Flow*flex*™ COVID-19 Antigen Home Test Package Insert for Healthcare Providers

REF L031-118B5	REF L031-125M5	REF L031-125N5	REF L031-125P5	English

A rapid test for the detection of SARS-CoV-2 nucleocapsid antigens in anterior nasal swab specimens.
For in vitro diagnostic use only. For Emergency Use Authorization only.

INTENDED USE

The Flow*flex* COVID-19 Antigen Home Test is a rapid lateral flow immunoassay intended for the qualitative detection of the nucleocapsid protein antigen from SARS-CoV-2.
This test is authorized for non-prescription home use with self-collected anterior nasal (nares) swab samples from individuals aged 14 years or older with symptoms of COVID-19 within 7 days of symptom onset. This test is also authorized for non-prescription home use with adult-collected anterior nasal (nares) swab samples from individuals aged 2 years or older with symptoms of COVID-19 within the first 7 days of symptom onset.
This test is also authorized for non-prescription home use with self-collected anterior nasal (nares) swab samples from individuals aged 14 years or older, or adult-collected anterior nasal swab samples from individuals aged 2 or older, with or without symptoms or other epidemiological reasons to suspect COVID-19 when tested twice over three days with at least 24 hours (and no more than 48 hours) between tests.
The Flow*flex* COVID-19 Antigen Home Test does not differentiate between SARS-CoV and SARS-CoV-2.
Results are for the identification of SARS-CoV-2 nucleocapsid protein antigen. Antigen is generally detectable in nasal (nares) 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 and the agent detected may not be the definite cause of disease.
Individuals who test positive with the Flow*flex* COVID-19 Antigen Home Test should self-isolate and consult their physician or healthcare provider as additional testing may be necessary.
Negative results are presumptive, and confirmation with a molecular assay, if necessary for patient management, may be performed. 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 an individual's recent exposures, history and the presence of clinical signs and symptoms consistent with COVID-19.
For serial testing programs, additional confirmatory testing with a molecular test for negative results may be necessary, if there is a high likelihood of SARS-CoV-2 infection, such as in an individual with a close contact with COVID-19 or with suspected exposure to COVID-19 or in communities with high prevalence of infection. Additional confirmatory testing with a molecular test for positive results may also be necessary, if there is a low likelihood of SARS-CoV-2 infection, such as in individuals without known exposures to COVID-19 or residing in communities with low prevalence of infection.
Individuals who test negative and continue to experience COVID-19 like symptoms of fever, cough and/or shortness of breath may still have SARS-CoV-2 infection and should seek follow up care from their healthcare provider.
Individuals should provide all results obtained with this product to their healthcare provider for public health reporting.
Healthcare providers will report all test results they received from individuals who use the authorized product to relevant public health authorities in accordance with local, state, and federal requirements using appropriate LOINC and SNOMED codes, as defined by the Laboratory In Vitro Diagnostics (LIVD) Test Code Mapping for SARS-CoV-2 Tests provided by CDC.
The Flow*flex* COVID-19 Antigen Home Test is intended for non-prescription self-use and/or as applicable an adult lay user testing another aged 2 or older. The Flow*flex* COVID-19 Antigen Home Test is only for use under the Food and Drug Administration's Emergency Use Authorization.

SUMMARY AND EXPLANATION

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 Flow*flex* COVID-19 Antigen Home Test is a qualitative membrane based chromatographic immunoassay for the qualitative detection of the nucleocapsid protein antigen from SARS-CoV-2 in human anterior nasal 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 colored anti-SARS-CoV-2 antibody-coated particles, which have been pre-coated on the test strip. The antigen-antibody complex then migrates toward the membrane by capillary action. This complex is then captured by anti-SARS-CoV-2 monoclonal antibodies immobilized at the test line region, and a colored line appears 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 red or pink line will always appear in the control line region after proper volume of specimen has been added, and membrane wicking has occurred.

REAGENTS

The test cassette contains anti-SARS-CoV-2 antibodies.

