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QIAstat-Dx® Respiratory SARS-CoV-2 Panel Instructions for Use (Handbook)

Version 1

For in vitro diagnostic use

IVD

CE

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R1

Sample to Insight

QIAGEN

Contents

Intended Use

The QIAstat-Dx Respiratory SARS-CoV-2 Panel is a qualitative test intended for analyzing nasopharyngeal swab (NPS) samples for the presence of viral or bacterial nucleic acids. The QIAstat-Dx Respiratory SARS-CoV-2 Panel is able to accept both dry swabs and transport medium liquid samples. The assay is designed for use with the QIAstat-Dx Analyzer 1.0 for integrated nucleic acid extraction and multiplex real-time RT-PCR detection.

The QIAstat-Dx Respiratory SARS-CoV-2 Panel detects SARS-CoV-2 and 21 additional pathogens (Influenza A, Influenza A subtype H1N1/2009, Influenza A subtype H1, Influenza A subtype H3, Influenza B, Coronavirus 229E, Coronavirus HKU1, Coronavirus NL63, Coronavirus OC43, Parainfluenza virus 1, Parainfluenza virus 2, Parainfluenza virus 3, Parainfluenza virus 4, Respiratory Syncytial virus A/B, human Metapneumovirus A/B, Adenovirus, Bocavirus, Rhinovirus/Enterovirus*, *Mycoplasma pneumoniae*, *Legionella pneumophila*, and *Bordetella pertussis*).

The results from the QIAstat-Dx Respiratory SARS-CoV-2 Panel must be interpreted within the context of all relevant clinical and laboratory findings.

The QIAstat-Dx Respiratory SARS-CoV-2 Panel is intended for professional use only and is not intended for self-testing.

For in vitro diagnostic use.

* Enterovirus and Rhinovirus are both detected, but not differentiated, with the QIAstat-Dx® Respiratory SARS-CoV-2 Panel.

Summary and Explanation

QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge description

The QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge is a disposable plastic device that allows performance of fully automated molecular assays for the detection of respiratory pathogens. The main features of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge include compatibility with the respiratory dry swabs (Copan® FLOQSwabs®, cat. no. 503CS01) and transport medium liquid samples, hermetical containment of the pre-loaded reagents necessary for testing, and true walk-away operation. All sample preparation and assay testing steps are performed within the cartridge.

All reagents required for the complete execution of a test run are pre-loaded and self-contained in the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge. The user does not need to come in contact with and/or manipulate any reagents. During the test, reagents are handled within the cartridge in the Analytical Module of the QIAstat-Dx Analyzer 1.0 by pneumatically-operated microfluidics and make no direct contact with the actuators. The QIAstat-Dx Analyzer 1.0 houses air filters for both incoming and outgoing air, further safeguarding the environment. After testing, the cartridge stays hermetically closed at all times, greatly enhancing its safe disposal.

Within the cartridge, multiple steps are automatically performed in sequence using pneumatic pressure to transfer samples and fluids via the transfer chamber to their intended destinations.

After the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge containing the sample is introduced into the QIAstat-Dx Analyzer 1.0, the following assay steps occur automatically:

- Resuspension of Internal Control
- Cell lysis using mechanical and/or chemical means
- Membrane-based nucleic acid purification
- Mixing of the purified nucleic acid with lyophilized master mix reagents
- Transfer of defined aliquots of eluate/master mix to different reaction chambers
- Performance of multiplex real-time RT-PCR testing within each reaction chamber.

  Note: An increase in fluorescence, indicating detection of the target analyte, is detected directly within each reaction chamber.

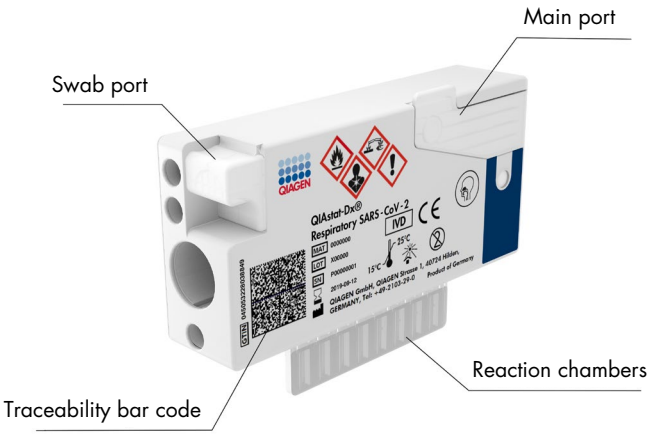

Figure 1. Layout of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge and its features.

Pathogen Information

Acute respiratory infections can be caused by a variety of pathogens, including bacteria and viruses, and generally present with nearly indistinguishable clinical signs and symptoms. The rapid and accurate determination of the presence or absence of potential causative agent(s) helps make timely decisions regarding treatment, hospital admission, infection control, and return of the patient to work and family. It may also greatly support improved antimicrobial stewardship and other important public health initiatives.

The QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge is a single-use cartridge that includes all reagents needed for nucleic acid extraction, nucleic acid amplification, and detection of 22 bacteria and viruses (or their subtypes), including SARS-CoV-2* that cause respiratory symptoms. Testing requires a small sample volume and minimal hands-on time, and the results are available in approximately one hour.

Pathogens (and subtypes) that can be detected and identified with the QIAstat-Dx Respiratory SARS-CoV-2 Panel are listed in Table 1 (next page).

* The SARS-CoV-2 target in the QIAstat-Dx Respiratory SARS-CoV-2 Panel has been designed upon alignment of more than 170 genomic sequences available in public databases from the SARS-CoV-2 identified as the causative agent of the viral pneumonia (COVID-19) outbreak that originated in Wuhan, Hubei, China. The SARS-CoV-2 in this panel targets 2 genes of the virus genome (Orf1b poly gen (Rdrp gene) and E genes) detected with the same fluorescent channel.

Table 1. Pathogens detected by the QIAstat-Dx Respiratory SARS-CoV-2 Panel

Pathogen	Classification (genome type)
Influenza A	Orthomyxovirus (RNA)
Influenza A, subtype H1N1/2009	Orthomyxovirus (RNA)
Influenza A subtype H1	Orthomyxovirus (RNA)
Influenza A subtype H3	Orthomyxovirus (RNA)
Influenza B	Orthomyxovirus (RNA)
Coronavirus 229E	Coronavirus (RNA)
Coronavirus HKU1	Coronavirus (RNA)
Coronavirus NL63	Coronavirus (RNA)
Coronavirus OC43	Coronavirus (RNA)
SARS-CoV-2	Coronavirus (RNA)
Parainfluenza Virus 1	Paramyxovirus (RNA)
Parainfluenza Virus 2	Paramyxovirus (RNA)
Parainfluenza Virus 3	Paramyxovirus (RNA)
Parainfluenza Virus 4	Paramyxovirus (RNA)
Respiratory Syncytial Virus A/B	Paramyxovirus (RNA)
Human Metapneumovirus A/B	Paramyxovirus (RNA)
Adenovirus	Adenovirus (DNA)
Bocavirus	Parvovirus (DNA)
Rhinovirus/Enterovirus	Picornavirus (RNA)
Mycoplasma pneumoniae	Bacterium (DNA)
Legionella pneumophila	Bacterium (DNA)
Bordetella pertussis	Bacterium (DNA)

Note: Enterovirus and Rhinovirus are both detected, but not differentiated, with the QIAstat-Dx Respiratory SARS-CoV-2 Panel.

Principle of the Procedure

Description of the process

Diagnostic tests with the QIAstat-Dx Respiratory SARS-CoV-2 Panel are performed on the QIAstat-Dx Analyzer 1.0. All of the sample preparation and analysis steps are performed automatically by the QIAstat-Dx Analyzer 1.0. Samples are collected and loaded manually into the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge, depending on the sample type:

Option 1: Inserting the swab into the swab port when using a dry swab sample type (Figure 2).

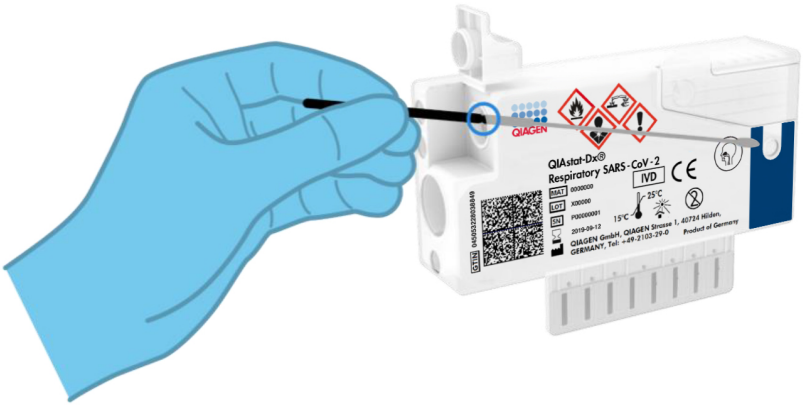

Figure 2. Loading the dry swab sample type into the swab port.

Option 2: A transfer pipette is used for dispensing transport medium liquid sample into the main port (Figure 3).

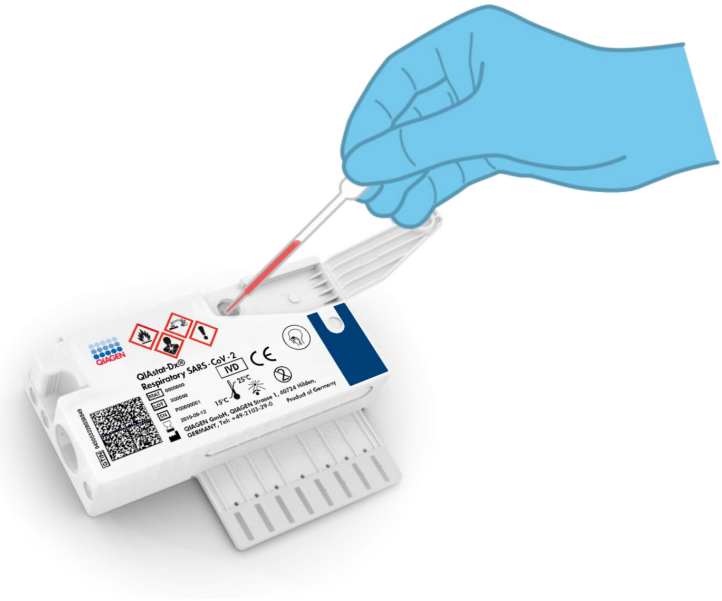

Figure 3. Dispensing transport medium liquid sample into the main port.

Sample collection and cartridge loading

The collection of samples and their subsequent loading into the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge should be performed by personnel trained in safe handling of biological samples.

The following steps are involved and must be executed by the user:

1. A nasopharyngeal swab sample is collected.
2. The nasopharyngeal swab is placed into transport medium only in the case of transport medium liquid sample type.
3. The sample information is manually written on or a sample label is affixed to the top of a QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.
4. Sample is loaded manually into the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge:
   - Dry swab sample type: The nasopharyngeal swab sample is inserted into the swab port of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.
   - Transport medium liquid sample type: 300 µl of sample is transferred into the main port of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge using one of the included transfer pipettes.

     IMPORTANT: When loading transport medium liquid sample, the user performs a visual check of the sample inspection window (see image below) to confirm that the liquid sample has been loaded (Figure 4, next page).

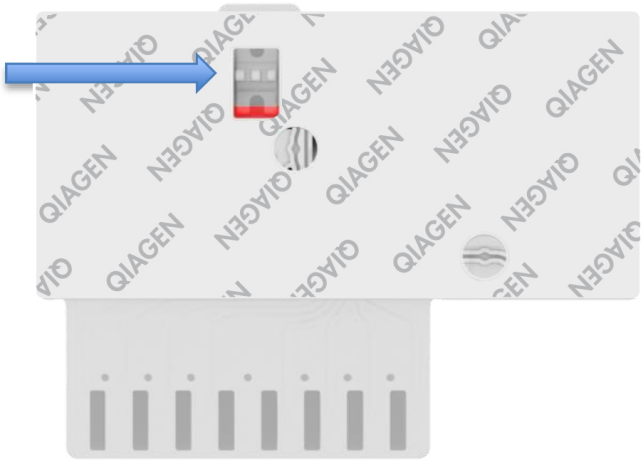

Figure 4. Sample inspection window (blue arrow).

5. The sample bar code and QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge bar code are scanned in the QIAstat-Dx Analyzer 1.0.
6. The QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge is introduced into the QIAstat-Dx Analyzer 1.0.
7. The test is started on the QIAstat-Dx Analyzer 1.0.

Sample preparation, nucleic acid amplification, and detection

The extraction, amplification, and detection of nucleic acids in the sample are performed automatically by the QIAstat-Dx Analyzer 1.0.

1. The liquid sample is homogenized and cells are lysed in the lysis chamber of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge, which includes a rotor that turns at high speed.
2. Nucleic acids are purified from the lysed sample via binding to a silica membrane in the purification chamber of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge in the presence of chaotropic salts and alcohol.
3. The purified nucleic acids are eluted from the membrane in the purification chamber and are mixed with the lyophilized PCR chemistry in the dried-chemistry chamber of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.
4. The mixture of sample and PCR reagents is dispensed into the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge PCR chambers, which contain lyophilized, assay-specific primers and probes.
5. The QIAstat-Dx Analyzer 1.0 creates the optimal temperature profiles to carry out effective multiplex real-time RT-PCR and performs real-time fluorescence measurements to generate amplification curves.
6. The QIAstat-Dx Analyzer 1.0 Software interprets the resulting data and process controls, and delivers a test report.

Materials Provided

Kit contents

QIAstat-Dx Respiratory SARS-CoV-2 Panel **Catalog no.** **Number of tests**	 **691214** **6**
QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge*	6
Transfer pipettes†	6

* 6 individually packaged cartridges containing all reagents needed for sample preparation and multiplex real-time RT-PCR, plus Internal Control.

† 6 individually packaged transfer pipettes for dispensing liquid sample into the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.

