Liferiver

Revision No.: ZJ0010R
Issue Date: May 9th, 2020

Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit (Detection for 3 Genes) Instructions for Use

REF RR-0479-02

For use with ABI Prism®7500/7900; Bio-Rad CFX96; Rotor Gene™6000; SLAN; MIC POC Dx48 Instrument

EC REP Obelis s.a.
Boulevard Général Wahis 53
1030 Brussels, Belgium
Tel: +(32) 2.732.59.54
Fax: +(32) 2.732.60.03
E-Mail : mail@obelis.net

Shanghai ZJ Bio-Tech Co., Ltd.
Building #26, 588 Xinjunhuan Road,
Pujiang High-tech Park, 201114 Shanghai, China
www.liferiverbiotech.com Tel: +86-21-34680598
info@liferiverbiotech.com Fax: +86-21-34680595

1. Intended Use

Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit (Detection for 3 Genes) is used for the in vitro qualitative detection of 2019 novel coronavirus (2019-nCoV) RNA in upper respiratory tract specimens (nasopharyngeal and oropharyngeal extracts) and lower respiratory tract specimens (bronchoalveolar lavage fluid (BALF) and deep cough sputum) by real time PCR systems. It is considered as an aid in the diagnosis of the 2019-nCoV infection.

2. Principle of Real-Time RT-PCR

The principle of the real-time detection is based on the fluorogenic 5'nuclease assay. During the PCR reaction, the DNA polymerase cleaves the probe at the 5' end and separates the reporter dye from the quencher dye only when the probe hybridizes to the target DNA. This cleavage results in the fluorescent signal generated by the cleaved reporter dye, which is monitored real-time by the PCR detection system. The PCR cycle at which an increase in the fluorescence signal is detected initially (Ct) is proportional to the amount of the specific PCR product. Monitoring the fluorescence intensities in real time allows the detection of the accumulating product without having to re-open the reaction tube after the amplification.

Real time reverse-transcription polymerase chain reaction (real-time RT-PCR) is used when the starting material is RNA. In this method, RNA is first transcribed into the complementary DNA (cDNA) by reverse transcriptase from total RNA. The cDNA is then used as a template for the real time PCR.

3. Product Description

Coronaviruses are a large family of viruses, some causing illness in human and others circulating among animals such as camels, cats and bats. 2019-nCoV is a novel coronavirus. The primer and probe design for this kit is based on the newly released strain (2019-nCoV) (GeneBank accession: MN908947) and covers six 2019-nCoV strains sequences (EPI_ISL_402119, EPI_ISL_402120, EPI_ISL_402121, EPI_ISL_402122, EPI_ISL_402123 and EPI_ISL_402124).

The kit contains a specific ready-to-use system for the detection of Novel Coronavirus (2019-nCoV) RNA by the real-time RT-PCR. The reaction is done in a one-step real time RT-PCR assay in a single tube. It includes a reverse transcription (RT) for the transcription of virus RNA into cDNA and real time PCR for the amplification and detection of the cDNA. Fluorescence is emitted and measured by the real time systems´optical unit during PCR. The detection of amplified virus DNA fragment is performed in fluorimeter **channel FAM, HEX/VIC/JOE** and **Cal Red 610/ ROX/TEXAS RED**.

4. Kit Contents

Ref.	Type of Reagent	Presentation 25rxns
1	Novel CoV (2019-nCoV) Super Mix	1 vial, 513μL
2	RT-PCR Enzyme Mix	1 vial, 27μL
3	Novel CoV (2019-nCoV) Internal Control	1 vial, 30μL
4	Novel CoV (2019-nCoV) Negative Control	1 vial, 400μL
5	Novel CoV (2019-nCoV) Positive Control	1 vial, 30μL

Analytical sensitivity: 1×10^3 copies/mL;

Note: Analytical sensitivity depends on the sample volume, elution volume, nucleic acid extraction method and other factors. If you use the RNA extraction kits recommended, the analysis sensitivity is the same as it declares. However, when the sample volume is dozens or even hundreds of times greater than elution volume by some concentrating method, the sensitivity can be much higher.

5. Storage

- All reagents should be stored at -20±5℃.
- All reagents can be used till the expiration date indicated on the kit label.
- Repeated thawing and freezing (> 3x) should be avoided as this may reduce the sensitivity of the assay.
- Cool all reagents during the working steps.
- Super Mix should be stored away from light.

6. Additionally Required Materials and Devices

- Biological cabinet
- Vortex mixer
- Cryo-container
- Sterile filter tips for micro pipets
- Disposable gloves, powderless
- Refrigerator and freezer
- Real time PCR system
- Real time PCR reaction tubes/plates
- Pipets (0.5μL – 1000μL)
- Sterile microtubes
- Biohazard waste container
- Tube racks
- Desktop microcentrifuge for "eppendorf" type tubes (RCF max. 16,000 x g)

7. ⚠Warnings and Precautions

Carefully read this instructions for use before starting the procedure.

- This assay needs to be carried out by skilled personnel.
- Clinical samples should be regarded as potentially infectious materials and be prepared in a laminar flow hood.
- This assay needs to be run according to Good Laboratory Practice.
- Do not use the kit after its expiration date.
- Avoid repeated thawing and freezing of reagents as this may reduce the sensitivity of the test.
- Once the reagents have been thawed, vortex and centrifuge briefly the tubes before use.
- Prepare quickly the reaction mix on ice or in the cooling block.
- Set up separate working areas for: 1) Reaction setup, 2) Isolation of the RNA and 3) Amplification/detection of amplification products.
- Pipets, vials and other working materials should not circulate among working units.
- Use always sterile pipette tips with filters.
- Wear separate coats and gloves in each area.
- Discard sample and assay waste according to your local safety regulations.
- Do not pipette by mouth. Do not eat, drink or smoke in laboratory.
- Avoid aerosols

8. Sample Collection, Storage and Transport

- Collect samples in sterile tubes;
- Specimens can be extracted immediately or stored at 2°C~8°C within 24 hours or frozen at -70℃ for long-term.
- Transportation of clinical specimens must comply with local regulations for the transport of etiologic agents

9. Procedure

9.1 RNA-Extraction

Different brand RNA extraction kits are available. You may use your own extraction systems or the commercial kits based on the yield. For the RNA extraction, please follow the manufacturer's instructions. The recommended extraction kits are as follows:

Nucleic Acid Isolation Kit	Cat. Number	Manufacturer
RNA Isolation Kit (Paramagnetic Beads Column)	ME-0010	Shanghai ZJ Bio-Tech
RNA Isolation Kit (for Auto-Extraction)	ME-0012	Shanghai ZJ Bio-Tech
Viral RNA Isolation Kit (Preloaded for Auto-Extraction)	ME-0044	Shanghai ZJ Bio-Tech
QIAamp Viral RNA Mini extraction Kit	52904/52906	QIAGEN

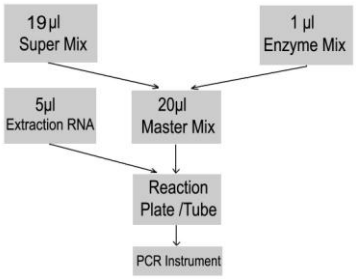
It is noted that the negative control in this kit should be extracted with the same protocol as for specimens. The positive control doesn't need to be extracted with the nucleic acid isolation kit.

9.2 Internal Control

The internal control (IC) in this kit should be added into the extraction mixture with 1μL/test to monitor the whole process.

