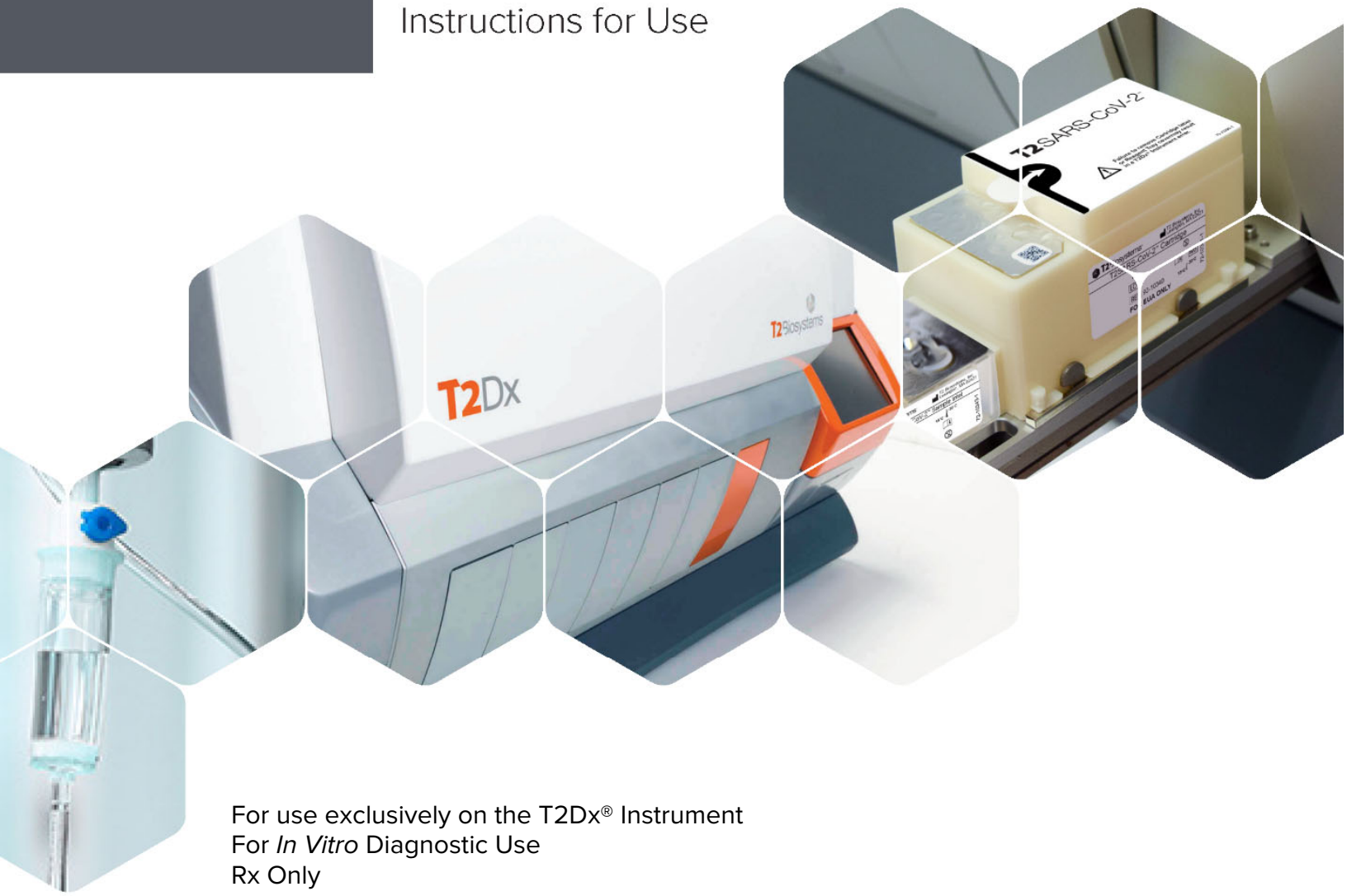
T2SARS-CoV-2™ Panel
Instructions for Use



For use exclusively on the T2Dx® Instrument
For *In Vitro* Diagnostic Use
Rx Only
For Emergency Use Authorization Only

T2Biosystems®

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The current revision of this manual is available at http://info.t2biosystems.com/instructions
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Proprietary Name

T2SARS-CoV-2™ Panel.

Intended Use

The T2SARS-CoV-2 Panel is a qualitative T2 magnetic resonance (T2MR) test for the direct detection of nucleic acid from SARS-CoV-2 in upper respiratory specimens (such as nasal, mid-turbinate, nasopharyngeal, and oropharyngeal swab specimens) and bronchoalveolar lavage specimens from individuals suspected of COVID-19 by their healthcare provider. Testing is limited to laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, that meet requirements to perform high or moderate complexity tests.

Results are for the identification of SARS-CoV-2 RNA. The SARS-CoV-2 RNA is generally detectable in respiratory specimens during the acute phase of infection. Positive results are indicative of the presence of SARS-CoV-2 RNA; clinical correlation with patient history and other diagnostic information is necessary to determine patient infective status. Positive results do not rule out bacterial co-infection with other viruses. Laboratories within the United States and its territories are required to report all positive results to the appropriate public health authorities.

Negative results do not preclude SARS-CoV-2 infection and should not be used as the sole basis for patient management decisions. Negative results must be combined with clinical observations, patient history, and epidemiological information.

The T2SARS-CoV-2 Panel is intended for use by qualified and trained clinical laboratory personnel specifically instructed and trained in the techniques of real-time PCR and in vitro diagnostic procedures. The T2SARS-CoV-2 Panel is only for use under a Food and Drug Administration's Emergency Use Authorization.

