomatically. Based on current epidemic knowledge, the asymptomatic infection may last for 1 day to 14 days, mainly 3 days to 7 days. Symptoms of COVID-19 include fever, fatigue, and cough. Some patients also complains about nasal obstruction, runny nose, score throat, muscle aches, and diarrhea.

In response to the emergent market needs, New Gene (Hangzhou) Bioengineering Co., Ltd. has developed the Novel Coronavirus Spike Glycoprotein Detection Kit (Ligand-receptor Competitive Chromatography). This product utilizes *in vitro* expressed human angiotensin I converting enzyme 2 (ACE2) protein to capture and visualize viral particles in test samples. Since ACE2 is the cellular receptor of SARS-COV-2 spike glycoprotein, it plays an indispensable role in the disease infection and

progression. Through various virus sub-types have emerged during the global pandemic, this product can reliably detect all contagious SARS-COV-2 sub-types, as long as they bind to ACE2 as the invasion target.

Production of the Novel Coronavirus Spike Glycoprotein Detection Kit (Ligand-receptor Competitive Chromatography) is implemented in Class 100,000 cleanrooms, by proficient operators. Multiple quality control processes are included in the manufacture procedures to examine the quality of raw materials, semi-finished products, and finished products. The construction of cleanrooms, personnel training, and manufacture practices are implemented under relevant laws and regulations.

To evaluate the clinical performance of the Novel Coronavirus Spike Glycoprotein Detection Kit (Ligand-receptor Competitive Chromatography), the current clinical trial is jointly carried out by the applicant and the Center for Disease Control and Prevention. The applicant is responsible for providing test reagents and training relevant personnel with the operating procedures and technical principles to minimize operational bias. The Center for Disease Control and Prevention is responsible for the collection and storage of clinical trial samples, the implementation of clinical trials, the compilation of clinical trial records, and the writing of clinical trial reports.

Trial objective

The objective of current trial is to evaluate the feasibility of test reagent in clinical applications. Data from test reagent are compared to those from a CFDA approved commercial SARS-COV-2 nucleic acid detection reagent, which is referred as the gold standard , to estimate the sensitive, specificity, and total agreement rate of test reagent.

Trail management

The Current clinical trial is completed in diagnosis laboratories managed by the Center for Disease Control and Prevention. Participants include a project leader, and professionally trained technical personnel. The clinical trial is carried out strictly in accordance with regulations in the "Principle Guidelines for Clinical Trail of In Vitro Diagnostic Reagents".

Trail design

Clinical samples for the current trial are collected by the Center for Disease Control and Prevention. Each sample is tested by both the test reagent and gold standard reagent. The clinical sensitivity, clinical specificity and total agreement rate of test reagent are calculated based on the test results.

Results and analysis

Determining the sample size.

With reference to relevant regulations, and considering the uncertainty of obtaining samples, the number of samples for this clinical trial shall be no less than 60, of which the number of positive samples shall not be less than 30.

Criteria for sample inclusion and exclusion .

Clinical samples for the present trial are all human sputum samples or stool samples, collected by the Center for Disease Control and Prevention.

Sample collection, storage, and transportation.

Clinical samples are collected from COVID-19 suspects, and preserved in virus preservation solution. Keep the solution frozen at -15°C~-25°C until used.

The gold standard reagent

Nucleic acid testing is currently the "gold standard" for COVID-19 diagnosis. A CFDA approved nucleic acid test reagent, namely the Novel Coronavirus (SARS-COV-2) Real Time Multiplex RT-PCR Kit produced by Shanghai ZJ Bio-Tech Co., Ltd. is chosen as the gold stardard reagent. It targets the ORF1ab gene, N gene and E gene of the SARS-COV-2, and is used as an auxiliary diagnosis and emergency reserve reagent for COVID-19.

Information of test reagent and the "gold standard" reagent.

Test reagent	Novel Coronavirus Spike Glycoprotein Detection Kit (Ligand-receptor Competitive Chromatography)		
Specification	25 Tests/Box	Lot:	200401
Period of Validity	2 years	Storage:	2°C~30°C
Manufacturer	New Gene (Hangzhou) Bioengineering Co., Ltd.		

Gold Standard reagent	Novel Coronavirus (SARS-COV-2) Real Time Multiplex RT-PCR Kit		
Approval Number	CFDA NO:20203400057		
Specification	50 Tests/Box		
Period of Validity	Six month	Storage:	Store at -20±5°C, keep away from light
Manufacturer	Shanghai ZJ Bio-Tech Co., Ltd.		

Quality control methods

The clinical trial is strictly implemented in accordance with the corresponding instruction manual.

Statistical analysis method of clinical trial data

		Gold standard reagent		Total
		Positive	Negative	
Test reagent	Positive	a	b	a + b

	Negative	c	d	c + d
Total		a + c	b + d	a + b + c + d

Clinical sensitivity (%) = [a / (a + c)] ×100%

Clinical specificity (%) = [d / (b + d)] ×100%

Total coincidence rate (%) = [(a + d) / (a + b + c + d)] ×100%

Clinical trial results and analysis

Trail Results

A collection of 120 samples were tested with test reagents; including 60 sputum samples, and 60 stool samples.

Test results are as follows:

		Gold standard reagent		Total
		Positive	Negative	
Test reagent	Positive	36	4	40
	Negative	4	76	80
Total		40	80	120

Result analysis

A total of 120 samples are tested in this trial. Both the test reagent and gold standard reagent find out 40 positive results, of which 36 samples are reported positive by both reagents. Four samples are reported positive only in test reagent, and another 4 samples are reported positive only in gold standard reagent. The other 76 samples are reported negative by both reagents.

Clinical sensitivity (%) = [36 / (36 + 4)] ×100% = 90.0%

Clinical specificity (%) = [76 / (4 + 76)] ×100% = 95.0%

Total agreement rate (%) = [(36 + 76) / (36 + 4 + 4 + 76)] ×100% = 93.3%

Discussion and conclusion

The Novel Coronavirus Spike Glycoprotein Detection Kit (Ligand-receptor Competitive Chromatography) developed by New Gene (Hangzhou) Bioengineering Co., Ltd. can quickly and qualitatively detect SARS-COV-2 in human sputum/stool samples. It can be used as a supplementary test for COVID-19 diagnosis.

In this clinic trial, the Novel Coronavirus (SARS-COV-2) Real Time Multiplex RT-PCR Kit produced by Shanghai ZJ Bio-Tech Co., Ltd., a commercial SARS-COV-2 kit approved by CFDA, is used as the "gold standard" reagent. In a collection of 120 clinical samples examined in this trial, the test reagent have shown clinical sensitivity of 90.0%, clinical specificity of 95.0%, and a total agreement rate of 93.3%.

In summary, the overall agreement rate between the test reagent and the gold standard reagent is relatively high, and the test reagent can be used clinically for the diagnosis of suspected cases of SARS-COV-2.