9.3 RT-PCR Protocol

The Master Mix volume for each reaction should be pipetted as follows:



1) The volumes of Super Mix and Enzyme Mix per reaction multiply with the number of samples, which includes the number of controls and samples prepared. Molecular Grade Water is used as the negative control. For reasons of imprecise pipetting, always add an extra virtual sample. Mix completely and then spin down briefly with a centrifuge.
2) Pipet **20μL** Master Mix with micropipets of sterile filter tips to each of the *Real Time* PCR reaction plate/tubes. Separately add **5μL** template (nucleic acid extracted from negative control and specimen, positive control without extraction) to different reaction plates/tubes. Immediately close the plates/tubes to avoid contamination.
3) Spin down briefly in order to collect the Master Mix and template in the bottom of the reaction tubes.
4) Perform the following protocol in the instrument of ***ABI Prism®7500/7900***; ***Bio-Rad CFX96***; ***Rotor Gene™6000***; ***SLAN***:

45 ℃ for 10min	1cycle
95 ℃ for 3min	1cycle
95 ℃ for 15sec, 58 ℃ for 30sec (Fluorescence measured at 58 ℃)	45cycles

Selection of Fluorescence Channels	
FAM	ORF1ab
HEX/VIC/JOE	Gene N
Cal Red 610/ROX/TEXAS RED	Gene E
Cy5	IC

⚠: Perform the following protocol in the instrument of ***MIC POC Dx48***:

45 ℃ for 10min	1cycle
95 ℃ for 90sec	1cycle
95 ℃ for 3sec, 58 ℃ for 20sec (Fluorescence measured at 58 ℃)	45cycles

Selection of Fluorescence Channels	
Green	ORF1ab
Yellow	Gene N
Orange	Gene E
Red	IC

5) ⚠If you use ABI Prism® system, please choose **"none"** as **passive reference** and **quencher.**

10. Threshold Setting: Just above the maximum level of molecular grade water.

11. Quality Control: Negative Control and Positive Control must be performed correctly; otherwise the sample results are invalid.

Control \ Channel	Ct Value			
	FAM (ORF1ab)	HEX/VIC/JOE (GeneN)	Cal Red 610 (Gene E)	Cy5 (IC)
Negative Control	UNDET	UNDET	UNDET	25~40
Positive Control	≤35	≤35	≤35	UNDET

12. Data Analysis and Interpretation

The table below lists the expected results for the 2019-nCoV Real-Time Multiplex RT-PCR Kit. If results are obtained that do not follow these guidelines, re-extract and re-test the sample. If repeat testing yields similar results, contact Liferiver for consultation.

Ct value				Result interpretation[a]
ORF1ab	N	E	IC	
+	+	+	/	2019-nCoV detected
+	—	+	/	
+	+	—	/	
—	+	+	/	
—	—	—	+	2019-nCoV not detected[b]
—	—	—	—	Invalid; Repeat testing or collect a new specimen from the patient.
+	—	—	/	Repeat testing. If it is still the same result, the sample is 2019-nCoV positive.
—	+	—	/	Repeat testing. If it is still the same result, the sample is 2019-nCoV RNA presumptive positive.
—	—	+	/	

"+"represents a positive detection signal, which is defined as Ct≤41;
"-" represents a negative detection signal, which is defined as Ct>41;
"/" represents no requirement. Detection of Internal Control is not required if result positive in any of the other three detection channels.

Note:

[a] Laboratories should report their diagnostic result as appropriate and in compliance with their specific reporting system.

[b] Optimum specimen types and timing for peak viral levels during infections caused by 2019-nCoV have not been determined. Collection of multiple specimens from the same patient may be necessary to detect the virus.

For further questions or problems，please contact our technical support at **info@liferiverbiotech.com**

CERTIFICATE OF IVD NOTIFICATION

Ref. No.: LM 8733-2020

Order No.: LM 8649-2020

BELGIUM

Date: 26/02/2020

THIS IS TO CERTIFY THAT, ACCORDING TO THE COUNCIL DIRECTIVE 98/79/EC, OBELIS S.A. (O.E.A.R.C.) PERFORMED ALL NOTIFICATION DUTIES AND RESPONSIBILITIES AS THE EUROPEAN AUTHORIZED REPRESENTATIVE (EC REP) OF:

NAME: SHANGHAI ZJ BIO-TECH CO. LTD.

ADDRESS: BUILDING #26, 588 XINJUNHUAN ROAD, PUJIANG HIGH-TECH PARK, 201114, SHANGHAI, CHINA

AS STIPULATED AND DEMANDED BY THE AFOREMENTIONED DIRECTIVE.

The Manufacturer declares that the IVD device complies with the Directive including all essential requirements.

The Manufacturer has provided Obelis s.a. (O.E.A.R.C.) with all the appropriate declarations according to the 98/79/EC Directive – article 10 requirements including the EC Declaration of Conformity confirming that his In-Vitro Diagnostics medical device, as stipulated here above, is fulfilling the applicable requirements of the European Council Directive 98/79/EC

The notification of the following In-Vitro Diagnostic medical device has been completed by Obelis s.a. (O.E.A.R.C.) on the 24/02/2020 in compliance with the European Council Directive 98/79/EC - article 10 requirements.

IN-VITRO DIAGNOSTIC MEDICAL DEVICES: PLEASE SEE ANNEX A - LIST OF DEVICES (1 PAGE, 1 DEVICE)

As of the 25/02/2020, and as long as the manufacturer will continue complying with the hereabove mentioned requirements* he therefore:

- Is required to affix the CE marking on this device;

- Place this device in the Territory of Belgium and/or the other EEA Member States (excluding territories not in alignment with Decision 2010/227/EU).

G. Elkayam C.E.O.

Obelis s.a. - O.E.A.R.C.
Registered Address :
Bd Général Wahis 53
1030 Bruxelles
Tel +32 2 732 59 54 - Fax +32 2 732 60 03

Mr. G. Elkayam CEO
Obelis sa

Obelis European Authorized Representative Center is a member of the European Association of Authorized Representatives (E.A.A.R.), ISO 9001 : 2015 and ISO 13485 : 2016 certified in accordance to the profession of a European Authorized Representative.

* This is not a CE mark and is only provided as a template for informational purposes.

** This Certificate will be automatically void if the notification is rejected by the EU Authorities or upon termination of the EAR agreement.

Order No.: LM 8649-2020
Ref No.: LM 8733-2020

Annex A - List of Devices

(Recital 29 of the Directive 98/79/EC on in Vitro Diagnostic Medical Devic

#	Catalogue reference number	Commercial Name	Generic Device Term	Short description and intended use	GMDN/ EDMS Code	Class
1.	RR-0479-02	Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit (Detection for 3 Genes)	Multiple respiratory virus nucleic acid IVD, kit, nucleic acid technique (NAT)	Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit (Detection for 3 Genes) is used for the in vitro qualitative detection of 2019 novel coronavirus (2019-nCoV) RNA in upper respiratory tract specimens (nasopharyngeal and oropharyngeal extracts) and lower respiratory tract specimens (bronchoalveolar lavage fluid (BALF) and deep cough sputum) by real time PCR systems. 1. Novel CoV (2019-nCoV) Super Mix 1 vial, 513µL 2. RT-PCR Enzyme Mix 1 vial, 27µL 3. Novel CoV (2019-nCoV) Internal Control 1 vial, 30µL 4. Novel CoV (2019-nCoV) Negative Control 1 vial, 400µL 5. Novel CoV (2019-nCoV) Positive Control 1 vial, 30µL	47922	

* Annex A is part of the Agreement.

** The here above product list classification is based on the classification claim of the manufacturer and under its sole responsibility (IVD 98/79/EC).