PRECAUTIONS

- Read the COVID-19 Antigen Home Test Package Insert carefully before performing a test. Follow directions for use. Failure to follow directions may produce inaccurate test results.
- For *in vitro* diagnostic use.
- This product has not been FDA cleared or approved but has been authorized by FDA under an EUA.
- This product has been authorized only for the detection of proteins from SARS-CoV-2, not for any other viruses or pathogens.
- The emergency use of this product is only authorized for the duration of the declaration that circumstances exist justifying the authorization of emergency use of in vitro diagnostics for detection and/or diagnosis of COVID-19 under Section 564(b)(1) of the Federal Food, Drug and Cosmetic Act, 21 U.S.C. § 360bbb-3(b)(1), unless the declaration is terminated, or authorization is revoked sooner.
- This product has been designed only for the detection of SARS-CoV-2 antigen, not for any other viruses or pathogens.
- False negative test results may occur if a specimen is incorrectly collected or handled.
- To obtain accurate results, the test must be performed as indicated in this Instructions for Use.
- **INVALID RESULTS**, indicated by no Control Line, can occur when an insufficient volume of sample solution is added to the test cassette. Gently squeeze the tube and dispense 4 drops of solution into the sample well of test cassette.
- Swabs in the kit are approved for use with Flow*flex* COVID-19 Antigen Home Test. Do not use other swabs.
- Do not use on anyone under two years of age. Keep test kit and materials out of the reach of children and pets, before and after use.
- Children aged 2 to 13 years of age should be tested by an adult.
- Wear a safely mask or other face-covering when collecting a specimen from a child or another individual.
- Leave the test cassette sealed in its pouch until just before use. Once opened, the test cassette should be used within 60 minutes.
- Do not use the test after the expiration date shown on the test cassette pouch.
- Do not use if any of the test kit contents or packaging is damaged or open.
- Test components are single use. Do not re-use. Do not use with multiple specimens.
- Make sure there is sufficient light when reading and interpreting test results.
- Do not use nasal sprays for at least 30 minutes before collecting a nasal sample.
- Remove any piercings from the nose before starting the test. Do not use on anyone who is prone to nosebleeds or has had facial injuries or head injuries/surgery in the past six months.
- Inadequate or improper nasal swab sample collection may result in false negative test results.
- Do not touch the swab tip when handling the swab.
- Test samples immediately after collection, and no more than one hour after the swab is added to the reagent solution, if stored at room temperature.
- The test is intended to be read at 15 minutes. If the test is read before 15 minutes or after 30 minutes, false negative or false positive results may occur, and the test should be repeated with a new test cassette.
- Avoid exposure of your skin, eyes, nose, or mouth to the solution in the extraction tube.
- Do not ingest any kit components.
- The reagent solution in the tube contains a harmful chemical (see table below). If the solution contacts the skin or eye, flush with plenty of water. If irritation persists, seek medical advice.
  https://www.poisonhelp.org or 1-800-222-1222

Hazardous Ingredients for the Reagent Solution		
Chemical Name/ Concentration	Harms (GHS) code for each ingredient	Concentration
TX-100	Acute toxicity, Oral (Category 4), H302 Skin irritation (Category 2), H315 Serious eye damage (Category 1), H318 Short-term (acute) aquatic hazard (Category 1), H400 Long-term (chronic) aquatic hazard (Category 1), H410	1%
Sodium Azide	Acute toxicity, Oral (Category 2), H300 Acute toxicity, Dermal (Category 1), H310 Specific target organ toxicity - repeated exposure, Oral (Category 2), Brain, H373 Short-term (acute) aquatic hazard (Category 1), H400 Long-term (chronic) aquatic hazard (Category 1), H410	0.02%

If INHALATION: Move to fresh air. If not breathing, give artificial respiration. Do not use mouth-to-mouth method if victim ingested or inhaled; give artificial respiration with the aid of a pocket mask equipped with a one-way valve or other proper respiratory medical device. Immediate medical attention is required.
If SKIN Contact: Take off immediately all contaminated clothing. Wash off immediately with plenty of water for at least 15 minutes. Immediate medical attention is required.
If EYE Contact: Immediately flush eyes with plenty of water for at least 15 minutes. Assure adequate flushing by separating the eyelids with fingers. Get medical attention immediately.
If INGESTION: Clean mouth with water. Do not induce vomiting. Risk of aspiration! Keep airways free. Pulmonary failure possible after aspiration of vomit. Call a physician or Poison Control Center immediately.