Principle of the Procedure

The T2SARS-CoV-2™ Panel is a qualitative test for the direct detection of the 2019 novel coronavirus (SARS-CoV-2) RNA in upper respiratory samples on the T2Dx Instrument (T2Dx). An upper respiratory sample in transport media is directly loaded into the T2SARS-CoV-2 Sample Inlet that has first been assembled with the T2SARS-CoV-2 Cartridge loaded with the T2SARS-CoV-2 Reagent Tray. The Panel contains all of the disposables and reagents required to detect SARS-CoV-2 RNA direct from sample. The assembled Panel is loaded onto the T2Dx, a benchtop, fully automated sample-to-result system, which performs all steps of the assay after sample loading.

During processing on the T2Dx, an aliquot of the patient sample is directly mixed with reverse transcriptase and DNA amplification reagents. After target amplification, amplicon is hybridized with target specific probes that are bound to superparamagnetic particles and then detected by T2MR[1]. The Internal Control on the Panel monitors performance for each patient sample or control.

Warnings & Precautions

- The Panel is intended for *in vitro* diagnostic use only.
- For prescription use only.
- The T2SARS-CoV-2™ Panel has not been FDA cleared or approved; the test has been authorized by FDA under an Emergency Use Authorization (EUA) for use by laboratories certified under the Clinical Laboratory Improvement Amendments (CLIA) of 1988, 42 U.S.C. §263a, that meet requirements to perform high or moderate complexity tests.
- The T2SARS-CoV-2™ Panel has been authorized only for the detection of nucleic acid from SARS-CoV-2, not for any other viruses or pathogens.
- The T2SARS-CoV-2™ Panel is only authorized for the duration of the declaration that circumstances exist justifying the authorization of emergency use of *in vitro* diagnostic tests for detection and/or diagnosis of COVID-19 under Section 564(b)(1) of the Federal Food, Drug, and Cosmetic Act, 21 U.S.C. § 360bbb-3(b)(1), unless the authorization is terminated or revoked sooner.
- The Panel is distributed in accordance with the FDA guidance: Policy for Coronavirus Disease-2019 Tests During the Public Health Emergency (Revised).
- The Panel is intended only for use with upper respiratory samples in transport media. Use of any other sample type or incorrect collection may lead to assay or instrument failure.
- Anterior nasal swabs, mid-turbinate swabs, oropharyngeal swabs, and bronchoalveolar lavage specimens are additional acceptable respiratory specimens that can be tested with the T2SARS-CoV-2 Panel; however, performance with these specimen types has not been determined.

- The T2SARS-CoV-2 Panel is a test for the direct detection of nucleic acid from SARS-CoV-2. Clinical correlation with patient history and other diagnostic information is necessary to determine patient infection status.
- Laboratories within the United States and its territories are required to report all positive results to the appropriate public health authorities.
- False-negative results may arise from degradation of the viral RNA during shipping and storage.
- As with any molecular test, mutations within the target regions of the T2SARS-CoV-2 Panel could affect primer and/or probe binding resulting in failure to detect the presence of the virus.
- Handle all specimens as if infectious using safe laboratory procedures. Refer to CDC Interim Laboratory Biosafety Guidelines for Handling and Processing Specimens Associated with Coronavirus Disease 2019 (COVID-19) https://www.cdc.gov/coronavirus/2019-nCoV/lab/lab-biosafety-guidelines.html
- Avoidance of contamination is critical for the effectiveness of the Panel. To avoid false positive results due to detection of contaminants from the workspace or from the operator, follow good laboratory practices including the following suggestions:
    - Establish a clean environment in which to perform the test as described in the *Work Area Preparation* section.
    - Change gloves as specified in the *T2SARS-CoV-2 Panel Instructions* section when preparing Panel components or processing samples. Operators may elect to change gloves more frequently at their discretion.
    - Wear protective clothing and disposable gloves when handling Panel components. Additional precautions include the use of surgical masks or face shields to further reduce the risk of exposure. Wash hands thoroughly prior to and upon completion of the Panel.
    - Handle all materials containing samples and controls according to good laboratory practices in order to prevent cross-contamination.
    - Assemble the Panel in a biosafety cabinet (BSC).
    - If any Panel component is dropped during the assembly process, discard the component and get a new component, changing gloves as needed to minimize contamination.
    - If the assembled Panel is dropped during the loading or unloading process, discard the Panel in a biohazard bag and follow your laboratory's established Exposure Plan. Decontamination may be required if the drop incident occurs while unloading a Panel. Contact T2 Biosystems Service for assistance.
- **Biohazard:** The Panel contains all waste, including human respiratory samples, generated by the test procedure. Samples should be handled as if infectious, using Universal Precautions and safe laboratory procedures.[2, 3]

- Do not smoke, eat, or drink in areas where samples or reagents are being handled.
- Do not use Panel components after their expiration date.
- When handling the Panel components, avoid contact with skin, eyes, or mucous membranes. Wash with water if contact occurs.
- Dispose of used and unused reagents and waste in accordance with Country, Federal, State, and Local requirements.
- Safety Data Sheets are available on request from T2 Biosystems Service.

Materials Provided

Reagents

The Panel is comprised of the T2SARS-CoV-2 Cartridge (Cartridge), the T2SARS-CoV-2 Sample Inlet (Sample Inlet), and the T2SARS-CoV-2 Reagent Tray (Reagent Tray), as depicted in Table 1. The Panel is packaged in two boxes: one box contains twelve (12) Reagent Trays, and a second box contains twelve (12) Cartridges and twelve (12) Sample Inlets.

