aptitude®

Metrix® COVID-19 Test

Instructions for Use
For Healthcare Providers

For *in vitro* diagnostic use.
For use under Emergency Use Authorization (EUA) only.
For use with the Metrix® Reader or Metrix Reader (Gen 2), sold separately.

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1. Intended Use

The Metrix® COVID-19 Test (Metrix) is a single-use molecular in vitro diagnostic test for the qualitative detection of nucleic acid from SARS-CoV-2, the virus that causes COVID-19. This test is authorized for non-prescription home use with anterior nasal (nares) swab and saliva specimens, self-collected from any individual aged 14 years or older, or adult-collected from any individual aged 2 years or older, including individuals without symptoms or other epidemiological reasons to suspect COVID-19.

This test utilizes nucleic acid amplification technology, similar to PCR, for the detection of SARS-CoV-2. SARS-CoV-2 viral RNA is generally detectable in anterior nasal (nares) swab and saliva samples during the acute phase of infection.

Positive results indicate the presence of viral RNA, but clinical correlation with past medical 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. Individuals who test positive with the Metrix COVID-19 Test should self-isolate and seek follow-up care with their physician or healthcare provider as additional testing may be necessary.

Negative results from saliva samples are presumptive and should be confirmed by molecular testing of an alternative sample type if clinically indicated. Negative results do not preclude SARS-CoV-2 infection and should not be used as the sole basis for treatment or management decisions for the individual, 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. 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 with their physician or healthcare provider.

Individuals should report all results obtained with this product to their healthcare provider and the Aptitude secure web portal. This Aptitude secure web portal will report all test results 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 the CDC.

The Metrix COVID-19 Test is only for use under the Food and Drug Administration's Emergency Use Authorization.

2. Summary and Explanation of the Test

Coronavirus disease 2019 (COVID-19) is a pandemic respiratory illness caused by a novel human coronavirus named severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) by the World Health Organization.[1-3] SARS-CoV-2 has spread globally and can cause mild to severe respiratory illness, including death. Patients can become infected with SARS-CoV-2 virus through contact with a contaminated environment or person.

The Metrix COVID-19 Test is a molecular *in vitro* diagnostic test for detecting SARS-CoV-2 RNA in saliva or nasal swab specimens from individuals with or without symptoms or other epidemiological reasons to suspect infection with SARS-CoV-2.

3. Principles of the Procedure

The Metrix COVID-19 Test is a compact nucleic acid amplification test (NAAT) that detects the genetic material of SARS-CoV-2 in unprocessed saliva or nasal swab specimens using an isothermal molecular amplification reaction that is an equivalent alternative to polymerase chain reaction (PCR). The Metrix COVID-19 test utilizes a multi-gene amplification method utilizing primers that target both the nucleocapsid (N) and open reading frame (ORF-1) genes of SARS-CoV-2. The Metrix COVID-19 Test also identifies nucleic acids from a human gene that serves as a control for proper assay execution, sample inhibition, amplification, and assay reagent function.

The consumable Metrix COVID-19 Test kit consists of four components: Collector, Cap, Sensor, and optional Nasal Swab. The Metrix COVID-19 Test kit is used with the reusable Metrix Reader or Metrix Reader (Gen 2), sold separately. To prepare for a test, a saliva or anterior nasal swab sample is first placed in the Collector. Then, the Cap is attached to the Collector to release neutralization buffer (NB) from the Cap to mix with the sample. The NB is designed to lyse SARS-CoV-2 and cells in the sample and release nucleic acids for downstream detection. The Collector is connected to the Sensor to allow the sample to mix with reagents stored in the Sensor. Finally, the Sensor is inserted into the Metrix Reader to initiate the reaction and detect the presence of SARS-CoV-2 RNA.

The system utilizes an electrochemical reporting technology to monitor the amplified double-stranded cDNA concentration in real-time in respective reaction chambers. The amplification creates a characteristic electrochemical signal analogous to the fluorescence signal of a laboratory PCR reaction. The electrochemical signal is analyzed automatically by the Metrix Reader, and the test result is reported as a combination of colors and positions of LED lights on the Metrix Reader. A result is obtained in 30 minutes or less.

A positive result occurs when the SARS-CoV-2 channel passes the detection threshold. A negative result occurs when only the internal control channel passes the detection threshold. An invalid result occurs when both SARS-CoV-2 and internal control channels fail to pass the detection threshold.

4. Materials

4.1 Materials Provided

Each Metrix COVID-19 Test comes with sufficient material to perform 1 or 25 tests (as specified on the outer packaging). Each of the following components is for single use only:

- Cap
- Collector
- Swab (optional)
- Sensor
- Quick Reference Instructions (QRI)

4.2 Materials Required but Not Provided in Test Kit

Each Metrix COVID-19 Test requires:

- Metrix Reader OR
- Metrix Reader (Gen 2)

5. Warnings and Precautions

5.1 General

- For *in vitro* diagnostic use.
- For non-prescription home use.

- For use under Emergency Use Authorization (EUA) only.
- This product has not been FDA cleared or approved, but has been authorized for emergency use by FDA under an EUA.
- This product has been authorized only for the detection of nucleic acid 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.
- Follow all instructions carefully. Correct use is required for accuracy.
- Only use the test components provided. Do not re-use any components for another test. Only the Metrix Reader and the Metrix Reader (Gen 2) may be re-used multiple times.
- Positive results are indicative of the presence of SARS-CoV-2 RNA.
- Do not use if kit is visibly damaged.

5.2 Storage and Handling

- Store all components at 59 °F to 86 °F (15 °C to 30 °C).
- Do not open kit components until you are ready to perform testing.
- Samples should be tested as quickly as possible after collection.
- Do not use the Metrix COVID-19 Test past the Use By date.
- Do not use components that are visibly damaged.
- All components other than the Metrix Reader and Metrix Reader (Gen 2) are single-use and should be disposed of after use.
- Treat all biological specimens, including used test components, as if capable of transmitting infectious agents. Because it is often impossible to know which might be infectious, all biological specimens should be handled using standard precautions.