STORAGE AND STABILITY

- The kit can be stored at temperatures between 36-86°F (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
- Disposable Nasal Swabs
- Extraction Buffer Tubes
- Tube Holder (only for 25 test quantity)
- Package Insert

***Note**: This test comes in a 1 test (REF: L031-118B5), 2 test (REF: L031-125M5), 5 test (REF: L031-125N5), 25 test (REF: L031-125P5) quantity. The number of items supplied in the kit will vary depending on which kit was purchased.*

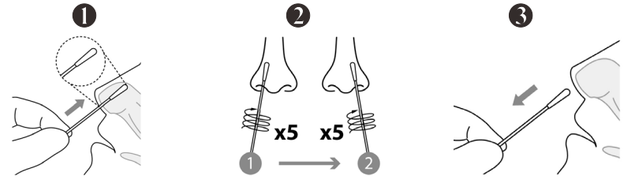
Materials Required But Not Provided

- Timer
- Flow*flex* Web App (Optional) - if using the Web App, ensure you have an internet connection and go to www.flowflexcovid.com prior to starting the test. Ensure you are using a compatible web browser (Chrome, Firefox, or Safari) and your electronic device has a camera.

SPECIMEN COLLECTION AND PREPARATION

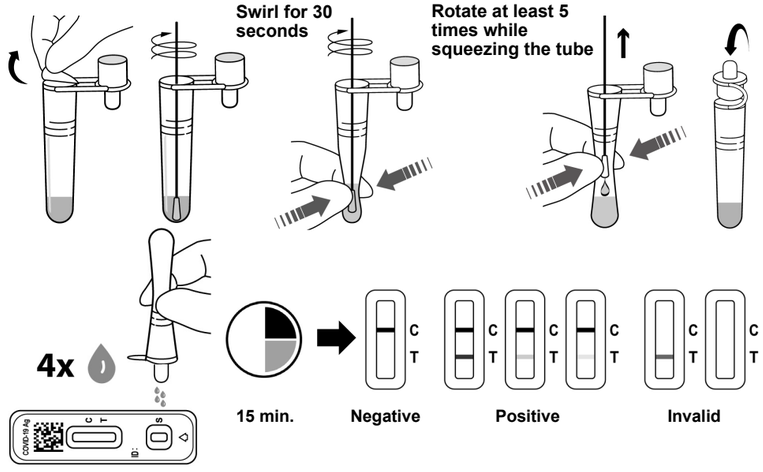
- The Flow*flex* COVID-19 Antigen Home Test is performed using anterior nasal swab specimens.
- Wash or sanitize your hands. Make sure they are dry before starting the test.
- Open the test cassette pouch and lay the cassette on a clean, flat surface.
- To collect an anterior nasal swab sample:

1. Gently insert the entire absorbent tip of the swab into 1 nostril (½ to ¾ of an inch). With children, the maximum depth of insertion into the nostril may be less than ¾ of an inch, and you may need to have a second person to hold the child's head while swabbing.
   Note: A false negative result may occur if the nasal swab specimen is not properly collected.
2. Firmly rub the swab in a circular motion around the inside wall of the nostril 5 times. Take approximately 15 seconds to collect the specimen. Be sure to collect any nasal drainage that may be present onto the swab. Repeat this in the other nostril using the same swab.
3. Remove the swab from the nostril and place into the extraction buffer tube.



DIRECTIONS FOR USE

1. Punch through the perforated circle on the kit box to form a tube holder. For 25 test quantity kit box the tube holder is included.
2. Remove the foil from the top of the extraction buffer tube. Place the tube in the tube holder.
3. Immediately place the swab into the tube and swirl for 30 seconds.
4. Rotate the swab 5 times **while squeezing the tube**.
5. Remove the swab **while squeezing the tube** to extract as much liquid as possible. Dispose the swab in the trash.
6. Attach the dropper tip firmly onto the tube. Mix thoroughly by swirling or flicking the bottom of the tube.
   Note: A false negative result may occur if the swab is not swirled at least 30 seconds or rotated 5 times.
7. Gently squeeze the tube and dispense **4 drops** of solution into the Sample Well. Dispose the tube in the trash.
   Note: A false negative or invalid result may occur if less than 4 drops of fluid are added to the Sample Well.
8. Set the timer for 15 minutes. Result should be read at 15 minutes. Do not read after 30 minutes. Dispose the test cassette in the trash.
   Note: A false negative or false positive result may occur if the test result is read before 15 minutes or after 30 minutes.