Table 1. Panel Components

Shipped Configuration	Storage Temperature	Component Name	Picture	Description
Cartridge Kit Catalog #: 90-10344	15-30°C	Sample Inlets (12 per box)		Component used to load the sample to be run.
		Cartridges (12 per box)		Component that contains disposables.
Reagent Trays Catalog #: 80-10284	-20°C ± 5°C	Reagent Trays (12 per box)		Component that contains the reagents.

Cartridges

Twelve (12) single-use Cartridges per box.

Sample Inlets

Twelve (12) single-use Sample Inlets per box.

Reagent Trays

Twelve (12) single-use Reagent Trays per pack. Each single use Reagent Tray is preloaded with all necessary solutions for the Panel, including:

Reaction Reagent
Aqueous buffered solution containing sequence-specific T2SARS-CoV-2 and Internal Control target primers, reverse transcriptase, and polymerase.

SARS-CoV-2 particles
Probe-coupled superparamagnetic particles that hybridize to SARS-CoV-2 amplicons in an aqueous buffered solution.

Internal Control particles
Probe-coupled superparamagnetic particles that hybridize to Internal Control amplicons in an aqueous buffered solution.

Reagent Storage & Stability

- Store Cartridges at 15-30°C. Note the expiration date present on the label.
- Store Reagent Trays at -20°C ± 5°C. Note the expiration date present on the label.

Materials Required but not Provided

- 1000 µL pipette (Eppendorf Research® plus 100 – 1000 µL single-channel pipette; catalog #3123000063 or equivalent)
- Sterile 1000 µL filter barrier pipette tips (Axygen 1000 µL Maxymum Recovery® Filter Barrier tips; catalog #TF-1000-L-R-S or equivalent)
- One (1) decontaminated tube rack that will hold the specific size of sample collection tube that will be used for testing inside of the biosafety cabinet
- Sample collection and storage consumables: Upper respiratory swab samples (e.g., nasopharyngeal or oropharyngeal swabs) in transport media
- Powderless disposable gloves
- Eye protection
- Lab coat
- Surface Decontaminant:
    - Pre-diluted, stabilized bleach solution consisting of ≥ 0.5% sodium hypochlorite (10% bleach),

      OR
    - Pre-diluted sodium dichloroisocyanurate (Troclosene sodium or NaDCC) solution consisting of ≥4,306 ppm chlorine (Brulin® BruTab 6S or equivalent).
- 70% isopropyl alcohol
- Lint-free wipes
- Biohazard waste bags
- Vortex Mixer (Fisher Scientific™ Analog Vortex Mixer, catalog #02-215-414, or equivalent)

Materials Available Separately from External Vendors

- Positive External Control:
    - AccuPlex SARS-CoV-2 Reference Material Kit (SeraCare #0505-0126)
    - SARS-Related Coronavirus 2 (SARS-CoV-2) External Run Control (ZeptoMetrix Corp., #NATSARS(COV2)-ERC)
- Negative External Control:
    - SARS-Related Coronavirus 2 (SARS-CoV-2) Negative Control (ZeptoMetrix Corp., #NATSARS(COV2)-NEG)

Sample Collection, Handling & Storage

⚠ *Inadequate or inappropriate specimen collection, storage, and transport are likely to yield false test results. Training in specimen collection is highly recommended due to the importance of specimen quality. Handle all samples and controls as if they are capable of transmitting infectious agents.*

- Refer to Interim Guidelines for Collecting and Handling of Clinical Specimens for COVID-19 Testing https://www.cdc.gov/coronavirus/2019-nCoV/lab/guidelines-clinical-specimens.html
- Follow specimen collection device manufacturer's instructions for proper collection methods.
- T2SARS-CoV-2 Panel performance characteristics have been determined with human upper respiratory specimens in transport media
- Specimens can be stored at 2-8°C for up to 72 hours after collection or at -70°C for extended durations.

T2SARS-CoV-2 Panel Instructions

Refer to *T2Dx Instrument Operator Manual* (DES-00405) for detailed instructions on T2Dx operation.

⚠ ***NOTE:*** *Should gloves become soiled during any of the steps of this procedure, remove and replace with a clean pair of gloves following standard lab procedures.*

⚠ ***NOTE:*** *Do not spray any liquid directly on the T2Dx. Do not allow cleaning solution to drip into the touchscreen bezel.*

Work Area Preparation

Appropriate precautions should be taken to avoid contamination of samples or cartridges with cellular or DNA material. Gloves should be changed at a minimum as indicated in the procedure below.

Prior to the first Panel(s) loaded each day and after each unload, all work surfaces and common touch points should be disinfected as outlined below using:

- Surface Decontaminant:
    - Pre-diluted, stabilized bleach solution consisting of ≥ 0.5% sodium hypochlorite (10% bleach or approximately 5,000 ppm sodium hypochlorite), OR
    - Pre-diluted sodium dichloroisocyanurate (Troclosene sodium or NaDCC) solution consisting of ≥4,306 ppm chlorine.
- 70% isopropyl alcohol
- Lint-free wipes

The following procedure is required for effective disinfection of the work surface in the BSC where the Panel(s) will be assembled, the T2Dx touchscreen and barcode scanner, the T2Dx drawer panel, and front cover, and the benchtop where the T2Dx is located:

Put on fresh gloves.

