# Validation of Surveillance Assay for SARS-COV-2 Testing of Pooled Patient Swab Samples

## (The Infinity BiologiX TaqMan SARS-CoV-2, Flu A, Flu B RT-PCR Surveillance Assay will be performed in the Infinity BiologiX LLC Clinical Genomics Laboratory, a Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, certified high-complexity laboratory).

## Test Summary:

The Infinity BiologiX TaqMan SARS-CoV-2, Flu A, Flu B RT-PCR Assay is a real-time reverse transcription polymerase chain reaction (rRT-PCR) test intended for the qualitative detection of nucleic acid from SARS-CoV-2, Influenza A, and Influenza B in saliva, oropharyngeal (throat) swab, nasopharyngeal swab, anterior nasal swab, mid-turbinate nasal swab, and bronchoalveolar lavage (BAL) fluid from individuals suspected of COVID-19 and/or Influenza A/B

Testing is limited to Infinity BiologiX LLC locations that are certified under Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, and meets requirements to perform high-complexity tests.

Results are for the detection and identification of SARS-CoV-2, Influenza A, and Influenza B RNA for survelliance purposes. The SARS-CoV-2, Influenza A, or Influenza B RNA are generally detectable in respiratory specimens collected using nasopharyngeal swabs during the acute phase of infection.

Specimen pooling for survelliance purposes can increase throughput and conserve testing resources, but validation is needed to ensure that the reduced sensitivity due to pooling does not increase the false-negative rate. We use of ThermoFisher's swab samples in a pooled fashion. Pooled specimen testing can help increase testing capacity for SARS-CoV-2 survelliance with a low risk of false-negative results. In a simple pooled testing scheme, a number of individually collected specimens are combined in a single well or tube and tested together. If the pooled test result is negative, results for all individual specimens may be immediately reported as negative. If a pool is positive, then each individual in the pool will be asked to quarantine until the invidual testing results are available. The optimal number of specimens that can be included in a pool to maximize efficiency is determined by the prevalence of positive specimens in the population being tested and is further constrained by the sensitivity of the test to reliably detect a positive signal in a diluted negative specimen pool. It is critical, therefore, to validate pooling strategies for diagnostics tests to ensure that the false-negative rate remains below an acceptable threshold

Testing with the TaqMan SARS-CoV-2/Flu A, Flu B RT-PCR test is intended for use by qualified laboratory personnel specifically instructed and trained in the techniques of real-time PCR and *in vitro* diagnostic procedures. The assay is intended for testing pooled patient samples for survelliance purposes.

## Assay Protocol Overview:

The Infinity BiologiX TaqMan SARS-CoV-2, Flu A, Flu B, RT-PCR Assay is a real-time reverse transcription assay designed to detect RNA from SARS-CoV-2, Flu A, Flu B in nasopharyngeal swabs collected from participants. The purpose of this validation is to enable testing of pooled COVID-19/Flu A/Flu B nasopharyngeal swab samples for survelliance purposes

Swab specimens must be collected, transported and stored using the Miraclean foam tip clean room swabs (ThomasScientific, Catalog # **20A00M411 / MFS-741)**. Swab specimens must be transported dry and stored at ambient temperature and tested within 72 hours of collection when stored at ambient temperature.

### Swab Pooling, Virus spike-in, and Reconstitution strategy

*Swab Pooling and virus spike-in*
Twenty-four (24) swab samples were collected using manufacturer's instructions from twenty-four (24) patients tested negative for SARS-CoV-2, which was confirmed via IBX SARS COV-2 RT-PCR assay. All 24 swabs collected from patients

are placed in one 50 ml Falcon tube. A 25th swab is then spiked with COVID19 virus (*BEI Resources* NR-52287, Lot # 70035888) at the concentrations representing pools with low viral concentrations (1,500 copies/ml – 3x Limit of Detection (LoD)), or weak positive sample in a pool (1,500 copies/mL, or strong positive sample in a pool (1,312,500 copies/mL, diluted in 5–10 µl phosphate buffered saline (PBS) and then the spiked swab is added to the same tube containing 24 negative swabs. Different swab pools were created to determine the performance across a wide variety of pooled specimens as laid out in **Table 1** below.

**Table 1:** Strategy for creating pooled swab samples

| Performance Criteria | Transcript copies/mL | Number of negative swabs per pool | Number of positive swabs per pool | Total number of pools | Total number of swabs |
|---|---|---|---|---|---|
| Confirmation of performance in a negative pool | 0 | 25 | 0 | 9 | 225 |
| Confirmation of performance at low viral concentrations | 1,500 (without clinical matrix) | 24 | 1 | 9 | 225 |
| Detection of weak positive sample in a pool | 1,500 (with clinical matrix) | 24 | 1 | 12 | 300 |
| Detection of strong positive sample in a pool | 1,312,500 | 24 | 1 | 6 | 150 |
| | **Total** | | | **36** | **900** |

*Reconstitution of dry swab samples:*
Prior to testing the pooled specimens using TaqMan SARS-CoV-2, Flu A, Flu B RT-PCR Assay, the dry pooled swabs are reconstituted in 6 ml of PBS followed by vortexing for 30 s with intermittent pulsing. The reconstituted samples are then incubated for 20 min at room temperature. After incubation, the samples are vortexed again for 30 s with intermittent pulsing. 300 µl of the reconstituted sample is used in subsequent steps.
RNA extraction for all swab samples is performed using the PerkinElmer Chemagic 360 automated specimen processing system with the Chemagic Viral DNA/RNA 300 Kit H96. The input sample volume is 300µL, the elution volume is 60µL. Reverse transcriptase-PCR (RT-PCR) is performed using the Applied Biosystems TaqMan SAS-CoV-2/Flu A/Flu B assay with 12.5µL of the extracted sample.

## Instruments used for the test:

The Infinity BiologiX TaqMan SARS-CoV-2, FluA, FluB Assay is for use with the ThermoFisher Applied Biosystems QuantStudio 5 Real-Time PCR System equipped with software v1.3, or the Applied Biosystems ViiA7 Real-Time PCR System with the Applied Biosystems QuantStudio 5 software v1.3 for data analysis, and Perkin Elmer Chemagic 360 extraction instrument (software v6.3.0.3).

**Table 2:** Reagents and materials required for use of the Infinity BiologiX TaqMan SARS-CoV-2, FluA, FluB Assay

| Reagents | Manufacturer | Catalog # |
|---|---|---|
| Chemagic Viral DNA/RNA 300 Kit H96 | PerkinElmer | CMG-1033-S |
| 96 well Deep Well Plates | PerkinElmer | 43001-0120 |
| TaqMan SARS-CoV-2/Flu A/Flu B Assay | ThermoFisher Scientific | A47701 |
| 384 well PCR plate | ThermoFisher Scientific | 4483273 |
| Optical adhesive PCR plate cover | ThermoFisher Scientific | 4311971 |
| Nuclease-free water | -- | -- |
| Ethanol (96-100%) | -- | -- |

## Controls:

The controls supplied with the ThermoFisher TaqMan COVID-19 Combo Kit are described in **Table 3**.