INTERPRETATION OF RESULTS

(Please refer to the illustration above)

NEGATIVE: Only the control line (C) and no test line (T) appears. This means that no SARS-CoV-2 antigen was detected. A negative test result indicates that antigens from the virus that causes COVID-19 were not detected from the specimen. Negative results do not rule out COVID-19. Individuals without symptoms that test negative should be tested again with at least 24 hours and no more than 48 hours between tests. All negative results are considered presumptive, and confirmation with a molecular assay, if necessary for patient management, may be performed.
POSITIVE:* Two red or pink lines appear in the test window, one on the test line position (T) and the other on the control line position (C). A positive test is interpreted as protein antigen from the virus that causes COVID-19 was detected in the specimen. The individual is positive for COVID-19. Test results should be considered in association with the patient's history and epidemiological data (such as local prevalence rates and current outbreak/epicenter locations).
*** NOTE:** The test line (red or pink line) may vary in shade and intensity (light or dark, weak or strong) depending on the concentration of antigen present in the sample. The intensity of the Control line should not be compared to that of the Test line for the interpretation of the test result. Any faint visible red or pink Test line should be interpreted as positive, when the control line (C) is also present.
INVALID: Control line fails to appear. If a line does not appear on the control line position (C) in 30 minutes, the test result is invalid. Re-test with a new Flowflex COVID-19 Antigen Home Test.

QUALITY CONTROL

Internal procedural controls are included in the test. A red or pink line appearing in the control line region (C) is an internal procedural control. The appearance of the procedural control line indicates that proper volume of specimen has been added and capillary flow occurred. If the procedural control line does not develop in 15 minutes, the test result is considered invalid, and retesting with a new cassette is recommended.

LIMITATIONS

1. The performance of this test was established based on the evaluation of a limited number of clinical specimens collected between March, 2021 and May, 2021. The clinical performance has not been established in 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.

2. Specimens should be tested as quickly as possible after specimen collection.
3. Failure to follow the instructions for use may adversely affect test performance and/or invalidate the test result.
4. A false negative result may occur if the level of antigen in a sample is below the detection limit of the test.
5. A false negative result may occur if the sample was collected incorrectly or handled.
6. A false negative result may occur if the swab is not swirled at least 30 seconds or rotated five times
7. A false negative or invalid result may occur if less than 4 drops of fluid are added to the Sample Well.
8. A false negative or false positive result may occur if the test result is read before 15 minutes or after 30 minutes
9. This test detects both viable (live) and non-viable, SARS-CoV, and SARS-CoV-2. Test performance depends on the amount of virus (antigen) in the sample and may or may not correlate with viral culture results performed on the same sample.
10. The Flow*flex* COVID-19 Antigen Home Test does not differentiate between SARS-CoV and SARS-CoV-2.
11. Test results should be correlated with other clinical data available to the physician.
12. A positive or negative test result does not rule out co-infections with other pathogens such as other viral or bacterial infections.
13. Negative results are presumptive, do not rule out COVID-19 and it may be necessary to obtain additional testing with a molecular assay, if needed for patient management.
14. A negative test result is not intended to rule out other viral or bacterial infections.
15. If the differentiation of specific SARS viruses and strains is needed, additional testing is required, in consultation with state or local public health departments, is required. The performance of this test was established based on the evaluation of a limited number of clinical specimens collected between March and May 2021. Clinical 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.
16. Testing for asymptomatic individuals should be performed at least twice over three days, with at least 24 hours and no more than 48 hours between tests. You may need to purchase additional tests to perform this serial (repeat) testing.
17. Serial testing (i.e., testing every day or every other day) is more likely to detect COVID-19, especially in individuals that do not have any symptoms.

