SARS-CoV-2 Antigen Rapid Test

Package Insert

Flowflex™

REF L031-11815 REF L031-129Z5	English

A rapid test for the qualitative detection of SARS-CoV-2 nucleocapsid antigens in nasal and nasopharyngeal swab specimens.
For professional in vitro diagnostic use only.

INTENDED USE

The SARS-CoV-2 Antigen Rapid Test is a lateral flow chromatographic immunoassay for the qualitative detection the nucleocapsid protein antigen from SARS-CoV-2 in nasal and nasopharyngeal swab specimens directly from individuals who are suspected of COVID-19 by their healthcare provider within the first seven days of the onset of symptoms. The SARS-CoV-2 Antigen Rapid Test can also test specimens from individuals without symptoms or other reasons to suspect COVID-19 infection when tested twice over two (or three days) with at least 24 hours (and no more than 36 hours) between tests. The SARS-CoV-2 Antigen Rapid Test does not differentiate between SARS-CoV and SARS-CoV-2.
Results are for the identification of SARS-CoV-2 nucleocapsid antigen. This antigen is generally detectable in upper respiratory samples during the acute phase of infection. Positive results indicate the presence of viral antigens, but clinical correlation with patient history and other diagnostic information is necessary to determine infection status. Positive results do not rule out bacterial infection or co-infection with other viruses. The agent detected may not be the definite cause of disease.
Negative results, from patients with symptom beyond seven days, should be treated as presumptive and confirmed with a molecular assay, if necessary, for patient management. Negative results do not rule out SARS-CoV-2 infection and should not be used as the sole basis for treatment or patient management decisions, including infection control decisions. Negative results should be considered in the context of a patient's recent exposures, history and the presence of clinical signs and symptoms consistent with COVID-19.
The SARS-CoV-2 Antigen Rapid Test is intended for use by trained clinical laboratory personnel and individuals trained in point of care settings.

SUMMARY

The novel coronaviruses belong to the β genus.[1] 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 SARS-CoV-2 Antigen Rapid Test is a qualitative membrane based chromatographic immunoassay for the qualitative detection of the nucleocapsid protein antigen from SARS-CoV-2 in human nasal and nasopharyngeal swab specimens.
When specimens are processed and added to the test cassette, SARS-CoV-2 antigens, if present in the specimen, will react with the anti-SARS-CoV-2 antibody-coated particles, which have been pre-coated on the test strip. The mixture then migrates upward on the membrane by capillary action. The antigen-conjugate complexes migrate across the test strip to the reaction area and are captured by a line of antibody bound on the membrane. Test results are interpreted visually at 15-30 minutes based on the presence or absence of visually colored lines.
To serve as a procedure control, a colored line will always appear in the control line region indicating that proper volume of specimen has been added and membrane wicking has occurred.

REAGENTS

The test cassette contains anti-SARS-CoV-2 antibodies. The positive control swab contains SARS-CoV-2 recombinant antigen pre-coated on the swab.

PRECAUTIONS

- For professional *in vitro* diagnostic use only. Do not use after the expiration date.
- Do not eat, drink, or smoke in the area where the specimens or kits are handled.
- Do not use the test if the pouch is damaged.
- Handle all specimens as if they contain infectious agents. Observe established precautions against biological hazards throughout testing and follow the standard procedures for proper disposal of specimens.
- Wear protective clothing such as laboratory coats, disposable gloves, mask and eye protection when specimens are being tested.
- The used test should be discarded according to local regulations. The used test should be considered potentially infectious and be discarded according to local regulations.
- Humidity and temperature can adversely affect results.
- This package insert must be read completely before performing the test. Failure to follow directions in insert may yield inaccurate test results.
- The test line for a high viral load sample may become visible within 15 minutes, or as soon as the sample passes the test line region.
- The test line for a low viral load sample may become visible within 30 minutes.
- Avoid exposure of your skin, eyes, nose, or mouth to the solution in the tube. The reagent solution in the tube contains hazardous ingredients (see table below). If the solution contacts the skin or eye, flush with plenty of water. If irritation persists, seek medical advice.

Chemical Name/ Concentration	Harms (GHS) code for each ingredient	Concentration
Triton X-100	Acute Tox. 4 (H302) Skin Irrit. 2 (H315) Eye Irrit. 2 (H319)	1%
Sodium Azide	Acute Tox. 2 * (H300) Aquatic Acute 1 (H400) Aquatic Chronic 1 (H410)	0.02%

STORAGE AND STABILITY

- The kit can be stored at temperatures between 2 - 30 °C.
- The test is stable until the expiration date printed on the sealed pouch.
- The test must remain in the sealed pouch until use.
- DO NOT FREEZE.
- Do not use after the expiration date.