**Table 3**. Controls supplied with the Applied Biosystems TaqMan COVID-19 Combo Kit

| Control Type | Purpose | Frequency of Testing |
|---|---|---|
| Negative | To monitor for cross-contamination during RNA extraction and RT-PCR | Once per batch of specimens |
| Positive | To monitor the integrity of the RT-PCR reagents and process | Once per run of RT-PCR |
| Internal (MS2 Phage) | To monitor the integrity of nucleic acid extraction and RT-PCR for each specimen | Added to each specimen and the Negative Control prior to extraction |

In addition to these controls, a No Template Control containing none of the SARS-CoV-2 targets or the Internal Control is included in every PCR run. The results from the controls are interpreted according to the criteria shown in **Table 4**. If the results obtained with the Positive, Negative and No Template Controls do not meet the criteria shown, the results from the entire batch of samples are considered invalid and repeat testing must be performed.

**Table 4:** Ct values for controls that must be observed to obtain valid results

| Control | Ct value (Optical Channel) | |
|---|---|---|
| | N/S Gene (VIC) | MS2 Phage (JUN) |
| Negative | >40 | <37 |
| Positive | <37 | Undetermined |
| No Template | Undetermined | Undetermined |
| Internal | Any | Any |

## Interpretation of Results:

The results from testing of pooled swab samples are interpreted according to the criteria described in **Table 5**.

**Table 5:** Result interpretation of pooled patient samples

| Ct Value (Optical Channel) | | Result Interpretation |
|---|---|---|
| N/S Gene (VIC) | MS2 Phage (JUN) | |
| Undetermined | <37 | Negative |
| <37 | <37 | Positive |
| <37 | Undetermined | Re-test |
| Undetermined | Undetermined | Re-test |

### Reportable range

The TaqMan SARS-CoV-2/Flu A/Flu B Assay for detection of SARS-CoV-2/FluA/FluB in pooled clinical swab samples is a qualitative test and therefore a reportable range does not apply.

### Reference range

The reference range will be "SARS-CoV-2/FluA/FluB not detected" and will not be validated by a clinical study.

## Performance Evaluation:

### *Accuracy*

To assess the test accuracy of detecting negative and positive contrived pooled swab specimens, we ran the TaqMan SARS-CoV-2, Flu A, Flu B RT-PCR Assay on four different swab pools as laid out in the **Table 6** below. After reconstituting the 25 dry swab pools in 6 mL of PBS, we obtained 10 extraction replicates of 300 µL each from the pool and ran RT-PCR on each replicate. 90/90 (100%, 95% CI [95.98% - 100%]) negative pooled swab specimens tested negative. 90/90 (100%, 95% CI [95.98% - 100%]) pooled swab specimens 97% - 100%]) pooled swab specimens with clinical matrix spiked with 1,500 copies/mL of the virus tested positive and 59/60 (95%, 95% CI [91% - 99.96%]) pooled swab specimens with clinical matrix spiked with 1,312,500 copies/mL of the virus tested positive. The results of the assay are summarized in **Table 6** below. The positive (PPA) and negative percent agreement (NPA) are summarized in **Table 7** below.

**Table 6:** Summary results of pooled swabs. *One pooled sample from the strong positive pooled swabs group tested inconclusive.

| Performance Metric | Transcript Copies/mL | Pool type | Swab Pools | Analysis | Positive | Negative |
|---|---|---|---|---|---|---|
| Negative pool | 0 | Negative | 9 | Negative (%) | 0 (0) | 90 (100) |
| Confirmation of performance at low viral concentration | 1,500 | Positive | 9 | Positive (%) | 90 (100) | 0 (0) |
| Detection of weak positive swabs in a pool | 1,500 | Positive | 12 | Positive (%) | 120 (100) | 0 (0) |
| Detection of strong positive swabs in a pool | 1,312,500 | Positive | 6 | Positive (%) | 59 (98.33) | 1* (1.67) |

**Table 7:** Positive percent (PPA) and negative percent agreement (NPA) for Pooled swab samples vs expected results tested across three different days with three sets of independent samples

| Samples Tested in 25-swab pool | Expected Results | |
|---|---|---|
| **Transcript Copies/mL** | **Positive** | **Negative** |
| Negative | 0 (0%) | 90 (100%) |
| 1,500 (without clinical matrix) | 90 (100%) | 0 (0) |
| 1,500 (with clinical matrix) | 120 (100% | 0 (0) |
| 1,312,500 (with clinical matrix) | 59 (98.33%) | 1 (1.67%) |
| All Positive | 269 (99.62%) | 1 (0.37%) |

***Acceptance criteria*:** > 95% percent agreement between replicate samples. **PASS**

***Precision***

***Repeatability***

To assess the repeatability (within-run precision) of pooled swab specimen testing, we performed ten replicate measurements of the contrived validation samples described in the section on Accuracy within a single batch. We then assessed the concordance between the ten replicate measurements and determined the percent of concordant results.

In total, 359/360 (98.33%, 95% CI [98.46% - 99.99%]) replicates were concordant between ten repeat measurements run on the same batch for 36 swab pools. See Table 8, 9, and 10 for between run variations across 3 different days. See **Appendix** for detailed run data.

***Acceptance criteria*:** > 95% percent agreement between duplicates. **PASS**

**Table 8:** Repeatability of pooled swab specimen testing (within in run precision)

| | Performance Criteria | Copies/mL | n | Analysis | Day 1 | | Day 2 | | Day 3 | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | Target (Optical Channel) | | Target (Optical Channel) | | Target (Optical Channel) | |
| | | | | | N/S Gene | MS2 | N/S Gene | MS2 | N/S Gene | MS2 |
| | | | | | (VIC) | (JUN) | (VIC) | (JUN) | (VIC) | (JUN) |
| 25-Swab Pools | Performance in a negative pool | 0 | 30 | **Mean** | NA | 19.01 | NA | 21.97 | NA | 22.84 |
| | | | | **SD** | NA | 1.15 | NA | 0.31 | NA | 0.67 |
| | | | | **CV** | NA | 6% | NA | 1% | NA | 3% |
| | Performance at low viral concentration | 1,500 | 30 | **Mean** | 31.66 | 18.94 | 33.56 | 22.16 | 32.62 | 22.58 |
| | | | | **SD** | 2.94 | 1.19 | 0.61 | 0.22 | 2.40 | 0.42 |
| | | | | **CV** | 9% | 6% | 2% | 1% | 7% | 2% |
| | Detection of weak positive swabs in a pool | 1,500 | 40 | **Mean** | 28.83 | 19.57 | 30.93 | 22.19 | 31.96 | 22.70 |
| | | | | **SD** | 2.88 | 0.81 | 0.93 | 0.33 | 2.53 | 0.52 |
| | | | | **CV** | 10% | 4% | 3% | 1% | 8% | 2% |
| | Detection of strong positive swabs in a pool | 1,312,500 | 20 | **Mean** | 20.69 | 20.37 | 20.90 | 22.22 | 22.29 | 22.89 |
| | | | | **SD** | 1.54 | 0.95 | 0.27 | 0.36 | 1.25 | 0.68 |
| | | | | **CV** | 0.07 | 0.05 | 0.01 | 2% | 6% | 3% |

**Table 9: Day 1** Positive percent (PPA) and negative percent agreement (NPA) for Pooled swab samples vs expected results

| Samples Tested in 25-swab pool | Expected Results | |
|---|---|---|
| **Transcript Copies/mL** | **Positive** | **Negative** |
| Negative | 0 (0%) | 30 (100%) |
| 1,500 (without clinical matrix) | 30 (100%) | 0 (0) |
| 1,500 (with clinical matrix) | 40 (100% | 0 (0) |
| 1,312,500 (with clinical matrix) | 19 (95.00%) | 1 (5%) |
| All Positive | 89 (98.88%) | 1 (1.12%) |