PERFORMANCE CHARACTERISTICS

Clinical Sensitivity, Specificity and Accuracy

The performance of Flow*flex* COVID-19 Antigen Home Test was established in an all-comers clinical study conducted between March 2021 and May 2021 with 108 nasal swabs self-collected or pair-collected by another study participant from symptomatic patients (within 7 days of onset) suspected of COVID-19. The study was conducted in a simulated home setting environment at two study sites in U.S. All study participants performed the test unassisted and interpreted the result, using only the product labeling. The Flow*flex* COVID-19 Antigen Home Test results were compared to an FDA EUA RT-PCR COVID-19 assay to determine test performance in the tables below:

Table 1. Performance of the Flow*flex* COVID-19 Antigen Home Test in Symptomatic subjects

Flow*flex* COVID-19 Antigen Home Test	RT-PCR method		
	Positive	Negative	Total
Positive	28	0	28
Negative	2	78	80
Total	30	78	108
Positive Percent Agreement (PPA)	93% (95%CI: 78% - 99%)		
Negative Percent Agreement (NPA)	100% (95%CI: 95% - 100%)		

Table 2. Cumulative PPA results by days since symptom onset

Days Since Symptom Onset	# Specimens Tested	# Cumulative Positive Flow*flex* COVID-19 Antigen Home Test	Cumulative Positive RT-PCR	Cumulative PPA
0 to 1 day	29	6	7	86%
0 to 2 days	64	15	16	94%
0 to 3 days	90	20	21	95%
0 to 4 days	96	21	22	95%
0 to 5 days	100	23	24	96%
0 to 6 days	106	26	28	93%
0 to 7 days	108	28	30	93%

Symptomatic Patient Age Distribution:

A total of 108 symptomatic patients participated in the study. Ages of symptomatic patients ranged from 2 years to 93 years. The table below shows age distribution and the positive results broken down by age of the symptomatic patient:

Table 3. Age distribution of patients and specimen positivity

Age Group	Flow*flex* COVID-19 Antigen Home Test (N=175)		
	Total	Total Positive	Prevalence
2-13 years	8	1	13%
14- 24 years	12	4	33%
25- 64 years	67	21	31%
⩾ 65 years	21	2	10%
Total	108	28	26%

Analytical Sensitivity: Limit of Detection (LoD)

The Limit of Detection (LoD) of the Flow*flex* COVID-19 Antigen Home Test was determined using limiting dilutions of the heat-inactivated SARS-CoV-2 virus (USA-WA1/2020). Nasal swabs from healthy donors were collected and eluted with PBS. The swab eluates were combined and mixed thoroughly to create a negative clinical matrix pool to be used as the diluent. Inactivated SARS-CoV-2 virus was diluted in this negative clinical matrix pool to generate virus dilutions for testing.

The contrived nasal swab samples were prepared by absorbing 50 µL of each virus dilution onto the swab. The contrived swab samples were processed and tested according to the package insert.

SARS-CoV-2 Concentration in nasal matrix	Number of Positives/Total	% Detected
2.5 x 10^3 $TCID_{50}$/mL	60/60	100%

LoD was determined as the lowest virus concentration that was detected ⩾ 95% of the time.

Based on this testing, the LoD in nasal matrix was confirmed to be 2.5 x 10^3 $TCID_{50}$/mL.

The performance of this test device in the detection of the Omicron variant of SARS-CoV-2 was evaluated in a dilution series of clinical specimens which were positive for the Omicron variant. This testing was conducted by the National Institutes of Health (NIH) as a component of the Rapid Acceleration of Diagnostics (RADx) initiative. The clinical specimens used to prepare this dilution series were not identical to the previous specimen pools prepared and tested by RADx to assess performance with the omicron variant. Results from this dilution series cannot be compared to other specimen pools and do not indicate that a test will have different clinical performance compared to other EUA authorized tests. Compared to an EUA authorized RT-PCR method, the Flow*flex* COVID-19 Antigen Home Test detected 100% of live virus Omicron samples at a Ct-value of 25.6 (n=5). Testing was also compared to two additional EUA-authorized OTC antigen tests (Assay #1 and Assay #2). Omicron dilutions at lower viral concentrations (Ct-values greater than 25.6) were not detected by the Flow*flex* COVID-19 Antigen Home Test in this study.