Materials Required but Not Provided

The QIAstat-Dx Respiratory SARS-CoV-2 Panel is designed for use with the QIAstat-Dx Analyzer 1.0. Before beginning a test, make sure the following are available:

- QIAstat-Dx Analyzer 1.0 (at least one Operational Module and one Analytical Module) with software version 1.2 or higher*
- *QIAstat-Dx Analyzer 1.0 User Manual* (for use with software version 1.2 or higher)
- QIAstat-Dx latest Assay Definition File software for Respiratory Panel installed on the Operational Module

* DiagCORE® Analyzer instruments running QIAstat-Dx software version 1.2 or higher can be used as an alternative to QIAstat-Dx Analyzer 1.0 instruments.

Warnings and Precautions

For in vitro diagnostic use.

The QIAstat-Dx Respiratory SARS-CoV-2 Panel is to be used by laboratory professionals trained in the use of QIAstat-Dx Analyzer 1.0.

Safety information

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, consult the appropriate safety data sheets (SDSs). These are available online in PDF format at **www.qiagen.com/safety** where you can find, view, and print the SDS for each QIAGEN kit and kit component.

Always wear appropriate personal protective equipment, including but not limited to disposable powder-free gloves, a lab coat, and protective eyewear. Protect skin, eyes and mucus membranes. Change gloves often when handling samples.

Handle all samples, used cartridges, and transfer pipettes as if they are capable of transmitting infectious agents. Always observe safety precautions as outlined in relevant guidelines, such as the Clinical and Laboratory Standards Institute® (CLSI) *Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline* (M29), or other appropriate documents provided by:

- OSHA®: Occupational Safety and Health Administration (United States of America)
- ACGIH®: American Conference of Government Industrial Hygienists (United States of America)
- COSHH: Control of Substances Hazardous to Health (United Kingdom)

Follow your institution's safety procedures for handling biological samples. Dispose of samples, QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges, and transfer pipettes according to the appropriate regulations.

The QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge is a closed, single-use device that contains all reagents needed for sample preparation and multiplex real-time RT-PCR within the QIAstat-Dx Analyzer 1.0. Do not use a QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge that is past its expiration date, appears damaged, or leaks fluid. Dispose of used or damaged cartridges in accordance with all national, state, and local health and safety regulations and laws.

Observe standard laboratory procedures for keeping the working area clean and contamination-free. Guidelines are outlined in publications such as the *Biosafety in Microbiological and Biomedical Laboratories* from the Centers for Disease Control and Prevention and the National Institutes of Health (**www.cdc.gov/od/ohs/biosfty/biosfty.htm**).

The following hazard and precautionary statements apply to components of the QIAstat-Dx Respiratory SARS-CoV-2 Panel.

QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge

Contains: ethanol; guanidine hydrochloride; guanidine thiocyanate; isopropanol; proteinase K; t-Octylphenoxypolyethoxyethanol. Danger! Highly flammable liquid and vapour. Harmful if swallowed or if inhaled. May be harmful in contact with skin. Causes severe skin burns and eye damage. May cause allergy or asthma symptoms or breathing difficulties if inhaled. May cause drowsiness or dizziness. Harmful to aquatic life with long lasting effects. Contact with acids liberates very toxic gas. Corrosive to the respiratory tract. Keep away from heat/sparks/open flames/hot surfaces. No smoking. Avoid breathing dust/fume/gas/mist/vapours/spray. Wear protective gloves/protective clothing/eye protection/face protection. Wear respiratory protection. IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. IF exposed or concerned: Immediately call a POISON CENTER or doctor/ physician. Remove person to fresh air and keep comfortable for breathing.

Reagent Storage and Handling

Store the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges in a dry, clean storage space at room temperature (15–25°C). Do not remove the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges or the transfer pipettes from their individual packaging until actual use. Under these conditions, QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges can be stored until the expiration date printed on the individual packaging. The expiration date is also included in the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge bar code and is read by the QIAstat-Dx Analyzer 1.0 when the cartridge is inserted into the instrument to run a test.

Specimen Handling, Storage and Preparation

Nasopharyngeal swab samples should be collected and handled according to the manufacturer's recommended procedures.

Recommended storage conditions for NPS (nasopharyngeal swab) resuspended in Universal Transport Medium (UTM) specimens are listed below:

- Room temperature up to 4 hours at 15–25°C
- Refrigerated up to 3 days at 2–8°C
- Frozen up to 30 days at –15 to –25°C

Procedure

Internal Control

The QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge includes a full process Internal Control, which is titered MS2 bacteriophage. The MS2 bacteriophage is a single-stranded RNA virus that is included in the cartridge in dried form and is rehydrated upon sample loading. This Internal Control material verifies all steps of the analysis process, including sample resuspension/homogenization, lysis, nucleic acid purification, reverse transcription, and PCR.

A positive signal for the Internal Control indicates that all processing steps performed by the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge were successful.

A negative signal of the Internal Control does not negate any positive results for detected and identified targets, but it does invalidate all negative results in the analysis. Therefore, the test should be repeated if the Internal Control signal is negative.

Protocol: Dry swab samples

Sample collection, transport, and storage

Collect nasopharyngeal swab samples using Copan FLOQSwabs (cat. no. 503CS01) according to the manufacturer's recommended procedures.

Loading a sample into the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge

1. Open the package of a QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge using the tear notches on the sides of the packaging (Figure 5).

   IMPORTANT: After the package is opened, sample should be introduced inside the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge and loaded into the QIAstat-Dx Analyzer 1.0 within 120 minutes.

Figure 5. Opening the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.

2. Remove the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge from the packaging and position it so that the bar code on the label faces you.
3. Manually write the sample information or place a sample information label on the top of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge. Make sure that the label is properly positioned and does not block the lid opening (Figure 6).

Figure 6. Sample information placement on top of QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.

4. Open the sample lid of the swab port on the left side of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge (Figure 7).

Figure 7. Opening the sample lid of swab port.

5. Insert the swab into the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge until the breakpoint is aligned with the access opening (i.e., the swab will go no further) (Figure 8).

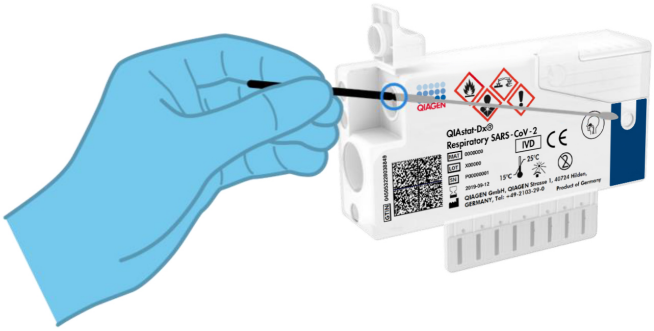

Figure 8. Inserting swab into the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.

6. Break the swab shaft at the breakpoint, leaving the rest of the swab in the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge (Figure 9).

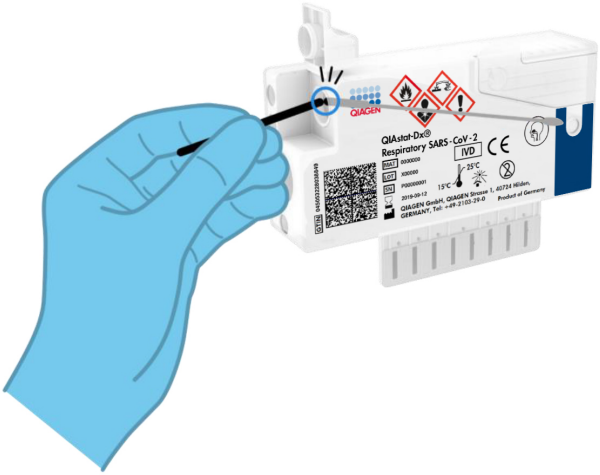

Figure 9. Breaking swab shaft.

7. Firmly close the sample lid of the swab port until it clicks (Figure 10).

   IMPORTANT: After the sample is placed inside the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge, the cartridge must be loaded into the QIAstat-Dx Analyzer 1.0 within 90 minutes.

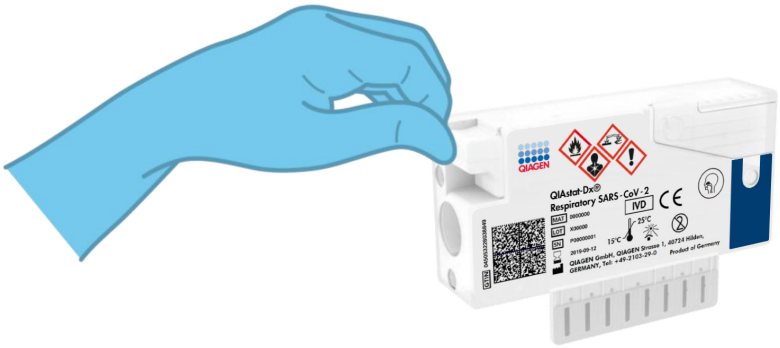

Figure 10. Closing the sample lid of the swab port.

Starting the QIAstat-Dx Analyzer 1.0

8. Power ON the QIAstat-Dx Analyzer 1.0 using the On/Off button on the front of the instrument.

   Note: The power switch on the back of the Analytical Module must be set in the "I" position. The QIAstat-Dx Analyzer 1.0 status indicators will turn blue.

9. Wait until the **Main** screen appears and the QIAstat-Dx Analyzer 1.0 status indicators turn green and stop blinking.

10. Log in to the QIAstat-Dx Analyzer 1.0 by entering the user name and password.

    Note: The **Login** screen will appear if **User Access Control** is activated. If the **User Access Control** is disabled, no user name/password will be required, and the **Main** screen will appear.

11. If the Assay Definition File software has not been installed on the QIAstat-Dx Analyzer 1.0, follow the installation instructions prior to running the test (see "Appendix A: Installing the Assay Definition File", page 81, for additional information).

Running a test

12. Press the **Run Test** button in the top right corner of the touchscreen of the QIAstat-Dx Analyzer 1.0.

13. When prompted, scan the sample ID bar code on the nasopharyngeal swab sample (located on the swab blister packaging), or scan the specimen information bar code located on the top of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge (see step 3) using the integrated front bar code reader of the QIAstat-Dx Analyzer 1.0 (Figure 11, next page).

    Note: It is also possible to enter the sample ID using the virtual keyboard of the touchscreen by selecting the **Sample ID** field.

    Note: Depending on the chosen system configuration, entering the patient ID may also be required at this point.

    Note: Instructions from the QIAstat-Dx Analyzer 1.0 appear in the **Instructions Bar** at the bottom of the touchscreen.

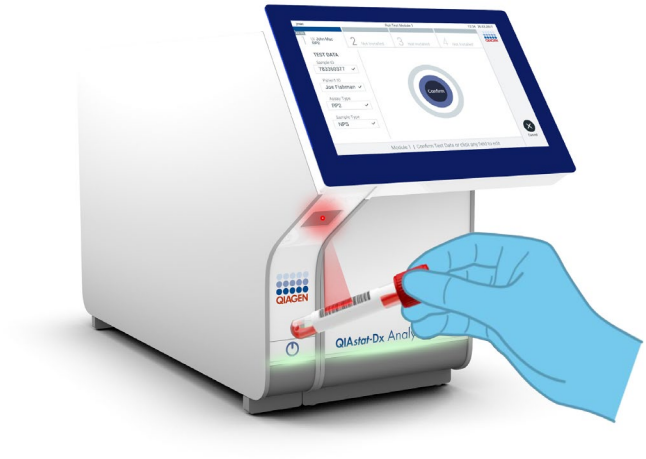

Figure 11. Scanning sample ID bar code.

14. When prompted, scan the bar code of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge to be used (Figure 12, next page). The QIAstat-Dx Analyzer 1.0 automatically recognizes the assay to be run based on the cartridge bar code.

    Note: The QIAstat-Dx Analyzer 1.0 will not accept QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges with lapsed expiration dates, previously used cartridges, or cartridges for assays that have not been installed on the unit. An error message will be shown in these cases, and the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge will be rejected. Refer to the *QIAstat-Dx Analyzer 1.0 User Manual* for further details on how to install assays.

Figure 12. Scanning QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge bar code.

15. Select the appropriate sample type from the list (Figure 13).

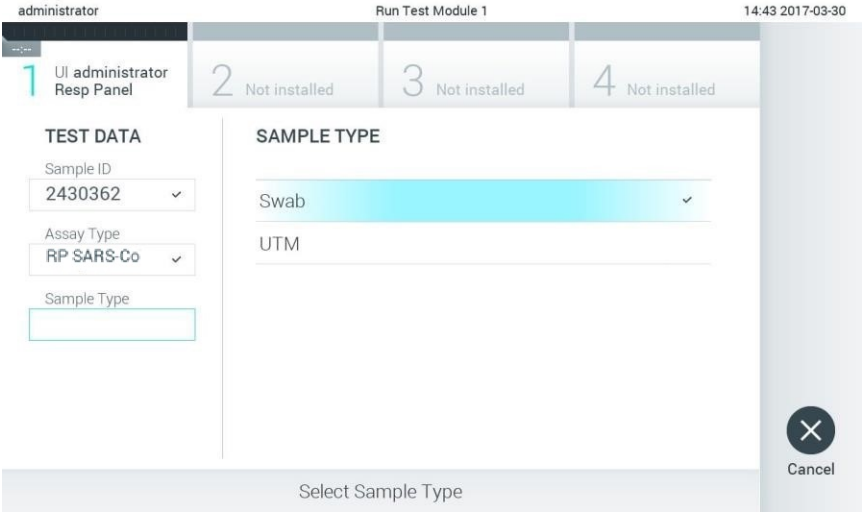

Figure 13. Selecting sample type.

16. The **Confirm** screen will appear. Review the entered data and make any necessary changes by selecting the relevant fields on the touchscreen and editing the information.
17. Press **Confirm** when all the displayed data are correct. If needed, select the appropriate field to edit its content, or press **Cancel** to cancel the test (Figure 14).