**Table 10: Day 1** Performance metrics

| Performance metric | Description | Performance |
|---|---|---|
| Confirmation of performance at low viral concentration | Percent of swab pools positive at 1500 copies/mL (without clinical matrix)* | 30 / 30 = 100%; 95% CI [88.4%-100%] |
| Detection of weak positive swabs in a pool | Percent of swab pools positive at 1500 copies/mL (with clinical matrix)* | 40 / 40 = 100%; 95% CI [91.2%-100%] |
| Detection of strong positive swabs in a pool | Percent of swab pools positive at 1,312,500 copies/mL (with clinical matrix)* | 19 / 20 = 95%; 95% CI [75.1%-100%] |

**Table 11: Day 2** Positive percent (PPA) and negative percent agreement (NPA) for Pooled swab samples vs expected results

| Samples Tested in 25-swab pool | Expected Results | |
|---|---|---|
| **Transcript Copies/mL** | **Positive** | **Negative** |
| Negative | 0 (0%) | 90 (100%) |
| 1,500 (without clinical matrix) | 30 (100%) | 0 (0) |
| 1,500 (with clinical matrix) | 40 (100% | 0 (0) |
| 1,312,500 (with clinical matrix) | 20 (100%) | 0 (0) |
| All Positive | 90 (100%) | 0 (0) |

**Table 12:** Day 2 Performance metrics

| Performance metric | Description | Performance |
|---|---|---|
| Confirmation of performance at low viral concentration | Percent of swab pools positive at 1500 copies/mL (without clinical matrix)* | 30 / 30 = 100%; 95% CI [88.4%-100%] |
| Detection of weak positive swabs in a pool | Percent of swab pools positive at 1500 copies/mL (with clinical matrix)* | 40 / 40 = 100%; 95% CI [91.2%-100%] |
| Detection of strong positive swabs in a pool | Percent of swab pools positive at 1,312,500 copies/mL (with clinical matrix)* | 20 / 20 = 100%; 95% CI [83.2%-100%] |

**Table 13: Day 3** Positive percent (PPA) and negative percent agreement (NPA) for Pooled swab samples vs expected results

| Samples Tested in 25-swab pool | Expected Results | |
|---|---|---|
| **Transcript Copies/mL** | **Positive** | **Negative** |
| Negative | 0 (0%) | 90 (100%) |
| 1,500 (without clinical matrix) | 30 (100%) | 0 (0) |
| 1,500 (with clinical matrix) | 40 (100% | 0 (0) |
| 1,312,500 (with clinical matrix) | 20 (100%) | 0 (0) |
| All Positive | 90 (100%) | 0 (0) |

**Table 14:** Day 3 Performance metrics

| Performance metric | Description | Performance |
|---|---|---|
| Confirmation of performance at low viral concentration | Percent of swab pools positive at 1500 copies/mL (without clinical matrix)* | 30 / 30 = 100%; 95% CI [88.4%-100%] |
| Detection of weak positive swabs in a pool | Percent of swab pools positive at 1500 copies/mL (with clinical matrix)* | 40 / 40 = 100%; 95% CI [91.2%-100%] |
| Detection of strong positive swabs in a pool | Percent of swab pools positive at 923,000 copies/mL (with clinical matrix)* | 20 / 20 = 100%; 95% CI [83.2%-100%] |

***Reproducibility***

To assess the reproducibility (between-run precision) of pooled saliva specimens, we performed three measurements of the clinical validation samples described in Accuracy on three separate days performed by three different operators on three different instruments. We then assessed the concordance between the three runs and determined the percent of concordant results to determine between run variability

In total, 119/120 (99.17%; 95% CI [95.44% - 99.98%]) replicates run each day were concordant between ten measurements run within the same batch across three different days. See Table 8, 9, and 10 for between run variations across 3 different days. See **Table 15** for summary of the results for assay reproducibility and **Appendix** for detailed run data.

***Acceptance criteria:*** > 95% concordance of samples between runs. **PASS**

**Table 15:** Reproducibility of pooled swab specimen testing (between run precision)

| | Performance Criteria | Description | Copies/mL | n | Analysis | Target (Optical Channel) | |
|---|---|---|---|---|---|---|---|
| | | | | | | N/S Gene (VIC) | MS2 (JUN) |
| 25-Swab Pools | Confirmation of performance in a negative pool | Pooled sample without virus spike-in representing a negative pool | 0 | 90 | **Mean** | NA | 21.27 |
| | | | | | **SD** | NA | 1.82 |
| | | | | | **CV** | NA | 9% |
| | Confirmation of performance at low viral concentration | Percent of swab pools positive at 1,500 copies/mL (without clinical matrix)* | 1,500 | 90 | **Mean** | 32.61 | 21.23 |
| | | | | | **SD** | 2.33 | 1.79 |
| | | | | | **CV** | 7% | 8% |
| | Detection of weak positive swabs in a pool | Percent of swab pools positive at 1,500 copies/mL (with clinical matrix)* | 1,500 | 120 | **Mean** | 30.57 | 21.49 |
| | | | | | **SD** | 2.61 | 1.50 |
| | | | | | **CV** | 9% | 7% |
| | Detection of strong positive swabs in a pool | Percent of swab pools positive at 1,312,500 copies/mL (with clinical matrix)* | 1,312,500 | 60 | **Mean** | 21.31 | 21.85 |
| | | | | | **SD** | 1.34 | 1.27 |
| | | | | | **CV** | 6% | 6% |

***Analytical Sensitivity***

The Limit of Detection (LoD) is defined as the lowest SARS-CoV-2, influenza A, influenza B RNA viral concentration that is successfully detected with a probability of 95% or greater. The LoD was determined using *in vitro* heat-inactivated SARS-CoV-2 virus (BEI NR-52287), Flu A (Zeptometrix, A/Perth/16/2009 and A/Brisbane/59/2007), Flu B (Zeptometrix, B/Florida/04/2006, and B/Wisconsin/01/2010) that was diluted in SARS-CoV-2, FluA, FluB negative single nasopharyngeal swab matrix. An initial estimate of the LoD with the Applied Biosystems QuantStudio 5 Real-Time PCR System was obtained by testing three replicates at each of four different target levels: 1000, 500, 200 and 100 copies/mL. The lowest level at which all three replicates were positive for SARS-CoV-2, FluA, Flu B targets was determined. The estimated LoD was confirmed by testing an additional 20 replicates at the same target level. All 20 NP swab replicates produced the expected results. The LoD was therefore confirmed to be as that shown in **Table 16** below.