G. ELKAYAM
C.E.O.

Obelis s.a. - O.E.A.R.C.
Registered Address :
Bld Général Wahis 53
1030 Bruxelles
Tél. +32 2 732 59 54 - Fax +32 2 732 60 03

APPROVAL FROM AUSTRALIA TGA

https://www.tga.gov.au/covid-19-test-kits-included-artg-legal-supply-australia

& public notices

st news & updates

Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit (Detection for 3 genes)	Laboratory	Nucleic Acid Test	Icon International Pty Ltd t/a Icon Medipharm	Shanghai ZJ Bio-Tech Co Ltd (China)	22 March 2020

APPROVAL FROM PHILIPPINES FDA

Republic of the Philippines
Department of Health
FOOD AND DRUG ADMINISTRATION

FDA
Food and Drug Administration
PHILIPPINES

List of Approved COVID-19 Test Kits for Commercial Use
as of 27 March 2020, 4:00PM

NO.	PRODUCT NAME	MANUFACTURER
14	NUCLEIC ACID REAGENT TEST KITS FOR NOVEL CORONAVIRUS 2019-NCOV (FLUOROMETRIC PCR)	Shanghai ZJ Bio-Tech Co., Ltd- – Floor 1, Building B, building 20, Floor 1, Building A, building 21, No. 528, Ruiqing Road, Zhangjiang high-tech Industry east district, Shanghai, China

Clinical Study Report

Generic Name of Diagnostic Kit under Clinical Study:

Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit

Clinical Study Performed by:

Guangdong Provincial Center for Disease Control and Prevention

The First Affiliated Hospital of Zhejiang University

Sponsor: Shanghai ZJ Bio-Tech Co., Ltd.

Contact Name: Lei CAO

Contact Phone Number: 021-34680599 ext. 8031

Date: 2020-1-25

Table of Contents

Clinical Study Summary

At the end of 2019, a mysterious pneumonia occurred in Wuhan. On January 8, 2020, novel coronavirus was initially identified as the pathogen causing this epidemic, whose sequence was disclosed subsequently. The World Health Organization (WHO) named this virus as 2019-nCoV (referred to as nCoV hereinafter) after receipt of the gene spectra from Chinese Center for Disease Control & Prevention.

The Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit, which is developed by Shanghai ZJ Bio-Tech Co., Ltd. (referred to as Liferiver hereafter), has gone through internal validation and passed the evaluation by Shanghai Medical Device Testing Institute.

The kit has been used in centers for disease control & prevention and some hospitals across the country. The clinical study data are summarized in this report, which are collected continuously during January 7~25, 2020 at Guangdong Provincial Center for Disease Control & Prevention ("GD CDC") as well as The First Affiliated Hospital of Zhejiang University ("1st Hospital").

The study data were collected from patients and their close contacts, with specimen types of nasal swab, pharyngeal swab, sputum, mouthwash, dry cough saliva, stool and bronchoalveolar lavage fluid.

In total, 252 specimens were tested at above two organizations, with 93 specimens at Guangdong Provincial Center for Disease Control & Prevention while 159 at The First Affiliated Hospital of Zhejiang University. Of all specimens, 112 were positive while 140 negative. Due to repeated specimens taken from some patients, there were 82 patients positive while 117 negative. With patient count used in the computation of coincidence rate between the testing results of Liferiver detection kit and the clinical diagnostic conclusions, the final results show 100% positive coincidence rate, 100% negative coincidence rate and 100% overall coincidence rate.

Acronyms

PCR: polymerase chain reaction

Liferiver: Shanghai ZJ Bio-Tech Co., Ltd.

1. Introduction

At the end of 2019, a mysterious pneumonia occurred in Wuhan. On January 8, 2020, novel coronavirus was initially identified as the pathogen causing this epidemic and its sequence was disclosed subsequently. The World Health Organization (WHO) received the gene spectra from the Chinese Center for Disease Control & Prevention and named this virus as 2019-nCoV.

On January 10, 2020, Shanghai Public Health Clinical Center, Wuhan Central Hospital, Huazhong University of Science & Technology, Wuhan Municipal Center for Disease Control & Prevention, Chinese Center for Disease Control & Prevention, Chinese Academy of Medical Sciences, The University of Sydney and other research institutions jointly released the gene sequence of the pathogen causing the mysterious pneumonia at Wuhan on Virological.org and GISAID, the gene library website. According to the article on GISAID website, this new coronavirus is similar to some coronaviruses found in bats but different from the previously found SARS / MERS.

Up to now, it is nucleic acid test and virus isolation & culture that experts and medical workers depend on for pathogen identification, and diagnostic kits distributed by government are used to test the confirmed cases later. In January of 2020, WHO issued *Clinical management of severe acute respiratory infection when novel coronavirus (nCoV) infection is suspected*, which provides guidance on laboratory testing of clinical samples.

The guidelines recommend that both upper respiratory tract (URT, including nasopharynx and oropharynx) and lower respiratory tract (LRT, including sputum, tracheal aspirate or bronchoalveolar lavage fluid) be collected at the same time and tested for nCoV with RT-PCR. Where lower respiratory tract samples are readily available (i.e. mechanically ventilated patients), only lower respiratory tract sample are collected; it is also only when RT-PCR is not available that serology is recommended for diagnosis.

The Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit, developed by Shanghai ZJ Bio-Tech Co., Ltd., is a multiplex test to detect three genes with one reaction, with internal control to monitor extraction. This test employs RNA reverse transcription reaction and polymerase chain reaction (PCR) combined with Taqman technology; the specific primer designed based on the nucleic acid sequence of the virus is to amplify the corresponding nucleic acid fragment, and the highly specific TaqMan probe is to bind the corresponding nucleic acid fragment and hydrolyze under the action of Taq exonuclease to generate fluorescent signal. Qualitative detection of 2019-nCoV nucleic acid can be realized

through the relation between fluorescent signal and amplification cycles.

This kit can be used for the qualitative detection of specific 2019-nCoV nucleic acid in human nasopharyngeal swabs, sputum and bronchoalveolar lavage fluid specimens. Applicable real-time PCR instruments include ABI 7500, CFX96, SLAN and MIC POC Dx48.

2. Objectives of Clinical Study

At the end of December 2019, cases of mysterious viral pneumonia were reported in Wuhan, which an expert group study preliminarily concluded was caused by novel coronavirus. The pathogen sequence information was disclosed subsequently. Shanghai ZJ Bio-Tech Co., Ltd. has developed a kit for the detection of this new coronavirus, which has undergone internal validation.

In order to examine its clinical performance, clinical study data are collected so as to evaluate its safety and efficacy in clinical use. The testing results with Liferiver assay are compared against the clinical diagnostic conclusions which are developed based on the methods recommended by China's Ministry of Health in Technical Guidance on Laboratory Test of Novel Coronavirus Infected Pneumonia and the Surveillance Protocol for Novel Coronavirus Infected Pneumonia Cases (2nd edition)[i].

3. Clinical Study Results and Analysis

3.1 Clinical Study Data

A total of 112 positive specimens from 82 patients and 140 negative specimens from 117 patients were collected by Guangdong Provincial Center for Disease Control & Prevention and The First Affiliated Hospital of Zhejiang University for this clinical study..

The number of positive and negative samples by each institute is shown in the table below:

Table 1. Summary of Clinical Study Samples

Clinical Study Institute	Specimen Count		Patient Count	
	Positive	Negative	Positive	Negative
GD CDC	79	14	69	10
1st Hospital	33	126	13	107
Total	112	140	82	117

3.2 Summary of Clinical Study Specimen Types

The table below shows the number of specimens of various types.

