KWORK

Novel Coronavirus Ag Rapid Test Device (Feces/Saliva/Sputum) Package Insert

Cat: NCP-F02 **Specimens:** Feces/Saliva/Sputum
Version: 06 **Effective Date:** 2020-11

For professional and in vitro diagnostic use only.

PRODUCT NAME
Novel Coronavirus Ag Rapid Test Device (Feces/Saliva/Sputum)

PACKING
1 Test/Kit, 10 Tests/Kit, 15 Tests/Kit, 20 Tests/Kit, 25 Tests/Kit.

INTENDED USE
This product is suitable for the qualitative detection of Novel Coronavirus in Feces/Saliva/Sputum samples. It provides an aid in the diagnosis of infection with Novel Coronavirus.

SUMMARY
The Novel Coronaviruses belong to the β genus. COVID-19 is an acute respiratory infectious disease. People are generally susceptible. Currently, the patients infected by the Novel Coronavirus are the main source of infection; asymptomatic infected people can also be an infectious source. Based on the current epidemiological investigation, the incubation period is 1 to 14 days, mostly 3 to 7 days. The main manifestations include fever, fatigue and dry cough. Nasal congestion, runny nose, sore throat, myalgia and diarrhea are found in a few cases.

PRINCIPLE
The Novel Coronavirus invades human cells by specifically binding of its spike glycoprotein (ligand) to ACE2 receptor on human cellular membrane with a high affinity (KD measured as 15 nM for the binding of the S protein to the ACE2 receptor), which is 10-20 times stronger than SARS-CoV. Accordingly, we substituted ACE2 receptor for antibody to establish a novel ligand-receptor chromatography test kit for rapid Novel Coronavirus detection. The sensitivity of the strip to S protein reached 30 nM. In clinical practice, this test strip can be used for rapid detection of Saliva, Sputum and Feces samples of patients, which only takes 10-15 minutes, much easier and faster than nucleic acid testing (RT-PCR). Therefore, this strip is suitable for detection of SARS-CoV-2 and all of its mutants. It has been found that this virus evolved into more contagious mutants through mutations in S1 proteins (such as D614G) that are stronger binding to ACE2 receptors. This means that our detection kit based on ACE2 receptor will be more sensitive to such mutants.

The test kit contains a test strip, in which, on the nitrocellulose (NC) membrane, the rabbit anti-S1 protein of Novel Coronavirus antibodies is coated at the test area (T), and the goat anti-rabbit IgG polyclonal antibody is coated at the control area (C). Latex-labeled ACE2 protein and Latex-labeled rabbit IgG are embedded in the reagent pad.

During the test, add three drops of the sample, and the sample laterally flows from the bottom to the top under the capillary effect. If the sample contains the virus, the latex-labeled ACE2 protein will be bound by the S1 protein of virus, and then captured by anti-S1 protein antibodies coated on the test area (T), the T line is appeared. If the sample does not contain the virus, the latex-labeled ACE2 protein cannot be captured by anti-S1 protein antibodies coated on the test area (T), therefore, no T line appear. No matter whether there is a Novel Coronavirus in the sample, the latex-labeled rabbit IgG will be combined with the goat anti-rabbit IgG polyclonal antibody coated on the control area (C). A red latex line will appear in the control area.

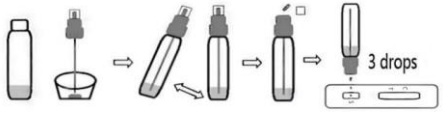
At the end of the test, the amount of latex-ACE2 protein bound on the T line is proportional to the concentration of Novel Coronavirus in the sample, while the amount of latex on the control line C bound is irrelevant to the amount of coronavirus in the sample.

COMPOSITION
1. Disposable test card
2. Disposable sample extraction tube
3. Cotton swab
4. Disposable paper cup
5. Dropper

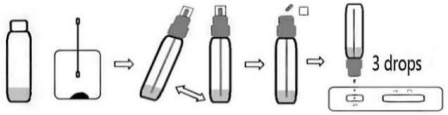
STORAGE AND STABILITY
1. Store as packaged in the hermetic bag at the temperature (2-30℃) and avoid direct sunshine. The kit is stable within the expiration date printed on the labeling.
2. Once open the hermetic bag, the test should be used within one hour. Prolonged exposure to hot and humid environment will cause product deterioration.
3. The lot number and the expiration date were printed on the labeling.