Guidelines for specimen handling are available from the U.S. Centers for Disease Control and Prevention and the Clinical and Laboratory Standards Institute.[4-6]

- Do not disassemble the Metrix COVID-19 Test Cap, Collector, Sensor, or any assemblies.
- Touch only the handle of the Swab with your hands to avoid contaminating the soft tip.
- Do not eat, drink, smoke or use oral hygiene products within 30 minutes of providing saliva a specimen for this test.
- Do not ingest any contents of this kit. Keep out of reach of children.
  Avoid contact with skin and eyes.

6. Operating Conditions

- The test should be used between 59 °F and 86 °F (15 °C and 30 °C).
- The test is best used in a room with adequate lighting away from glare.
- The Metrix Reader and Metrix Reader (Gen 2) should be used on a level surface without movement.
- If a power failure occurs or if the Metrix Reader is unplugged while the Sensor is inserted, the test result is invalid and the patient should be retested using a fresh sample.

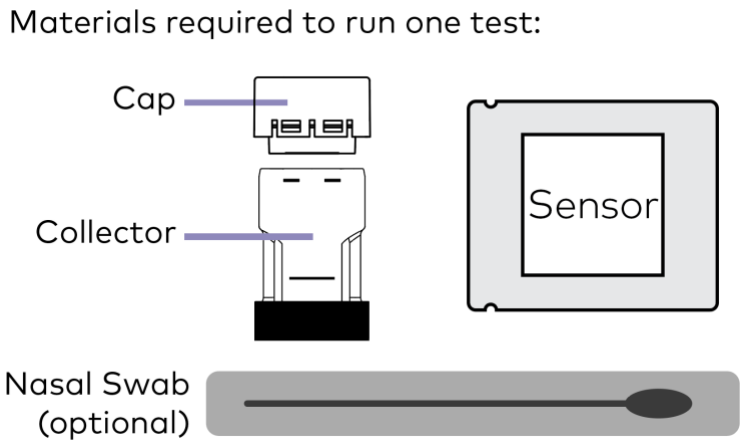
7. Procedure

7.1 Start Here

- Carefully read all instructions before beginning the test.

Scan QR code for interactive instructions and a video demonstration

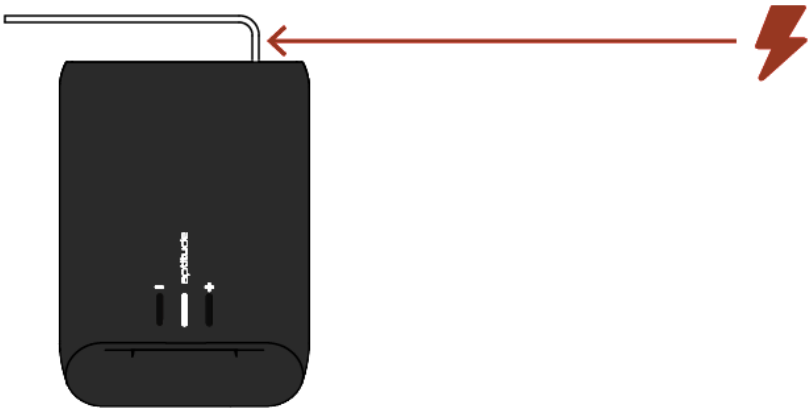
- Complete the entire procedure without delay between steps.
- No food/drink (including water)/oral hygiene products for at least 30 minutes before the test.
- Contents:



- Metrix Reader or Metrix Reader (Gen 2) required. Available separately.

7.2 STEP 1: Power Up the Metrix Reader or Metrix Reader (Gen 2)

- Connect the reader to the power supply. The center light will turn **solid white** (not flashing) when ready.

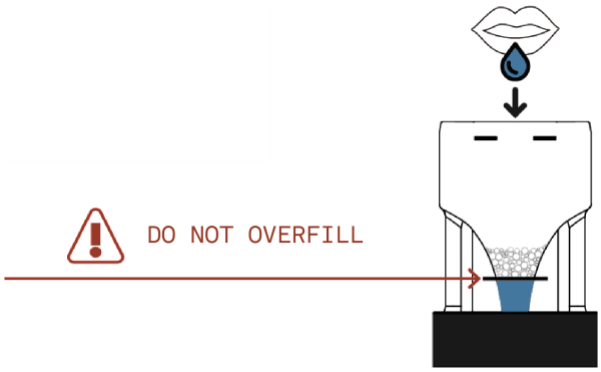


7.3 STEP 2: Collect Your Sample

Choose **one** preferred sample method: **Saliva** or **Nasal Swab.**

Option 1: Saliva Collection

- Deposit a **small amount** of saliva into the collector.
- Fluid level must **not** go above the **black line**. Ignore bubbles.

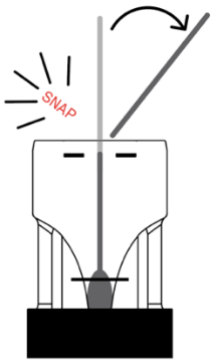


OR

Option 2: Nasal Swab Collection

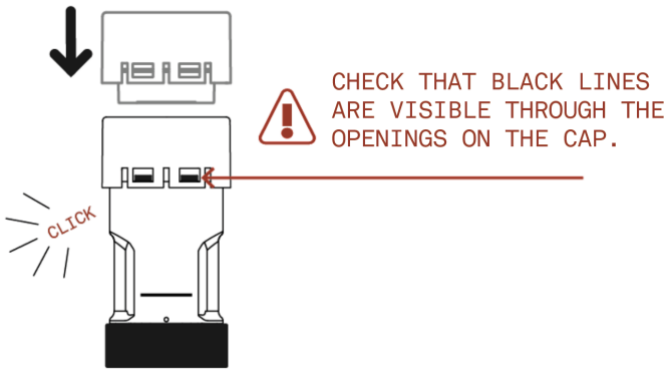
- Insert the nasal swab into your nostril until the tip is fully inside. Stop when you meet resistance (about 1 inch for adults, ½ inch for children).
- Roll the swab against the inside of your nostril 5 times

- Firmly insert the swab into collector until it cannot go any further.
- Snap off and discard the swab handle.