1. *Fully saturate* a lint-free wipe with surface decontaminant solution. Wipe the preparation area in the BSC in a unidirectional motion. Discard wipe.
2. *Dampen* a **new** lint-free wipe with surface decontaminant solution and wipe the following T2Dx components in a unidirectional motion (Figure 1) and in the sequence indicated below:
    - Touchscreen
    - Barcode scanner

3. *Fully saturate* the lint-free wipe with surface decontaminant solution. Wipe the following work areas in a unidirectional motion and in the sequence indicated below (Figure 1):
    - Drawer panel and front cover of the T2Dx
    - Benchtop around the T2Dx
4. Discard wipe.
5. Allow the surface decontaminant solution to sit for at least 3 minutes.
6. Repeat **Steps 1-5** replacing surface decontaminant solution with 70% isopropyl alcohol. Repeat until all surface decontaminant residue (visible as white film) is completely removed.

Figure 1. Areas to clean on the T2Dx and unidirectional wiping.

T2SARS-CoV-2 Panel Assembly

Put on fresh gloves.

1. Confirm that the T2Dx is operational and that a drawer is available for each sample to be loaded.

⚠ *NOTE: Up to 7 samples can be loaded onto the T2Dx.*

2. Obtain sample(s) and allow to equilibrate to room temperature (15-25°C) by storing upright in a tube rack as follows:
    a. Refrigerated samples: 10 minutes (± 5 minutes).
    b. Frozen samples: 30 minutes (± 10 minutes).
3. Ensure that there is at least 0.5 mL of sample volume in each tube and that the sample label and barcode are legible and undamaged.

⚠ ***NOTE: Do not proceed if sample does not meet the volume requirement.***

4. Print an extra, adhesive label for the sample(s) to be tested.

Put on fresh gloves.

5. Based on the number of samples to be tested, obtain the corresponding number of Reagent Tray(s) from bulk packaging, check the label and barcode for integrity, and place onto the Panel assembly work surface in the BSC. Return any unused Tray(s) to proper storage. Allow to equilibrate to room temperature (15-25°C) by storing upright for 20 minutes (± 10 minutes).
6. Based on the number of samples to be tested, obtain the corresponding number of Cartridge(s) from bulk packaging, check the label and barcode for integrity, and place onto the Panel assembly work surface in the BSC. Return any unused Cartridges to proper storage.
7. Based on the number of samples to be tested, obtain the corresponding number of Sample Inlet(s) from bulk packaging, check the label for integrity, and carefully place onto the Panel assembly work surface in the BSC, being careful to avoid touching the foil. Return any unused Sample Inlets to proper storage.
8. Ensure that the contents of the Reagent Trays, obtained in Step 5, are fully thawed. Mix the reagents in Reagent Tray by vortexing.
    - Set vortex speed to 2560-3200 RPM.
    - Securely hold the Reagent Tray upright with the barcode facing up.
    - Place the Reagent Tray onto the center of the 3 inch vortex head cover attachment.

- Press down on the Reagent Tray cover while supporting the sides of the Reagent Tray to activate the vortexer.
- Vortex the Reagent Tray for 5 seconds and then visually check the wells for homogeneity. If the contents do not appear homogenous, repeat vortexing up to two more times until wells are visually homogenous.

⚠ *NOTE: If homogeneity is not achieved after three 5 second intervals (15 seconds total of vortexing), discard Reagent Tray and repeat with a new Reagent Tray.*

9. Reagent mixing may result in reagent splashing onto the walls of the reagent tray wells and/or bubble forming in the wells. If necessary, the tray may be flicked to remove air bubbles and collect solutions at the bottom of the wells. Visually confirm that air bubbles are removed and volumes appear consistent.

⚠ *NOTE*: *The T2SARS-CoV-2 reagent tray has only 3 wells containing reagent. All other wells are empty*.

10. Prepare to insert the Reagent Tray onto the Cartridge by orienting the Reagent Tray such that the tab on the Reagent Tray cover is on the right and the notches on the Reagent Tray and Cartridge align.
11. Push down on the left side of the Reagent Tray cover to engage the tab on the left. Then push down on the right side of the Reagent Tray cover to engage the tab on the right. An audible click will be heard when each tab engages.
12. Obtain a sample inlet. Hold the back sides of the Sample Inlet assembly, taking care to avoid contact with the foil seal, and push it down onto the Cartridge until an audible snap is heard. Once snapped in, the Panel set-up is complete.
13. Repeat **Steps 9-12** for all Cartridges, Sample Inlets, and Reagent Trays to be used.

Adding Samples to the Sample Inlet

Put on fresh gloves.

14. Attach the extra sample label, printed in Step 4, to the empty/unlabeled side of the Cartridge body.
15. Obtain the corresponding room temperature specimen to be tested with the Panel that was thawed in Step 2.
16. Ensure that the specimen is adequately mixed.
    - Vortex the specimen for 5 seconds and visually inspect the contents for homogeneity. If the contents do not appear homogenous, invert the specimen collection tube and repeat vortexing up to four more times until sample appears homogenous.

17. In the BSC, uncap the sample tube following standard molecular practices. Carefully place the cap down so that it does not contaminate the preparation area. Exercise care not to spill or aerosolize the sample.

⚠ *WARNING: In the event of sample spillage, follow your institution's Exposure Plan for biohazardous spill cleanup procedures.*

18. Set the 1000 µL pipette to aspirate 500 µL.
19. Slowly aspirate 500 µL of sample, taking care to avoid drawing any clumps or concentrated material with the pipette.
20. Slowly dispense the sample into the pre-punctured well of the Sample Inlet.
21. Discard tip in biohazard waste according to institution procedures.
22. Carefully recap the sample collection tube and return it to the tube rack in the preparation area.

Loading the T2SARS-CoV-2 Panel onto the T2Dx

Put on fresh gloves.