**Table 16:** Limit of Detection of Infinity BiologiX TaqMan SARS Cov-2, FluA, FluB, combo assay

| Target | Strain/details | Vendor | Catalog # | Lot Number | Stock Viral GCE/mL | LoD in GCE/mL |
|---|---|---|---|---|---|---|
| SARS-CoV-2 | Isolate USA-WA1/2020 | BEI | NR-52287 | 70035888 | 1.75E+09 | **100** |
| Flu A | A/Perth/16/2009 | ZeptoMetrix | 0810251CF | 324079 | 3.11E+09 | **200** |
| Flu A | A/Brisbane/59/2007 | ZeptoMetrix | 0810244CF | 323919 | 1.58E+11 | **500** |
| Flu B | B/Florida/04/2006 | ZeptoMetrix | 0810255CF | 312479 | 1.07E+10 | **500** |
| Flu B | B/Wisconsin/01/2010 | ZeptoMetrix | 0810241CF | 323015 | 1.37E+10 | **1000** |

Furthermore, an initial estimate to determine the limit of detection in a pooled swab sample with the Applied Biosystems QuantStudio 5 Real-Time PCR System was obtained by testing three replicates of 25 dry pooled swab samples (24 negative samples, and one sample spiked at 1,200 or 1,500 copies/mL). The lowest level at which all three pooled replicates were positive for SARS-CoV-2 N/S Gene was 1,500 copies/mL. All 20 replicates produced the expected results for each SARS-CoV-2 target, and the LoD was therefore confirmed to be 1,500 copies/mL for 25 dry swabs per pool specimen.

Validated results of the 25 dry swabs per pool across three days are summarized in **Table 17** below.

**Table 17.** Validation of pooling test performance with 25 dry swabs per pool across three days

| Performance Criteria | | Description | Copies /mL | Swab pools | Analysis | Target (Optical Channel) | |
|---|---|---|---|---|---|---|---|
| | | | | | | N/S Gene (VIC) | MS2 (JUN) |
| 25-Swab Pool | Confirmation of performance in a negative pool | Pooled sample without virus spike-in representing a negative pool | 0 | 9 | Negative (%) | 0 (0) | 90 (100) |
| | | | | | Mean (SD) | NA (NA) | 21.27 (1.82) |
| | | | | | CI (95%) | 96% - 100% | |
| | Confirmation of performance at low viral concentration | Percent of swab pools positive at 1,500 copies/mL (without clinical matrix)* | 1,500 | 9 | Positive (%) | 90 (100) | 90 (100) |
| | | | | | Mean (SD) | 32.61 (2.33) | 21.23 (1.79) |
| | | | | | CI (95%) | 96% - 100% | |
| | Detection of weak positive swabs in a pool | Percent of swab pools positive at 1,500 copies/mL (with clinical matrix)* | 1,500 | 12 | Positive (%) | 120 (100) | 120 (100) |
| | | | | | Mean (SD) | 30.57 (2.60) | 21.48 (1.49) |
| | | | | | CI (95%) | 97% - 100% | |
| | Detection of strong positive swabs in a pool | Percent of swab pools positive at 1,312,500 copies/mL (with clinical matrix)* | 1,312,500 | 6 | Positive (%) | 59 (98.33) | 1 (1.67) |
| | | | | | Mean (SD) | 21.30 (1.33) | 21.84 (1.27) |
| | | | | | CI (95%) | 91.06% - 99.96% | |

**Virus used to prepare swab samples: BEI Resources* NR-52287, Lot # 70035888

***Analytical Specificity***

***Inclusivity***

The Infinity BiologiX TaqPath SARS-CoV-2 Assay is a modification of the previously authorized ThermoFisher Applied Biosystems TaqPath COVID-19 Combo Kit. The assay targets specific genomic regions of the SARS-CoV-2 nucleocapsid (N) gene, spike (S) gene, and ORF1ab region, influenza A and Influenza B. Inclusivity was demonstrated by mapping the primers and probes to complete SARS-CoV-2, Influenza A, Influenza B genomes that were available in the GenBank and GISAID (Global Initiative on Sharing All Influenza Data) databases as of March 5, 2020. For all primers and probes, there was 100% homology to each of the SARS-CoV-2, Flu A, and Flu Bsequences analyzed

***Acceptance criteria:*** Greater than 80% homology with each sequence analyzed. **PASS**

***Cross reactivity***

The Infinity BiologiX TaqMan SARS-CoV-2/Flu A/Flu B Assay is a modification of the previously authorized ThermoFisher Applied Biosystems TaqMan COVID-19 Combo Kit.In silico analysis was performed to predict cross-reactivity between the TaqPath™ RT-PCR COVID-19, Flu A, Flu B Combo kit (COVID/Flu test) primer/probe sequences and a series of microbes commonly found in respiratory specimens per [the] TaqPath™ RT-PCR COVID, Flu A, Flu B Kit Cross-Reactivity Study Protocol. Based upon BLAST analysis, 33 isolates representing 21 total bacterial and fungal organisms as shown in **Table 18** had >=80% homology to either the reverse primer, forward primer or probe sequence of the COVID/Flu test SARS-

CoV-2 N gene, Influenza A, or MS2 internal positive control assays; no primer or probe homology of >=80% was identified with the Influenza B assay. For the select isolates that shared >=80% identity with a given assay primer or probe sequence, sequence identity with other assay components was not detected. Given that both assay primers are required to produce an amplicon and all three assay components are required to generate signal, it was determined that there is little to no chance of cross-reactivity between assays and other respiratory microbes, or interference with detection of SARS-CoV-2, influenza A and influenza B viruses, or the MS2 internal control. Based on this analysis, significant amplification of non-target sequences that could result in cross-reaction (false-positive results) or interference (false-negative results) was considered unlikely to occur.
Because cross reactivity was assessed for the SARS-CoV-2 S gene assay used in this test under [under previous EUA for TaqPath COVID-19 assay], it was not required to evaluate the SARS-CoV-2 S gene assay in this study.

***Acceptance criteria:*** *0% Cross* reactivity or interference with detection. **PASS**

**Table 18:** Organisms and viruses evaluated for potential cross-reaction and/or interference with the TaqMan-SARS-Cov-2/Flu A/Flu B RT-PCR assay

| | |
|---|---|
| *Bacillus anthracis* | *Adenovirus* |
| *Bordetella pertussis* | *Enterovirus* |
| *Chlamydia psittaci*[1] | *Human Coronavirus 229E* |
| *Chlamydophila pneumoniae* | *Human Coronavirus HKU1* |
| *Corynebacterium diphtheriae* | *Human Coronavirus NL63* |
| *Coxiella burnetii*[1] | *Human Coronavirus OC43* |
| *Haemophilus influenzae* | *Human Metapneumovirus* |
| *Legionella non-pneumophila (Legionella longbeachae)* | *Influenza C virus* |
| *Legionella pneumophila* | *MERS Coronavirus* |
| *Leptospira spp.* | *Parainfluenza 1 virus* |
| *Moraxella catarrhalis* | *Parainfluenza 2 virus* |
| *Mycobacterium tuberculosis* | *Parainfluenza 3 virus* |
| *Mycoplasma pneumoniae* | *Parainfluenza 4 virus* |
| *Neisseria elongata* | *Parechovirus* |
| *Neisseria meningitidis* | *Respiratory Syncytial Virus A* |
| *Pseudomonas aeruginosa* | *Respiratory Syncytial Virus B* |
| *Staphylococcus aureus* | *Rhinovirus* |
| *Staphylococcus epidermidis* | *SARS Coronavirus* |
| *Streptococcus pneumoniae* | *COVID-BEI GIV (SARS-CoV-2)* |
| *Streptococcus pyogenes* | *Candida albicans* |
| *Streptococcus salivarius* | *Pneumocystis jirovecii* |