Omicron BA.2 Pool 1 Dilutions	Avg (N=9)	Assay #1 Percent Positive N=5	ACON Flow*flex* Percent Positive N=5	Assay #2 Percent Positive N=5
Dilution 1	19.4	100	100	100
Dilution 2	20.6	100	100	100
Dilution 3	21.6	100	100	100
Dilution 4	22.4	100	100	100
Dilution 5	23.3	100	100	100
Dilution 6	24.5	0	100	100
Dilution 7	25.6	0	100	100
Dilution 8	26.5	0	0	0
Dilution 9	27.7	0	0	0
Dilution 10	28.5	0	0	0
Dilution 11	29.4	0	0	0
Dilution 12	30.3	0	0	0

Analytical Specificity: Cross-Reactivity 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 (USA-WA1/2020) at a low concentration.

No cross-reactivity or interference was observed with the following organisms when tested at the concentration presented in the table below.

Potential Cross Reactant		Test Concentration	Cross-Reactivity Results	Interference Results
Virus	Adenovirus	1.14 x 10^6 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Enterovirus	9.50 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Human coronavirus 229E	1.04 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Human coronavirus OC43	2.63 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Human coronavirus NL63	1.0 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Human Metapneumovirus	1.25 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	MERS-coronavirus	7.90 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Influenza A	1.04 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Influenza B	1.04 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Parainfluenza virus 1	1.25 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Parainfluenza virus 2	3.78 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Parainfluenza virus 3	1.0 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Parainfluenza virus 4	2.88 x 10^6 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Respiratory syncytial virus	3.15 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
	Rhinovirus	3.15 x 10^5 $TCID_{50}$/mL	No cross-reactivity	No Interference
Bacteria	*Bordetella pertussis*	2.83 x 10^9 CFU/mL	No cross-reactivity	No Interference
	Chlamydia pneumonia	3.5 x 10^7 IFU/mL	No cross-reactivity	No Interference
	Chlamydia trachomatis	3.13 x 10^8 CFU/mL	No cross-reactivity	No Interference
	Haemophilus influenzae	1.36 x 10^8 CFU/mL	No cross-reactivity	No Interference
	Legionella pneumophila	4.08 x 10^9 CFU/mL	No cross-reactivity	No Interference
	Mycobacterium tuberculosis	1.72 x 10^7 CFU/mL	No cross-reactivity	No Interference
	Mycoplasma pneumoniae	7.90 x 10^7 CFU/mL	No cross-reactivity	No Interference
	Staphylococcus aureus	1.38 x 10^7 CFU/mL	No cross-reactivity	No Interference
	Staphylococcus epidermidis	2.32 x 10^9 CFU/mL	No cross-reactivity	No Interference
	Streptococcus pneumoniae	1.04 x 10^8 CFU/mL	No cross-reactivity	No Interference
	Streptococcus pyogenes	4.10 x 10^6 CFU/mL	No cross-reactivity	No Interference
	Pneumocystis jirovecii-S. cerevisiae	8.63 x 10^7 CFU/mL	No cross-reactivity	No Interference
	Pseudomonas aeruginosa	1.87 x 10^8 CFU/mL	No cross-reactivity	No Interference
Yeast	*Candida albicans*	1.57 x 10^8 CFU/mL	No cross-reactivity	No Interference

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.

Compared the sequence homology between the SARS-CoV-2 nucleocapsid protein and the structural proteins of SARS coronavirus (SARS-CoV) and with given the substantial homology rate (91.5%), there is high probability of cross-reactivity between the nucleocapsid proteins of SARS-CoV-2 and SARS-CoV. The Flow*flex* COVID-19 Antigen Home Test does not differentiate between SARS-CoV and SARS-CoV-2.

Endogenous Interfering Substances

The following substances, naturally present in respiratory specimens or that may be artificially introduced into the nasal cavity or nasopharynx, were evaluated. In addition to the materials that are found in the nasal cavity, substances that are commonly found on the hands were also tested. Each substance was tested in the absence or presence of SARS-CoV-2 virus (USA-WA1/2020) at a low concentration. The performance of Flow*flex* COVID-19 Antigen Home Test was not affected by any of the potentially interfering substances listed in the table below at the concentrations tested.