23. Transport the assembled Panel from the BSC to the T2Dx, using secondary containment as dictated by laboratory protocol.
24. Press "**Load**" on the T2Dx touchscreen.
25. When prompted by the instrument, use the handheld barcode scanner on the T2Dx to scan the sample barcode label attached to the side of the Cartridge. When dictated by laboratory protocol, you may enter a unique sample identifier using the touchscreen keyboard instead of scanning the sample barcode label.
26. When prompted by the instrument, scan the Reagent Tray barcode and the Cartridge barcode.
27. The T2Dx will open an available drawer and prompt the user to load the Panel. Place the Panel in the drawer, ensuring that it is level, fully in contact with the metal rails and seated on the location pins (Figure 2). Press "**Next**" on the touchscreen.

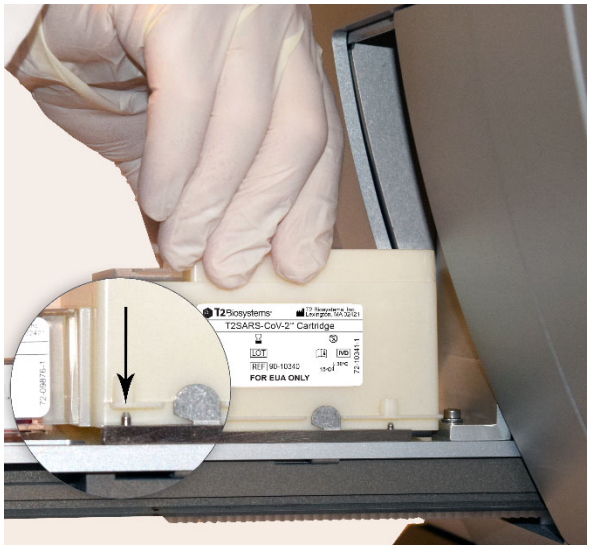

Figure 2. Panel should be level and seated on location pins.

28. When prompted to "**Tear off Label**," hold the Panel with one hand for stability and remove the top seal from the Cartridge by gently pulling back on the label tab (Figure 3).

⚠ *NOTE: Failure to remove Cartridge label will result in a T2Dx error.*

Figure 3. Remove top seal.

29. Hold the Panel with one hand for stability and remove the plastic Reagent Tray cover without touching the foil seal or any of the exposed Cartridge components (Figure 4).

Figure 4. Remove Reagent Tray cover

⚠ *NOTE: Failure to remove Cartridge label or Reagent Tray cover will result in a T2Dx error.*

30. Visually confirm that the Reagent Tray cover and Cartridge top seal are removed and all components are present (Figure 5):
    - Small tubes with caps (3)
    - Pipette tips (5)

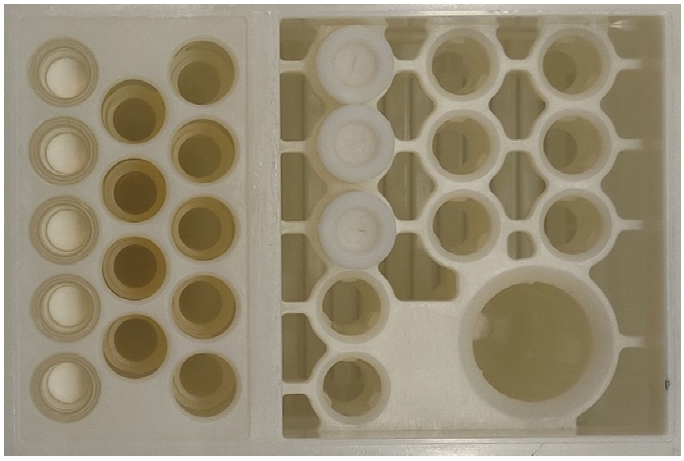

Figure 5. Confirm all components are present.

⚠ *NOTE: If any components are missing from the Panel, remove the Panel from the drawer, cancel its load by pressing "Cancel" on the T2Dx touchscreen, and contact T2 Biosystems Service. The sample should be discarded.*

31. Press "**Next**" on the touchscreen.
32. Scan or enter personal user ID and ensure that the Sample ID is present and the Script is "T2SARS-CoV-2".
33. Press "**Confirm**" on the touchscreen, which will close the drawer and initiate the panel.
34. Repeat **Steps 14-34** to load all assembled Panels onto the T2Dx Instrument.

Unloading the T2Dx

Once the Panel is finished, the "Run Complete" indicator will appear on the T2Dx touchscreen. All the used and unused disposables along with reagents, sample, and liquid waste are contained in the Panel.

⚠ *CAUTION: The used Panel may contain SARS-CoV-2 virus. Handle with care.*

Put on fresh gloves.

1. Obtain a biohazard bag.
2. Press "**Unload**" on the T2Dx touchscreen. When prompted, press "**Next**" which will open the drawer. Visually confirm that all of the Panel components are present. If any components are missing, contact T2 Biosystems Service.
3. Using the biohazard waste bag as a glove, remove the used Panel from the drawer keeping it and its contents upright to avoid spilling. Use your other hand to invert the biohazard waste bag over the Panel (Figure 6).

Figure 6. Proper Technique for Unloading the T2SARS-CoV-2 Panel.

⚠ *NOTE: Be careful when removing the used Panel to avoid spilling of reagents, samples, and disposables. Do not tip or invert the used Panel. Ensure that the Panel remains upright while unloading.*

4. Immediately close and secure the biohazard waste bag with a twist tie, and then discard in a biohazardous waste container in accordance with local biohazard waste disposal regulations.
5. Press "**Next**" twice on the T2Dx touchscreen to close the drawer.
6. Repeat **Steps 1-5** for all other drawers that display a "**Run Complete**" indication.

Put on fresh gloves.