Table 2. Summary of Clinical Study Specimen Types

Specimen Type	Specimen Total		
	Positive	Negative	Total
Pharyngeal swab	69	17	86
Nasal swab	6	0	6
Sputum	31	121	152
Dry cough saliva	2	0	2
Mouthwash	2	0	2
Stool	0	1	1
Bronchoalveolar lavage fluid	2	0	2
Nasopharyngeal swab	0	1	1
Total	112	140	252

3.3 Coincidence Rate

The clinical study results, along with clinical diagnostic conclusions, are recorded in the table below. The Kappa analysis is then performed and coincidence rate computed respectively by specimen count and patient count.

Table 3. Clinical Study Coincidence Summary (specimen count)

		Comparative Method (clinical diagnostic conclusion)		Total
		Positive	Negative	
Study Assay (Liferiver)	Positive	112	0	112
	Negative	0	140	140
Total		112	140	252

The coincidence rate is computed according to the following formula:

$$\text{Positive coincidence rate} = \frac{a}{(a+c)} \times 100\% = 100\%$$
$$\text{Negative coincidence rate} = \frac{d}{(b+d)} \times 100\% = 100\%$$
$$\text{Overall coincidence rate} = \frac{(a+d)}{N} \times 100\% = 100\%$$

Kappa test:

$$P0 = \frac{(a+d)}{N}$$
$$Pe = \frac{(a+b)(a+c)+(c+d)(b+d)}{N \times N}$$
$$\text{Kappa} = \frac{P0-Pe}{1-Pe}$$

Comparing the testing results of Liferiver assay against the clinical diagnostic conclusions, positive coincidence rate is computed as 100%, negative coincidence rate 100%, and overall coincidence rate 100%.

The two sets of data are completely in agreement so there is no need for Kappa analysis; it is demonstrated that Liferiver assay and the clinical diagnostic conclusion are in full agreement.

In total, 82 positive patients and 117 negative patients are identified in the study data through checking different specimens collected from same patient. Fourfold table below is used to calculate the coincidence rate by patient count with the same aforementioned formula.

Table 4. Clinical Study Coincidence Summary (patient count)

		Comparative Method (clinical diagnostic conclusion)		Total
		Positive	Negative	
Study Assay (Liferiver)	Positive	82	0	82
	Negative	0	117	117
Total		82	117	199

Positive coincidence rate = 100%
Negative coincidence rate = 100%
Overall coincidence rate = 100%

The testing results of total 199 samples with Liferiver assay are completely in agreement with the clinical diagnostic conclusions, with positive coincidence rate, negative coincidence rate and overall coincidence rate all as 100% . Since the two sets of data are completely in agreement, there is no need for Kappa analysis; it is proven that the two diagnostic methods are in full agreement.

3.4 Analysis of Demographic Data

Analysis is performed on the 199 patients by gender, age and clinical symptom in this clinical study. Distribution of gender is shown in the following table.

Table 5. Summary of Study Patient Gender

Gender	Confirmed Case Count	Excluded Case Count	Total Cases
Male	41	42	83
Female	40	50	90
Not Included in Analysis	1	25	26

Distribution of age of these 199 patients is shown in the table below.

Table 6. Summary of Study Patient Age

Age	Confirmed Case Count	Excluded Case Count	Total Cases
⩽20	1	5	6
21-30	5	36	41
31-40	13	30	43
41-50	7	4	11
51-61	24	5	29
⩾61	31	5	36
Not Included in Analysis	1	32	33
Total	82	117	199

Clinical symptoms of these 199 patients are shown in the table below.

Table 7. Clinical Symptom Summary of Study Patients

Clinical Symptom	Confirmed Case Count	Excluded Case Count	Total Cases
Fever	8	48	56
Pulmonary Infection	1	1	2
Hepatolithiasis	0	1	1
Liver Transplantation Status	0	1	1
Cough	0	5	5
Intercostal Neuralgia	1	0	1
Upper Respiratory Tract Infection	0	2	2
Pancreatic Tumor	0	1	1
Not Included in Analysis	72	58	130
Total	82	117	199

No significant difference is noticed in different genders with regard to confirmed cases. But among confirmed cases, senior people take an obviously higher proportion, which indicates that the elderly are the susceptible population in this epidemic. The clinical symptomatic information obtained is very limited, and quite a number of people are included as suspected cases for confirmation by pathogen because of their fever symptoms. Due to the limited size of sample size collected, no statistically significant conclusion can be drawn.

4. Discussion and Conclusion

This clinical study uses real clinical data of this assay, analyzes negative & positive cases and examines specimen types. Negative coincidence rate, positive coincidence rate and overall coincidence rate are computed by comparing testing results of Liferiver assay against clinical diagnostic conclusion.

A total of 252 specimens are studied at Guangdong Provincial Center for Disease Control & Prevention and The First Affiliated Hospital of Zhejiang University, with 112 specimens positive and 140 negative. Considering repeated specimens collected from same patient, a total of 82 positive cases and 117 negative cases are included in this study.

Among the 252 specimens collected, 86 are of pharyngeal swab, 6 nasal swab, 152 of sputum, 2 of dry cough saliva, 2 of mouthwash, 1 of stool, 2 of bronchoalveolar lavage fluid and 1 of nasopharyngeal swab.

The specimen types which have gone through our company's systematic performance validation include nasopharyngeal swab, sputum and bronchoalveolar lavage fluid. All of the three specimen types have been tested in this clinical study with no abnormalities.

The testing results of Liferiver assay are consistent with the clinical diagnostic conclusions. The clinical diagnostic conclusions, confirmed cases and excluded cases, are made based on the criteria defined in Technical Guidance on Laboratory Test of Novel Coronavirus Infected Pneumonia and the Surveillance Protocol for Novel Coronavirus Infected Pneumonia Cases (2nd edition). The positive coincidence rate, negative coincidence rate and overall coincidence rate of this test compared against the clinical diagnosis method are all 100%, indicating concordance of the two diagnostic methods. With consistent accuracy, Liferiver detection kit applies one reaction and multi-channel, involves simple operation, and delivers testing result in about 80 minutes, which significantly enhances testing efficiency in actual use.

Annex

Annex I. Clinical Study Data Sheet of Guangdong Provincial Center for Disease Control & Prevention ("GD CDC")
Annex II. Clinical Study Data Sheet of The First Affiliated Hospital of Zhejiang University ("1st Hospital")

Annex I: Summary of Testing Results (GD CDC)

Ref.	Name of Clinical Study Institute	Patient ID	Gender	Age	Specimen ID	Specimen Type	Sampling Date	Clinical Background	Clinical Diagnostic Conclusion	Testing Result of Liferiver Assay	Note
	GD CDC	A	Male	78	GD010	Pharyngeal swab	1- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	B	Female	75	GD011	Pharyngeal swab	1- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	C	Female	48	GD012	Nasal swab	1- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	D	Male	75	GD013	Nasal swab	1- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	E	Male	51	GD014	Pharyngeal swab	1- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	F	Female	66	GD015	Nasal swab	1- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	F	Female	66	GD016	Pharyngeal swab	1- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	F	Male	79	GD017	Pharyngeal swab	1- 19	Patient for Triage	Confirmed	Positive	
	GD CDC	G	Male	79	GD018	Dry cough saliva	1- 19	Patient for Triage	Confirmed	Positive	
	GD CDC	G	Male	79	GD019	Mouthwash	1- 19	Patient for Triage	Confirmed	Positive	
	GD CDC	H	Female	56	GD020	Pharyngeal swab	1- 20	Patient for Triage	Confirmed	Positive	
	GD CDC	I	Female	58	GD021	Pharyngeal swab	1- 20	Patient for Triage	Confirmed	Positive	
	GD CDC	I	Female	62	GD022	Pharyngeal swab	1- 20	Patient for Triage	Confirmed	Positive	
	GD CDC	J	Female	63	GD023	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	J	Female	63	GD024	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	J	Female	63	GD025	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	K	Male	58	GD026	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	L	Female	64	GD027	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	M	Male	69	GD028	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	N	Female	64	GD029	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	O	Male	71	GD030	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	P	Male	73	GD031	Pharyngeal swab		Patient for Triage	Excluded	Negative	
	GD CDC	Q	Female	54	GD032	Pharyngeal swab	1- 17	Patient for Triage	Excluded	Negative	
	GD CDC	Q	Female	54	GD033	Sputum	1- 17	Patient for Triage	Excluded	Negative	
	GD CDC	R	Male	35	GD034	Pharyngeal swab	1- 19	Close contact	Released from quarantine	Negative	
	GD CDC	R	Male	35	GD035	Sputum	1- 19	Close contact	Released from quarantine	Negative	
	GD CDC	S	Male		GD036	Pharyngeal swab	1- 20	Close contact	Released from quarantine	Negative	
	GD CDC	T	Male		GD037	Pharyngeal swab	1- 20	Close contact	Released from quarantine	Negative	