TEST PROCEDURE
Allow the test device and specimens to equilibrate to temperature (15-30℃) prior to testing.

Feces Sample:

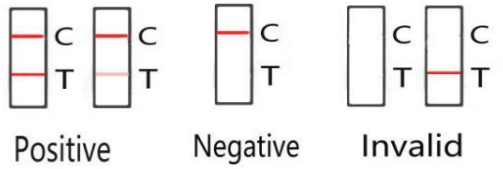


Saliva/Sputum Sample:



1. **Feces Sample:** Unscrew the sampling tube, use the sampling rod to pick up 100-200mg of fresh Feces samples (equivalent to the size of a match head); or swab the Feces with a cotton swab. Put them into the tube and shake and mix completely.
2. **Retropharyngeal Saliva Sample:** Clean the hand and open Disposable paper cup, clean the throat, spilt the saliva from the retropharynx into the Disposable paper cup (repeat the action make the sample above 2 ml). Avoid the sampling tube contaminated by the saliva. The optimal sampling time is after waking up and before brushing teeth or drinking. Unscrew the sampling tube, use the dropper or a cotton swab to pick up 200-300uL of fresh Saliva samples. Put them into the tube and shake and mix completely.
3. **Sputum Sample:** Unscrew the sampling tube, use the dropper or a cotton swab to pick up 200-300uL of fresh sputum samples. Put them into the tube and shake and mix completely.
4. Take the test card from the packaging bag, place it on a table, cut off the protrusion of the sampling tube, and add 3 drops of the sample into the test well vertically.
5. Wait for the appearance of the red stripe on T line, read the result in 15 minutes, and judge it invalid after 20 minutes.

INTERPRETATION OF RESULTS



Positive (+): Both of T and C lines are appeared in 3-15minutes.
Negative (-): C line is appeared while no T line appeared in 15 minutes after the sample added.
Invalid: As long as the C line does not appear, it indicates that the test result is invalid, and should retest with another test card.

PERFORMANCE CHARACTERISTICS
Limit of detection
The Limit of detection (LOD) of the Novel Coronavirus Ag Rapid Test Device (Feces/Saliva/Sputum) is 5 ng/ml SARS-COV-2 spike glycoprotein.

Sensitivity and Specificity
60 samples of the Novel Coronavirus and 120 samples of healthy people were tested with test reagents. Includes 60 Saliva samples, 60 Sputum samples and 60 Feces samples. Compared with the Novel Coronavirus (SARS-COV-2) real-time multiplex RT-PCR kit, the sensitivity and specificity of the Novel Coronavirus Ag Rapid Test Device.

Saliva Sample		RT-PCR		Total
		Positive	Negative	
Test reagent	Positive	54	0	54
	Negative	6	120	126
Total		60	120	180

Feces Sample		RT-PCR		Total
		Positive	Negative	
Test reagent	Positive	57	0	57
	Negative	3	120	123
Total		60	120	180

Sputum Sample		RT-PCR		Total
		Positive	Negative	
Test reagent	Positive	56	0	56
	Negative	4	120	124
Total		60	120	180

*** 95% Confidence Interval = (% ～ %)**

Saliva samples: Novel Coronavirus Ag Rapid Test Device showed 90% sensitivity and 100% specificity in Saliva samples.
Clinical sensitivity (%) = [54/ (54+ 6)] ×100% = 90% (79.49%～96.24%)
Clinical specificity (%) = [120/ (0+ 120)] ×100% = 100% (96.97%～100%)
Total agreement rate (%) = [(54+120) / (54+ 6+ 0+ 120)] ×100% =96.7% (92.89%～98.77%)

Feces samples: Novel Coronavirus Ag Rapid Test Device showed 95% sensitivity and 100% specificity in Feces samples.
Clinical sensitivity (%) = [57/ (57+ 3)] ×100% = 95% (86.08%～98.96%)
Clinical specificity (%) = [120/ (0+ 120)] ×100% = 100% (96.97%～100%)
Total agreement rate (%) = [(57+120) / (57+3+ 0+ 120)] ×100% =98.3% (95.21%～99.66%)