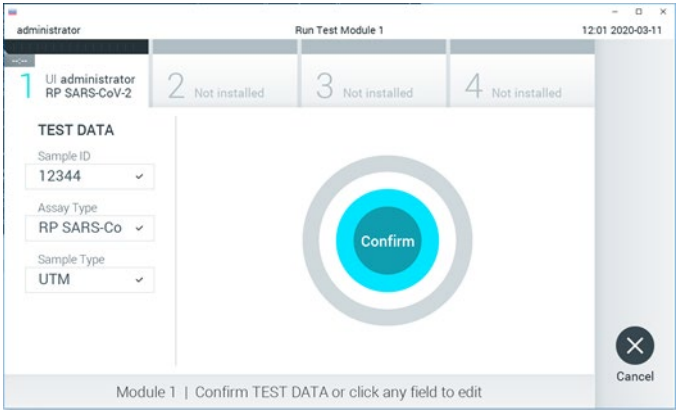

Figure 14. Confirming data entry.

18. Make sure that both sample lids of the swab port and main port of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge are firmly closed. When the cartridge entrance port on the top of the QIAstat-Dx Analyzer 1.0 automatically opens, insert the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge with the bar code facing to the left and the reaction chambers facing down (Figure 15, next page).

    Note: There is no need to push the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge into the QIAstat-Dx Analyzer 1.0. Position it correctly into the cartridge entrance port and the QIAstat-Dx Analyzer 1.0 will automatically move the cartridge into the Analytical Module.

Figure 15. Inserting QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge into QIAstat-Dx Analyzer 1.0.

19. Upon detecting the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge, the QIAstat-Dx Analyzer 1.0 will automatically close the lid of the cartridge entrance port and start the test run. No further action from the operator is required to start the run.

    Note: The QIAstat-Dx Analyzer 1.0 will not accept a QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge other than the one used and scanned during the test setup. If a cartridge other than the one scanned is inserted, an error will be generated and the cartridge will be automatically ejected.

    Note: Up to this point, it is possible to cancel the test run by pressing the **Cancel** button in the bottom right corner of the touchscreen.

    Note: Depending on the system configuration, the operator may be required to re-enter their user password to start the test run.

    Note: The lid of the cartridge entrance port will close automatically after 30 seconds if a QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge is not positioned in the port. If this occurs, repeat the procedure starting with step 16.

20. While the test is running, the remaining run time is displayed on the touchscreen.

21. After the test run is completed, the **Eject** screen will appear (Figure 16, next page) and the Module status bar will display the test result as one of the following options:

    - **TEST COMPLETED:** The test was completed successfully
    - **TEST FAILED**: An error occurred during the test
    - **TEST CANCELED**: The user canceled the test

    IMPORTANT: If the test fails, refer to the "Troubleshooting" section in the *QIAstat-Dx Analyzer 1.0 User Manual* for possible reasons and instructions on how to proceed.

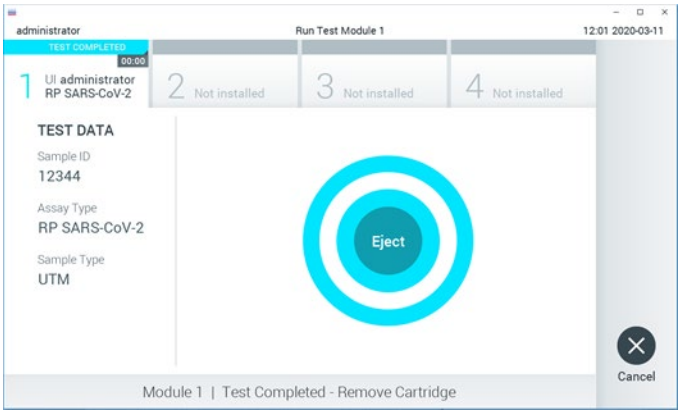

Figure 16. Eject screen display.

22. Press **Eject** on the touchscreen to remove the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge and dispose of it as biohazardous waste in accordance with all national, state, and local health and safety regulations and laws. The QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge should be removed when the cartridge entrance port opens and ejects the cartridge. If the cartridge is not removed after 30 seconds, it will automatically move back into the QIAstat-Dx Analyzer 1.0 and the cartridge entrance port lid will close. If this occurs, press **Eject** to open the lid of the cartridge entrance port again and then remove the cartridge.

    IMPORTANT: Used QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges must be discarded. It is not possible to re-use cartridges for tests for which the execution was started but then subsequently canceled by the operator, or for which an error was detected.

23. After the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge has been ejected, the results **Summary** screen will appear. Refer to "Interpretation of Results", page 42, for further details. To begin the process for running another test, press **Run Test**.

    Note: For further information on the use of the QIAstat-Dx Analyzer 1.0, refer to the *QIAstat-Dx Analyzer 1.0 User Manual*.

Protocol: Transport medium liquid samples

Sample collection, transport and storage

Collect nasopharyngeal swab samples according to the swab manufacturer's recommended procedures and place the swab into UTM.

Loading a sample into the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge

1. Open the package of a QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge using the tear notches on the sides of the packaging (Figure 17).

   IMPORTANT: After the package is open, sample should be introduced inside the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge and loaded into the QIAstat-Dx Analyzer 1.0 within 120 minutes.

Figure 17. Opening the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.

2. Remove the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge from the packaging and position it so that the bar code on the label faces you.
3. Manually write the sample information, or place a sample information label, on the top of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge. Make sure that the label is properly positioned and does not block the lid opening (Figure 18).

Figure 18. Sample information placement on top of QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.

4. Open the sample lid of the main port on the front of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge (Figure 19).

Figure 19. Opening the sample lid of main port.

5. Open the tube with the sample to be tested. Use the supplied transfer pipette to draw up fluid to the third fill line on the pipette (i.e., 300 µl) (Figure 20).

   IMPORTANT: Take care to avoid drawing air into the pipette. If Copan UTM® Universal Transport Medium is used as transport medium, take care not to aspirate any of the beads present in the tube. If air or beads are drawn into the pipette, carefully expel the sample fluid in the pipette back into the sample tube and draw up fluid again.

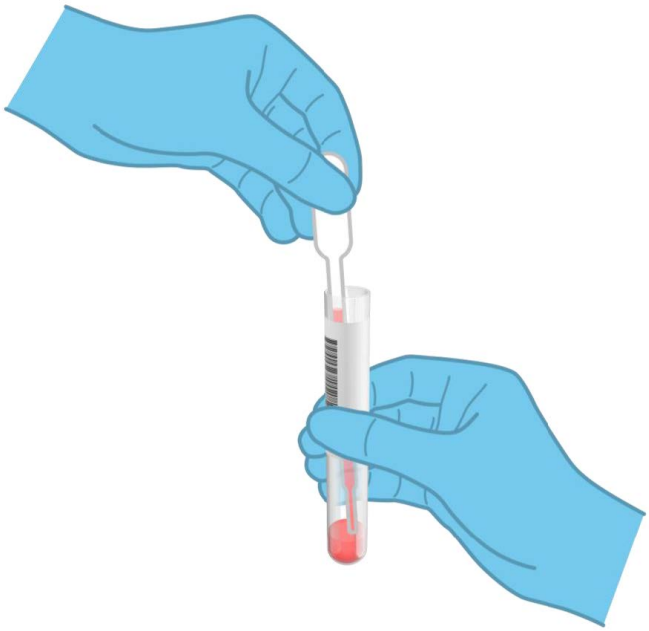

Figure 20. Drawing up sample into the supplied transfer pipette.

6. Carefully transfer 300 µl of sample volume into the main port of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge using the supplied single-use transfer pipette (Figure 21, next page).

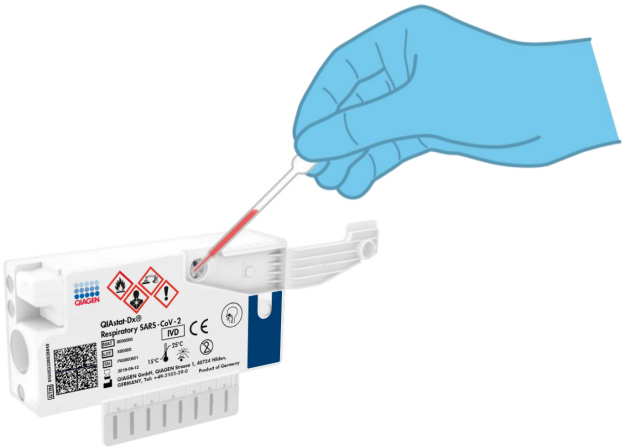

Figure 21. Transferring sample to main port of QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.

7. Firmly close the sample lid of the main port until it clicks (Figure 22).

Figure 22. Closing the sample lid of the main port.

8. Visually confirm that the sample has been loaded by checking the sample inspection window of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge (Figure 23).

   IMPORTANT: After the sample is placed inside the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge, the cartridge must be loaded into the QIAstat-Dx Analyzer 1.0 within 90 minutes.

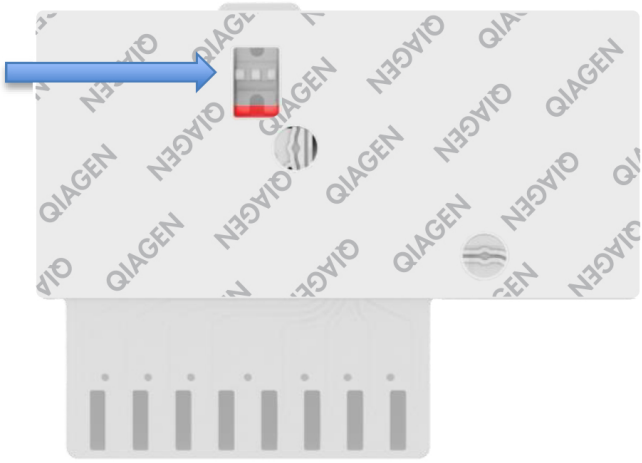

Figure 23. Sample inspection window (blue arrow).

Starting the QIAstat-Dx Analyzer 1.0

9. Power ON the QIAstat-Dx Analyzer 1.0 using the On/Off button on the front of the instrument.

   Note: The power switch on the back of the Analytical Module must be set in the "I" position. The QIAstat-Dx Analyzer 1.0 status indicators will turn blue.

10. Wait until the **Main** screen appears and the QIAstat-Dx Analyzer 1.0 status indicators turn green and stop blinking.

11. Log in to the QIAstat-Dx Analyzer 1.0 by entering the user name and password.

    Note: The **Login** screen will appear if **User Access Control** is activated. If the **User Access Control** is disabled, no user name/password will be required and the **Main** screen will appear.

12. If the Assay Definition File software has not been installed on the QIAstat-Dx Analyzer 1.0, follow the installation instructions prior to running the test (see Appendix A: Installing the Assay Definition File, page 81, for additional information).

Running a test

13. Press the **Run Test** button in the top right corner of the touchscreen of the QIAstat-Dx Analyzer 1.0.
14. When prompted, scan the sample ID bar code on the UTM tube containing the sample, or scan the specimen information bar code located on the top of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge (see step 3), using the integrated front bar code reader of the QIAstat-Dx Analyzer 1.0 (Figure 24).

    Note: It is also possible to enter the sample ID using the virtual keyboard of the touchscreen by selecting the **Sample ID** field.

    Note: Depending on the chosen system configuration, entering the patient ID may also be required at this point.

    Note: Instructions from the QIAstat-Dx Analyzer 1.0 appear in the **Instructions Bar** at the bottom of the touchscreen.

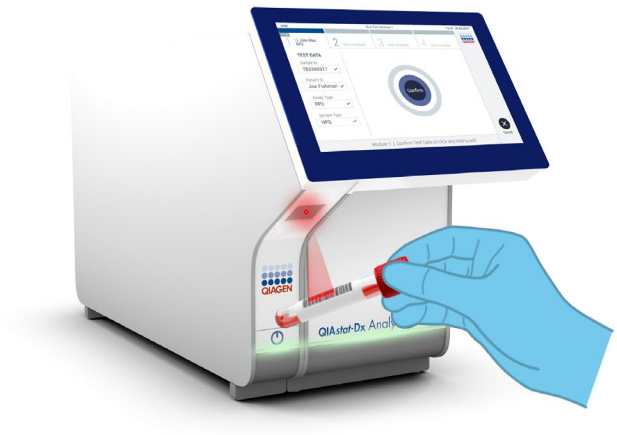

Figure 24. Scanning sample ID bar code.

15. When prompted, scan the bar code of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge to be used (Figure 25). The QIAstat-Dx Analyzer 1.0 automatically recognizes the assay to be run based on the cartridge bar code.

    Note: The QIAstat-Dx Analyzer 1.0 will not accept QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges with lapsed expiration dates, previously used cartridges or cartridges for assays that have not been installed on the unit. An error message will be shown in these cases and the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge will be rejected. Refer to the *QIAstat-Dx Analyzer 1.0 User Manual* for further details on how to install assays.

Figure 25. Scanning QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge bar code.

16. Select the appropriate sample type from the list (Figure 26, next page).

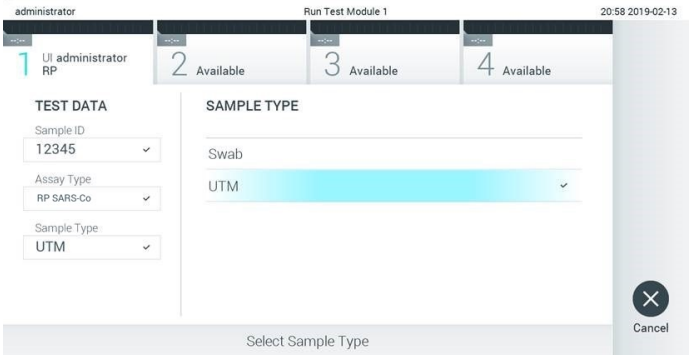

Figure 26. Selecting sample type.

17. The **Confirm** screen will appear. Review the entered data and make any necessary changes by selecting the relevant fields on the touchscreen and editing the information.
18. Press **Confirm** when all the displayed data are correct. If needed, select the appropriate field to edit its content, or press **Cancel** to cancel the test (Figure 27).