The complete performance characteristics of the TaqMan-SARS-Cov-2/Flu A/Flu B RT-PCR assay for 25 dry swabs per pool testing strategy is listed in **Table 19** below

**Table 19:** Performance characteristics of the TaqMan-SARS-Cov-2/Flu A/Flu B RT-PCR assay

| **Performance Metric** | | **Description** | **Performance** |
|---|---|---|---|
| Accuracy | PPA | TP/ (TP + FN) | 269 / 270 = 99.62%;<br>95% CI [97.95% - 99.99%] |
| | PNA | TN/ (TN + FP) | 90 / 90 = 100%;<br>95% CI [95.98% - 100%] |
| Precision | Repeatability | Within run replicate agreement | 359 / 360 = 99.72%;<br>95% CI [98.46% – 99.99%] |
| | Reproducibility | Between run replicate agreement | 119 / 120 = 99.16%;<br>95% CI [95.44% - 99.98%] |
| Analytical Sensitivity | Limit of Detection | Lowest concentration of virus detected with 95% probability of obtaining a correct result | 1,500 copies/mL (ATCC VR-1986HK) |
| | Inclusivity | Amplification of all possible SARS-CoV-2/FluA/FluB genomes | 100% homology to each of the sequences analyzed |
| | Cross-Reactivity | Amplification of non-target sequences | 0% false positive and 0% false negative |

**Validation Report and Data Review**

All data and analysis were reviewed by laboratory director and validation report was compiled by the laboratory director listed below

Sameer Kalghatgi, Ph.D.
Sr. Director, Laboratory Operations
Infinity BiologiX

## Appendix: Detailed run data:

All raw data related to all the qPCR runs for obtaining the data in the appendix are stored securely in the ThermoFisher cloud platform under a common filename format – RUO-Gingko_Concentric Pooling_ExptDate**.** The cloud account is only accessible to authorized users with login credentials. Login information is changed every 6 months.

**Day 1**

| IBX Sample | MS2 | COVID N/S | IBX Result | Expected Call | COVID copies/mL |
|---|---|---|---|---|---|
| 603299_1 | 15.1 | 32 | Detected | Positive | 1500 |
| 603299_10 | 20.87 | 34.67 | Detected | Positive | 1500 |
| 603299_2 | 18.76 | 31.85 | Detected | Positive | 1500 |
| 603299_3 | 19.56 | 21.72 | Detected | Positive | 1500 |
| 603299_4 | 18.6 | 33.93 | Detected | Positive | 1500 |
| 603299_5 | 16.29 | 32.95 | Detected | Positive | 1500 |
| 603299_6 | 17.37 | 32.28 | Detected | Positive | 1500 |
| 603299_7 | 16.98 | 32.14 | Detected | Positive | 1500 |
| 603299_8 | 20.34 | 33.95 | Detected | Positive | 1500 |
| 603299_9 | 19.86 | 34.22 | Detected | Positive | 1500 |
| 603300_1 | 19.07 | 34.14 | Detected | Positive | 1500 |
| 603300_10 | 19.31 | 28.98 | Detected | Positive | 1500 |
| 603300_2 | 19.37 | 33.85 | Detected | Positive | 1500 |
| 603300_3 | 19.23 | 34.38 | Detected | Positive | 1500 |
| 603300_4 | 19.78 | 35.39 | Detected | Positive | 1500 |
| 603300_5 | 19.68 | 34.34 | Detected | Positive | 1500 |
| 603300_6 | 19.84 | 34.59 | Detected | Positive | 1500 |
| 603300_7 | 19.95 | 34.95 | Detected | Positive | 1500 |
| 603300_8 | 19.37 | 32.46 | Detected | Positive | 1500 |
| 603300_9 | 19.19 | 29.54 | Detected | Positive | 1500 |
| 603302_1 | 19.48 | 29.12 | Detected | Positive | 1500 |
| 603302_10 | 19.24 | 29.62 | Detected | Positive | 1500 |
| 603302_2 | 19.38 | 27.88 | Detected | Positive | 1500 |
| 603302_3 | 19.54 | 28.51 | Detected | Positive | 1500 |
| 603302_4 | 18.83 | 29 | Detected | Positive | 1500 |
| 603302_5 | 18.57 | 28.69 | Detected | Positive | 1500 |
| 603302_6 | 18.11 | 31.56 | Detected | Positive | 1500 |
| 603302_7 | 18.44 | 30.86 | Detected | Positive | 1500 |
| 603302_8 | 18.7 | 31.17 | Detected | Positive | 1500 |
| 603302_9 | 19.49 | 31.19 | Detected | Positive | 1500 |
| 603297_1 | 19.93 | 31.86 | Detected | Positive | 1500 |
| 603297_10 | 18.9 | 30.51 | Detected | Positive | 1500 |

| | | | | | |
|---|---|---|---|---|---|
| 603297_2 | 19.6 | 30.85 | Detected | Positive | 1500 |
| 603297_3 | 19.79 | 31.52 | Detected | Positive | 1500 |
| 603297_4 | 19.8 | 30.75 | Detected | Positive | 1500 |
| 603297_5 | 20.56 | 32.97 | Detected | Positive | 1500 |
| 603297_6 | 20.07 | 31.74 | Detected | Positive | 1500 |
| 603297_7 | 20.1 | 31.82 | Detected | Positive | 1500 |
| 603297_8 | 20.1 | 30.98 | Detected | Positive | 1500 |
| 603297_9 | 18.5 | 30.43 | Detected | Positive | 1500 |
| 603301_1 | 19.23 | 25.93 | Detected | Positive | 1500 |
| 603301_10 | 20.17 | 30.3 | Detected | Positive | 1500 |
| 603301_2 | 19.23 | 24.86 | Detected | Positive | 1500 |
| 603301_3 | 20.09 | 28.99 | Detected | Positive | 1500 |
| 603301_4 | 19.17 | 29.3 | Detected | Positive | 1500 |
| 603301_5 | 18.82 | 25.67 | Detected | Positive | 1500 |
| 603301_6 | 18.47 | 29.45 | Detected | Positive | 1500 |
| 603301_7 | 20.01 | 30.3 | Detected | Positive | 1500 |
| 603301_8 | 19.67 | 31.66 | Detected | Positive | 1500 |
| 603301_9 | 20.22 | 32.4 | Detected | Positive | 1500 |
| 603303_1 | 18.6 | 29.08 | Detected | Positive | 1500 |
| 603303_10 | 18.88 | 31.75 | Detected | Positive | 1500 |
| 603303_2 | 19.31 | 22.43 | Detected | Positive | 1500 |
| 603303_3 | 18.33 | 26.95 | Detected | Positive | 1500 |
| 603303_4 | 19.45 | 25.41 | Detected | Positive | 1500 |
| 603303_5 | 19.91 | 24.45 | Detected | Positive | 1500 |
| 603303_6 | 19.68 | 26.45 | Detected | Positive | 1500 |
| 603303_7 | 19.11 | 24.12 | Detected | Positive | 1500 |
| 603303_8 | 19.4 | 26.51 | Detected | Positive | 1500 |
| 603303_9 | 19.49 | 28.58 | Detected | Positive | 1500 |
| 603304_1 | 18.35 | 30.52 | Detected | Positive | 1500 |
| 603304_10 | 20.88 | 26.75 | Detected | Positive | 1500 |
| 603304_2 | 19.41 | 31.86 | Detected | Positive | 1500 |
| 603304_3 | 17.87 | 32.2 | Detected | Positive | 1500 |
| 603304_4 | 18.22 | 28.79 | Detected | Positive | 1500 |
| 603304_5 | 19.88 | 22.67 | Detected | Positive | 1500 |
| 603304_6 | 21.22 | 27.89 | Detected | Positive | 1500 |
| 603304_7 | 20.38 | 30.42 | Detected | Positive | 1500 |
| 603304_8 | 20.81 | 25.95 | Detected | Positive | 1500 |
| 603304_9 | 21 | 28.12 | Detected | Positive | 1500 |
| 603298_1 | 20.14 | 20.39 | Detected | Positive | 1312500 |
| 603298_10 | 20.65 | 21.67 | Detected | Positive | 1312500 |