7.4 STEP 3: Cap Your Sample

- Put the cap onto the collector and press down **firmly** until the cap clicks into place.

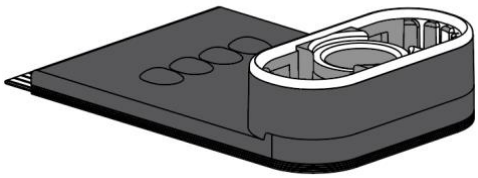


7.5 STEP 4: Shake to Mix the Sample

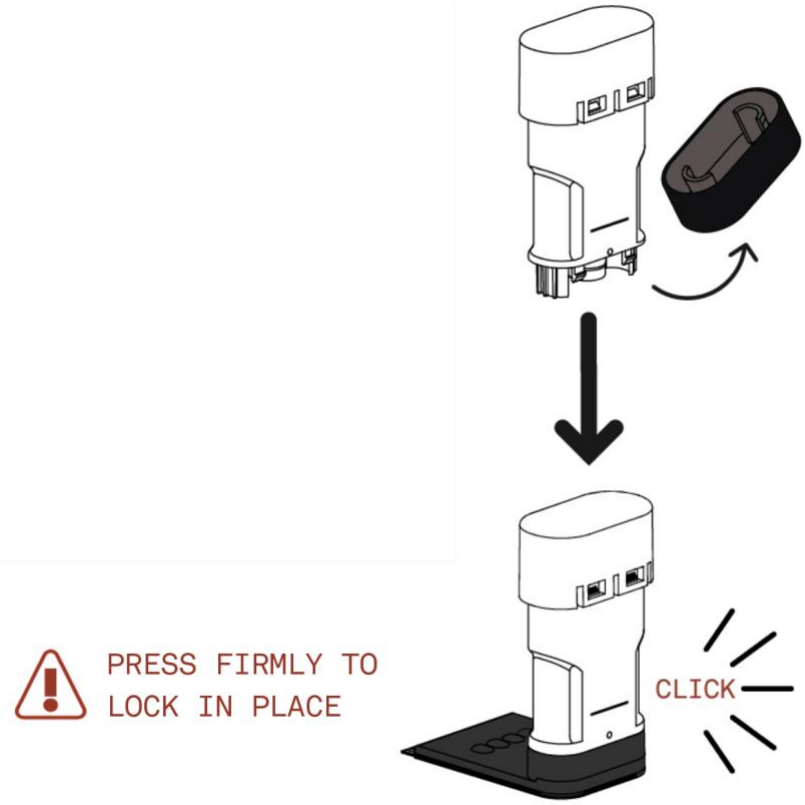
- Shake the collector **very hard** for 20 seconds to mix.

7.6 STEP 5: Attach Collector to the Sensor

- Open the sensor pouch and place the sensor on a flat surface.



- Remove the **black plastic cover** from the bottom of the collector. **Firmly** insert the collector into the sensor until it **clicks** and locks in place.

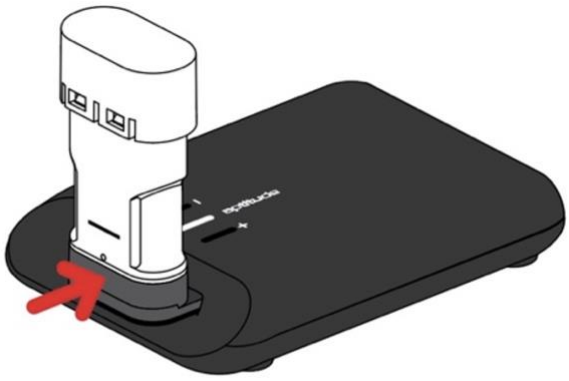


7.7 STEP 6: Run the Test

- Check the reader status

CONFIRM THAT THE READER IS READY.
THE CENTER LIGHT WILL BE SOLID
(NOT FLASHING).

- Insert the sensor into reader until it cannot go further. The test will begin automatically and will take 30 minutes to complete.



Flashing White

Test in progress

Flashing Red

Test error, see below for troubleshooting

7.8 STEP 7: Read Your Results

- Use the following visual signifiers to note the results of the test:

Solid green = COVID Negative
SARS-CoV-2 was not present.

Solid red = COVID Positive
SARS-CoV-2 was present.

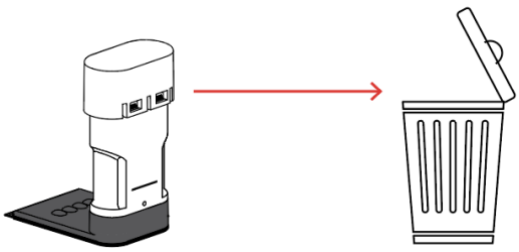
Solid purple = Invalid
Repeat test with a new kit.

IF PROOF OF TEST IS NEEDED, TAKE A PHOTO OF YOUR RESULT.
THE RESULT WILL DISPLAY UNTIL THE SENSOR IS REMOVED.

7.9 STEP 8: Discard Your Sensor

- Pull the sensor out of the reader. Discard the sensor**. Do not disassemble**.

- The reader is now ready to begin another test.



7.10 After Your Test

- Please seek follow up care from a healthcare physician if your symptoms persist or if you are concerned about your health.

- To report your Metrix COVID-19 Test results to public health agencies, please visit: aptitudemetrix.com/publichealth/reporting.

7.11 Reader Statuses and Troubleshooting

- All Metrix Reader/Metrix Reader (Gen 2) statuses for normal and abnormal operation are shown below. If troubleshooting fails to resolve any problem, please contact support. If your Metrix Reader needs to be disposed of, please place in electronic waste.

Reader Statuses

Starting Up
The reader is starting up. Wait until the center light is solid white before inserting a sensor

Ready
The reader is ready to start a test.

Test Running
The reader is running a test.
Do not remove the sensor or unplug the reader.

Positive Result
The test is complete and SARS-CoV-2 **was** detected in the sample.

Negative Result
The test is complete and SARS-CoV-2 **was not** detected in the sample.

Troubleshooting

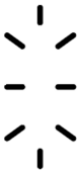
Invalid Result
The test is complete and the result is invalid. Repeat with a new Metrix COVID-19 Test kit.