Interfering Substance	Source/Item	Test Concentration	Cross-Reactivity Results	Interference Results
Biotin	Sigma/ B4501	3500 ng/mL	No cross-reactivity	No interference
Chloraseptic Throat Lozenge (Menthol/Benzocaine)	Chloraseptic	1.5 mg/mL	No cross-reactivity	No interference
Cough Lozenge (Menthol)	Ricola	1.5 mg/mL	No cross-reactivity	No interference
Dyclonine Hydrochloride	Sigma/PHR1849	1.5mg/mL	No cross-reactivity	No interference
Fluticasone propionate	Flonase	5% v/v	No cross-reactivity	No interference
Mucin	Sigma/M3895	0.5% w/v	No cross-reactivity	No interference
Mupirocin	Sigma/M7694	10 mg/mL	No cross-reactivity	No interference
Nasal Drops (Phenylephrine)	Equate (Walmart)	15% v/v	No cross-reactivity	No interference
Nasal Spray (Cromolyn)	NasalCrom	15% v/v	No cross-reactivity	No interference
Nasal Spray (Homeopathic)	ALKALOL	1:10 Dilution	No cross-reactivity	No interference
Nasal Spray (Oxymetazoline HCl)	Afrin	15% v/v	No cross-reactivity	No interference
Naso GEL (NeilMed)	NeilMed	5% v/v	No cross-reactivity	No interference
Sore Throat Phenol Spray	Equate (Walmart)	15% v/v	No cross-reactivity	No interference
Tamiflu (Oseltamivir Phosphate)	Tamiflu	5 mg/mL	No cross-reactivity	No interference
Tobramycin	Sigma/LRAC4285	4 µg/mL	No cross-reactivity	No interference
Whole Blood	In-house	4% v/v	No cross-reactivity	No interference
Zicam	Zicam	5% v/v	No cross-reactivity	No interference

Potential Interfering Household Items	Source /Item	Test Concentration	Cross-Reactivity Results	Interference Results
Body & Hand Lotion	Aveeno	0.5% w/v	No cross-reactivity	No interference
Body Lotion, with 1.2% dimethicone	Aveeno	0.5% w/v	No cross-reactivity	No interference
Hand Lotion	Bath & Body	5% w/v	No cross-reactivity	No interference
Hand Sanitizer with Aloe, 62% ethyl alcohol	Hand in Hand	5% v/v	No cross-reactivity	No interference
Hand Sanitizer cream lotion	Dove	15% v/v	No cross-reactivity	No interference
Hand Sanitizer, 80% ethanol, fast drying	Allied Photo Chemical	15% w/v	No cross-reactivity	No interference
Hand soap liquid gel	SoftSoap	10% w/v	No cross-reactivity	No interference

High Dose Hook Effect

No high dose hook effect was observed when tested with up to a concentration of 1.0 x 10^6 $TCID_{50}$/mL of heat-inactivated SARS-CoV-2 virus (USA-WA1/2020) with the Flow*flex* COVID-19 Antigen Home Test.

Usability Study

A total of 431 subjects were enrolled in the study and were instructed to self-collect or collect a sample from a child, complete the required procedural steps, and interpret the test results unassisted in a simulated home-setting. The overall success of every task completed by all subjects enrolled was determined by unassisted professional observation. Subjects performed 96.2% (409/425) of steps/tasks correctly compared to healthcare professional users. After the completion of the test, the subject (or Parent/Legal Guardian) completed a test usability and satisfaction questionnaire. Specifically, 98.8% of subjects indicated that it was easy to see and understand the test results. Untrained lay users missed 7.9% of results compared to a healthcare provider, suggesting that lay users should carefully inspect the test cassette for faint lines. The Invalid Test Rate for the clinical study: the overall invalid result rate was 0% (0/172), this indicated that all the users had added sufficient sample volume (4 drops) onto the test cassettes.

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

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Number: 1151390304
Effective Date: 2022-06-21