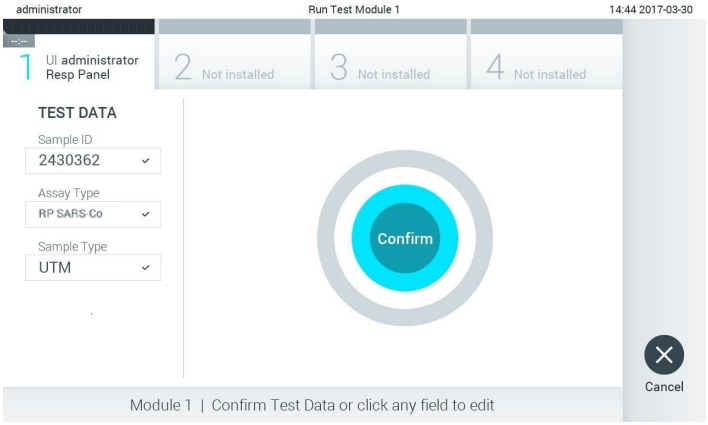

Figure 27. Confirming data entry.

19. Make sure that both sample lids of the swab port and main port of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge are firmly closed. When the cartridge entrance port on the top of the QIAstat-Dx Analyzer 1.0 automatically opens, insert the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge with the bar code facing to the left and the reaction chambers facing down (Figure 28).

    Note: There is no need to push the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge into the QIAstat-Dx Analyzer 1.0. Position it correctly into the cartridge entrance port and the QIAstat-Dx Analyzer 1.0 will automatically move the cartridge into the Analytical Module.

Figure 28. Inserting QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge into QIAstat-Dx Analyzer 1.0.

20. Upon detecting the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge, the QIAstat-Dx Analyzer 1.0 will automatically close the lid of the cartridge entrance port and start the test run. No further action from the operator is required to start the run.

    Note: The QIAstat-Dx Analyzer 1.0 will not accept a QIAstat-Dx Respiratory SARS CoV-2 Panel Cartridge other than the one used and scanned during the test setup. If a cartridge other than the one scanned is inserted, an error will be generated and the cartridge will be automatically ejected.

    Note: Up to this point, it is possible to cancel the test run by pressing the **Cancel** button in the bottom right corner of the touchscreen.

    Note: Depending on the system configuration, the operator may be required to re-enter their user password to start the test run.

    Note: The lid of the cartridge entrance port will close automatically after 30 seconds if a QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge is not positioned in the port. If this occurs, repeat the procedure starting with step 17.

21. While the test is running, the remaining run time is displayed on the touchscreen.

22. After the test run is completed, the **Eject** screen will appear (Figure 29, next page) and the Module status bar will display the test result as one of the following options:

    - **TEST COMPLETED**: The test was completed successfully
    - **TEST FAILED**: An error occurred during the test
    - **TEST CANCELED**: The user canceled the test

    IMPORTANT: If the test fails, refer to the "Troubleshooting" section in the *QIAstat-Dx Analyzer 1.0 User Manual* for possible reasons and instructions on how to proceed.

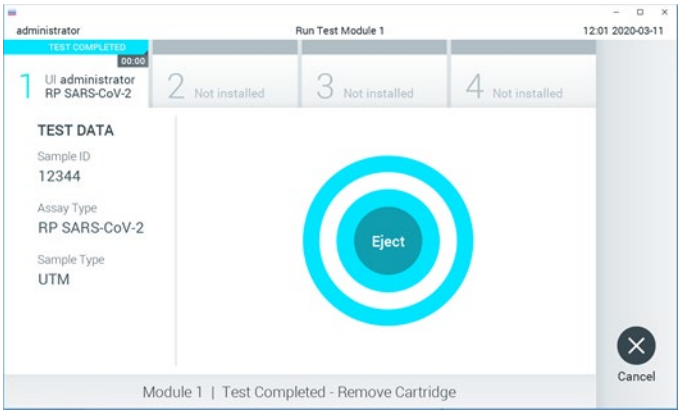

Figure 29. Eject screen display.

23. Press **Eject** on the touchscreen to remove the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge and dispose of it as biohazardous waste in accordance with all national, state and local health and safety regulations and laws. The QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge should be removed when the cartridge entrance port opens and ejects the cartridge. If the cartridge is not removed after 30 seconds, it will automatically move back into the QIAstat-Dx Analyzer 1.0 and cartridge entrance port lid will close. If this occurs, press **Eject** to open the lid of the cartridge entrance port again and then remove the cartridge.

    IMPORTANT: Used QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges must be discarded. It is not possible to re-use cartridges for tests for which the execution was started but then subsequently canceled by the operator, or for which an error was detected.

24. After the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge has been ejected, the results **Summary** screen will appear. Refer to "Interpretation of Results", page 42, for further details. To begin the process for running another test, press **Run Test**.

    Note: For further information on the use of the QIAstat-Dx Analyzer 1.0, refer to the *QIAstat-Dx Analyzer 1.0 User Manual*.

Interpretation of Results

Viewing results

The QIAstat-Dx Analyzer 1.0 automatically interprets and saves test results. After ejecting the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge, the results **Summary** screen is automatically displayed (Figure 30).

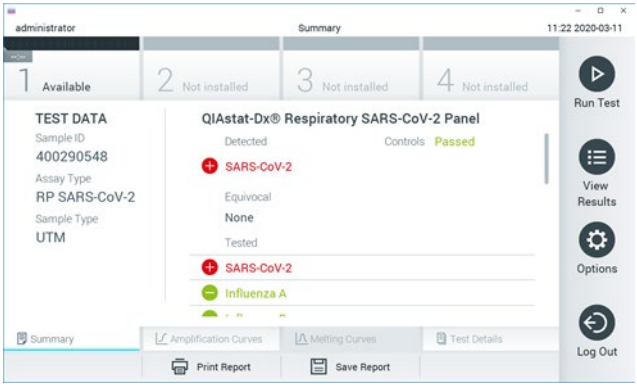

Figure 30. Results Summary screen example showing Test Data on the left panel and Test Summary in the main panel.

The main part of the screen provides the following three lists and uses color-coding and symbols to indicate the results:

- The first list, under the heading "Detected", includes all pathogens detected and identified in the sample, which are preceded by a ⊕ sign and are colored red.
- The second list, under the heading "Equivocal" is not used. "Equivocal" results are not applicable for the QIAstat-Dx Respiratory SARS-CoV-2 Panel. Therefore, the "Equivocal" list will always be empty.

- The third list, under the heading "Tested", includes all pathogens tested in the sample. Pathogens detected and identified in the sample are preceded by a  sign and are colored red. Pathogens that were tested but not detected are preceded by a  sign and are colored green.

Note: Pathogens detected and identified in the sample are shown in both the "Detected" and "Tested" lists.

If the test failed to complete successfully, a message will indicate "Failed" followed by the specific Error Code.

The following Test Data is shown on the left side of the screen:

- Sample ID
- Assay Type
- Sample Type

Further data about the assay is available, depending on the operator's access rights, through the tabs at the bottom of the screen (e.g., amplification plots and test details).

A report with the assay data can be exported to an external USB storage device. Insert the USB storage device into one of the USB ports of the QIAstat-Dx Analyzer 1.0 and press **Save Report** in the bottom bar of the screen. This report can be exported later at any time by selecting the test from the **View Result List**.

The report can also be sent to the printer by pressing **Print Report** in the bottom bar of the screen.

Viewing amplification curves

To view test amplification curves of pathogens detected, press the **Amplification Curves** tab (Figure 31).

Figure 31. Amplification Curves screen (PATHOGENS tab).

Details about the tested pathogens and controls are shown on the left and the amplification curves are shown in the center.

Note: If **User Access Control** is enabled on the QIAstat-Dx Analyzer 1.0 the **Amplification Curves** screen is only available for operators with access rights.

Press the **PATHOGENS** tab on the left side to display the plots corresponding to the tested pathogens. Press on the pathogen name to select which pathogens are shown in the amplification plot. It is possible to select single, multiple or no pathogens. Each pathogen in the selected list will be assigned a color corresponding to the amplification curve associated with the pathogen. Unselected pathogens will be shown in gray.

The corresponding C_T and endpoint fluorescence (EP) values are shown below each pathogen name.

Press the **CONTROLS** tab on the left side to view the controls in the amplification plot. Press the circle next to the control name to select or deselect it (Figure 32).

Figure 32. Amplification Curves screen (CONTROLS tab).

The amplification plot displays the data curve for the selected pathogens or controls. To alternate between logarithmic or linear scale for the Y-axis, press the **Lin** or **Log** button at the bottom left corner of the plot.

The scale of the X-axis and Y-axis can be adjusted using the ● **blue pickers** on each axis. Press and hold a **blue picker** and then move it to the desired location on the axis. Move a **blue picker** to the axis origin to return to the default values.

Viewing test details

Press **Test Details** in the Tab Menu bar at the bottom of the touchscreen to review the results in more detail. Scroll down to see the complete report.

The following Test Details are shown in the center of the screen (Figure 33, next page):

- User ID
- Cartridge SN (serial number)
- Cartridge Expiration Date
- Module SN (serial number)
- Test Status (Completed, Failed, or Canceled by operator)
- Error Code (if applicable)
- Test Start Date and Time
- Test Execution Time
- Assay Name
- Test ID
- Test Result:
  - Positive (if at least one respiratory pathogen is detected/identified)
  - Negative (no respiratory pathogen is detected)
  - Invalid
- List of analytes tested in the assay, with C_T and endpoint fluorescence in the event of a positive signal
- Internal Control, with C_T and endpoint fluorescence

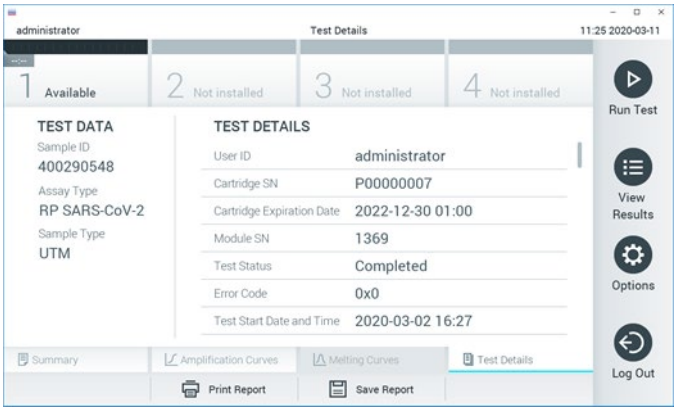

Figure 33. Example screen showing Test Data on the left panel and Test Details in the main panel.

Browsing results from previous tests

To view results from previous tests that are stored in the results repository, press **View Results** on the Main Menu bar (Figure 34).

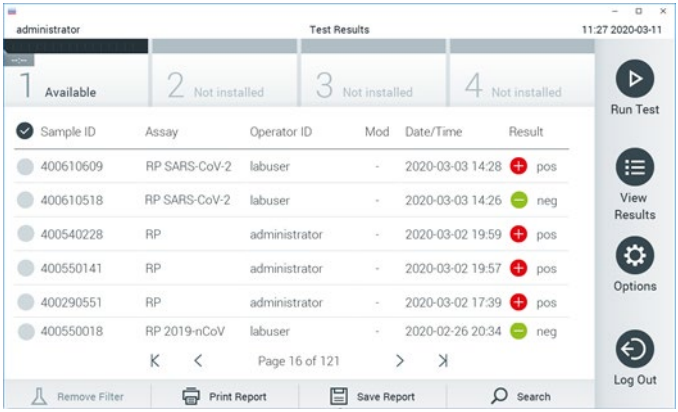

Figure 34. Example View Results screen.

The following information is available for every executed test (Figure 35):

- Sample ID
- Assay (name of test assay, which is "RP" for Respiratory Panel)
- Operator ID
- Mod (Analytical Module on which the test was executed)
- Date/Time (date and time when the test was finished)
- Result (outcome of the test: positive [pos], negative [neg], failed [fail] or successful [suc])

Note: If **User Access Control** is enabled on the QIAstat-Dx Analyzer 1.0, the data for which the user has no access rights will be hidden with asterisks.

Select one or more test results by pressing the **gray circle** to left of the sample ID. A **checkmark** will appear next to selected results. Unselect test results by pressing this **checkmark**. The entire list of results can be selected by pressing the **checkmark circle** in the top row (Figure 35).

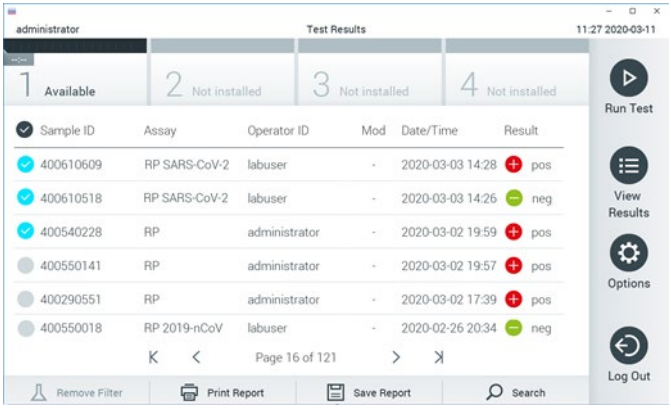

Figure 35. Example of selecting Test Results in the View Results screen.

Press anywhere in the test row to view the result for a particular test.

Press a column headline (e.g., **Sample ID**) to sort the list in ascending or descending order according to that parameter. The list can be sorted according to only one column at a time.

The **Result** column shows the outcome of each test (Table 2):

Table 2. Descriptions of test results

Outcome	Result	Description
Positive	pos	At least one pathogen is positive
Negative	neg	No pathogens were detected
Failed	fail	The test failed because either an error occurred or the test was canceled by the user
Successful	suc	The test is either positive or negative, but the user does not have the access rights to view the test results

Make sure a printer is connected to the QIAstat-Dx Analyzer 1.0 and the proper driver is installed. Press **Print Report** to print the report(s) for the selected result(s).