7. Repeat Steps 1-6 of Work Area Preparation to decontaminate the T2Dx and workspace.

Quality Control

T2Dx Instrument

The T2Dx incorporates multiple in-line sensor checks to monitor and ensure proper Panel loading, system operation and sub-system functionality. Messages and error codes may be generated by the T2Dx during the performance of the Panel. The user must check Panel result printouts to verify the test result is valid. Refer to the *T2Dx Instrument Operator Manual* (DES-00405) for further instructions.

Internal Control

The Panel includes an Internal Control that monitors the sample processing, amplification and detection process. The Internal Control is designed to ensure that samples do not contain inhibitors that would interfere with the detection of SARS-CoV-2 targeted by the Panel. The Internal Control also ensures that, when SARS-CoV-2 is not detected, the sample collection, amplification and detection process has functioned properly to ensure accurate reporting of a negative result. If the Internal Control is Invalid and no target SARS-CoV-2 is detected, the result cannot be determined and "Invalid" will be displayed as the result.

Process Quality Controls

T2 Biosystems recommends the ZeptoMetrix SARS-Related Coronavirus 2 (SARS-CoV-2) External Run Control (#NATSARS(COV2)-ERC) or SeraCare's AccuPlex SARS-CoV-2 Reference Material Kit (#0505-0126) as a positive control and ZeptoMetrix Negative Control (#NATSARS(COV2)-NEG) to be used for periodic quality checks with the Panel and the T2Dx. All external controls should be prepared following manufacturer instruction, including vortexing to ensure the samples are sufficiently mixed. Users should follow all laboratory procedures, local, state, and/or national requirements and accrediting organizations' guidelines for the testing of all positive and negative controls.

It is recommended that external control testing be run during initial T2SARS-CoV-2 Panel software installation, to establish laboratory validation procedures, and with the receipt of a new lot of T2SARS-CoV-2 reagents.

Each Panel result generated by the Panel reagents is an independent analysis and each result has an internal control to ensure the integrity of the result. As such, there are no recommendations regarding the necessity to validate patient sample results solely on the basis of the results obtained from external controls. The laboratory should follow its own established standard operating procedure for appropriate action(s).

⚠ *NOTE: In the event of two successive failed or invalid controls, or in the event that system contamination is suspected, immediately contact T2 Biosystems Service.*

Data Retrieval & Interpreting Results

Panel results are accessible via printed form (Figure 7a) or via the T2Dx touchscreen display (Figure 7b).

T2SARS-CoV-2 Panel

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Run Summary

Run Result:	**Positive**
Sample ID:	PTS156489497516
Sample Type:	Patient Sample

Result Details

SARS-CoV-2	Positive
Internal Control	Valid

Run Details

Operator:	John Smith	Drawer Number:	1
Reagent Lot:	WO01234	Expiration Date:	12/31/2020
Cartridge Lot:	WO012345	Expiration Date:	12/19/2020
Error Codes:	N/A		
Panel Version:	1.6.4.14		
T2Dx Version:	1.6.4.14	T2Dx ID:	ISA0814000025
Run ID:	42c59469-b475-4510-99d9-c20af1c8f70d		
Load Time:	5/8/20 05:48 PM		
End Time:	5/8/20 08:03 PM		

EXAMPLE

t2service@t2biosystems.com +1-877-504-T2T2 (8282)

Figure 7a. Panel results via printed form

Figure 7b. Panel results via the T2Dx touchscreen display

Messages and error codes may be generated by the T2Dx during the performance of the Panel. The operator must check the results printout to verify the Panel result is valid.

To print the Panel results, press "**Results**" on the T2Dx touchscreen, choose the box next to the result to print, and press "**Print**".

Each valid Panel will yield one reportable result (Positive or Target Not Detected) for SARS-CoV-2. In addition, an Internal Control (IC) result will be reported as Valid or Invalid. Table 2 outlines possible results for each target resistance marker and the Internal Control as well as the interpretation of results.

Table 2. T2SARS-CoV-2 Panel Results

Target Species	Target Result	IC Result	Interpretation
SARS-CoV-2	Positive	Valid	SARS-CoV-2 detected.
	Target Not Detected	Valid	SARS-CoV-2 **NOT** detected. The Internal Control is valid.
All	Invalid	Invalid	Target results are invalid. The Internal Control is invalid.

Limitations of the Procedure

- The performance of the T2SARS-CoV-2™ Panel was established using nasopharyngeal swab specimens. Oropharyngeal swabs, anterior nasal swabs, midturbinate nasal swabs and BAL are also considered acceptable specimen types for use with the T2SARS-CoV-2™ Panel but performance has not been established.

- Samples must be collected, transported, and stored using appropriate procedures and conditions. Improper collection, transport, or storage of specimens may hinder the ability of the assay to detect the target sequences.

- The following conditions can negatively impact the performance of the test:
  - Sample is not at room temperature
  - Contaminated sample or contaminated T2Dx Instrument
  - Interfering substances. The effect of interfering substances has only been evaluated for those listed in the labeling. Interference by substances other than those described in the Interference section could lead to erroneous results.

- If the virus mutates in the RT-qPCR target region, SARS-CoV-2 may not be detected or may be detected less predictably.

- False Positive results may arise from the contamination during specimen handling or preparation, or between patient samples.

- Negative results do not preclude SARS-CoV-2 infection and should not be used as the sole basis for treatment or other patient management decisions. Optimum specimen types and timing for peak viral levels during infections caused by 2019-nCoV have not been determined.