| | | | | | |
|---|---|---|---|---|---|
| 603298_2 | 21.22 | 22.11 | Detected | Positive | 1312500 |
| 603298_3 | 19.77 | 18.13 | Detected | Positive | 1312500 |
| 603298_4 | 19.39 | 20.12 | Detected | Positive | 1312500 |
| 603298_5 | 18.91 | 16.46 | Detected | Positive | 1312500 |
| 603298_6 | 19.47 | 21.19 | Detected | Positive | 1312500 |
| 603298_7 | Undetermined | Undetermined | Inconclusive | Positive | 1312500 |
| 603298_8 | 19.89 | 20.95 | Detected | Positive | 1312500 |
| 603298_9 | 19.6 | 20.12 | Detected | Positive | 1312500 |
| 603308_1 | 20.63 | 21.28 | Detected | Positive | 1312500 |
| 603308_10 | 20.41 | 20.59 | Detected | Positive | 1312500 |
| 603308_2 | 23.21 | 23.73 | Detected | Positive | 1312500 |
| 603308_3 | 21.05 | 21.78 | Detected | Positive | 1312500 |
| 603308_4 | 20.89 | 20.9 | Detected | Positive | 1312500 |
| 603308_5 | 20.46 | 21.05 | Detected | Positive | 1312500 |
| 603308_6 | 20.49 | 21.16 | Detected | Positive | 1312500 |
| 603308_7 | 20.95 | 21.27 | Detected | Positive | 1312500 |
| 603308_8 | 19.3 | 19.16 | Detected | Positive | 1312500 |
| 603308_9 | 20.56 | 21.07 | Detected | Positive | 1312500 |
| 603305_1 | 19.38 | Undetermined | Not Detected | Negative | 0 |
| 603305_10 | 18.67 | Undetermined | Not Detected | Negative | 0 |
| 603305_2 | 20.05 | Undetermined | Not Detected | Negative | 0 |
| 603305_3 | 19.22 | Undetermined | Not Detected | Negative | 0 |
| 603305_4 | 18.95 | Undetermined | Not Detected | Negative | 0 |
| 603305_5 | 19.15 | Undetermined | Not Detected | Negative | 0 |
| 603305_6 | 18.84 | Undetermined | Not Detected | Negative | 0 |
| 603305_7 | 18.87 | Undetermined | Not Detected | Negative | 0 |
| 603305_8 | 19.32 | Undetermined | Not Detected | Negative | 0 |
| 603305_9 | 19.24 | Undetermined | Not Detected | Negative | 0 |
| 603306_1 | 20.6 | Undetermined | Not Detected | Negative | 0 |
| 603306_10 | 19.81 | Undetermined | Not Detected | Negative | 0 |
| 603306_2 | 17.74 | Undetermined | Not Detected | Negative | 0 |
| 603306_3 | 14.55 | Undetermined | Not Detected | Negative | 0 |
| 603306_4 | 18.21 | Undetermined | Not Detected | Negative | 0 |
| 603306_5 | 16.95 | Undetermined | Not Detected | Negative | 0 |
| 603306_6 | 18.14 | Undetermined | Not Detected | Negative | 0 |
| 603306_7 | 18.1 | Undetermined | Not Detected | Negative | 0 |
| 603306_8 | 18.6 | Undetermined | Not Detected | Negative | 0 |
| 603306_9 | 19.89 | Undetermined | Not Detected | Negative | 0 |
| 603307_1 | 19.4 | Undetermined | Not Detected | Negative | 0 |
| 603307_10 | 20.08 | Undetermined | Not Detected | Negative | 0 |

| | | | | | |
|---|---|---|---|---|---|
| 603307_2 | 19.47 | Undetermined | Not Detected | Negative | 0 |
| 603307_3 | 19.59 | Undetermined | Not Detected | Negative | 0 |
| 603307_4 | 19.04 | Undetermined | Not Detected | Negative | 0 |
| 603307_5 | 20.22 | Undetermined | Not Detected | Negative | 0 |
| 603307_6 | 19.54 | Undetermined | Not Detected | Negative | 0 |
| 603307_7 | 19.33 | Undetermined | Not Detected | Negative | 0 |
| 603307_8 | 19.68 | Undetermined | Not Detected | Negative | 0 |
| 603307_9 | 19.65 | Undetermined | Not Detected | Negative | 0 |

**Day 2**

| IBX Sample | MS2 | COVID N/S | IBX Result | Expected Call | COVID copies/mL |
|---|---|---|---|---|---|
| 606490-1 | 22.36 | 33.24 | Detected | Positive | 1500 |
| 606490-10 | 22.11 | 31.30 | Detected | Positive | 1500 |
| 606490-2 | 22.21 | 33.17 | Detected | Positive | 1500 |
| 606490-3 | 22.00 | 33.49 | Detected | Positive | 1500 |
| 606490-4 | 22.20 | 33.67 | Detected | Positive | 1500 |
| 606490-5 | 22.21 | 33.84 | Detected | Positive | 1500 |
| 606490-6 | 22.29 | 33.97 | Detected | Positive | 1500 |
| 606490-7 | 22.33 | 33.89 | Detected | Positive | 1500 |
| 606490-8 | 22.06 | 33.59 | Detected | Positive | 1500 |
| 606490-9 | 22.43 | 33.94 | Detected | Positive | 1500 |
| 606491-1 | 21.84 | 32.38 | Detected | Positive | 1500 |
| 606491-10 | 21.97 | 32.90 | Detected | Positive | 1500 |
| 606491-2 | 22.00 | 33.67 | Detected | Positive | 1500 |
| 606491-3 | 22.27 | 33.06 | Detected | Positive | 1500 |
| 606491-4 | 21.98 | 33.06 | Detected | Positive | 1500 |
| 606491-5 | 21.85 | 33.93 | Detected | Positive | 1500 |
| 606491-6 | 21.47 | 34.13 | Detected | Positive | 1500 |
| 606491-7 | 22.30 | 33.12 | Detected | Positive | 1500 |
| 606491-8 | 22.00 | 33.58 | Detected | Positive | 1500 |
| 606491-9 | 21.97 | 33.34 | Detected | Positive | 1500 |
| 606494-1 | 22.29 | 33.98 | Detected | Positive | 1500 |
| 606494-10 | 22.03 | 34.39 | Detected | Positive | 1500 |
| 606494-2 | 22.53 | 33.77 | Detected | Positive | 1500 |
| 606494-3 | 22.28 | 34.00 | Detected | Positive | 1500 |
| 606494-4 | 22.25 | 33.91 | Detected | Positive | 1500 |
| 606494-5 | 22.37 | 34.00 | Detected | Positive | 1500 |
| 606494-6 | 22.28 | 33.50 | Detected | Positive | 1500 |