Press **Save Report** to save the report(s) for the selected result(s) in PDF format to an external USB storage device.

Select the report type: List of Tests or Test Reports.

Press **Search** to search the test results by Sample ID, Assay and Operator ID. Enter the search string using the virtual keyboard and press **Enter** to start the search. Only the records containing the search text will be displayed in the search results.

If the results list has been filtered, the search will only apply to the filtered list.

Press and hold a column headline to apply a filter based on that parameter. For some parameters, such as **Sample ID**, the virtual keyboard will appear so the search string for the filter can be entered.

For other parameters, such as **Assay**, a dialog will open with a list of assays stored in the repository. Select one or more assays to filter only the tests that were performed with the selected assays.

The T symbol to the left of a column headline indicates that the column's filter is active.

A filter can be removed by pressing **Remove Filter** in the Submenu bar.

Exporting results to a USB drive

From any tab of the **View Results** screen, select **Save Report** to export and save a copy of the test results in PDF format to a USB drive. The USB port is located on the front of the QIAstat-Dx Analyzer 1.0.

Printing results

Make sure a printer is connected to the QIAstat-Dx Analyzer 1.0 and the proper driver is installed. Press **Print Report** to send a copy of the test results to the printer.

Result interpretation

A result for a respiratory organism is interpreted as "Positive" when the corresponding PCR assay is positive, except for Influenza A. The Influenza A assay in the QIAstat-Dx Respiratory SARS-CoV-2 Panel is designed to detect Influenza A as well as Influenza A subtype H1N1/2009, Influenza A subtype H1 or Influenza A subtype H3. In particular, this means:

- If seasonal Influenza A H1 strain is detected by the QIAstat-Dx Respiratory SARS-CoV-2 Panel assay, two signals will be generated and displayed on the QIAstat-Dx Analyzer 1.0 screen: one for Influenza A and a second one for H1 strain.
- If seasonal Influenza A H3 strain is detected by the QIAstat-Dx Respiratory SARS-CoV-2 Panel assay, two signals will be generated and displayed on the QIAstat-Dx Analyzer 1.0 screen: one for Influenza A and a second one for H3 strain.
- If a pandemic Influenza A/H1N1/2009 strain is detected, two signals will be generated and displayed on the QIAstat-Dx Analyzer 1.0 screen: one for Influenza A and a second one for H1N1/2009.

For every other pathogen that can be detected with the QIAstat-Dx Respiratory SARS-CoV-2 Panel, only one signal will be generated if the pathogen is present in the sample.

Internal Control interpretation

Internal Control results are to be interpreted according to Table 3.

Table 3. Interpretation of Internal Control results

Control result	Explanation	Action
Passed	The Internal Control amplified successfully	The run was completed with success. All results are validated and can be reported. Detected pathogens are reported as "positive" and undetected pathogens are reported as "negative".
Failed	The Internal Control failed	Positively detected pathogen(s) are reported, but all negative results (tested but not detected pathogen[s]) are invalid. Repeat the testing using a new QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge.

Quality Control

In accordance with QIAGEN's ISO-certified Quality Management System, each lot of QIAstat-Dx Respiratory SARS-CoV-2 Panel is tested against predetermined specifications to ensure consistent product quality.

Limitations

- Results from the QIAstat-Dx Respiratory SARS-CoV-2 Panel are not intended to be used as the sole basis for diagnosis, treatment, or other patient management decisions.
- Positive results do not rule out co-infection with organisms not included in the QIAstat-Dx Respiratory SARS-CoV-2 Panel. The agent detected may not be the definitive cause of the disease.
- Negative results do not preclude infection of the upper respiratory tract. Not all agents of acute respiratory infection are detected by this assay and sensitivity in some clinical settings may differ from that described in the package insert.
- A negative result with the QIAstat-Dx Respiratory SARS-CoV-2 Panel does not exclude the infectious nature of the syndrome. Negative assay results may originate from several factors and their combinations, including sample handling mistakes, variation in the nucleic acid sequences targeted by the assay, infection by organisms not included in the assay, organism levels of included organisms that are below the limit of detection for the assay and use of certain medications, therapies, or agents.
- The QIAstat-Dx Respiratory SARS-CoV-2 Panel is not intended for testing of samples other than those described in these Instructions for Use. Test performance characteristics have been established only with nasopharyngeal swab samples collected in transport medium, from individuals with acute respiratory symptoms.

- The QIAstat-Dx Respiratory SARS-CoV-2 Panel is intended to be used in conjunction with standard of care culture for organism recovery, serotyping and/or antimicrobial susceptibility testing where applicable.
- The results from the QIAstat-Dx Respiratory SARS-CoV-2 Panel must be interpreted by a trained healthcare professional within the context of all relevant clinical, laboratory, and epidemiological findings.
- The QIAstat-Dx Respiratory SARS-CoV-2 Panel can be used only with the QIAstat-Dx Analyzer 1.0.*
- The QIAstat-Dx Respiratory SARS-CoV-2 Panel is a qualitative assay and does not provide a quantitative value for detected organisms.
- Viral and bacterial nucleic acids may persist in vivo, even if the organism is not viable or infectious. Detection of a target marker does not imply that the corresponding organism is the causative agent of the infection or the clinical symptoms.
- Detection of viral and bacterial nucleic acids depends on proper sample collection, handling, transportation, storage, and loading into the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge. Improper operations for any of the aforementioned processes can cause incorrect results, including false-positive or false-negative results.
- The assay sensitivity and specificity for the specific organisms and for all organisms combined are intrinsic performance parameters of a given assay and do not vary depending on prevalence. In contrast, both the negative and positive predictive values of a test result are dependent on the disease/organism prevalence. Please note that a higher prevalence favors the positive predictive value of a test result, while a lower prevalence favors the negative predictive value of a test result.

* DiagCORE Analyzer instruments running QIAstat-Dx software version 1.2 or higher can be used as an alternative to QIAstat-Dx Analyzer 1.0 instruments.

Performance Characteristics

The QIAstat-Dx Respiratory SARS-CoV-2 Panel (Cat. no. 691214) assay was developed by introducing the SARS-CoV-2 target in a separate reaction chamber of the QIAstat-Dx Respiratory Panel assay (Cat. No. 691211) leaving all other targets unchanged. It is known that sample preparation and RT-qPCR in the QIAstat-Dx Respiratory SARS-CoV-2 Panel cartridge are steps common to all target organisms. In the cartridge, the pooled sample and PCR enzyme mixture is equally allocated to each reaction chamber. As a result of this and/or availability of SARS-CoV-2 clinical samples, certain studies shown below were not done or repeated using the QIAstat-Dx Respiratory SARS-CoV-2 Panel.

Clinical performance

Clinical performance of the SARS-CoV-2 target

Clinical testing with retrospective nasopharyngeal swab specimens in transport medium was conducted at a hospital in Paris (France). Samples tested by the QIAstat-Dx Respiratory SARS-CoV-2 Panel were compared with the results of the standard of care (SOC) method(s) at the site (Corman et al. workflow developed at the Institute of Virology, Charité University Hospital, Berlin and endorsed by the World Health Organization (WHO)).

A total of 16 NPS samples were tested. Positive Percentage Agreement (PPA%) and Negative Percentage Agreement (NPA%) were calculated to measure the concordance between the 2 methods (Table 4).

Table 4. QIAstat-Dx Respiratory SARS-CoV-2 Panel

Nasopharyngeal swab samples		Corman et al. Workflow	
		Positive	**Negative**
QIAstat-Dx Respiratory SARS-CoV-2 Panel Result	**Positive**	11	0
	Negative	0	5
		PPA%	**NPA**
		100%	100%

Extensive clinical performance was established using the QIAstat-Dx Respiratory Panel (Cat. no. 691211) assay, which does not include SARS-CoV-2, and data shown below for clinical performance, unless where specifically noted, were established using this assay.

Clinical performance of the QIAstat-Dx Respiratory Panel

The performance characteristics of the QIAstat-Dx Respiratory Panel assay (excluding SARS-CoV-2) were assessed in a multicenter clinical trial. The performances of the universal transport medium of a nasopharyngeal swab specimen (UTM) and a dry nasopharyngeal swab specimen (FLOQSwabs, Copan ref 503CS01) (SWAB) were both assessed. In the latter case, a swab is directly introduced in the QIAstat-Dx Respiratory Panel Cartridge after collection, avoiding the transfer into a liquid medium. This testing approach may greatly support safe and error-free sample management, especially at the point of care setting.

The study was designed as observational and prospective-retrospective using left-over samples obtained from subjects with signs and symptoms of an acute respiratory infection. Participating sites were asked to test fresh and/or frozen clinical samples, according to a protocol and site/specific instructions.

Three (3) hospital laboratories located in Copenhagen (Denmark), Bonn (Germany), and Paris (France) participated in the study. Samples tested by the QIAstat-Dx Respiratory Panel were compared with the results of the standard of care (SOC) method(s) at the sites, as well as with a range of validated and commercially available molecular methods. This approach provided results for pathogens not detected by SOC and/or allowed for final discrepancy resolution of discordant results. As such, the QIAstat-Dx Respiratory Panel assay results were compared against FilmArray® Respiratory Panel 1.7 & 2 and the Allplex® Respiratory Panel assay.

A total of 578 clinical UTM patient samples were enrolled into the study. One (1) sample was excluded from the analysis due to the sample being misplaced between QIAstat-Dx and comparator testing. Seven (7) out of 577 samples failed initial testing, resulting in a first testing success rate of 98.8%.

The failure rate includes the failure rate of the Internal Control, which was 0.17% (1/577). Two (2) samples could not be retested due to insufficient remaining specimen volume. The sample that showed an initial Internal Control failure was successful upon retesting.

Fifteen (15) pathogen results could not be resolved because there was no SOC result (10 results) or no resolution method result available (5 results). This resulted in the exclusion of 2 samples; the remaining unresolvable results were in samples with multiple pathogens detected (coinfection samples).

Clinical Sensitivity or Positive Percent Agreement (PPA) was calculated as 100% x (TP/[TP + FN]). True positive (TP) indicates that both the QIAstat-Dx Respiratory Panel and comparator(s) methods had a positive result for the organism, and false negative (FN) indicates that the QIAstat-Dx Respiratory Panel result was negative while the comparator methods results were positive. Specificity or Negative Percent Agreement (NPA) was calculated as 100% x (TN/[TN + FP]). True negative (TN) indicates that both the QIAstat-Dx Respiratory Panel and the comparator method had negative results, and a false positive (FP) indicates that the QIAstat-Dx Respiratory Panel result was positive but the comparator methods results were negative. For the calculation of the clinical specificity of the individual pathogens, the total available results were used with the concerning true and false positive organism results subtracted. The exact binomial two-sided 95% confidence interval was calculated for each point estimate.

A total of 698 results were available for analysis.* Overall Clinical Sensitivity or PPA could be calculated from 475 results. The overall Clinical Specificity or NPA was calculated from 190 full negative samples.

In total, 462 true positive and 204 true negative QIAstat-Dx Respiratory Panel results were found, as well as 13 false negative and 17 false positive results.

Table 5 displays QIAstat-Dx Respiratory Panel Clinical Sensitivity (or Positive Percent Agreement) and Clinical Specificity (or Negative Percent Agreement) with 95% Confidence Intervals.

* There were 7 *Chlamydophila pneumoniae* pathogens found by the comparator methods in the study samples. These were all correctly detected by the QIAstat-Dx Respiratory Panel but are not subject to this CE mark, and the sensitivity performance is therefore not reported. The 7 results were however included in the specificity calculation for the individual panel pathogens.

Table 5. QIAstat-Dx Respiratory Panel performance data

	TP/(TP+FN)	Sensitivity /PPA	95% CI	TN/(TN+FP)	Specificity /NPA	95% CI
Overall	462/475	97.3%	95.4%–98.4%	187/190	98.4%	95.5%–99.5%
Viruses						
Adenovirus	35/36	97.2%	85.8%–99.5%	659/662	99.5%	98.7%–99.8%
Bocavirus	4/4	100%	51.0%–100%	693/694	99.9%	99.2%–100%
Coronavirus 229E	4/5	80.0%	37.6%–96.4%	693/693	100%	99.4%–100%
Coronavirus HKU1	8/8	100%	67.6%–100%	690/690	100%	99.4%–100%
Coronavirus OC43	10/10	100%	72.2%–100%	688/688	100%	99.4%–100%
Coronavirus NL63	22/24	91.7%	74.2%–97.7%	674/674	100%	99.4%–100%
Human Rhinovirus/ Enterovirus	56/59	94.9%	86.1%–98.3%	629/639	98.4%	97.1%–99.1%
Human Metapneumovirus	22/22	100%	85.1%–100%	676/676	100%	99.4%–100%
Influenza A H3N2	36/36	100%	90.4%–100%	662/662	100%	99.4%–100%
Influenza A H1N1	29/29	100%	88.3%–100%	669/669	100%	99.4%–100%
Influenza A H1-2009 strain (pandemic)	11/12	91.7%	64.5%–98.5%	688/688	100%	99.4%–100%
Influenza B	55/56	98.2%	90.6%–99.7%	642/642	100%	99.4%–100%
Parainfluenza Virus 1 (PIV 1)	19/19	100%	83.2%–100%	696/696	100%	99.5%–100%
Parainfluenza Virus 2 (PIV 2)	3/3	100%	43.8%–100%	695/695	100%	99.5%–100%
Parainfluenza Virus 3 (PIV 3)	9/9	100%	70.1%–100%	689/689	100%	99.4%–100%
Parainfluenza Virus 4 (PIV 4)	5/6	83.3%	43.6%–97.0%	691/692	99.9%	99.2%–100%
Respiratory Syncytial Virus	100/103	97.1%	91.8%–99.0%	595/595	100%	99.4%–100%
Bacteria						
Bordetella pertussis	29/29	100%	88.3%–100%	693/693	100%	99.4%–100%
Mycoplasma pneumoniae	21/21	100%	84.5%–100%	676/677	99.8%	99.2%–100%

Note: There were no evaluable results available for *Legionella pneumophila*, because this pathogen was found in a low number in the study (2 detections) and because of the absence of comparator method results.