Test Error
Remove sensor and **firmly** press down on collector. **Firmly** reinsert sensor into reader. If error persists, discard sensor and use a new test kit.

Canceled Test
The test did not complete. Discard the sensor and run the test with a new Metrix COVID-19 Test Kit.

Hardware Failure
There is an error with the reader. Disconnect and reconnect the power.

No Power
Check all electrical connections. The reader is not receiving power.



Indicates flashing light

8. Quality Control

8.1 Internal Control

The Metrix COVID-19 Test contains an internal control to assess the presence of a human sample and ensure adequate amplification.

8.2 External Controls

When using the Metrix COVID-19 Test in laboratory settings, good laboratory practice suggests the use of external positive and negative control materials to ensure that reagents are functional and the test is performed correctly.

Aptitude recommends the following commercially available positive and negative control materials from Microbiologics:

- HE0065N Inactivated SARS-CoV-2 Whole Virus (Pellet),
- HE0058N Negative Cellularity Control (Pellet).

HE0065N acts as a positive control material for evaluating the successful detection of N genes and O genes of SARS-CoV-2. HE0058N acts as a negative control material for evaluating the successful detection of human genes. The control material is resuspended (500 µL) and diluted (8-fold) in molecular biology-grade water, added to Collector (300 µL) in place of the clinical specimen, and processed per the instructions.

Aptitude recommends that the external controls be run:

- once for each new lot or shipment of tests received,
- once for each new operator,
- when problems with testing are suspected or identified,
- as deemed additionally necessary in order to conform with the laboratory's internal quality control procedures, with local, state and/or federal regulations, or accrediting groups.

If the correct control results are not obtained, repeat the control tests. Results from the repeated assay(s) must be acceptable to proceed. If acceptable results are not obtained, do not perform patient tests or report patient results, and contact your distributor for Technical Support before testing new patient specimens.

9. Limitations

- Performance was evaluated with saliva and anterior nasal (nares) swab specimens only, using the instructions provided in this document. Failure to follow these procedures may alter test performance.
- False negative results may occur if a specimen is improperly collected or handled. False negative results may occur if inadequate levels of virus are present in the specimen. False negative results may occur if the virus mutates in the regions targeted by the test.
- The test is a qualitative test and does not provide the quantitative value of detected organism present.
- Results from this test are read visually. Color-blind users may be unable to differentiate between all color status lights. Users with conditions affecting their vision, including color blind users should seek assistance to interpret results accurately.
- Cross-reactivity with respiratory tract organisms other than those tested in the Analytical Specificity Study may lead to erroneous results.
- This test cannot rule out diseases caused by other bacterial or viral pathogens.
- Analyte targets (viral sequences) may persist *in vivo*, independent of virus viability. Detection of analyte target(s) does not imply that the corresponding virus(es) are infectious, nor are they the causative agents for clinical symptoms.
- Positive and negative predictive values are dependent upon disease prevalence. False negative results are more likely during peak activity when disease prevalence is high and false positive results are more likely during periods of low activity.
- The performance of this test was established based on the evaluation of a limited number of clinical specimens. 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.

- Negative results for SARS-CoV-2 RNA from saliva should be confirmed by testing of an alternative sample type if clinically indicated.

10. Performance Characteristics

10.1 Analytical Sensitivity (Limit of Detection)

(A) Determination of Limit of Detection

The LoD studies for saliva and swab samples were conducted to determine the lowest detectable concentration of the Metrix COVID-19 Test at which 95% of all true positive replicates yielded a positive result in the test. LoDs were determined via a range-finding study followed by a limiting dilution study. Specific viral loads were set (i.e., genome equivalents, GE) using contrived positive samples created by serially diluting γ-irradiation-inactivated SARS-CoV-2 virus into pooled saliva or nasal matrix that was verified to be SARS-CoV-2-free with RT-PCR. For each sample type, the range-finding study was conducted to explore a wide range of viral concentrations to find a preliminary LoD at which 3/3 samples showed positive results in the Metrix COVID-19 Test. The preliminary LoD for each sample type was then used in the limiting dilution study to test positive samples at a narrower concentration range at 20 replicates per sample. The LoD was then confirmed across multiple lots of devices by repeating another 20 replicates at the LoD concentration in a second lot of devices (see tables below for each sample type). Replicate negative samples were also run and all showed negative results in the tests as expected.

The LoD of nasal swab samples was determined to be 2000 GE/swab, which is equivalent to 667 copies per mL of VTM assuming 100% elution of the swab in 3 mL of viral transport media (VTM). The LoD of saliva samples was determined to be 20 GE/µL of saliva.

Limit of Detection: Anterior Nasal (Nares) Swab Samples

Lot	Swab Viral Load (GE/swab)	#Positive/#Tested	% Positive
1	4000	20/20	100
1	2000	20/20	100
1	1000	14/20	70
2	2000	20/20	100

Limit of Detection: Saliva Samples

Lot	Saliva Viral Load (GE/µL)	#Positive/#Tested	% Positive
1	40	20/20	100
1	20	19/20	95
1	10	15/20	75
2	20	20/20	100

(B) Evaluation of Metrix Reader (Gen 2)

In a subsequent study, use of the Metrix COVID-19 Test in combination with the Metrix Reader (Gen 2) was evaluated in a LoD confirmation study that tested 20 replicates at 0.5x, 1x and 2x LoD concentration of inactivated SARS-CoV-2 (USA-WA1/2020) in both nasal swab and saliva clinical matrix. In addition, 20 negative test control (NTC) replicates were run for each clinical matrix.

The LoD for nasal swabs and saliva spiked with inactivated SARS-CoV-2 was determined to be 2000 GE/swab and 20 GE/µL, respectively, using the Metrix Reader (Gen 2), demonstrating equivalent performance to the original Metrix Reader.