Sputum samples: Novel Coronavirus Ag Rapid Test Device showed 93.3% sensitivity and 100% specificity in Sputum samples.
Clinical sensitivity (%) = [56/ (56+ 4)] ×100% = 93.3% (83.80%～98.15%)
Clinical specificity (%) = [120/ (0+ 120)] ×100% = 100% (96.97%～100%)
Total agreement rate (%) = [(56+120) / (56+4+ 0+ 120)] ×100% =97.7% (94.41%～99.39%)

Cross-Reactivity
The antibody used in the Novel Coronavirus Ag Rapid Test Device has been shown to detect all known Coronavirus. Coronavirus strains have been tested with the Novel Coronavirus Ag Rapid Test Device, and were not shown to cross-react when tested in suspensions of 50 ug/mL.
The following organisms were found negative when tested with the Novel Coronavirus Ag Rapid Test Device:

Analytes	Concentration	Cross-Reactivity (Yes/No)
SARS-CoV S1	50 ug/mL	No
MERS- CoV S1	50 ug/mL	No
HCoV-NL63 S1	50 ug/mL	No
HCoV-HKU1 S1	50 ug/mL	No
HCoV-229E S1	50 ug/mL	No
HCoV-OC43 S1	50 ug/mL	No
Influenza A H1N1 Protein	50 ug/mL	No
Influenza B Protein	50 ug/mL	No
Human RSV(B1) G Protein	50 ug/mL	No

Interference
Interferences of common exogenous potential interfering substances of some kinds of samples were tested, it showed that no interference was found in these tests.

Effect of the potentially cross-reactive endogenous substances:

Analytes	Concentration	Interference (Yes/No)
Albumin	20 mg/mL	No
Bilirubin	20 ug/mL	No
Hemoglobin	15 mg/mL	No
Glucose	20 mg/mL	No
Uric Acid	200 ug/mL	No
Lipids	20 mg/mL	No

Effect of some other common biological analytes:

Analytes	Concentration	Interference (Yes/No)
Acetaminophen	200 ug/mL	No
Acetoacetic Acid	200 ug/mL	No
Acetylsalicylic Acid	200 ug/mL	No
Benzoylecgonine	100 ug/mL	No
Caffeine	200 ug/mL	No
EDTA	800 ug/mL	No
Ethanol	1.0%	No
Gentisic Acid	200 ug/mL	No
P- Hydroxybutyrate	200,000 ug/mL	No
Methanol	10.0%	No
Phenothiazine	200 ug/mL	No
Phenylpropanolamine	200 ug/mL	No
Salicylic Acid	200 ug/mL	No

NOTES

1. Novel Coronavirus Ag Rapid Test Device (Feces/Saliva/Sputum) is only applicable to Feces/Saliva/Sputum samples. Blood, Serum, Plasma, Urine and other samples may cause abnormal results. Due to the large sampling difference of oropharyngeal swabs, abnormal results may also appear in the test using the oropharyngeal swabs. We recommend using Feces samples first. If the Sputum sample is negative, it is recommended to use Feces examination again, because the Sputum sample varies greatly among individuals and there are many factors affecting the examination. If any sample tests positive, please go to the hospital for further clinical diagnosis.
2. Please make sure that a proper amount of sample is added for testing. Too much or too little sample may cause deviations in results.
3. For positive judgement, it can be confirmed as soon as both T and C line appeared. That may be in 3-15 minutes after the sample added. For negative judgement, please wait for 15 minutes after the sample added, C line is appeared while no T line appeared. The result is invalid after 20 minutes after sample added.
4. The test card is a disposable product. Please dispose properly after use in accordance with the relevant local medical waste disposal guideline.
5. This test device is disposable, please use within the validity period. After use, the test reagent, sample and other waste should be treated in accordance with the relevant local medical waste disposal guideline.
6. If part of the test strip is out of the test window, do not use. Otherwise, the test result is invalid and should replace with another new test card.

INDEX OF SYMBOLS

	Consult instructions for use	Σ	Tests per kit	EC REP	Authorized Representative
IVD	For in vitro diagnostic use only		Use by		Do not reuse
2°C 30°C	Store between 2-30°C	LOT	Lot Number	REF	Catalogue number

Zhejiang Quark Biotechnology Co., Ltd
No.1 Jiangdong Road,
Zhejiang Xinchang High-tech Industrial Park,
Xinchang District, 312500, Shaoxing, Zhejiang,
PEOPLE'S REPUBLIC OF CHINA

CE

EC REP
Wellkang Ltd.
16 Castle St., Dover, Kent,
CT16 1PW, England, UK