- False Negative results may arise from:
  - Improper sample collection
  - Degradation of the viral RNA during shipping/storage
  - The presence of RT-PCR inhibitors
  - Mutation(s) in the sequence of SARS-CoV-2 virus

Conditions of Authorization for the Laboratory

The T2SARS-CoV-2™ Panel Letter of Authorization, along with the authorized Fact Sheet for Healthcare Providers, the authorized Fact Sheet for Patients and other authorized labeling are available on the FDA website: https://www.fda.gov/medical-devices/coronavirus-disease-2019-covid-19-emergency-use-authorizations-medical-devices/vitro-diagnostics-euas

Use of the T2SARS-CoV-2™ Panel must follow the procedures outlined in these manufacturer's Instructions for Use and the conditions of authorization outlined in the Letter of Authorization. Deviations from the procedures outlined are not permitted under the Emergency Use Authorization (EUA). To assist clinical laboratories running the T2SARS-CoV-2™ Panel, the relevant Conditions of Authorization are listed verbatim below, and are required to be met by laboratories performing the EUA test.

- Authorized laboratories[1] using the T2SARS-CoV-2™ Panel will include with test result reports, all authorized Fact Sheets. Under exigent circumstances, other appropriate methods for disseminating these Fact Sheets may be used, which may include mass media.

- Authorized laboratories will perform the T2SARS-CoV-2™ Panel as outlined in the authorized labeling. Deviations from the authorized procedures, including authorized extraction methods, authorized clinical specimen types, authorized control materials, authorized other ancillary reagents and authorized materials required to perform the T2SARS-CoV-2™ Panel are not permitted.

- Authorized laboratories that receive the T2SARS-CoV-2™ Panel will notify the relevant public health authorities of their intent to run the Panel prior to initiating testing.

- Authorized laboratories will have a process in place for reporting test results to healthcare providers and relevant public health authorities, as appropriate.

- Authorized laboratories will collect information on the performance of the test and report to: DMD/OHT7-OIR/OPEQ/CDRH (via email: CDRH-EUA- Reporting@fda.hhs.gov) and to T2 Biosystems, Inc (via phone: 781-457-1200 or via email: t2service@t2biosystems.com) any suspected occurrence of false positive or false negative results and significant deviations from the established performance characteristics of the test of which they become aware.

- All laboratory personnel using the test must be appropriately trained/experienced in molecular in vitro diagnostic test techniques, use appropriate laboratory and personal protective equipment when handling this kit, and use the test in accordance with the authorized labeling.

- T2 Biosystems, Inc authorized distributors, and authorized laboratories will ensure that any records associated with this EUA are maintained until otherwise notified by FDA. Such records will be made available to FDA for inspection upon request.

[1] The letter of authorization refers to, "Laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, that meet requirements to perform high or moderate complexity tests" as "authorized laboratories."

Performance Characteristics - Summary

Limit of Detection

Limit of Detection (LoD) was defined as the minimum concentration at which a ≥95% positivity rate was achieved from replicate samples. LoD screening was conducted by testing dilutions of inactive SARS-CoV-2 virus (BEI #NR-52286) prepared in negative nasopharyngeal swab media with 4 replicates for each concentration. The lowest concentration resulting in 100% detection was used as a starting point for confirmation studies. Table 3 shows the outcomes of LoD screening. LoD confirmation was performed with at least 20 independent replicates and determined as the lowest concentration where ≥ 95% (19/20) of the replicates test positive. Table 4 shows the outcomes of LoD confirmation testing. Based on these data, 2,000 GE/mL was determined as the LoD for the T2SARS-CoV-2 Panel.

Table 3. Limit of Detection Screening Outcomes

SARS-CoV-2 Concentration (GE/mL)	Replicates	Percent Detection
2,000	4	100% (4/4)
1,800	4	100% (4/4)
1,500	4	75% (3/4)
900	4	0% (0/4)

Table 4. Limit of Detection Confirmation Outcomes

SARS-CoV-2 Concentration (GE/mL)	Replicates	Percent Detection
2,000	20	100% (20/20)
1,800	20	70% (14/20)

Analytical Reactivity (Inclusivity)

To establish the analytical reactivity of the Panel, *in silico* analysis was performed against the National Center for Biotechnology (NCBI) nt database. Of 7,695 SARS-CoV-2 sequences analyzed, 99% had 100% identity with both T2SARS-CoV-2 primers and probes. Sequences that had mismatches with either primers or probes were further analyzed using thermodynamic simulations and selected wet testing. None of the mismatches were predicted to prevent binding, amplification or detection.

Analytical Specificity (Exclusivity)

Analytical specificity of the T2SARS-CoV-2 primers and probes were examined *in silico* by filtering the NCBI nt database for near full length records from FDA's Recommended List of Organisms for Cross-Reactivity (Table 5). T2MR detection requires >80% homology for both primers and both detection probes for a positive result to occur. None of the listed species have >80% homology for both primers and both detection probes, and thus no amplicon is expected to be produced. Based on

this analysis and selected wet testing, none of the members of the list of organisms are expected to be cross-reactive with this assay.

Table 5: T2SARS-CoV-2 Panel in silico exclusivity

Other high priority pathogens from the same genetic family	**High priority organisms likely in the circulating area**
Human coronavirus 229E	Adenovirus
Human coronavirus OC43	Human Metapneumovirus (hMPV)
Human coronavirus HKU1	Parainfluenza virus 1-4
Human coronavirus NL63	Influenza A & B
SARS-coronavirus	Enterovirus
MERS-coronavirus	Respiratory syncytial virus
	Rhinovirus
	Chlamydia pneumonia
	Haemophilus influenza
	Leionella pneumophila
	Mycobacterium tuberculosis
	Streptococcus pneumonia
	Streptococcus pyogenes
	Bordatella pertussis
	Mycoplasma penumoniae
	Pneumocystis jirovecii (PJP)
	Pooled human nasal wash
	Candida albicans
	Pseudomonas aeruginosa
	Staphlococcus epidermis
	Staphlococcus salivarius

Interfering Substances

To determine and characterize the effects of potential interfering substances on the performance of the Panel, 2 endogenous and 9 exogenous substances were tested. Substances were added to SARS-CoV-2 spiked positive samples that were tested by the Panel on the T2Dx and compared to spiked positive samples with the vehicle control used to solubilize each respective substance. Potentially interfering substances were tested above clinically relevant concentrations. No interference was detectable at the specified test concentration for all substances listed below (Table 6).