Evaluation of Metrix Reader (Gen 2): Anterior Nasal (Nares) Swab Samples

Swab Viral Load (GE/swab)	#Positive/#Tested	% Positive
0 (NTC)	0/20	0
4000	20/20	100
2000	19/20	95
1000	10/20	50

Evaluation of Metrix Reader (Gen 2): Saliva Samples

Saliva Viral Load (GE/µL)	#Positive/#Tested	% Positive
0 (NTC)	0/20	0
40	20/20	100
20	19/20	95
10	9/20	45

10.2 Analytical Reactivity (Inclusivity)

An inclusivity study was conducted to demonstrate the *in silico* reactivity of the primer sets used in the Metrix COVID-19 Test against all SARS-CoV-2 variants. A total of 9,903,024 complete sequences of all known SARS-CoV-2 variants from human hosts were compiled from

GISAID database (https://www.gisaid.org) up to May 17th of 2022, and analyzed with a modified BLAST protocol. The primer sets in the Metrix COVID-19 Test were found to have an overall zero-mismatching rate of 99.81% against all analyzed SARS-CoV-2 variants, and 99.95% against the group of variants that had been reported between Feb 17 and May 17 of 2022 (i.e., Omicron, Delta, and Alpha), indicating high *in silico* reactivity of the Metrix COVID-19 Test to the SARS-CoV-2 variants identified up to May 17 of 2022. Our latest analysis (August 2022) indicated that the primer sets used in Metrix COVID-19 Test can detect all BA.4 and BA.5 variants worldwide reported in GISAID prior June. The Metrix COVID-19 Test is capable of detecting all known variants and subvariants of concern, including recent BA.4 and BA.5.

10.3 Analytical Specificity/Exclusivity (Cross-Reactivity)

The analytical specificity of the Metrix COVID-19 Test was demonstrated by testing cross-reactivity with and interference from other microorganisms as well as endogenous substances.

For the following tests with saliva samples, a positive sample refers to negative pooled saliva spiked with 2x LoD of gamma-irradiated SARS-CoV-2 virus, and a negative sample refers to negative pooled saliva. For the following tests with swab samples, a positive sample refers to a swab sample from negative pooled nasal matrix (NP swab matrix) spiked with 2x LoD of gamma-irradiated SARS-CoV-2 virus, and a negative sample refers to a swab sample from negative pooled swab matrix (NP swab matrix).

In addition, *in silico* analysis was conducted to verify the assay does not cross-react with other high prevalence disease agents and normal or pathogenic flora that are reasonably likely to be encountered in a clinical specimen. Whole genome sequences were downloaded from NCBI. BLAST alignments showed that only SARS-CoV-1 has > 80% homology on individual primers.

(C) Cross-reactivity and Microbial Interference – Swab samples

The cross-reactivity of microorganisms was evaluated via running Metrix COVID-19 Tests with triplicates of negative swab samples spiked with each of the 30 commensal microorganisms, tabulated below, at a high concentration to represent the worst-case scenario (all organisms were tested with concentrations $\geq 10^6$ GE/mL or 10^6 $TCID_{50}$/mL for viruses and 10^6 CFU/mL for bacteria). The cross-reactivity testing confirmed that none of the 30 organisms were cross-reactive with the Metrix COVID-19 Test at the concentrations tested.

The interference from microorganisms was evaluated via running Metrix COVID-19 Test with triplicates of positive swab samples (2x LoD, 4000 GE/swab) spiked with each of the commensal microorganisms at a high concentration to represent the worst-case scenario. The interference testing confirmed that none of the 30 organisms interfere with the Metrix COVID-19 Test at the concentrations tested.

Microorganism	Cross Reactivity		Microbial Interference	
	#Positive/ #Tested	% Positive	#Positive / #Tested	% Positive
Adenovirus (e.g., C1 Ad. 71)	0/3	0	3/3	100
Bordetella pertussis	0/3	0	3/3	100
Candida albicans	0/3	0	3/3	100
Enterovirus (e.g., EV68, Genomic RNA)	0/3	0	3/3	100
Chlamydia pneumoniae	0/3	0	3/3	100
Haemophilus influenzae	0/3	0	3/3	100

Microorganism	Cross Reactivity		Microbial Interference	
	#Positive/ #Tested	% Positive	#Positive / #Tested	% Positive
Human coronavirus 229E	0/3	0	3/3	100
Human coronavirus HKU1 (Synthetic RNA)	0/3	0	3/3	100
Human coronavirus NL63 (Synthetic RNA)	0/3	0	3/3	100
Human coronavirus OC43	0/3	0	3/3	100
Human Metapneumovirus (hMPV)	0/3	0	3/3	100
Influenza A	0/3	0	3/3	100
Influenza B	0/3	0	3/3	100
Legionella pneumophila	0/3	0	3/3	100
MERS-coronavirus (Synthetic RNA)	0/3	0	3/3	100
Mycobacterium tuberculosis	0/3	0	3/3	100
Mycoplasma pneumoniae	0/3	0	3/3	100
Parainfluenza 1	0/3	0	3/3	100
Parainfluenza 2	0/3	0	3/3	100
Parainfluenza 3	0/3	0	3/3	100
Parainfluenza 4	0/3	0	3/3	100

Microorganism	Cross Reactivity		Microbial Interference	
	#Positive/ #Tested	% Positive	#Positive / #Tested	% Positive
Pneumocystis jirovecii (PJP)	0/3	0	3/3	100
Pseudomonas aeruginosa	0/3	0	3/3	100
Respiratory syncytial virus	0/3	0	3/3	100
Rhinovirus	0/3	0	3/3	100
SARS-coronavirus (Synthetic RNA)	0/3	0	3/3	100
Staphylococcus epidermidis	0/3	0	3/3	100
Streptococcus pneumoniae	0/3	0	3/3	100
Streptococcus pyogenes	0/3	0	3/3	100
Streptococcus salivarius	0/3	0	3/3	100

(D) Cross-reactivity and Microbial Interference – Saliva samples

The cross-reactivity of microorganisms was evaluated via running Metrix COVID-19 Tests with triplicates of negative saliva samples spiked with each of the 41 commensal microorganisms, tabulated below, at a high concentration to represent the worst-case scenario. The cross-reactivity testing confirmed that none of the 42 organisms were cross-reactive with the Metrix COVID-19 Test at the concentrations tested.