Ref.	Name of Clinical Study Institute	Patient ID	Gender	Age	Specimen ID	Specimen Type	Sampling Date	Clinical Background	Clinical Diagnostic Conclusion	Testing Result of Liferiver Assay	Note
	GD CDC	case l	Male	66	GDOl	Bronchoalveolar lavage fluid	l -11	Patient for Triage	Confirmed	Positive	
	GD CDC	case 2	Female	65	GD02	Pharyngeal swab	l -11	Patient for Triage	Confirmed	Positive	
	GD CDC	case 3	Male	36	GD03	Pharyngeal swab	1-11	Close contact	Confirmed	Positive	
	GD CDC	case 4	Male	10	GD01	Pharyngeal swab	1-15	Close contact	Confirmed	Positive	
	GD CDC	case 5	Female	63	GD05	Pharyngeal swab	1-15	Close contact	Confirmed	Positive	
	GD CDC	case 6	Male	63	GD06	Bronchoalveolar lavage fluid	l-15	Patient for Triage	Confirmed	Positive	
	GD CDC	case 7	Female	67	GD07	Pharyngeal swab	1-15	Patient for Triage	Confirmed	Positive	
	GD CDC	case 8	Male	62	GD08	Pharyngeal swab	1- 16	Patient for Triage	Confirmed	Positive	
	GD CDC	case 9	Male	35	GD09	Pharyngeal swab	1-17	Patient for Triage	Confirmed	Positive	
	GD CDC	case 10	Male	78	GDOl0	Pharyngeal swab	1- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	case 11	Female	75	GDO11	Pharyngeal swab	l -18	Patient for Triage	Confirmed	Positive	
	GD CDC	case 12	Female	18	GD012	Nasal swab	l- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	case 13	Male	75	GD013	Nasal swab	l- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	case 11	Male	51	GD014	Pharyngeal swab	1- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	case 15	Female	66	GD015-1	Nasal swab	1-18	Patient for Triage	Confirmed	Positive	
	GD CDC	case 15	Female	66	GD015-2	Pharyngeal swab	l -18	Patient for Triage	Confirmed	Positive	
	GD CDC	case 16	Male	79	GD016-l	Pharyngeal swab	l -19	Patient for Triage	Confirmed	Positive	
	GD CDC	case 16	Male	79	GD016-2	Dry cough saliva	l -19	Patient for Triage	Confirmed	Positive	
	GD CDC	case 16	Male	79	GD016-3	Mouthwash	1-19	Patient for Triage	Confirmed	Positive	
	GD CDC	case 17	Female	56	GD017	Pharyngeal swab	l-20	Patient for Triage	Confirmed	Positive	
	GD CDC	case 18	Female	58	GD018	Pharyngeal swab	l - 20	Patient for Triage	Confirmed	Positive	
	GD CDC	case 19	Female	62	GD019	Pharyngeal swab	l - 20	Patient for Triage	Confirmed	Positive	
	GD CDC	case 20	Female	63	GD020-1	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 20	Female	63	GD020-2	Pharyngeal swab	l-21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 20	Female	63	GD020-3	Pharyngeal swab	1-21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 21	Male	58	GD021	Pharyngeal swab	l-21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 22	Female	61	GD022	Pharyngeal swab	l-21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 23	Male	69	GD023	Pharyngeal swab	1-21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 21	Female	61	GD021	Pharyngeal swab	l-21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 25	Male	71	GD025	Pharyngeal swab	1-21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 26	Male	10	GD026	Pharyngeal swab	1- 10	Patient for Triage	Confirmed	Positive	
	GD CDC	case 27	Male	36	GD027	Pharyngeal swab	1-20	Patient for Triage	Confirmed	Positive	
	GD CDC	case 28	Female	25	GD028	Pharyngeal swab	1- 11	Patient for Triage	Confirmed	Positive	
	GD CDC	case 29	Male	53	GD029	Pharyngeal swab	1-13	Patient for Triage	Confirmed	Positive	

Ref.	Name of Clinical Study Institute	Patient ID	Gender	Age	Specimen ID	Specimen Type	Sampling Date	Clinical Background	Clinical Diagnostic Conclusion	Testing Result of Liferiver Assay	Note
	GD CDC	case 30	Female	51	GD030	Pharyngeal swab	1-13	Patient for Triage	Confirmed	Positive	
	GD CDC	case 31	Female	81	GD031	Pharyngeal swab	1-7	Patient for Triage	Confirmed	Positive	
	GD CDC	case 32	Male	46	GD032	Pharyngeal swab	1-20	Patient for Triage	Confirmed	Positive	
	GD CDC	case 33	Male	57	GD033	Pharyngeal swab	1-19	Patient for Triage	Confirmed	Positive	
	GD CDC	case 34	Male	59	GD034	Pharyngeal swab	1-13	Patient for Triage	Confirmed	Positive	
	GD CDC	case 35	Female	31	GD035	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 36	Female	57	GD036	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 37	Male	56	GD037	Pharyngeal swab	1- 22	Patient for Triage	Confirmed	Positive	
	GD CDC	case 38	Female	73	GD038	Pharyngeal swab	1- 20	Patient for Triage	Confirmed	Positive	
	GD CDC	case 39	Female	55	GD039	Pharyngeal swab	1- 21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 40	Female	50	GD040	Pharyngeal swab	1- 18	Patient for Triage	Confirmed	Positive	
	GD CDC	case 41	Male	51	GD041	Pharyngeal swab	1- 20	Patient for Triage	Confirmed	Positive	
	GD CDC	case 42	Female	58	GD042	Pharyngeal swab	1- 22	Patient for Triage	Confirmed	Positive	
	GD CDC	case 43	Male	51	GD043	Pharyngeal swab	1- 10	Patient for Triage	Confirmed	Positive	
	GD CDC	case 44	Female	51	GD044	Pharyngeal swab	1- 15	Patient for Triage	Confirmed	Positive	
	GD CDC	case 45	Male	64	GD045	Pharyngeal swab	1- 22	Patient for Triage	Confirmed	Positive	
	GD CDC	case 46	Male	35	GD046	Pharyngeal swab	1- 22	Patient for Triage	Confirmed	Positive	
	GD CDC	case 47	Male	48	GD047	Pharyngeal swab	1- 22	Patient for Triage	Confirmed	Positive	
	GD CDC	case 48	Female	56	GD048	Pharyngeal swab	1-22	Patient for Triage	Confirmed	Positive	
	GD CDC	case 49	Male	34	GD049	Pharyngeal swab	1-21	Patient for Triage	Confirmed	Positive	
	GD CDC	case 50	Male	56	GD050	Pharyngeal swab	1-19	Patient for Triage	Confirmed	Positive	
	GD CDC	case 51	Female	38	GD051	Pharyngeal swab	1- 17	Patient for Triage	Confirmed	Positive	
	GD CDC	case 52	Male	36	GD052	Pharyngeal swab	1- 20	Patient for Triage	Confirmed	Positive	
	GD CDC	case 53	Female	50	GD053	Pharyngeal swab		Patient for Triage	Confirmed	Positive	
	GD CDC	case 54	Male	73		Pharyngeal swab		Patient for Triage	Excluded	Negative	
	GD CDC	case 55	Female	54		Pharyngeal swab	1- 17	Patient for Triage	Excluded	Negative	
	GD CDC	case 55	Female	54		Sputum	1- 17	Patient for Triage	Excluded	Negative	
	GD CDC	Close contact1	Male			Pharyngeal swab •	1-20	Close contact	Excluded	Negative	
	GD CDC	Close contact2	Male			Pharyngeal swab	1- 20	Close contact	Excluded	Negative	
	GD CDC	Close contact3	Male	35		Pharyngeal swab	43851	Close contact	Excluded	Negative	
	GD CDC	Close contact3	Male	35		Sputum	43852	Close contact	Excluded	Negative	