MATERIALS

Materials Provided

- Test Cassettes
- Positive Control Swab
- Disposable Swabs*
- Specimen Collection Guide
- Extraction Buffer Tubes
- Negative Control Swab
- Package Insert

* *The Disposable Swabs are produced by another manufacturer. Either Nasal swabs or nasopharyngeal swabs are supplied in the kit depending on the package you ordered.*

Materials Required But Not Provided

- Personal Protective Equipment
- Timer

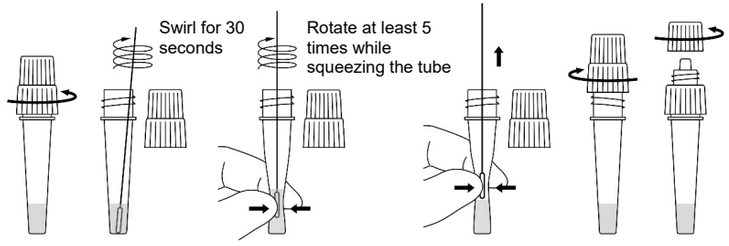
SPECIMEN COLLECTION AND PREPARATION

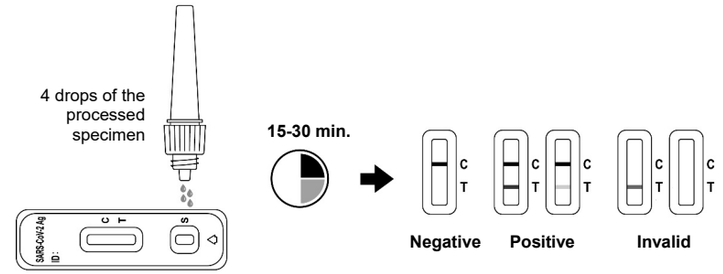
- The SARS-CoV-2 Antigen Rapid Test can be performed using nasal and nasopharyngeal swab specimens.
- Testing should be performed immediately after specimen collection, or at most within one (1) hour after specimen collection, if stored at room temperature (15-30°C).
- Please refer to the Specimen Collection Guide provided with the kit for specimen collection details.

DIRECTIONS FOR USE

Allow the test and extraction buffer to reach room temperature (15-30 °C) prior to testing.

1. Use an extraction buffer tube for each specimen to be tested and label each tube appropriately.
2. Unscrew the dropper cap from the extraction buffer tube without squeezing.
3. Insert the swab into the tube and swirl it for 30 seconds. Then rotate the swab at least 5 times while squeezing the sides of the tube. Take care to avoid splashing contents out of the tube.
4. Remove the swab while squeezing the sides of the tube to extract the liquid from the swab.
5. Screw the dropper cap firmly onto the extraction buffer tube containing the sample. Mix thoroughly by swirling or flicking the bottom of the tube.
6. Remove the test cassette from the foil pouch and use it as soon as possible.
7. Place the test cassette on a flat and clean surface.
8. Add the processed specimen to the sample well of the test cassette.
   a. Unscrew the small cap from the dropper tip.
   b. Invert the extraction buffer tube with the dropper tip pointing downwards and hold it vertically.
   c. Gently squeeze the tube, dispensing 4 drops of the processed specimen into the sample well.
9. Wait for the colored line(s) to appear. The result should be read at 15-30 minutes. **Do not read the result after 30 minutes.**





INTERPRETATION OF RESULTS

(Please refer to the illustration above)
NEGATIVE: Only one colored control line appears in the control region (C). No apparent colored line appears in the test line region (T). This means that no SARS-CoV-2 antigen was detected.
POSITIVE:* Two distinct colored lines appear. One line in the control line region (C) and the other line-in the test line region (T). This means that the presence of SARS-CoV-2 antigen was detected.
***NOTE:** The intensity of the color in the test line (T) may vary depending on the level of the SARS-CoV-2 antigen present in the specimen. Therefore, any shade of color in the test line region (T) should be considered positive.
INVALID: Control line fails to appear. Insufficient specimen volume or incorrect operation are the most likely reasons for control line failure. Review the procedure and repeat the test with a new test cassette. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

QUALITY CONTROL

Internal procedural controls are included in the test. A colored line appearing in the control line region (C) is an internal procedural control. It confirms sufficient specimen volume and correct procedural technique.
Positive and Negative control swabs are supplied with each kit. These control swabs should be used to ensure that the test cassette and that the test procedure is performed correctly. Follow the "**DIRECTIONS FOR USE**" section to perform the control test.
The control swabs can be tested under any of the following circumstances:
1. When new lot of tests are used and/or when a new operator performs the test.
2. At periodic intervals as dictated by local requirements, and/or by the user's Quality Control procedures.