The interference from microorganisms was evaluated via running Metrix COVID-19 Test with triplicates of positive saliva samples (2x LoD, 40 GE/

µL) spiked with each of the 42 commensal microorganisms at a high concentration to represent the worst-case scenario. The interference testing confirmed that none of the 41 organisms interfere with the Metrix COVID-19 Test at the concentrations tested.

Microorganism	Cross Reactivity		Microbial Interference	
	#Positive/ #Tested	% Positive	#Positive / #Tested	% Positive
Adenovirus C1	0/3	0	3/3	100
Bordetella pertussis	0/3	0	3/3	100
Candida albicans	0/3	0	3/3	100
Chlamydia pneumoniae	0/3	0	3/3	100
Cytomegalovirus (CMV)	0/3	0	3/3	100
Eikenella sp.	0/3	0	3/3	100
Haemophilus influenzae	0/3	0	3/3	100
Herpes simplex virus type 1 (HSV-1)	0/3	0	3/3	100
Human coronavirus 229E	0/3	0	3/3	100
Human coronavirus OC43	0/3	0	3/3	100
Human Metapneumovirus (hMPV)	0/3	0	3/3	100
Influenza A	0/3	0	3/3	100
Influenza B	0/3	0	3/3	100
Lactobacillus sp.	0/3	0	3/3	100

Microorganism	Cross Reactivity		Microbial Interference	
	#Positive/ #Tested	% Positive	#Positive / #Tested	% Positive
Legionella pneumophila	0/3	0	3/3	100
Moraxella catarrhalis	0/3	0	3/3	100
Mycobacterium tuberculosis	0/3	0	3/3	100
Mycoplasma pneumoniae	0/3	0	3/3	100
Neisseria sp.	0/3	0	3/3	100
Nocardia sp.	0/3	0	3/3	100
Parainfluenza 1	0/3	0	3/3	100
Parainfluenza 2	0/3	0	3/3	100
Parainfluenza 3	0/3	0	3/3	100
Parainfluenza 4	0/3	0	3/3	100
Pneumocystis jirovecii (PJP)	0/3	0	3/3	100
Pseudomonas aeruginosa	0/3	0	3/3	100
Respiratory syncytial virus	0/3	0	3/3	100
Rhinovirus	0/3	0	3/3	100
Staphylococcus epidermidis	0/3	0	3/3	100
Streptococcus mitis	0/3	0	3/3	100

Microorganism	Cross Reactivity		Microbial Interference	
	#Positive/ #Tested	% Positive	#Positive / #Tested	% Positive
Streptococcus mutans	0/3	0	3/3	100
Streptococcus pneumoniae	0/3	0	3/3	100
Streptococcus pyogenes	0/3	0	3/3	100
Streptococcus salivarius	0/3	0	3/3	100
Bacteroides oralis	0/3	0	3/3	100
Human coronavirus NL63 (Synthetic RNA)	0/3	0	3/3	100
SARS-coronavirus (Synthetic RNA)	0/3	0	3/3	100
Enterovirus (68)	0/3	0	3/3	100
Human coronavirus HKU1 (Synthetic RNA)	0/3	0	3/3	100
MERS-coronavirus (Synthetic RNA)	0/3	0	3/3	100
Porphyromonas gingivalis (DNA)	0/3	0	3/3	100

10.4 Analytical Specificity (Interfering Substances)

(A) Interfering Substances – Swab Samples

Endogenous interference studies were conducted to assess potential interference effects on the Metrix COVID-19 Test from 14 substances that

may naturally be present in respiratory specimens or artificially introduced into the oral or nasal cavity, including common household items that may be present in the testing environment. The interfering substances were tested by running Metrix COVID-19 Test with triplicates of positive (2x LoD, 4000 GE/swab) and negative swab samples spiked with each of the 16 substances listed in the table below at the highest medically relevant concentration. The interference testing confirmed that none of the 16 substances interfere with the Metrix COVID-19 Test at the concentrations tested.

Potential Interfering Substance	Concen-tration	Positive		Negative	
		# Positive/ #Tested	% Positive	# Positive/ #Tested	% Positive
Afrin	5% v/v	3/3	100	0/3	0
Human whole blood	1% v/v	3/3	100	0/3	0
Chloraseptic Sore Throat Spray	5% v/v	3/3	100	0/3	0
Flonase allergy relief	5% v/v	3/3	100	0/3	0
Mucin	1 mg/mL	3/3	100	0/3	0
NeoSynepthrine Cold and Sinus Extra Strength Spray	5% v/v	3/3	100	0/3	0
NeilMed NasoGel	1.25% v/v	3/3	100	0/3	0
Relenza (Zanamivir)	1 mg/mL	3/3	100	0/3	0
Tamiflu	6 mg/mL	3/3	100	0/3	0
Tobramycin	2.5 mg/ mL	3/3	100	0/3	0

Potential Interfering Substance	Concen-tration	Positive		Negative	
		# Positive/ #Tested	% Positive	# Positive/ #Tested	% Positive
Flunisolide	7.5% v/v	3/3	100	0/3	0
Zicam Allergy Relief	5% v/v	3/3	100	0/3	0
Method All-Purpose Surface Cleaner	5% v/v	3/3	100	0/3	0
Mupirocin	1 mg/mL	3/3	100	0/3	0

(B) Interfering Substances - Saliva

Endogenous interference studies were conducted to assess potential interference effects on the Metrix COVID-19 Test from sixteen substances that may naturally be present in respiratory specimens or artificially introduced into the oral or nasal cavity, including common household items that may be present in the testing environment. The interfering substances were tested by running Metrix COVID-19 Test with triplicates of positive (2x LoD, 40 GE/μL) and negative saliva samples spiked with each of the 16 substances listed in the table below at the highest medically relevant concentration. The interference testing confirmed that none of the 16 substances interfere with the Metrix COVID-19 Test at the concentrations tested.