Note: The sensitivity and specificity performance results for Parainfluenza Virus 1 (17 of 19 results) and for *Bordetella pertussis* (24 of 29 results) include results from a previous study (DiagCORE® [now called QIAstat-Dx] Respiratory Panel assay study). This is a true reflection of the performance for these pathogens because no design or other changes were made for these pathogens between these 2 assays. Except for the sensitivity and specificity calculation of these respective organisms, these 41 results are not part of the 698 results used to calculate the specificity performance for the remaining QIAstat-Dx Respiratory Panel assay pathogens.

The QIAstat-Dx Respiratory Panel assay detected multiple organisms in 101 samples for a total of 228 organism results. This represents 26.3% of the total positive specimens (101/385). Eighty-two (82) samples were double infections, 15 were triple infections, and the remaining coinfection samples had 4 (3 samples) or more pathogens (1 sample had 7 pathogens).

Dry swab specimen

A total of 448 clinical samples were tested to assess the ability to test swabs as dry swabs and to assess the clinical performance characteristics of the dry swab specimens when introduced directly into the QIAstat-Dx Respiratory Panel Cartridge. This testing was conducted at 2 of the 3 sites that participated in the performance evaluation of the UTM specimen. The objective was to demonstrate equivalency between performance characteristics of the dry swab and the UTM specimens.

One clinical site had requested and obtained Institutional Review Board (IRB) approval to enroll patients for this part of the study. Patients consenting to participate in the study provided 2 nasopharyngeal swabs (one from each nostril). One swab was transferred into UTM and the other swab was directly entered into the QIAstat-Dx Respiratory Panel Cartridge. Ninety-eight (98) swab samples were enrolled following this approach. To augment the number of dry swab results and to ensure that all QIAstat-Dx Respiratory Panel pathogens were

represented in the dry swab testing, an additional 350 swabs were dipped in UTM. Because each swab holds approximately 0.1 ml of liquid after dipping, two (2) swabs were simultaneously dipped in UTM and introduced into the QIAstat-Dx Respiratory Panel Cartridge. For all swab specimens, the simultaneously tested UTM specimen served as the comparator method.

A minimum of 5 dry swab results were available for each QIAstat-Dx Respiratory Panel pathogen. Parainfluenza Virus 4 and *Legionella pneumophila* were exceptions as only 3 and 2 results, respectively, were available.

The Clinical Sensitivity (or PPA) was calculated as 100% x (TP/[TP + FN]). True positive (TP) indicates that both the dry swab and the UTM specimen had a positive result for a specific organism and false negative (FN) indicates that the dry swab result was negative while the UTM specimen result was positive. Specificity (or NPA) was calculated as 100% x (TN/[TN + FP]). True negative (TN) indicates that both the dry swab and UTM specimen had negative results and a false positive (FP) indicates that the dry swab result was positive but the UTM specimen result was negative. The exact binomial two-sided 95% confidence interval was calculated for each point estimate.

For all dry swabs, a total of 440 results were available for analysis. Overall Clinical Sensitivity (or PPA) could be calculated from 244 results. The overall Clinical Specificity (or NPA) was calculated from 196 results. In total, 241 true positive and 188 true negative dry swab results were found, as well as 3 false negative and 8 false positive dry swab results.

Because the same sample is tested, the dipped swab may be considered to be most relevant with regard to assessing equivalency of the dry swab with the UTM specimen performance. Testing of the dry swab involves taking 2 samples from the same patient and, although paired, a bias may occur due to this approach. Also, as the nasopharyngeal swab collection does present a level of discomfort for the patient, it is likely that the yields obtained between the 2 collections may differ.

For all dipped swabs, a total of 337 results were available for analysis. Overall Clinical Sensitivity (or PPA) could be calculated from 178 results. The overall Clinical Specificity (or NPA) was calculated from 159 results. In total, 177 true positive and 156 true negative dipped swab results were found, as well as 1 false negative and 3 false positive dipped swab results.

Table 6 shows QIAstat-Dx Respiratory Panel Sensitivity and Specificity characteristics with 95% Confidence Intervals for dry swab specimens.

Table 6. QIAstat-Dx Respiratory SARS-CoV-2 Panel performance data for dry swab specimens

	TP/(TP+FN)	Sensitivity/PPA	95% CI	TN/(TN+FP)	Specificity/NPA	95% CI
All dry swabs	241/244	98.8%	96.4%–99.6%	188/196	95.9%	92.2%–97.9%
Dipped swabs	177/178	99.4%	96.9%–99.9%	156/159	98.1%	94.6%–99.4%

Conclusion

This extensive multicenter study sought to assess the performance of the UTM specimen, as well as the equivalency of the dry swab, with the UTM specimen performance in the QIAstat-Dx Respiratory Panel assay.

The overall Clinical Sensitivity of the UTM specimen was found to be 97.3% (95% CI, 95.4%–98.4%). The overall Clinical Specificity in 190 full negative samples was 98.4% (95% CI, 95.5%–99.5%).

The overall Clinical Sensitivity of the dry swab specimen was found to be 98.8% (95% CI, 96.4%–99.6%). The overall Clinical Specificity for the dry swab specimen was 95.9% (95% CI, 92.2%–97.9%).

The dry swab study results supported the ability to test swabs entered directly into the QIAstat-Dx Respiratory Panel Cartridges as dry swabs. The dry swab specimen was found to show excellent concordance with the UTM specimen, as demonstrated by the overall agreement between the UTM specimen and dipped swabs, which was 98.5% (95% CI, 97%–99.5 %).

Analytical performance

Sensitivity (Limit of Detection)

The Analytical Sensitivity, or Limit of Detection (LoD), is defined as the lowest concentration at which ≥95% of the tested samples generate a positive call.

The LoD per analyte was determined using selected strains* representing individual pathogens that are possible to detect with the QIAstat-Dx Respiratory SARS-CoV-2 Panel. Simulated NPS sample matrix (cultured human cells in Copan UTM) was spiked with one (1) or more pathogens and tested in 20 replicates.

Individual LoD values for each target are shown in Table 7.

Table 7. LoD values obtained for the different respiratory target strains tested with the QIAstat-Dx Respiratory SARS-CoV-2 Panel

Pathogen	Strain	Source	Concentration	Detection rate
Influenza A H1N1	A/New Jersey/8/76	ATCC® VR-897	28.1 $CEID_{50}$/ml	20/20
	A/Brisbane/59/07	ZeptoMetrix® 0810244CFHI	0.04 $TCID_{50}$/ml	19/20
	A/New Caledonia/20/99	ZeptoMetrix 0810036CFHI	4.6 $TCID_{50}$/ml	19/20
Influenza A H3N2	A/Virginia/ATCC6/2012	ATCC VR-1811	0.4 PFU/ml	19/20
	A/Wisconsin/67/2005	ZeptoMetrix 0810252CFHI	2.5 $TCID_{50}$/ml	20/20
	A/Port Chalmers/1/73	ATCC VR-810	791.1 $CEID_{50}$/ml	20/20
Influenza A, subtype H1N1/2009	A/Virginia/ATCC1/2009	ATCC VR-1736	2.6 PFU/ml	20/20
	A/SwineNY/03/2009	ZeptoMetrix 0810249CFHI	14.1 $TCID_{50}$/ml	20/20
Influenza B	B/Virginia/ATCC5/2012	ATCC VR-1807	0.08 PFU/ml	20/20
	B/FL/04/06	ATCC VR-1804	34.8 $CEID_{50}$/ml	19/20
	B/Taiwan/2/62	ATCC VR-295	28.1 $CEID_{50}$/ml	20/20

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* Due to limited access to cultured virus, synthetic material (gBlock) was used to determine LoD spiked in clinical negative matrix for the SARS-CoV-2 target.

(Table 7 continued)

Pathogen	Strain	Source	Concentration	Detection rate
Coronavirus 229E	–	ATCC VR-740	0.3 $TCID_{50}$/ml	20/20
Coronavirus OC43	–	ATCC-1558	0.1 $TCID_{50}$/ml	20/20
Coronavirus NL63	–	ZeptoMetrix 0810228CFHI	0.01 $TCID_{50}$/ml	20/20
Coronavirus HKU1	–	ZeptoMetrix NATRVP-IDI	1/300*	19/20
SARS-CoV-2	-	IDT (gBlock)	500 copies/ml	19/20
Parainfluenza Virus 1 (PIV 1)	C35	ATCC VR-94	23.4 $TCID_{50}$/ml	20/20
Parainfluenza Virus 2 (PIV 2)	Greer	ATCC VR-92	5.0 $TCID_{50}$/ml	19/20
Parainfluenza Virus 3 (PIV 3)	C 243	ATCC VR-93	15.8 $TCID_{50}$/ml	20/20
Parainfluenza Virus 4 (PIV 4)	M-25	ATCC VR-1378	2.8 $TCID_{50}$/ml	20/20
Respiratory Syncytial Virus A	A2	ATCC VR-1540	2.8 $TCID_{50}$/ml	20/20
Respiratory Syncytial Virus B	9320	ATCC VR-955	0.02 $TCID_{50}$/ml	20/20
Human Metapneumovirus	Peru6-2003 (type B2)	ZeptoMetrix 0810159CFHI	1.1 $TCID_{50}$/ml	19/20
	hMPV-16, IA10-2003	ZeptoMetrix 0810161CFHI	3.0 $TCID_{50}$/ml	20/20
Adenovirus	GB (Adenovirus B3)	ATCC VR-3	50.0 $TCID_{50}$/ml	20/20
	RI-67 (Adenovirus E4)	ATCC VR-1572	15.8 $TCID_{50}$/ml	20/20
	Adenoid 75 (Adenovirus C5)	ATCC VR-5	5.0 $TCID_{50}$/ml	20/20
	Adenoid 71 (Adenovirus C1)	ATCC VR-1	5.0 $TCID_{50}$/ml	19/20
	Adenovirus C2	ATCC VR-846	28.1 $TCID_{50}$/ml	20/20
	Adenovirus C6	ATCC VR-6	505.6 $TCID_{50}$/ml	20/20
Bocavirus	Clinical sample	–	>1.0 copies/ml	20/20

* Relative dilution from stock concentration.

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(Table 7 continued)

Pathogen	Strain	Source	Concentration	Detection rate
Enterovirus	/US/IL/14-18952 (Enterovirus D68)	ATCC VR-1824	50.0 $TCID_{50}$/ml	19/20
	Echovirus 6 (D-1 (Cox))	ATCC VR-241	0.001 $TCID_{50}$/ml	19/20
Rhinovirus	1059 (Rhinovirus B14)	ATCC VR-284	28.1 $TCID_{50}$/ml	20/20
	HGP (Rhinovirus A2)	ATCC VR-482	0.3 $TCID_{50}$/ml	19/20
	11757 (Rhinovirus A16)	ATCC VR-283	8.9 $TCID_{50}$/ml	20/20
	Type 1A	ATCC VR-1559	5.0 $TCID_{50}$/ml	20/20
Mycoplasma pneumoniae	M129-B7	ATCC 29342	0.1 CFU/ml	20/20
Legionella pneumophila	CA1	ATCC 700711	>0.01 CFU/ml	20/20
Bordetella pertussis	I028	ATCC BAA-2707	>0.001 CFU/ml	20/20
	A639	ZeptoMetrix NATRVP-IDI	1/10000*	19/20

* Relative dilution from stock concentration.

Assay robustness

The verification of robust assay performance was assessed by analyzing the Internal Control performance in clinical nasopharyngeal swab samples. Thirty (30) individual nasopharyngeal swab samples, negative for all pathogens possible to detect, were analyzed with the QIAstat-Dx Respiratory Panel.

All samples tested showed a positive result and valid performance for the Internal Control of the QIAstat-Dx Respiratory Panel.

Exclusivity (Analytical Specificity)

The exclusivity study was carried out by in silico analysis and in vitro testing to assess the Analytical Specificity for respiratory or non-respiratory organisms that are not covered by the panel. These organisms included specimens which are related to, but distinct from, respiratory panel organisms or that could be present in specimens collected from the intended test population. Selected organisms are clinically relevant (colonizing the upper respiratory tract or causing respiratory symptoms), are common skin flora or laboratory contaminants, or are microorganisms for which much of the population may have been infected.

Samples were prepared by spiking potential cross-reactive organisms into simulated nasopharyngeal swab sample matrix at the highest concentration possible based on the organism stock, preferably 10^5 $TCID_{50}$/ml for viral targets and 10^6 CFU/ml for bacterial targets.

A certain level of cross-reactivity with Bordetella species was predicted by preliminary sequence analysis and was observed when high concentrations of *Bordetella holmesii* were tested. No cross-reactivity was observed with *Bordetella bronchiseptica* and *Bordetella parapertussis* at high concentrations. The target gene used for *Bordetella pertussis* detection (insertion element IS481) is a transposon also present in other Bordetella species. Table 8 shows the list of pathogens tested.

Table 8. List of Analytical Specificity pathogens tested

Type	Pathogen
Bacteria	*Bordetella bronchiseptica* *Bordetella holmesii* *Bordetella parapertussis* *Chlamydia trachomatis* *Enterobacter aerogenes* *Escherichia coli* (0157) *Haemophilus influenzae* *Klebsiella oxytoca* *Klebsiella pneumoniae* *Lactobacillus acidophilus* *Moraxella catarrhalis* *Mycoplasma genitalium* *Mycoplasma hominis* *Neisseria elongata* *Neisseria gonorrhoeae* *Neisseria meningitidis* *Pseudomonas aeruginosa* *Serratia marcescens* *Staphylococcus aureus* *Staphylococcus epidermidis* *Stenotrophomonas maltophilia* *Streptococcus agalactiae* *Streptococcus pneumoniae* *Streptococcus pyogenes* *Streptococcus salivarus*
Viruses	Cytomegalovirus Epstein-Barr Virus Herpes Simplex Virus 1 Herpes Simplex Virus 2 Measles Virus Mumps
Fungi	*Aspergillus fumigatus* *Candida albicans* *Cryptococcus neoformans*

All pathogens tested showed a negative result and no cross-reactivity was observed for the organisms tested in the QIAstat-Dx Respiratory SARS-CoV-2 Panel (except for *Bordetella holmesii* as described above).