LIMITATIONS

1. The SARS-CoV-2 Antigen Rapid Test is for *in vitro* diagnostic use only. The test should be used for the detection of SARS-CoV-2 antigens in nasal and nasopharyngeal swab specimens only. The intensity of the test line does not necessarily correlate to SARS-CoV-2 viral titer in the specimen.
2. Specimens should be tested as quickly as possible after specimen collection and at most within the hour following collection.
3. Use of viral transport media may result in decreased test sensitivity.
4. A false-negative test may result if the level of antigen in a sample is below the detection limit of the test or if the sample was collected incorrectly.
5. Test results should be correlated with other clinical data available to the physician.
6. A positive test result does not rule out co-infections with other pathogens.
7. A positive test result does not differentiate between SARS-CoV and SARS-CoV-2.
8. A negative test result is not intended to rule out other viral or bacterial infections.
9. The performance of the SARS-CoV-2 Antigen Rapid Test has not been assessed in a population vaccinated against COVID-19.
10. Laboratories may be required to report all positive results in accordance with any country-specific or public health authority requirements.
11. Use in conjunction with the testing strategy outlined by public health authorities in your area.
12. This test is not intended for home testing (or self-testing).
13. The performance has not been established with all circulating variants but is anticipated to be reflective of the prevalent variants in circulation at the time and location of the clinical evaluation. Performance at the time of testing may vary depending on the variants circulating, including newly emerging strains of SARS-CoV-2 and their prevalence, which change over time.
14. For asymptomatic patients:
    a) Clinical studies in asymptomatic patients using serial testing are ongoing to establish clinical performance.
    b) The performance of this test has not yet been clinically validated for use in patients without signs and symptoms of respiratory infection or for serial screening applications.
    c) Note that performance may differ in these populations.
15. The SARS-CoV-2 Antigen Rapid Test can detect both viable and non-viable SARS-CoV-2 material.

PERFORMANCE CHARACTERISTICS

Clinical Sensitivity, Specificity and Accuracy

Nasal Swab Specimens

The performance of SARS-CoV-2 Antigen Rapid Test was established with 605 nasal swabs collected from individual patients who were suspected of COVID-19. The results show that the relative sensitivity and the

relative specificity are as follows:

Method		RT-PCR (Nasopharyngeal Swab Specimens)		Total Results
SARS-CoV-2 Antigen Rapid Test (Nasal Swab Specimens)	**Results**	Negative	Positive	
	Negative	433	5	438
	Positive	2	165	167
Total Results		435	170	605

Relative Sensitivity: 97.1% (93.1%-98.9%)* Relative Specificity: 99.5% (98.2%-99.9%)*
Accuracy: 98.8% (97.6%-99.5%)* *95% Confidence Intervals

Stratification of the prospective positive samples post onset of symptoms between 0-3 days has a positive percent agreement (PPA) of 98.3% (n=60) and 4-7 days has a PPA of 96.0% (n=25).

Prospective positive samples with Ct value ⩽30 has a positive percent agreement (PPA) of 100% (n=73) and Ct value >30 has a positive percent agreement (PPA) of 81.0% (n=21).

Nasopharyngeal Swab Specimens

The performance of SARS-CoV-2 Antigen Rapid Test was established with 299 nasopharyngeal swabs collected from individual patients who were suspected of COVID-19. The results show that the relative sensitivity and the relative specificity are as follows:

Method		RT-PCR (Nasopharyngeal Swab Specimens)		Total Results
SARS-CoV-2 Antigen Rapid Test (Nasopharyngeal Swab Specimens)	**Results**	Negative	Positive	
	Negative	175	3	178
	Positive	1	120	121
Total Results		176	123	299

Relative Sensitivity: 97.6% (92.8% - 99.5%)* Relative Specificity: 99.4% (96.5% - 99.9%)*
Accuracy: 98.7% (96.5% - 99.6%)* *95% Confidence Intervals

Stratification of the prospective positive samples post onset of symptoms between 0-3 days has a positive percent agreement (PPA) of 100% (n=20) and 4-7 days has a PPA of 100% (n=24).