Potential Interfering Substance	Concen-tration	Positive		Negative	
		# Positive/ #Tested	% Positive	# Positive/ #Tested	% Positive
Mucin: bovine	2.5 mg/mL	3/3	100	0/3	0

Potential Interfering Substance	Concen-tration	Positive		Negative	
		# Positive/ #Tested	% Positive	# Positive/ #Tested	% Positive
submax-illary gland, type I-S					
Human Whole blood cells	~5×10^6 cells/mL	3/3	100	0/3	0
Afrin Original nasal spray	15% v/v	3/3	100	0/3	0
NeilMed NasoGel	1.25% v/v	3/3	100	0/3	0
Cepacol (benzocaine/ menthol lozenges)	3 mg/mL	3/3	100	0/3	0
Chloraseptic Sore Throat spray/solution	5% v/v	3/3	100	0/3	0
Toothpaste (Colgate)	0.5% v/v	3/3	100	0/3	0
Crest Mouth Wash (Hydrogen Peroxide)	5% v/v	3/3	100	0/3	0
Cepacol Mouth Wash	5% v/v	3/3	100	0/3	0
Nicotine	0.03 mg/mL	3/3	100	0/3	0
Human Genomic DNA	10 ng/µL	3/3	100	0/3	0
Robitussin	5% v/v	3/3	100	0/3	0
Emergen-C (zinc, magnesium, riboflavin)	5% v/v	3/3	100	0/3	0
Zinc (liquid	5% v/v	3/3	100	0/3	0

Potential Interfering Substance	Concen-tration	Positive		Negative	
		# Positive/ #Tested	% Positive	# Positive/ #Tested	% Positive
supplement)					
ACT dry mouth lozenges (isomalt, xylitol, glycerin)	2 mg/mL	3/3	100	0/3	0
Nyquil (Acetaminophen, doxylamine)	5% v/v	3/3	100	0/3	0

10.5 Human Factors Study

Aptitude conducted a human factors usability study to evaluate the ability of users to successfully complete the Metrix COVID-19 Test. Forty-two subjects were recruited with a wide range of age and education demographics. Twenty-five subjects between the ages of 2 and 14 were assisted by a parent or a guardian during the test (ten of these subjects were aged 2 or 3 years). The study was conducted in a simulated home environment; subjects were provided with standard items found in a common home setting such as a sturdy work surface, electrical power outlets, tissues, handwashing area, and a trash can. Subjects were asked to perform both saliva and nasal swab Metrix COVID-19 Test following printed instructions found in the test kits, or a step-by-step video guide accessible on smart devices via a QR code in the printed instructions. To eliminate bias from subjects' familiarity towards the general testing procedure, the order of saliva or nasal swab tests alternated for every other subject. For both the saliva and nasal swab tests, 100% (42/42) of subjects successfully completed all critical steps and 100% (42/42) correctly interpreted their result.

Swab Assay Participant	Completed All Critical Steps	%
Self-test by lay-user	17/17	100%
Assisted test by lay-user	25/25	100%
Total	**42/42**	**100%**

Saliva Assay Participant	Completed All Critical Steps	%
Self-test by lay-user	17/17	100%
Assisted test by lay-user	25/25	100%
Total	**42/42**	**100%**

Ease of use was assessed via a 16-point questionnaire on a Likert scale for agreement against a comprehensive set of product features and user interactions. A mean score of 4.7/5 was determined, demonstrating a high degree of ease of use and user comprehension.

10.6 Clinical Evaluation

A prospective multi-site clinical study was conducted in the United States by lay users in simulated home environments. Sample collection and testing was performed by the untrained subject (ages 14 years and older) or the subject's parent/guardian for children between 2 and 14 years of age. The study enrolled a total of 358 evaluable symptomatic and asymptomatic participants on an all-comers basis and included a traditional screening cohort. The anterior nasal swab sample of one individual was excluded due to a protocol deviation.

Asymptomatic individuals were enrolled as part of school/college COVID-19 screening programs and who were receiving tests from the clinical sites as part of the standard of care. Such asymptomatic screening programs included periodic testing of unvaccinated individuals, as well as random sampling of the student and staff population. Individuals from the traditional screening cohort were invited into the study and enrolled accordingly.

Each participant, by themselves or with the assistance of their parent/guardian, performed the entire test from sample to interpretation of results following the QRI without guidance or training. To evaluate the performance of the device with saliva and nasal swabs, participants first performed the test with one specimen and then performed another test with the second specimen. For each subject, a comparator NP swab was collected and sent to a reference laboratory for testing with a high sensitivity FDA EUA-authorized SARS-CoV-2 RT-PCR comparator test. The study included 5.3% (19/358) of pediatric subjects below the age of 6, and 15.9% (57/358) of pediatric subjects between the ages of 7 and 13.

Summary Clinical Performance

The Metrix COVID-19 Test results were compared to those produced by the high-sensitivity FDA-authorized SARS-CoV-2 RT-PCR test which uses nasopharyngeal swab samples.

Anterior Nasal (Nares) Swab Samples:
For anterior nasal (nares) swab samples, the Positive Percent Agreement (PPA) was 95.0% (38/40) and the Negative Percent Agreement (NPA) was 97.1% (34/35) in the symptomatic group. In the asymptomatic group, the PPA was 100.0% (21/21) and the NPA was 99.2% (259/261). The PPA was 96.7% (59/61) and the NPA was 99.0% (293/296) in the combined total study population. There were 5 invalid tests (1.4%) and no canceled tests. The initial candidate invalid results were retested with concordant results. When excluding the single sample

(Days Post Symptom Onset (DPSO) = 4) with a viral load below the comparator LoD, the Metrix COVID-19 Test correctly identified 59/60 positive nasal swab samples.

Subject Type	Metrix COVID-19 Test – Swab Assay vs. Comparator EUA SARS-CoV-2 RT-PCR Assay	
	PPA	NPA
Symptomatic	95.0% (38/40)	97.1% (34/35)
Asymptomatic	100.0% (21/21)	99.2% (259/261)
Total	**96.7% (59/61)**	**99.0% (293/296)**

Saliva Samples:
Comparing the Metrix COVID-19 Test results, when using saliva samples, the symptomatic group PPA was 90.2% (37/41) and the NPA was 100.0% (35/35). The asymptomatic group PPA was 90.5% (19/21) and the NPA was 99.2% (258/260). For the combined study population, the PPA was 90.3% (56/62) and the NPA was 99.3% (293/295). There were 10 invalid tests (2.8%) and 2 canceled tests (0.9%) due to device errors requiring re-tests. Samples were retested per the IFU with a valid result.