| | | | | | |
|---|---|---|---|---|---|
| 606494-7 | 22.29 | 34.00 | Detected | Positive | 1500 |
| 606494-8 | 22.29 | 34.09 | Detected | Positive | 1500 |
| 606494-9 | 22.24 | 33.82 | Detected | Positive | 1500 |
| 606488-1 | 21.80 | 30.57 | Detected | Positive | 1500 |
| 606488-10 | 21.83 | 30.12 | Detected | Positive | 1500 |
| 606488-2 | 22.27 | 30.83 | Detected | Positive | 1500 |
| 606488-3 | 22.09 | 31.26 | Detected | Positive | 1500 |
| 606488-4 | 22.07 | 30.75 | Detected | Positive | 1500 |
| 606488-5 | 22.06 | 30.53 | Detected | Positive | 1500 |
| 606488-6 | 22.39 | 31.50 | Detected | Positive | 1500 |
| 606488-7 | 22.11 | 30.42 | Detected | Positive | 1500 |
| 606488-8 | 22.16 | 30.49 | Detected | Positive | 1500 |
| 606488-9 | 21.98 | 30.54 | Detected | Positive | 1500 |
| 606495-1 | 22.44 | 30.55 | Detected | Positive | 1500 |
| 606495-10 | 22.75 | 31.33 | Detected | Positive | 1500 |
| 606495-2 | 22.46 | 30.70 | Detected | Positive | 1500 |
| 606495-3 | 22.21 | 30.28 | Detected | Positive | 1500 |
| 606495-4 | 22.08 | 30.53 | Detected | Positive | 1500 |
| 606495-5 | 22.20 | 30.49 | Detected | Positive | 1500 |
| 606495-6 | 21.86 | 30.39 | Detected | Positive | 1500 |
| 606495-7 | 21.95 | 30.14 | Detected | Positive | 1500 |
| 606495-8 | 22.06 | 30.41 | Detected | Positive | 1500 |
| 606495-9 | 22.10 | 30.44 | Detected | Positive | 1500 |
| 606496-1 | 22.16 | 31.12 | Detected | Positive | 1500 |
| 606496-10 | 22.04 | 30.71 | Detected | Positive | 1500 |
| 606496-2 | 23.07 | 33.46 | Detected | Positive | 1500 |
| 606496-3 | 22.58 | 31.04 | Detected | Positive | 1500 |
| 606496-4 | 23.47 | 35.13 | Detected | Positive | 1500 |
| 606496-5 | 22.30 | 32.20 | Detected | Positive | 1500 |
| 606496-6 | 22.37 | 32.10 | Detected | Positive | 1500 |
| 606496-7 | 22.06 | 30.81 | Detected | Positive | 1500 |
| 606496-8 | 22.24 | 30.92 | Detected | Positive | 1500 |
| 606496-9 | 22.27 | 31.15 | Detected | Positive | 1500 |
| 606497-1 | 22.06 | 30.65 | Detected | Positive | 1500 |
| 606497-10 | 21.80 | 30.32 | Detected | Positive | 1500 |
| 606497-2 | 21.90 | 30.44 | Detected | Positive | 1500 |
| 606497-3 | 22.06 | 30.26 | Detected | Positive | 1500 |
| 606497-4 | 21.98 | 30.52 | Detected | Positive | 1500 |
| 606497-5 | 22.02 | 31.07 | Detected | Positive | 1500 |

| | | | | | |
|---|---|---|---|---|---|
| 606497-6 | 22.35 | 30.70 | Detected | Positive | 1500 |
| 606497-7 | 21.84 | 30.43 | Detected | Positive | 1500 |
| 606497-8 | 22.23 | 30.58 | Detected | Positive | 1500 |
| 606497-9 | 22.04 | 31.20 | Detected | Positive | 1500 |
| 606489-1 | 21.88 | 20.88 | Detected | Positive | 1312500 |
| 606489-10 | 21.91 | 20.71 | Detected | Positive | 1312500 |
| 606489-2 | 22.18 | 21.50 | Detected | Positive | 1312500 |
| 606489-3 | 21.71 | 20.73 | Detected | Positive | 1312500 |
| 606489-4 | 22.91 | 21.23 | Detected | Positive | 1312500 |
| 606489-5 | 21.67 | 20.88 | Detected | Positive | 1312500 |
| 606489-6 | 21.91 | 20.77 | Detected | Positive | 1312500 |
| 606489-7 | 21.80 | 20.78 | Detected | Positive | 1312500 |
| 606489-8 | 22.60 | 21.73 | Detected | Positive | 1312500 |
| 606489-9 | 21.87 | 20.78 | Detected | Positive | 1312500 |
| 606498-1 | 22.34 | 20.76 | Detected | Positive | 1312500 |
| 606498-10 | 22.44 | 20.86 | Detected | Positive | 1312500 |
| 606498-2 | 22.38 | 20.89 | Detected | Positive | 1312500 |
| 606498-3 | 22.33 | 20.82 | Detected | Positive | 1312500 |
| 606498-4 | 22.95 | 20.85 | Detected | Positive | 1312500 |
| 606498-5 | 22.15 | 20.73 | Detected | Positive | 1312500 |
| 606498-6 | 22.25 | 20.71 | Detected | Positive | 1312500 |
| 606498-7 | 22.30 | 20.67 | Detected | Positive | 1312500 |
| 606498-8 | 22.41 | 20.83 | Detected | Positive | 1312500 |
| 606498-9 | 22.43 | 20.97 | Detected | Positive | 1312500 |
| 606492-1 | 22.19 | Undetermined | Not Detected | Negative | 0 |
| 606492-10 | 21.84 | Undetermined | Not Detected | Negative | 0 |
| 606492-2 | 21.63 | Undetermined | Not Detected | Negative | 0 |
| 606492-3 | 22.50 | Undetermined | Not Detected | Negative | 0 |
| 606492-4 | 21.98 | Undetermined | Not Detected | Negative | 0 |
| 606492-5 | 21.62 | Undetermined | Not Detected | Negative | 0 |
| 606492-6 | 21.50 | Undetermined | Not Detected | Negative | 0 |
| 606492-7 | 21.63 | Undetermined | Not Detected | Negative | 0 |
| 606492-8 | 21.61 | Undetermined | Not Detected | Negative | 0 |
| 606492-9 | 22.15 | Undetermined | Not Detected | Negative | 0 |
| 606493-1 | 21.80 | Undetermined | Not Detected | Negative | 0 |
| 606493-10 | 22.65 | Undetermined | Not Detected | Negative | 0 |
| 606493-2 | 21.82 | Undetermined | Not Detected | Negative | 0 |
| 606493-3 | 21.32 | Undetermined | Not Detected | Negative | 0 |
| 606493-4 | 22.05 | Undetermined | Not Detected | Negative | 0 |