Tested by:

Reviewed by:

Statistician:

Principal Investigator:

Annex II: Summary of Testing Results (1st Hospital)

Ref.	Serial #	Patient ID	Gender	Age	Specimen Type	Name of Clinical Study Institute	Sample Collection Date	Clinical Background	Testing Results of Liferiver Assay	Clinical Diagnostic Conclusion	Note
1	20200123MLA901	LCL			Sputum	1st Hospital	2020.1.23	2019-nCoV RNA detection	Negative	Excluded	
2	20200123MLA902	YXL			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
3	20200123MLA903	TQY			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
4	20200123MLA904	LXY			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
5	20200123MLA905	WJQ			Sputum	1st Hospital	2020.1.23		Positive	Confirmed	
6	20200123MLA906	LH			Sputum	1st Hospital	2020. 1.23		Positive	Confirmed	
7	20200123MLA907	HSJ			Sputum	1st Hospital	2020.1.23		Positive	Confirmed	.
8	20200123MLA908	YYQ			Sputum	1st Hospital	2020.1.23		Positive	Confirmed	
9	i 20200123MLA909	WMM			Sputum	1st Hospital	2020.1.23		Positive	Confirmed	
10	20200123MLA910	HSJ			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
11	20200123MLA911	YQ			Sputum	1st Hospital	2020. 1.23		Negative	Excluded	
12	20200123MLA912	HSJ			Stool	1st Hospital	2020.1.23		Negative	Excluded	
13	20200123MLA913	ZYW			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
14	20200123MLA914	SJF			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
15	20200123MLA915	YL			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
16	20200123MLA916	LHD			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
17	20200123MLA917	ZM			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
18	20200123MLA918	YYC	Female	47	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	

19	20200123MLA919	HJC	Male	75	Sputum	1st Hospital	2020.1.23	Pancreatic tumor	Negative	Excluded	
20	20200123MLA920	SWD	Male	50	Sputum	1st Hospital	2020.1.23		Positive	Confirmed	
21	20200123MLA921	ZB	Male	18	Sputum	1st Hospital	2020.1.23	Cough	Negative	Excluded	
22	20200123MLA922	HML	Female		Sputum	1st Hospital	2020.1.23		Negative	Excluded	
23	20200123MLA923	ZSF	Female	45	Sputum	1st Hospital	2020.1.23		Negative	Excluded	
24	20200123MLA924	YT	Male	24	Sputum	1st Hospital	2020.1.23		Negative	Excluded	
25	20200123MLA925	GCQ			Sputum	1st Hospital.	2020.1.23		Negative	Excluded	
26	20200123MLA926	BHB			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
27	20200123MLA927	HHZ	Female	55	Sputum	1st Hospital	2020.1.23	Pulmonary infection	Positive	Confirmed	
28	20200123MLA928	XM	Female	35	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
29	20200123MLA929	MLL	Male	29	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
30	20200123MLA930	XJQ	Male	30	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
31	20200123MLA931	YYQ	Male	35	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
32	20200123MLA932	WJQ	Female	24	Sputum	1st Hospital	2020.1.23	Fever	Positive	Confirmed	
33	20200123MLA933	YX	Female	21	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
34.	20200123MLA934	WMM	Female	34	Sputum	1st Hospital	2020.1.23	Fever to be checked	Positive	Confirmed	
35	20200123MLA935	WTG	Male	22	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
36	20200123MLA936	ZKL	Male	25	Sputum	1st Hospital	2020.1.23		Negative	Excluded	
37	20200123MLA937	LHT	Female	23	Sputum	1st Hospital	2020.1.23		Negative	Excluded	

38	20200123MLA938	WYM	Female	26	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
39	20200123MLA939	FBH	Male	29	Sputum	1st Hospital	2020.1.23		Negative	Excluded	
40	20200123MLA940	FBH	Male	29	Nasopharyngeal swab	1st Hospital	2020.1.23		Negative	Excluded	
41	20200123MLA941	LH	Male	30	Sputum	1st Hospital	2020.1.23	Fever	Positive	Confirmed	
42	20200123MLA942	BSF	Female	24	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
43	20200123MLA943	FY	Female	30	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
44	20200123MLA944	ZT	Female	38	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
45	20200123MLA945	YXS	Male	23	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
46	20200123MLA946	LCE	Female	71	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
47	20200123MLA947	LMX	Male	53	Sputum	1st Hospital	2020.1.23	Fever	Positive	Confirmed	
48	20200123MLA948	WZH	Male		Pharyngeal swab	1st Hospital	2020.1.23	Fever	Negative	Excluded	
49	20200123MLA949	XF	Female	29	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
50	20200123MLA950	SFF	Female	31	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
51	20200123MLA951	LHY	Female	35	Sputum	1st Hospital	2020.1. 23		Negative	Excluded	
52	20200123MLA952	LQW	Male	30	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
53	20200123MLA953	HYM	Male	39	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
54	20200123MLA954	SWP	Female	54	Sputum	1st Hospital	2020.1. 23	Fever	Positive	Confirmed	
55	20200123MLA955	ZXP	Female	30	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
56	20200123MLA956	DXX	Female	57	Sputum	1st Hospital	2020.1. 23	Hepatolithiasis	Negative	Excluded	

57	20200123MLA957	SH	Male	23	Sputum	1st Hospital	2020.1. 23	Cough	Negative	Excluded	
58	20200123MLA958	ZXH	Female	26	Sputum	1st Hospital	2020.1. 23	Cough	Negative	Excluded	
59	20200123MLA959	CSY	Female	22	Sputum	1st Hospital	2020. 1.23	Fever	Negative	Excluded	
60	20200123MLA960	DXY	Female	22	Sputum	1st Hospital	2020. 1. 23	Fever	Negative	Excluded	
61	20200123MLA961	ZW	Male	19	Sputum	1st Hospital	2020.1. 23	Viral infection	Negative	Excluded	
62	20200123MLA962	GYY	Female	35	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
63	20200123MLA963	CQ	Female	22	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
64	20200123MLA964	SSG	Male	31	Sputum	1st Hospital	2020. 1. 23	Upper respiratory tract infection	Negative	Excluded	
65	20200123MLA965	JYH	Female	57	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
66	20200123MLA966	ZEY			Sputum	1st Hospital	2020. 1. 23		Negative	Excluded	
67	20200123MLA967	HHZ		55	Sputum	1st Hospital	2020.1. 23		Positive	Confirmed	
68	20200123MLA968	SWP	Female	54	Sputum	1st Hospital	2020.1. 23	Pulmonary infection	Positive	Confirmed	
69	20200123MLA969	MLL	Male	29	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
70	20200123MLA970	LQQ			Sputum	1st Hospital	2020.1. 23		Negative	Excluded	
71	20200123MLA971	FJ	Male	41	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
72	20200123MLA972	XSB	Male	33	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
73	20200123MLA973	LRH	Male	40	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
74	20200123MLA974	WJ			Sputum	1st Hospital	2020.1. 23		Negative	Excluded	
75	20200123MLA975	XZJ	Female	30	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	