In silico analysis was performed for all primer/probe designs included in the QIAstat-Dx Respiratory SARS-CoV-2 Panel, proving specific amplification and detection of targets without cross-reactivity.

For the SARS-CoV-2 target, only a limited number of organisms were tested in vitro (*Haemophilus influenzae*, *Streptococcus pyogenes*, *Chlamidophila pneumoniae*, *Streptococcus pneumoniae*, *Mycobacterium tuberculosis*, MERS Coronavirus, SARS Coronavirus). No cross-reactivity was observed, both *in silico* and *in vitro*, with any clinically relevant pathogens (colonizing the upper respiratory tract or causing respiratory symptoms), or common skin flora or laboratory contaminants, or microorganisms.

Inclusivity (Analytical Reactivity)*

An inclusivity study was performed to analyze the detection of a variety of strains that represent the genetic diversity of each respiratory panel target organism ("inclusivity strains"). Inclusivity strains for all analytes were included in the study, representative of the species/types for the different organisms (e.g., a range of Influenza A strains isolated from different geographical areas and in different calendar years were included). Table 9 (next page) shows the list of respiratory pathogens tested in this study.

* Not applicable to the SARS-CoV-2 target due to the presence of a single strain at time of study.

Table 9. List of Analytical Reactivity pathogens tested

Pathogen	Subtype/serotype	Strain	Source
Influenza A	H1N1	A/PR/8/34	ATCC VR-1469
		A/New Jersey/8/76	ATCC VR-897
		A/Brisbane/59/07	ZeptoMetrix 0810244CFHI
		A/New Caledonia/20/99	ZeptoMetrix 0810036CFHI
		A/Virginia/ATCC6/2012	ATCC VR-1811
		A/Wisconsin/67/2005	ZeptoMetrix 0810252CFHI
	H3N2	A/Port Chalmers/1/73	ATCC VR-810
		A/Victoria/3/75	ATCC VR-822
		A/Brisbane/10/07	ZeptoMetrix NATRVP-IDI
		A/Virginia/ATCC2/2009	ATCC VR-1737
		A/Virginia/ATCC3/2009	ATCC VR-1738
	H1N1 (pandemic)	A/Virginia/ATCC1/2009	ATCC VR-1736
		A/SwineNY/03/2009	ZeptoMetrix 0810249CFHI
		H1N1/NY/02/09	ZeptoMetrix NATRVP-IDI
Influenza B	Not available	B/Virginia/ATCC5/2012	ATCC VR-1807
		B/FL/04/06	ATCC VR-1804
		B/Taiwan/2/62	ATCC VR-295
		B/Panama/45/90	ZeptoMetrix NATFLUB-ERCM
		B/Florida/02/06	ZeptoMetrix 810037CFHI
		B/Maryland/1/59	ATCC VR-296
Coronavirus 229E	Not available	Not available	ATCC VR-740
		Not available	ZeptoMetrix NATRVP-IDI
Coronavirus OC43	Not available	Not available	ATCC-1558
		Not available	ZeptoMetrix 0810024CFHI
		Not available	ZeptoMetrix NATRVP-IDI
Coronavirus NL63	Not available	Not available	ZeptoMetrix 0810228CFHI
		Not available	ZeptoMetrix NATRVP-IDI
Coronavirus HKU1	Not available	Not available	ZeptoMetrix NATRVP-IDI

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(Table 9 continued)

Pathogen	Subtype/serotype	Strain	Source
Parainfluenza 1	Not available	C35	ATCC VR-94
		n/a	ZeptoMetrix NATPARA1-ST
		n/a	ZeptoMetrix NATRVP-IDI
Parainfluenza 2	Not available	Greer	ATCC VR-92
		Not available	ZeptoMetrix 0810015CFHI
		Not available	ZeptoMetrix NATRVP-IDI
Parainfluenza 3	Not available	C 243	ATCC VR-93
		Not available	ZeptoMetrix NATPARA3-ST
		Not available	ZeptoMetrix NATRVP-IDI
Parainfluenza 4	A	M-25	ATCC VR-1378
	B	CH 19503	ATCC VR-1377
	B	Not available	ZeptoMetrix NATRVP-IDI
RSV A	Not available	A2	ATCC VR-1540
		Long	ATCC VR-26
		Not available	ZeptoMetrix NATRVP-IDI
RSV B	Not available	9320	ATCC VR-955
		18537	ATCC VR-1580
		WV/14617/85	ATCC VR-1400
		Not available	ZeptoMetrix NATRSVB-ST
Human Metapneumovirus	B1	Peru2-2002	ZeptoMetrix 0810156CFHI
	B1	IA18-2003	ZeptoMetrix 0810162CFH
	B1	Peru3-2003	ZeptoMetrix 0810158CFHI
	B2	Peru6-2003	ZeptoMetrix 0810159CFHI
	B2	Peru1-2002	ZeptoMetrix 0810157CFHI
	A1	hMPV-16, IA10-2003	ZeptoMetrix 0810161CFHI
	A1	IA3-2002	ZeptoMetrix 0810160CFHI
	A2	IA14-2003	ZeptoMetrix 0810163CFH

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(Table 9 continued)

Pathogen	Subtype/serotype	Strain	Source
Adenovirus B	B21	AV-1645 [128]	ATCC VR-256
	B7	Gomen	ATCC VR-7
	B3	GB	ATCC VR-3
	B3	Not available	ZeptoMetrix NATADV3-ST
Adenovirus C	C1	Adenoid 71	ATCC VR-1
	C2	Not available	ATCC VR-846
	C5	Adenoid 75	ATCC VR-5
	C6	Not available	ATCC VR-6
Adenovirus E	E4	RI-67	ATCC VR-1572
Bocavirus	Not available	Not available	ZeptoMetrix 0601178NTS
		Not available	ZeptoMetrix MB-004
Enterovirus A	EV-A71	EV-A71	ZeptoMetrix 0810236CFHI
Enterovirus B	E-11	Gregory	ATCC VR-41
	E-30	Bastianni	ATCC VR-1660
	CV-A9	Griggs	ATCC VR-1311
	CV-B1	Conn-5	ATCC VR-28
	CV-B2	Ohio-1	ATCC VR-29
	CV-B3	Nancy	ATCC VR-30
	E-17	CHHE-29	ATCC VR-47
	Not available	Echovirus 6 (D-1 (Cox))	ATCC VR-241
Enterovirus C	CV-A21	Kuykendall [V-024-001-012]	ATCC VR-850
Enterovirus D	D68	US/IL/14-18952	ATCC VR-1824
	EV-D68	US/MO/14-18947	ATCC VR-1823

(Continued on next page)

(Table 9 continued)

Pathogen	Subtype/serotype	Strain	Source
Rhinovirus A	A1	Not available	ZeptoMetrix NATRVP-IDI
	1A	Not available	ATCC VR-1559
	A2	HGP	ATCC VR-482
	A16	11757	ATCC VR-283
	HRV-1B	B632	ATCC VR-1645
	HRV-A39	209	ATCC VR-340
Rhinovirus B	B14	1059	ATCC VR-284
M. pneumoniae	1	PI 1428	ATCC 29085
	Not available	M129	ZeptoMetrix NATMPN(M129)-ERCM
	Not available	M129-B7	ATCC 29342
	Not available	FH strain of Eaton Agent [NCTC 10119]	ATCC 15531
L. pneumophila	Not available	CA1	ATCC 700711
		Legionella pneumophila subsp. *Pneumophila*/169-MN-H	ATCC 43703
		Not available	ZeptoMetrix 0601645NTS
		subsp. *Pneumophila/ Philadelphia-1*	ATCC 33152
B. pertussis	Not available	I028	ATCC BAA-2707
		A639	ZeptoMetrix NATRVP-IDI
		18323 [NCTC 10739]	ATCC 9797

All pathogens tested showed positive results at the concentration tested.

Co-Infections

A co-infections study was performed to verify that multiple QIAstat-Dx Respiratory SARS-CoV-2 Panel analytes included in one nasopharyngeal swab sample can be detected.

High and low concentrations of different organisms were combined in one sample. Selection of organisms was made based on relevance, prevalence, and layout of the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge (distribution of targets in different reaction chambers).

Analytes were spiked into simulated NPS sample matrix (cultured human cells in UTM) in high (50x LoD concentration) and low concentrations (5x LoD concentration) and tested in different combinations. Table 10 shows the combination of co-infections tested in this study.

Table 10. List of co-infections combinations tested

Pathogens	Strain	Concentration
Influenza A/H3N2	A/Virginia/ATCC6/2012	50x LoD
Adenovirus C5	Adenoid 75	5x LoD
Influenza A/H3N2	A/Virginia/ATCC6/2012	5x LoD
Adenovirus C5	Adenoid 75	50x LoD
Parainfluenza 3	C243	50x LoD
Influenza A/H1N1/2009	NY/03/09	5x LoD
Parainfluenza 3	C243	5x LoD
Influenza A/H1N1/2009	NY/03/09	50x LoD
Respiratory Syncytial Virus A	A2	50x LoD
Influenza B	B/FL/04/06	5x LoD
Respiratory Syncytial Virus A	A2	5x LoD
Influenza B	B/FL/04/06	50x LoD
Adenovirus C5	Adenoid 75	50x LoD
Rhinovirus B, Type HRV-B14	1059	5x LoD
Adenovirus C5	Adenoid 75	5x LoD
Rhinovirus B, Type HRV-B14	1059	50x LoD

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(Table 10 continued)

Pathogens	Strain	Concentration
Respiratory Syncytial Virus A **Rhinovirus B, Type HRV-B14**	A2 1059	50x LoD 5x LoD
Respiratory Syncytial Virus A **Rhinovirus B, Type HRV-B14**	A2 1059	5x LoD 50x LoD
Respiratory Syncytial Virus B **Bocavirus**	9320 Not available	50x LoD 5x LoD
Respiratory Syncytial Virus B **Bocavirus**	9320 Not available	5x LoD 50x LoD
Coronavirus OC43 **Rhinovirus B, Type HRV-B14**	Not available 1059	50x LoD 5x LoD
Coronavirus OC43 **Rhinovirus B, Type HRV-B14**	Not available 1059	5x LoD 50x LoD
Human Metapneumovirus B2 **Parainfluenza 1**	Peru6-2003 C-35	50x LoD 5x LoD
Human Metapneumovirus B2 **Parainfluenza 1**	Peru6-2003 C-35	5x LoD 50x LoD
Coronavirus 229E **Respiratory Syncytial Virus A**	Not available A2	50x LoD 5x LoD
Coronavirus 229E **Respiratory Syncytial Virus A**	Not available A2	5x LoD 50x LoD
Respiratory Syncytial Virus B **Coronavirus NL63**	9320 Not available	50x LoD 5x LoD
Respiratory Syncytial Virus B **Coronavirus NL63**	9320 Not available	5x LoD 50x LoD

All co-infections tested gave a positive result for the two pathogens combined at low and high concentrations. No effect in results are observed due to the presence of co-infections.

Interfering substances

The influence of potential interfering substances on the performance of the QIAstat-Dx Respiratory Panel was evaluated in this study. The interfering substances include endogenous as well as exogenous substances that are normally found in the nasopharynx or may be introduced into NPS specimens during specimen collection, respectively.

A set of selected samples that cover all the respiratory pathogens from the panel were used for the interfering substances testing. Interfering substances were spiked into the selected samples at a level predicted to be above the concentration of the substance likely to be found in an authentic nasopharyngeal swab specimen. The selected samples were tested with and without addition of the potential inhibitory substance for direct sample-to-sample comparison. Additionally, pathogen-negative samples were spiked with the potential inhibitory substances.

None of the tested substances showed interference with the Internal Control or the pathogens included in the combined sample.

Tables 11, 12 and 13 (below and next page) show concentrations of the interfering substances tested for the QIAstat-Dx Respiratory Panel.

Table 11. Endogenous substances tested

Substance	Concentration
Human genomic DNA	50 ng/µl
Human whole blood	10% v/v
Human mucin	0.5% v/v

Table 12. Competitive microorganisms tested

Microorganism (source)	Concentration
Staphylococcus aureus (ATCC CRM-6538)	1.70E+08 CFU/ml
Streptococcus pneumoniae (ATCC 6303)	1.25E+07 CFU/ml
Haemophilus influenzae (ATCC 49766)	6.20E+08 CFU/ml
Candida albicans (ATCC CRM-10231)	1.00E+06 CFU/ml
Herpes Simplex Virus 1 (ATCC VR-1789)	1.60E+07 $TCID_{50}$/ml
Human Cytomegalovirus (ATCC NATCMV-0005)	2.0E+04 $TCID_{50}$/ml

Table 13. Exogenous substances tested

Substance	Concentration
Utabon® Nasal spray (decongestant)	10% v/v
Rhinomer® Nasal spray (salt water solutions)	10% v/v
Tobramycin	6 mg/ml
Mupirocin	2.5% w/v

Carryover

A carryover study was performed to evaluate the potential occurrence of cross-contamination between consecutive runs when using the QIAstat-Dx Respiratory SARS-CoV-2 Panel on the QIAstat-Dx Analyzer 1.0.

Samples of simulated NPS matrix, with alternating high-positive and negative samples, were conducted on one QIAstat-Dx Analyzer 1.0.

No carryover between samples was observed in the QIAstat-Dx Respiratory SARS-CoV-2 Panel.

Reproducibility

To prove reproducible performance of the QIAstat-Dx Respiratory Panel on the QIAstat-Dx Analyzer 1.0, a set of selected samples composed of low-concentrated analytes (3x LoD and 1x LoD) and negative samples was tested. Samples were tested in replicates using different lots of QIAstat-Dx Respiratory Panel Cartridges and tests were executed on different QIAstat-Dx Analyzers 1.0 by different operators on different days.

Reproducibility and repeatability will impact the SARS-CoV-2 target in the same manner as other target organisms verified in the QIAstat-Dx Respiratory Panel.