Prospective positive samples with Ct value ⩽30 has a positive percent agreement (PPA) of 100% (n=39) and Ct value >30 has a positive percent agreement (PPA) of 88.9% (n=9).

Limit of Detection (LOD)

The LOD of SARS-CoV-2 Antigen Rapid Test was established using limiting dilutions of an inactivated viral sample. The viral sample was spiked with negative human nasal and nasopharyngeal sample pool into a series of concentrations. Each level was tested for 30 replicates. The results show that the LOD is $1.6*10^2$ $TCID_{50}$/mL.*

* Base on the concentration of virus in extraction buffer

Cross-Reactivity (Analytical Specificity) and Microbial Interference

Cross-reactivity was evaluated by testing a panel of related pathogens and microorganisms that are likely to be present in the nasal cavity. Each organism and virus were tested in the absence or presence of heat-inactivated SARS-CoV-2 virus at low positive level. Both nasal swab specimens and nasopharyngeal swab specimens were tested.

No cross-reactivity or interference was observed with the following microorganisms when tested at the concentration presented in the table below. The SARS-CoV-2 Antigen Rapid Test does not differentiate between SARS-CoV and SARS-CoV-2.

Potential Cross-Reactant		Test Concentration	Cross-Reactivity (in the absence of SARS-CoV-2 virus)	Interference (in the presence of SARS-CoV-2 virus)
Virus	Adenovirus	1.14 x 10^6 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Enterovirus	9.50 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Human coronavirus 229E	1.04 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Human coronavirus OC43	2.63 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Human coronavirus NL63	1.0 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Human Metapneumovirus	1.25 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	MERS-coronavirus	7.90 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Influenza A	1.04 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Influenza B	1.04 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Parainfluenza virus 1	1.25 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Parainfluenza virus 2	3.78 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Parainfluenza virus 3	1.0 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Parainfluenza virus 4	2.88 x 10^6 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Respiratory syncytial virus	3.15 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
	Rhinovirus	3.15 x 10^5 $TCID_{50}$/mL	No 3/3 negative	No 3/3 positive
Bacteria	Bordetella pertussis	2.83 x 10^9 CFU/mL	No 3/3 negative	No 3/3 positive
	Chlamydia trachomatis	3.13 x 10^8 CFU/mL	No 3/3 negative	No 3/3 positive
	Haemophilus influenza	1.36 x 10^8 CFU/mL	No 3/3 negative	No 3/3 positive
	Legionella pneumophila	4.08 x 10^9 CFU/mL	No 3/3 negative	No 3/3 positive
	Mycobacterium tuberculosis	1.72 x 10^7 CFU/mL	No 3/3 negative	No 3/3 positive
	Mycoplasma pneumoniae	7.90 x 10^7 CFU/mL	No 3/3 negative	No 3/3 positive
	Staphylococcus aureus	1.38 x 10^7 CFU/mL	No 3/3 negative	No 3/3 positive
	Staphylococcus epidermidis	2.32 x 10^9 CFU/mL	No 3/3 negative	No 3/3 positive
	Streptococcus pneumoniae	1.04 x 10^8 CFU/mL	No 3/3 negative	No 3/3 positive
	Streptococcus pyogenes	4.10 x 10^6 CFU/mL	No 3/3 negative	No 3/3 positive
	Pneumocystis jirovecii-S. cerevisiae	8.63 x 10^7 CFU/mL	No 3/3 negative	No 3/3 positive
	Pseudomonas aeruginosa	1.87 x 10^8 CFU/mL	No 3/3 negative	No 3/3 positive
	Chlamydia pneumoniae	1×10^6 IFU/ml	No 3/3 negative	No 3/3 positive
Yeast	Candida albicans	1.57 x 10^8 CFU/mL	No 3/3 negative	No 3/3 positive
Pooled human nasal wash			No 3/3 negative	No 3/3 positive

To estimate the likelihood of cross-reactivity with SARS-CoV-2 of organisms that were not available for wet testing, in-silico analysis was used to assess the degree of protein sequence homology. The comparison between SARS-CoV-2 nucleocapsid protein and human coronavirus HKU1 revealed a low homology of 36.7% across 82.8% of the SARS-CoV-2 nucleocapsid sequence. The result suggests that cross-reactivity with human coronavirus HKU1 cannot be completely ruled out.