76	20200123MLA976	TY	Female	32	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
77	20200123MLA977	YKY	Female	25	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
78	20200123MLA978	LL	Female	38	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
79	20200123MLA979	HYP	Female	40	Sputum	1st Hospital	2020.1. 23	Cough	Negative	Excluded	
80	； 20200123MLA980	CNY			Sputum	1st Hospital	2020.1. 23		Negative	Excluded	
81	20200123MLA981	**YW**			Sputum	1st Hospital	2020.1. 23		Negative	Excluded	
82	20200123MLA982	SSY	Female	22	Sputum	1st Hospital	2020.1. 23	Fever	Negative	Excluded	
83	20200123MLA983	WQY	Male	31	Sputum	1st Hospital	2020. 1.23	Fever to be determined	Negative	Excluded	
84	20200123MLA984	ZR	Female	30	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
85	20200123MLA985	ZY	Female	34	Sputum	1st Hospital	2020. 1.23	Fever	Negative	Excluded	
86	20200123MLA986	LQ	Female	26	Sputum	1st Hospital	2020.1.23	Cough	Negative	Excluded	
87	20200123MLA987	HXJ	Female	26	Sputum	1st Hospital	2020.1 23	Fever	Negative	Excluded	
88	20200123MLA988	HK	Male	31	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded[1]	
89	20200123MLA989	XQT	Male	28	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
90	20200123MLA990	ZXY	Female	27	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
91	20200123MLA991	ZW	Male	32	Sputum	1st Hospital	2020.1.23	Upper respiratory tract infection	Negative	Excluded	
92	20200123MLA992	ZSY	Female	29	Sputum	1st Hospital	2020.1.23	General medical examination	Negative	Excluded	
93	20200123MLA993	WHY	Female	27	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
94	20200123MLA994	YD	Male	31	Sputum	1st Hospital	2020.1.23	General medical examination	Negative	Excluded	

95	20200123MLA995	=ZXK	Male	34	Sputum	1st Hospital	2020.1.23	Fever	Negative	Excluded	
96	20200123MLA996	SY			Sputum	1st Hospital	2020.1.23		Negative	Excluded	
97	20200124MLA941	GJ	Male	30	Sputum	1st Hospital	2020.1.24	Intercostal neuralgia	Positive	Confirmed	
98	20200124MLA946	SHF	Female	43	Sputum	1st Hospital	2020.1.24	Fever	Negative	Excluded	
99	20200124MLA947	ZLQ	Male	36	Sputum	1st Hospital	2020.1.24	Fever	Negative	Excluded	
100	20200125MLA901	CHC	Male	40	Sputum	1st Hospital	2020.1.25	Liver transplantation status	Negative	Excluded	
101	20200125MLA902	SHX	Female	35	Sputum	1st Hospital	2020.1.25	Fever	Negative	Excluded	
102	20200125MLA903	HHZ	Female	55	Sputum	1st Hospital	2020.1.25	Pulmonary infection	Positive	Confirmed	
103	20200125MLA904	SWP	Female	54	Sputum	1st Hospital	2020.1.25	Pulmonary infection	Positive	Confirmed	
104	20200125MLA905	ZWL	Male	53	Sputum	1st Hospital	2020.1.25	Aortic dissection	Negative	Excluded	
105	20200125MLA906	ZL			Sputum	1st Hospital	2020.1.25		Negative	Excluded	
106	20200125MLA907	YYQ	Male	35	Sputum	1st Hospital	2020.1.25	Fever	Positive	Confirmed	
107	20200125MLA908	WMM	Female	34	Sputum	1st Hospital	2020.1.25	Fever	Positive	Confirmed	
108	20200125MLA909	WJQ	Female	24	Sputum	1st Hospital	2020.1.25	Fever	Positive	Confirmed	
109	20200125MLA910	WMM	Female	34	Sputum	1st Hospital	2020.1.25	Fever	Positive	Confirmed	
110	20200125MLA911	LH	Male	30	Sputum	1st Hospital	2020.1.25	Fever to be determined	Positive	Confirmed	
111	20200125MLA912	WJQ	Female	24	Sputum	1st Hospital	2020.1.25	Fever	Positive	Confirmed	
112	20200125MLA913	YYQ	Male	35	Sputum	1st Hospital	2020.1.25	Fever	Positive	Confirmed	
113	20200125MLA914	GXM	Female	39	Sputum	1st Hospital	2020.1.25	Fever	Positive	Confirmed	

114	20200125MLA915	HHZ	Female	55	Sputum	1st Hospital	2020.1.25	Pulmonary infection	Positive	Confirmed	
115	20200125MLA916	HSJ	Male	32	Sputum	1st Hospital	2020.1.25	Fever	Positive	Confirmed	
116	20200125MLA917	CZ	Male	34	Sputum	1st Hospital	2020.1.25	Fever	Positive	Confirmed	
117	20200125MLA918	LH	Male	30	Sputum	1st Hospital	2020.1.25	Fever	Positive	Confirmed	
118	20200125MLA919	DML	Male	31	Sputum	1st Hospital	2020.1.25	Pulmonary infection	Negative	Excluded	
119	20200125MLA920	CF	Male	33	Sputum	1st Hospital	2020.1.25	Fever	Negative	Excluded	
120	20200125MLA921	ZFF	Female	25	Sputum	1st Hospital	2020.1.25	Fever	Negative	Excluded	
121	20200125MLA922	HZC	Female	40	Sputum	1st Hospital	2020.1.25	Fever	Negative	Excluded	
122	20200125MLA923	FJE	Female	65	Sputum	1st Hospital	2020.1.25	Fever	Negative	Excluded	
123	20200122MLA908	TP			Sputum	1st Hospital	2020.1.22		Negative	Excluded	
124	20200122MLA909	DEC			Sputum	1st Hospital	2020.1.22		Negative	Excluded	
125	20200122MLA910	LHT			Pharyngeal swab	1st Hospital	2020.1.22		Negative	Excluded	
126	20200122MLA911	WYM			Pharyngeal swab	1st Hospital	2020.1.22		Negative	Excluded	
127	20200122MLA912	GZH			Sputum	1st Hospital	2020.1.22		Negative	Excluded	
128	20200122MLA913	YHB			Sputum	1st Hospital	2020.1.22		Negative	Excluded	
129	20200122MLA914	LHT			Sputum	1st Hospital	2020.1.22		Negative	Excluded	-
130	20200122MLA915	LH			Sputum	1st Hospital	2020.1.22		Negative	Excluded	
131	20200122MLA916	LL			Sputum	1st Hospital	2020.1.22		Negative	Excluded	
132	20200122MLA917	YN			Sputum	1st Hospital	2020.1.22		Negative	Excluded	