Table 14. List of respiratory pathogens tested for performance reproducibility

Pathogen	Strain
Influenza A H1	A/New Jersey/8/76
Influenza A H3	A/Virginia/ATCC6/2012
Influenza A H1N1 pdm	A/SwineNY/03/2009
Influenza B	B/FL/04/06
Coronavirus 229E	Not available
Coronavirus OC43	Not available
Coronavirus NL63	Not available
Coronavirus HKU1	Not available

(Continued on next page)

(Table 14 continued)

Pathogen	Strain
Parainfluenza Virus 1	C35
Parainfluenza Virus 2	Greer
Parainfluenza Virus 3	C 243
Parainfluenza Virus 4a	M-25
Rhinovirus	A16
Enterovirus	/US/IL/14-18952 (enterovirus D68)
Adenovirus	RI-67 (adenovirus E4)
RSV B	9320
hMPV	Peru6-2003 (type B2)
Bocavirus	Clinical sample
Mycoplasma pneumoniae	M129-B7 (type 1)
Chlamydophila pneumoniae	TW183
Legionella pneumophila	CA1
Bordetella pertussis	I028

Table 15. Summary of Positive Agreement/Negative Agreement for reproducibility testing

Concentration	Pathogen	Expected result	Detection rate	% Agreement with Expected Result
3x LoD	Influenza A H1*	Positive	20/20	100%
	Coronavirus HKU1	Positive	20/20	100%
	PIV-2	Positive	20/20	100%
	RSVB	Positive	20/20	100%
1x LoD	Influenza A H1*	Positive	20/20	100%
	Coronavirus HKU1	Positive	19/20	95%
	PIV-2	Positive	19/20	95%
	RSVB	Positive	20/20	100%

* Detection rate applies for both targets, Influenza A and H1.

(Continued on next page)

(Table 15 continued)

Concentration	Pathogen	Expected result	Detection rate	% Agreement with Expected Result
Negative	Influenza A H1*	Negative	80/80	100%
	Coronavirus HKU1	Negative	80/80	100%
	PIV-2	Negative	80/80	100%
	RSVB	Negative	80/80	100%
3x LoD	Bocavirus	Positive	20/20	100%
1x LoD	Bocavirus	Positive	20/20	100%
Negative	Bocavirus	Negative	80/80	100%
3x LoD	Influenza B	Positive	20/20	100%
	Coronavirus 229E	Positive	20/20	100%
	PIV-4a	Positive	20/20	100%
	Enterovirus D68	Positive	20/20	100%
	hMPV B2	Positive	20/20	100%
	B. pertussis	Positive	20/20	100%
1x LoD	Influenza B	Positive	19/20	95%
	Coronavirus 229E	Positive	20/20	100%
	PIV-4a	Positive	20/20	100%
	Enterovirus D68	Positive	19/20	95%
	hMPV B2	Positive	19/20	95%
	B. pertussis	Positive	20/20	100%
Negative	Influenza B	Negative	80/80	100%
	Coronavirus 229E	Negative	80/80	100%
	PIV-4a	Negative	80/80	100%
	Enterovirus D68	Negative	80/80	100%
	hMPV B2	Negative	80/80	100%
	B. pertussis	Negative	80/80	100%

* Detection rate applies for both targets, Influenza A and H1.

(Continued on next page)

(Table 15 continued)

Concentration	Pathogen	Expected result	Detection rate	% Agreement with Expected Result
	Influenza H1N1 (pdm)†	Positive	20/20	100%
	Coronavirus OC43	Positive	20/20	100%
3x LoD	PIV-3	Positive	20/20	100%
	Rhinovirus A16	Positive	20/20	100%
	M. pneumoniae	Positive	20/20	100%
	Influenza H1N1 (pdm)†	Positive	20/20	100%
	Coronavirus OC43	Positive	20/20	100%
1x LoD	PIV-3	Positive	20/20	100%
	Rhinovirus A16	Positive	20/20	100%
	M. pneumoniae	Positive	20/20	100%
	Influenza H1N1 (pdm)†	Negative	80/80	100%
	Coronavirus OC43	Negative	80/80	100%
Negative	PIV-3	Negative	80/80	100%
	Rhinovirus A16	Negative	80/80	100%
	M. pneumoniae	Negative	80/80	100%
	Influenza A H3‡	Positive	20/20	100%
	Coronavirus NL63	Positive	20/20	100%
3x LoD	PIV-1	Positive	20/20	100%
	Adenovirus E4	Positive	20/20	100%
	L. pneumophila	Positive	20/20	100%
	Influenza A H3‡	Positive	19/20	95%
	Coronavirus NL63	Positive	20/20	100%
1x LoD	PIV-1	Positive	20/20	100%
	Adenovirus E4	Positive	20/20	100%
	L. pneumophila	Positive	20/20	100%
	Influenza A H3‡	Negative	80/80	100%
	Coronavirus NL63	Negative	80/80	100%
Negative	PIV-1	Negative	80/80	100%
	Adenovirus E4	Negative	80/80	100%
	L. pneumophila	Negative	80/80	100%

† Detection rate applies for both targets, Influenza A and H1/pandemic.

‡ Detection rate applies for both targets, Influenza A and H3.

All samples tested generated the expected result (95–100% agreement) showing reproducible performance of the QIAstat-Dx Respiratory Panel.

Reproducibility testing demonstrated that the QIAstat-Dx Respiratory Panel running in the QIAstat-Dx Analyzer 1.0 provides highly reproducible test results when the same samples are tested in multiple runs, on multiple days and with various operators using different QIAstat-Dx Analyzers 1.0, and multiple lots of QIAstat-Dx Respiratory Panel Cartridges.

Sample stability

A sample stability study was executed to analyze storage conditions for clinical samples to be tested with the QIAstat-Dx Respiratory SARS-CoV-2 Panel. Simulated NPS sample matrix (cultured human cells in Copan UTM) was spiked with viral or bacterial culture material of low concentration (e.g., 3x LoD). Samples were stored at the following conditions for testing:

- 15°C to 25°C for 4 hours
- 2°C to 8°C for 3 days
- –15°C to –25°C for 30 days
- –70°C to –80°C for 30 days

All pathogens were successfully detected at the different storage temperatures and durations showing that samples were stable at the indicated storage conditions and durations.

Sample stability was not performed for SARS-CoV-2 specifically. However, specimen stability testing was performed with Coronavirus 229E, HKU1, OC43 and NL63, pathogens from the same virus subfamily, with no impact on performance caused by storage of the samples prior to analysis under conditions stated above.

Appendices

Appendix A: Installing the Assay Definition File

The Assay Definition File of the QIAstat-Dx Respiratory SARS-CoV-2 Panel must be installed on the QIAstat-Dx Analyzer 1.0 prior to testing with QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges.

Note: Whenever a new version of the QIAstat-Dx Respiratory SARS-CoV-2 Panel assay is released, the new QIAstat-Dx Respiratory SARS-CoV-2 Panel Assay Definition File must be installed prior to testing.

Note: Assay Definition Files are available at **www.qiagen.com**. The Assay Definition File (**.asy** file type) must be saved onto a USB Drive prior to installation on the QIAstat-Dx Analyzer 1.0. This USB Drive must be formatted with a FAT32 file system.

To import new assays from the USB to the QIAstat-Dx Analyzer 1.0, proceed with the following steps:

1. Insert the USB stick containing the Assay Definition File into one of the USB ports on the QIAstat-Dx Analyzer 1.0.
2. Press the **Options** button and then select **Assay Management**. The Assay Management screen appears in the Content area of the display (Figure 36, next page).

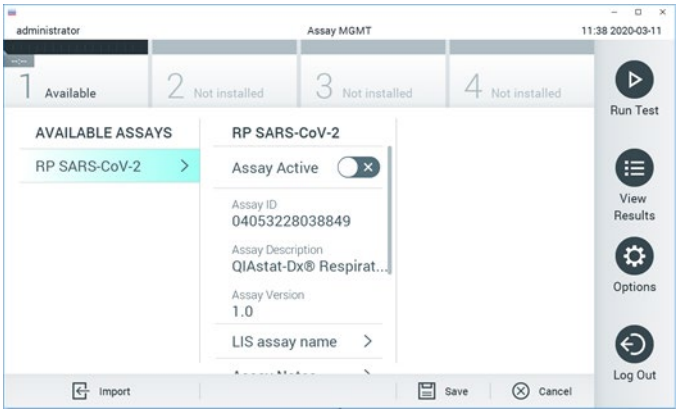

Figure 36. Assay Management screen.

3. Press the **Import** icon in the bottom left of the screen.
4. Select the file corresponding to the assay to be imported from the USB drive.
5. A dialog will appear to confirm upload of the file.
6. A dialog may appear to override the current version by a new one. Press **yes** to override.
7. The assay becomes active by selecting **Assay Active** (Figure 37).

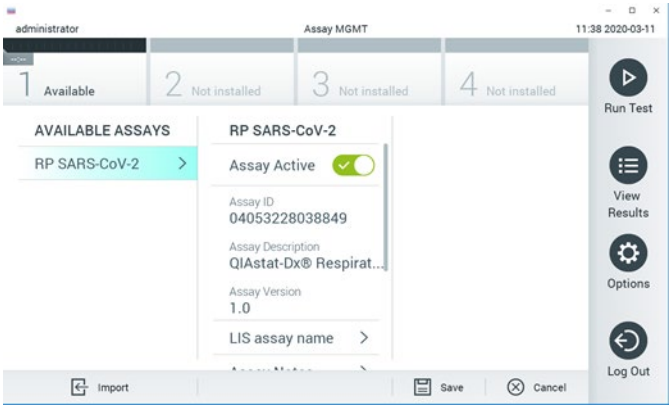

Figure 37. Activating the assay.

8. Assign the active assay to the user by pressing the **Options** button and then the **User Management** button. Select the user who should be allowed to run the assay. Next, select **Assign Assays** from the "User Options". Enable the assay and press the **Save** button (Figure 38).

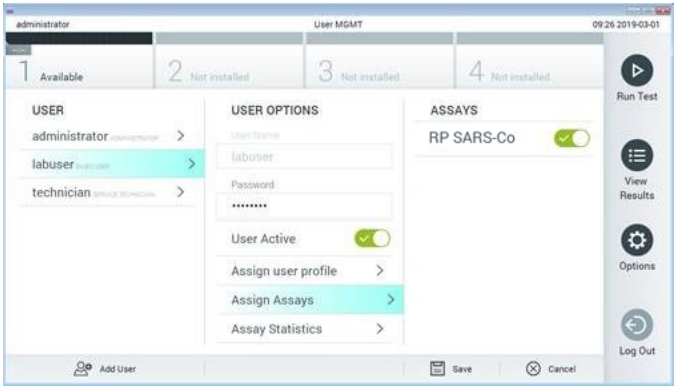

Figure 38. Assigning the active assay.

Appendix B: Glossary

Amplification curve: Graphical representation of the multiplex real-time RT-PCR amplification data.

Analytical Module (AM): The main QIAstat-Dx Analyzer 1.0 hardware module, in charge of executing tests on QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges. It is controlled by the Operational Module. Several Analytical Modules can be connected to one Operational Module.

QIAstat-Dx Analyzer 1.0: The QIAstat-Dx Analyzer 1.0 consists of an Operational Module and an Analytical Module. The Operational Module includes elements that provide connectivity to the Analytical Module and enables user interaction with the QIAstat-Dx Analyzer 1.0. The Analytical Module contains the hardware and software for sample testing and analysis.

QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge: A self-contained disposable plastic device with all pre-loaded reagents required for the complete execution of fully automated molecular assays for the detection of respiratory pathogens.

IFU: Instructions For Use.

Main port: In the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge, inlet for transport medium liquid samples.

Nucleic acids: Biopolymers, or small biomolecules composed of nucleotides, which are monomers made of three components: a 5-carbon sugar, a phosphate group and a nitrogenous base.

Operational Module (OM): The dedicated QIAstat-Dx Analyzer 1.0 hardware that provides the user interface for 1–4 Analytical Modules (AM).

PCR: Polymerase Chain Reaction

RT: Reverse Transcription

Swab port: In the QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge, inlet for dry swabs.

User: A person who operates the QIAstat-Dx Analyzer 1.0/QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge in the intended way.

Appendix C: Disclaimer of warranties

EXCEPT AS PROVIDED IN QIAGEN TERMS AND CONDITIONS OF SALE FOR THE QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge, QIAGEN ASSUMES NO LIABILITY WHATSOEVER AND DISCLAIMS ANY EXPRESS OR IMPLIED WARRANTY RELATING TO THE USE OF THE QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridge INCLUDING LIABILITY OR WARRANTIES RELATING TO MERCHANTABILITY, FITNESS FOR A PARTICULAR PURPOSE, OR INFRINGEMENT OF ANY PATENT, COPYRIGHT, OR OTHER INTELLECTUAL PROPERTY RIGHT ANYWHERE IN THE WORLD.

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Symbols

The following table describes the symbols that may appear on the labeling or in this document.

 <N>	Contains reagents sufficient for <N> reactions
	Use by
	In vitro diagnostic medical device
	Catalog number
	Lot number
	Material number (i.e., component labeling)
	Upper respiratory application
Rn	R is for revision of the Handbook and n is the revision number
	Temperature limitation
	Manufacturer
	Consult instructions for use
	Caution
	CE marking for European Conformity
	Serial number
	Do not reuse
	Keep away from sunlight
	Do not use if package is damaged
	Global Trade Item Number

Ordering Information

Product	Contents	Cat. no.
QIAstat-Dx Respiratory SARS-CoV-2 Panel	For 6 tests: 6 individually packaged QIAstat-Dx Respiratory SARS-CoV-2 Panel Cartridges and 6 individually packaged transfer pipettes	691214
Related Products		
QIAstat-Dx Analyzer 1.0	1 QIAstat-Dx Analytical Module, 1 QIAstat-Dx Operational Module and related hardware and software to run molecular diagnostic QIAstat-Dx assay cartridges	9002824

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at **www.qiagen.com** or can be requested from QIAGEN Technical Services or your local distributor.

Document Revision History

Date	Changes
Revision 1 03/2020	Initial release.

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