Subject Type	Metrix COVID-19 Test – Saliva Assay vs. Comparator EUA SARS-CoV-2 RT-PCR Assay	
	PPA	NPA
Symptomatic	90.2% (37/41)	100.0% (35/35)
Asymptomatic	90.5% (19/21)	99.2% (258/260)
Total*	**90.3% (56/62)**	**99.3% (293/295)**

* The Metrix COVID-19 Test with saliva specimens was also evaluated against a saliva-based EUA RT-PCR test (SalivaDirect). 353/357 results were in agreement with the saliva-based EUA test, which is 3 fewer discordant results relative to the comparator that used NP samples (7 discordant results). The saliva-based EUA test characterized these 3 samples as negative while the NP comparator characterized them as positive. One of these 3 subjects was asymptomatic while the DPSO of the other 2 subjects were 4 and 7 days.

The individual comparator tables across all subject groups and assay configurations are summarized below.

(A) Metrix COVID-19 Test – Nasal Swab Assay

All Subjects		Comparator: NP Swab on High Sensitivity EUA SARS-CoV-2 RT-PCR Assay		
		Positive	Negative	Total
Candidate: Metrix COVID-19 Test Swab Assay	Positive	59	3	62
	Negative	2	293	295
	Total	61	296	357
PPA: 96.7% (59/61), (95% CI: 88.8%-99.1%)				
NPA: 99.0% (293/296), (95% CI: 97.1%-99.7%)				

Symptomatic Subjects		Comparator: NP Swab on High Sensitivity EUA SARS-CoV-2 RT-PCR Assay		
		Positive	Negative	Total
Candidate: Metrix COVID-19 Test Swab Assay	Positive	38	1	39
	Negative	2	34	36
	Total	40	35	75
PPA: 95.0% (38/40), (95% CI: 83.5%-98.6%)				
NPA: 97.1% (34/35), (95% CI: 85.5%-99.5%)				

Asymptomatic Subjects		Comparator: NP Swab on High Sensitivity EUA SARS-CoV-2 RT-PCR Assay		
		Positive	Negative	Total
Candidate: Metrix COVID-19 Test Swab Assay	Positive	21	2	23
	Negative	0	259	259
	Total	21	261	282
PPA: 100.0% (21/21), (95% CI: 84.5%-100.0%)				
NPA: 99.2% (259/261), (95% CI: 97.2%-99.8%)				

(B) Metrix COVID-19 Test – Saliva Assay

All Subjects		Comparator: NP Swab on High Sensitivity EUA SARS-CoV-2 RT-PCR Assay		
		Positive	Negative	Total
Candidate: Metrix COVID-19 Test Saliva Assay	Positive	56	2	58
	Negative	6	293	299
	Total	62	295	357
PPA: 90.3% (56/62), (95% CI: 80.5%-95.5%)				
NPA: 99.3% (293/295), (95% CI: 97.6%-99.8%)				

Symptomatic Subjects		Comparator: NP Swab on High Sensitivity EUA SARS-CoV-2 RT-PCR Assay		
		Positive	Negative	Total
Candidate: Metrix COVID-19 Test Saliva Assay	Positive	37	0	37
	Negative	4	35	39
	Total	41	35	76
PPA: 90.2% (37/41), (95% CI: 77.5%-96.1%)				
NPA: 100.0% (35/35), (95% CI: 90.1%-100.0%)				

Asymptomatic Subjects		Comparator: NP Swab on High Sensitivity EUA SARS-CoV-2 RT-PCR Assay		
		Positive	Negative	Total
Candidate: Metrix COVID-19 Test Saliva Assay	Positive	19	2	21
	Negative	2	258	260
	Total	21	260	281
PPA: 90.5% (19/21), (95% CI: 71.1%-97.3%)				
NPA: 99.2% (258/260), (95% CI: 97.2%-99.8%)				

(C) Near-Cutoff Results Summary

An additional evaluation was conducted to test the ability of untrained users to correctly run and interpret near-cutoff positive samples with the Metrix COVID-19 test. Subjects were each given three blinded contrived saliva samples, a mix of negative and low-positive (2x LoD) to test. Comparing their results to those expected, the PPA was 100.0% (15/15) and the NPA was 100.0% (15/15), demonstrating that in the hands of untrained users the Metrix COVID-19 Test can reliably identify low-positive samples.

11. References

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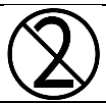
5. Centers for Disease Control and Prevention. “Biosafety in Microbiological and Biomedical Laboratories (BMBL) 5th Edition.” https://www.cdc.gov/labs/BMBL.html
6. Clinical and Laboratory Standards Institute. “Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline.” Document M29 https://clsi.org/standards/products/microbiology/documents/m29/

12. Table of Symbols

The following symbols are used throughout this manual or on the product:

Symbol	Definition
	This symbol references the product's catalog number.
	This symbol indicates the product's batch/lot code.
	This symbol indicates the product's serial number.
	For In Vitro Diagnostic Use.
	This symbol indicates the name and location of the product manufacturer.
	This symbol indicates the date that the product was manufactured.
	This symbol indicates that you should consult the instructions for use.
	This symbol indicates the product's upper and lower temperature limitations.
	This symbol indicates the product's expiration date.
	This symbol indicates that you should not use the package if it is damaged.
	This symbol indicates that the product is intended for single use only. Do no reuse.
	This symbol indicates that the product should not be disposed of in a municipal trash bin when it has reached the end of its lifetime.
	This symbol indicates that the product should be kept dry.
	This symbol certifies that the electromagnetic interference from the device is under limits approved by the Federal Communications Commission.
	This symbol indicates direct current (DC) voltage.

13. Manufacturer, Support, and Ordering Information

Manufacturer

Aptitude Medical Systems, Inc.
125 Cremona Drive, Suite 100
Goleta, CA 93117

Customer Support and Ordering Information
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