133	20200122MLA918	XM			Sputum	1st Hospital	2020.1.22		Negative	Excluded	
134	20200122MLA919	LMX			Pharyngeal swab	1st Hospital	2020.1.22		Positive	Confirmed	
135	20200122MLA920	XJQ			Pharyngeal swab	1st Hospital	2020.1 22		Negative	Excluded	
136	20200122MLA921	MLL			Pharyngeal swab	1st Hospital	2020.1.22		Negative	Excluded	
137	20200122MLA922	YYQ			Pharyngeal swab	1st Hospital	2020. 1.22		Positive	Confirmed	
138	20200122MLA923	YX			Pharyngeal swab	1st Hospital	2020.1.22		Negative	Excluded	
139	20200122MLA924	CQ			Pharyngeal swab	1st Hospital	2020.1.22		Negative	Excluded	
140	20200122MLA925	SFF			Sputum	1st Hospital	2020.1.22		Negative	Excluded	
141	20200122MLA926	XF			Sputum	1st Hospital	2020.1. 22		Negative	Excluded	
142	20200122MLA927	JYH	Female	57	Sputum	1st Hospital	2020.1.22		Negative	Excluded	
143	20200122MLA928	LQW	Male	30	Sputum	1st Hospital	2020. 1. 22		Negative	Excluded	
144	20200122MLA929	ZJQ	Male	27	Sputum	1st Hospital	2020.1. 22		Negative	Excluded	
145	20200122MLA930	WYX	Female	18	Sputum	1st Hospital	2020.1. 22		Negative	Excluded	
146	20200122MLA931	GYY	Female		Sputum	1st Hospital	2020.1. 22		Negative	Excluded	
147	20200122MLA932	ZW	Male		Sputum	1st Hospital	2020.1. 22		Negative	Excluded	
148	20200122MLA933	HC	Male	31	Sputum	1st Hospital	2020. 1. 22		Negative	Excluded	
149	20200122MLA934	LSH	Male	23	Sputum	1st Hospital	2020. 1.22		Negative	Excluded	
150	20200122MLA935	HJ	Female	20	Sputum	1st Hospital	2020. 1.22		Negative	Excluded	
151	20200122MLA936	LMX	Male		Sputum	1st Hospital	2020. 1. 22		Positive	Confirmed	

152	20200122MLA937	ZZF	Female	23	Sputum	1st Hospital	2020.1. 22		Negative	Excluded	
153	20200122MLA938	LL	Female	37	Sputum	1st Hospital	2020.1.22		Negative	Excluded	
154	20200122MLA939	YKY	Female	24	Sputum	1st Hospital	2020.1.22		Negative	Excluded	
155	20200122MLA940	WJQ	Female	23	Sputum	1st Hospital	2020.1.22		Positive	Confirmed	
156	20200122MLA941	DML	Female	15	Sputum	1st Hospital	2020.1.22		Negative	Excluded	
157	20200122MLA942	WQM	Male		Sputum	1st Hospital	2020.1.22		Negative	Excluded	
158	20200122MLA943	HXJ	Female	25	Sputum	1st Hospital	2020. 1.22		Negative	Excluded	
159	20200122MLA944	LHY	Female	34	Sputum	1st Hospital	2020.1.22		Negative	Excluded	

1. [i] Suspected Patients
   a) Epidemiological history
      i. Stay or travel in Wuhan or other territories with continuous local spread of disease within last 14 days before onset
      ii. Contact with patients with febrile or respiratory symptoms from Wuhan or other territories with continuous local spread of disease within last 14 days before onset
      iii. Epidemiological connection with group sick or confirmed patient, mild patient or patient without symptoms
   b) Clinical symptoms
      i. Fever
      ii. With pneumonia imaging features
      iii. Normal or lower white cell count at early stage, or reduced lymphocyte count

   Suspected patients refer to those who meet any one of those epidemiological history criteria combined with b.i & b.ii or b.ii &b.iii and those who have no definite epidemiological history but meet b.iii.
2. Confirmed Patients

   Confirmed patients refer to suspected patients who also demonstrate one of the following etiological evidences--
   a) Real time RT-PCT test positive for novel coronavirus on respiratory or blood specimen
   b) High-degree match in origin with the known novel coronavirus by virus gene sequencing

India Contact:
Life Technologies (India) Pvt. Ltd.
306, Aggarwal City Mall, Opposite M2K Pitampura,
Delhi – 110034 (INDIA).
Ph: +91-11-42208000, 42208111, 42208222
Mobile: +91-9810521400 Fax: +91-11-42208444
Email: customerservice@lifetechindia.com
Web: www.lifetechindia.com

ICMR-National Institute for Implementation Research on Non-Communicable Diseases (NIIRNCD), Jodhpur

20-08-2020

M/s Life Technologies India Private Ltd

Subject : Performance evaluation report for LifeRiver Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit manufactured by Shanghai ZJ Bio-Tech Co., Ltd.

Sir,
We have evaluated the LifeRiver Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit manufactured by Shanghai ZJ Bio-Tech Co., Ltd. (Lot No. : 20200601).The final report is attached for your information.

Yours sincerely

Prof (H). Dr. G. S. Toteja
Addl. DG-ICMR &
Director,NIIRNCD, Jodhpur.

ICMR-National Institute for Implementation Research on Non-Communicable Diseases (NIIRNCD), Jodhpur

Performance evaluation report for RT-PCR diagnostic kit

Name of the kit : LifeRiver Novel Coronavirus (2019-nCoV) Real Time Multiplex RT-PCR Kit (Ref: RR-0479-02)

Name of the manufacturer : Shanghai ZJ Bio-Tech Co., Ltd

Lot / batch number : 20200601

Date of expiry : 05/12/2020

Application :In Vitro Testing kit for detection of novel corona virus infection (COVID-19) by qualitatively detecting ORF1ab, N gene and E gene of SARS-CoV-2 virus along with Internal Control.

Kit components : Novel CoV (2019-nCoV) Super Mix, RT-PCR Enzyme Mix, Novel CoV (2019-nCoV) Internal Control, Novel CoV (2019-nCoV) Negative Control, Novel CoV (2019-nCoV) Positive Control

Sample Panel

1. SARS CoV-2 Positive samples (n=75)(25 High, 25 Med, 25 Low Ct values as per ICMR-NIV Kit)
2. SARS CoV-2 Negative samples (n=75)
3. SARS CoV-2 Negative samples but positive for other respiratory viruses (n=10; Influenza A H1N1 positive samples)

Results

		ICMR NIV RT-PCR Kit Results (as Gold Standard)		
		Positive	**Negative**	**Total**
LifeRiver kit	Positive	72	0	72
	Negative	3	85	88
	Total	75	85	160

Parameter	Estimate (%)	95% CI
Sensitivity	96%	88.8% to 99.2%
Specificity	100%	95.8% to 100%

Conclusion:

Sensitivity for detection of SARS-CoV-2	**: 96%**
Specificity for detection of SARS-CoV-2	**: 100 %**
Performance	**: SATISFACTORY**

(Sensitivity and specificity have been assessed in controlled lab setting using kits provided by the manufacturer from the batch mentioned above)

Disclaimers

1. ICMR's validation process does not approve / disapprove the kit design
2. ICMR's validation process does not certify user friendliness of the kit / assay
3. Validation of a kit by ICMR is not an assurance that the kit specifications would be included in the tendering process.

Note: This report is exclusively for RT-PCR Kit (Lot No 20200601) manufactured by Shanghai ZJ Bio-Tech Co., Ltd. (supplied by Life Technologies India Private Ltd.)

The company shall not use or publish information or report for advertising or promotional purposes

Evaluation Done on .....

Evaluation Done by .....

Prof (H). Dr. G. S. Toteja
Addl. DG-ICMR &
Director,NIIRNCD, Jodhpur.