Table 6. Substances tested for interference with the T2SARS-CoV-2 Panel –No interference observed

Exogenous Substances & Concentrations			
Human genomic DNA	3e5 cells/mL	Zanavir	3.3 mg/mL
Human blood	0.5% v/v	Tobramycin	0.1 mg/mL
Mucin (Bovine submaxillary gland)	0.1 mg/mL	Homeopathic allergy relief medications	10% v/v

Exogenous Substances & Concentrations			
Nasal spray	5% v/v	Mupirocin	0.2 mg/mL
Nasal gel	10 mg/mL	Benzocaine/Menthol throat lozenges	5 mg/mL
Nasal corticosteroids	5% v/v		

Clinical Performance

The clinical performance of the T2SARS-CoV-2 Panel was established by evaluating clinical nasopharyngeal swab specimens. A total of 101 clinical nasopharyngeal swab samples (60 positive and 41 negative for SARS-CoV-2) were tested with the T2SARS-CoV-2 Panel and the results were compared to results obtained with an FDA EUA RT-PCR test. Table 7 outlines the overall Positive Percent Agreement (PPA) and Negative Percent Agreement (NPA).

Table 7. T2SARS-CoV-2 Panel Clinical Performance

PPA		NPA	
Sensitivity	**95% CI**	**Specificity**	**95% CI**
95.0% (57/60)	86.3% - 98.3%	100.0% (41/41)	91.4% - 100.0%

FDA SARS-CoV-2 Reference Panel Testing

The evaluation of sensitivity and MERS-CoV cross-reactivity was performed using reference material (T1), blinded samples and a standard protocol provided by the FDA. The study included a range finding study and a confirmatory study for LoD. Blinded sample testing was used to establish specificity and to confirm the LoD. The samples were tested using the T2Dx instrument and the SARS-CoV-2 Panel. The results are summarized in Table 8.

Table 8. Summary of LoD Confirmation Result using the FDA SARS-CoV-2 Reference Panel

Reference Material Provided by FDA	Specimen Type	Product LoD (NDU/mL)	Cross-Reactivity
SARS-CoV-2	NP Swab	$1.8x10^4$	N/A
MERS-CoV		N/A	ND

NDU/mL = RNA NAAT detectable units/mL
N/A: Not Applicable
ND: Not Detected

Bibliography

1. Mylonakis E, Clancy CJ, Ostrosky-Zeichner L, Garey KW, Alangaden GJ, Vazquez JA, Groeger JS7, Judson MA, Vinagre YM, Heard SO, Zervou FN, Zacharioudakis IM, Kontoyiannis DP, Pappas PG. T2 magnetic resonance assay for the rapid diagnosis of candidemia in whole blood: a clinical trial. *Clin Infect Dis.* 2015 Mar 15;60(6):892-9.
2. U.S. Department of Health and Human Services, Centers for Disease Control and Prevention, & National Institutes of Health. (2009). Biosafety in microbiological and biomedical laboratories (5th ed.). Chosewood LC & Wilson DE (Eds.). Washington, DC: U.S. Government Printing Office.
3. Protection of Laboratory Workers from Occupationally Acquired Infections. Approved Guideline-Third Edition. CLSI Document. Clinical and Laboratory Standards Institute (CLSI). M29-A3 Wayne,PA: CLSI, 2005.

Table of Abbreviations

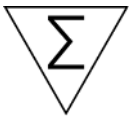
Table 8. Abbreviations that appear in the T2SARS-CoV-2 IFU

Abbreviations	Definitions
Panel	T2SARS-CoV-2 Panel
T2Dx	T2Dx Instrument
IFU	Instructions for Use
Cartridge	T2SARS-CoV-2 Cartridge
Reagent Tray	T2SARS-CoV-2 Reagent Tray
Sample Inlet	T2SARS-CoV-2 Sample Inlet
BSC	Biosafety Cabinet
IC	Internal Control
NaDCC	Sodium dichloroisocyanurate
uL	Microliter
mL	Milliliter
GE/mL	Genomic Equivalents per Milliliter
LoD	Limit of Detection
RPM	Revolutions Per Minute

Understanding the Symbols

Table 9. Symbols that may appear on the packaging and labeling

Symbol	Meaning
	*In Vitro* Diagnostic Medical Device
	Use By <YYYY-MM-DD>
	Reference Number or Catalog Number
	Manufacturer
	Do Not Reuse
	Batch Code or Lot Number

Symbol	Meaning
	Sufficient For <N tests>
	Consult *Instructions For Use*
	Caution
	Temperature Limitation
	For prescription use only
	Control

Notice to Purchaser

The Panel is an *in vitro* diagnostic for prescription use only. Distributed in accordance with the FDA guidance: Policy for Coronavirus Disease-2019 Tests During the Public Health Emergency (Revised)

See http://info.t2biosystems.com/instructions to download the latest version of the *T2SARS-CoV-2 Panel Instructions for Use*.

Technical Support & Contact Information

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