Interfering Substances

The following substances, naturally present in respiratory specimens or that may be artificially introduced into the nasal cavity or nasopharynx, were evaluated. Each substance was tested in the absence or presence of SARS-CoV-2 virus at low positive level. The final concentration of the substances tested are listed below and were found not to affect test performance.

Interfering Substance	Active Ingredient	Concentration	Results (in the absence of SARS-CoV-2 virus)	Results (in the presence of SARS-CoV-2 virus)
Endogenous	Biotin	2.4 mg/mL	3/3 negative	3/3 positive
	Mucin	0.5% w/v	3/3 negative	3/3 positive
	Whole Blood	4% v/v	3/3 negative	3/3 positive
Afrin Original Nasal Spray	Oxymetazoline	15% v/v	3/3 negative	3/3 positive
ALKALOL Allergy Relief Nasal Spray	Homeopathic	1:10 Dilution	3/3 negative	3/3 positive
Chloraseptic Max Sore Throat Lozenges	Menthol, Benzocaine	1.5 mg/mL	3/3 negative	3/3 positive
CVS Health Fluticasone Propionate Nasal Spray	Fluticasone propionate	5% v/v	3/3 negative	3/3 positive
Equate Fast-Acting Nasal Spray	Phenylephrine	15% v/v	3/3 negative	3/3 positive
Equate Sore Throat Phenol Oral Anesthetic Spray	Phenol	15% v/v	3/3 negative	3/3 positive
Original Extra Strong Menthol Cough Lozenges	Menthol	1.5 mg/mL	3/3 negative	3/3 positive
NasalCrom Nasal Spray	Cromolyn	15% v/v	3/3 negative	3/3 positive
NeilMed NasoGel for Dry Noses	Sodium Hyaluronate	5% v/v	3/3 negative	3/3 positive
Throat Lozenge	Dyclonine Hydrochloride	1.5mg/mL	3/3 negative	3/3 positive
Zicam Cold Remedy	Galphimia glauca, Luffa operculata, Sabadilla	5% v/v	3/3 negative	3/3 positive
Antibiotic	Mupirocin	10 mg/mL	3/3 negative	3/3 positive
Tamiflu	Oseltamivir Phosphate	5 mg/mL	3/3 negative	3/3 positive
Antibiotic	Tobramycin	4 µg/mL	3/3 negative	3/3 positive
Mometasone Furoate Nasal Spray	Mometasone Furoate	5%v/v	3/3 negative	3/3 positive
Physiological Seawater Nasal Cleaner	NaCl	15%v/v	3/3 negative	3/3 positive

PRECISION

Intra-Assay

Within-run precision was determined using 60 replicates of specimens: negative control and SARS-CoV-2 antigen positive controls. The specimens were correctly identified >99% of the time.

Inter-Assay

Between-run precision was determined using 60 independent assays on the same specimen: negative specimen and SARS-CoV-2 antigen positive specimen. Three different lots of the SARS-CoV-2 Antigen Rapid Test were tested using these specimens. The specimens were correctly identified >99% of the time.

High Dose Hook Effect

No high dose hook effect was observed when tested with up to a concentration of 1.43 x 10^5 $TCID_{50}$/mL of heat-inactivated SARS-CoV-2 virus with the SARS-CoV-2 Antigen Rapid Test.

POC STUDY

A total of 9 operators from 3 sites performed the SARS-CoV-2 Antigen Rapid Test on 60 blinded labeled specimens by following the instructions of the package insert and recorded the results on data sheet. Based on the results of this POC clinical evaluation, untrained operators with various background and experience levels can perform the SARS-COV-2 Antigen Rapid Test correctly after read the product package insert without other training. The untrained operators found that the test procedure described in the package insert is simple to follow.

BIBLIOGRAPHY

1. Shuo Su, Gary Wong, Weifeng Shi, et al. Epidemiology, Genetic recombination, and pathogenesis of coronaviruses. Trends in Microbiology, June 2016, vol. 24, No. 6: 490-502
2. Susan R. Weiss, Julian L. Leibowitz, Coronavirus Pathogenesis, Advances in Virus Research, Volume 81: 85-164

Index of Symbols

	Manufacturer	Σ	Contains sufficient for <*n*> tests		Temperature limit
IVD	*In vitro* diagnostic medical device		Use-by date		Do not reuse
	Consult instructions for use	LOT	Batch code	REF	Catalogue number
	Date of manufacture				

***ACON* Biotech (Hangzhou) Co., Ltd.**
No.210 Zhenzhong Road, West Lake District
Hangzhou, P.R. China, 310030

Number: 1151430502
Effective Date: 2022-02-28