| | | | | | |
|---|---|---|---|---|---|
| 606493-5 | 21.62 | Undetermined | Not Detected | Negative | 0 |
| 606493-6 | 21.74 | Undetermined | Not Detected | Negative | 0 |
| 606493-7 | 22.00 | Undetermined | Not Detected | Negative | 0 |
| 606493-8 | 21.87 | Undetermined | Not Detected | Negative | 0 |
| 606493-9 | 22.09 | Undetermined | Not Detected | Negative | 0 |
| 606499-1 | 22.16 | Undetermined | Not Detected | Negative | 0 |
| 606499-10 | 22.13 | Undetermined | Not Detected | Negative | 0 |
| 606499-2 | 22.06 | Undetermined | Not Detected | Negative | 0 |
| 606499-3 | 22.10 | Undetermined | Not Detected | Negative | 0 |
| 606499-4 | 21.88 | Undetermined | Not Detected | Negative | 0 |
| 606499-5 | 22.07 | Undetermined | Not Detected | Negative | 0 |
| 606499-6 | 22.27 | Undetermined | Not Detected | Negative | 0 |
| 606499-7 | 22.32 | Undetermined | Not Detected | Negative | 0 |
| 606499-8 | 22.06 | Undetermined | Not Detected | Negative | 0 |
| 606499-9 | 22.46 | Undetermined | Not Detected | Negative | 0 |

**Day 3**

| IBX Sample | MS2 | COVID N/S | IBX Result | Expected Call | COVID copies/mL |
|---|---|---|---|---|---|
| 608813-1 | 23.0 | 34.5 | Detected | Positive | 1500 |
| 608813-10 | 22.5 | 29.8 | Detected | Positive | 1500 |
| 608813-2 | 22.6 | 33.4 | Detected | Positive | 1500 |
| 608813-3 | 22.8 | 34.5 | Detected | Positive | 1500 |
| 608813-4 | 22.3 | 33.1 | Detected | Positive | 1500 |
| 608813-5 | 22.4 | 32.5 | Detected | Positive | 1500 |
| 608813-6 | 22.2 | 32.7 | Detected | Positive | 1500 |
| 608813-7 | 22.6 | 33.1 | Detected | Positive | 1500 |
| 608813-8 | 22.3 | 33.6 | Detected | Positive | 1500 |
| 608813-9 | 22.3 | 34.0 | Detected | Positive | 1500 |
| 608815-1 | 22.0 | 33.7 | Detected | Positive | 1500 |
| 608815-10 | 22.7 | 29.7 | Detected | Positive | 1500 |
| 608815-2 | 22.3 | 35.3 | Detected | Positive | 1500 |
| 608815-3 | 22.9 | 35.1 | Detected | Positive | 1500 |
| 608815-4 | 22.7 | 32.0 | Detected | Positive | 1500 |
| 608815-5 | 22.1 | 32.9 | Detected | Positive | 1500 |
| 608815-6 | 22.5 | 34.3 | Detected | Positive | 1500 |
| 608815-7 | 22.0 | 33.7 | Detected | Positive | 1500 |
| 608815-8 | 22.8 | 34.4 | Detected | Positive | 1500 |
| 608815-9 | 22.7 | 34.3 | Detected | Positive | 1500 |
| 608821-1 | 23.8 | 32.6 | Detected | Positive | 1500 |

| | | | | | |
|---|---|---|---|---|---|
| 608821-10 | 22.9 | 34.0 | Detected | Positive | 1500 |
| 608821-2 | 22.2 | 23.5 | Detected | Positive | 1500 |
| 608821-3 | 23.5 | 28.6 | Detected | Positive | 1500 |
| 608821-4 | 22.9 | 29.7 | Detected | Positive | 1500 |
| 608821-5 | 22.6 | 31.5 | Detected | Positive | 1500 |
| 608821-6 | 22.1 | 32.4 | Detected | Positive | 1500 |
| 608821-7 | 23.1 | 34.2 | Detected | Positive | 1500 |
| 608821-8 | 22.8 | 32.9 | Detected | Positive | 1500 |
| 608821-9 | 22.1 | 32.6 | Detected | Positive | 1500 |
| 608811-1 | 22.6 | 33.7 | Detected | Positive | 1500 |
| 608811-10 | 22.4 | 33.1 | Detected | Positive | 1500 |
| 608811-2 | 22.3 | 33.2 | Detected | Positive | 1500 |
| 608811-3 | 21.9 | 26.9 | Detected | Positive | 1500 |
| 608811-4 | 21.7 | 30.3 | Detected | Positive | 1500 |
| 608811-5 | 22.4 | 32.6 | Detected | Positive | 1500 |
| 608811-6 | 22.2 | 32.0 | Detected | Positive | 1500 |
| 608811-7 | 22.3 | 32.8 | Detected | Positive | 1500 |
| 608811-8 | 22.8 | 32.6 | Detected | Positive | 1500 |
| 608811-9 | 22.8 | 34.2 | Detected | Positive | 1500 |
| 608818-1 | 22.4 | 26.5 | Detected | Positive | 1500 |
| 608818-10 | 22.8 | 33.5 | Detected | Positive | 1500 |
| 608818-2 | 22.6 | 27.6 | Detected | Positive | 1500 |
| 608818-3 | 22.5 | 29.4 | Detected | Positive | 1500 |
| 608818-4 | 23.2 | 32.4 | Detected | Positive | 1500 |
| 608818-5 | 22.6 | 29.8 | Detected | Positive | 1500 |
| 608818-6 | 22.5 | 28.7 | Detected | Positive | 1500 |
| 608818-7 | 22.8 | 34.0 | Detected | Positive | 1500 |
| 608818-8 | 22.4 | 33.7 | Detected | Positive | 1500 |
| 608818-9 | 22.8 | 33.2 | Detected | Positive | 1500 |
| 608819-1 | 23.7 | 34.8 | Detected | Positive | 1500 |
| 608819-10 | 22.6 | 31.8 | Detected | Positive | 1500 |
| 608819-2 | 23.3 | 34.4 | Detected | Positive | 1500 |
| 608819-3 | 23.2 | 35.0 | Detected | Positive | 1500 |
| 608819-4 | 22.2 | 32.6 | Detected | Positive | 1500 |
| 608819-5 | 23.2 | 34.1 | Detected | Positive | 1500 |
| 608819-6 | 24.3 | 38.0 | Detected | Positive | 1500 |
| 608819-7 | 22.0 | 32.9 | Detected | Positive | 1500 |
| 608819-8 | 22.0 | 32.3 | Detected | Positive | 1500 |
| 608819-9 | 22.3 | 31.7 | Detected | Positive | 1500 |

| | | | | | |
|---|---|---|---|---|---|
| 608820-1 | 22.8 | 30.7 | Detected | Positive | 1500 |
| 608820-10 | 22.1 | 29.8 | Detected | Positive | 1500 |
| 608820-2 | 22.6 | 31.3 | Detected | Positive | 1500 |
| 608820-3 | 23.5 | 33.2 | Detected | Positive | 1500 |
| 608820-4 | 22.7 | 32.6 | Detected | Positive | 1500 |
| 608820-5 | 22.9 | 32.6 | Detected | Positive | 1500 |
| 608820-6 | 23.1 | 24.9 | Detected | Positive | 1500 |
| 608820-7 | 22.8 | 32.7 | Detected | Positive | 1500 |
| 608820-8 | 23.3 | 32.4 | Detected | Positive | 1500 |
| 608820-9 | 23.2 | 30.6 | Detected | Positive | 1500 |
| 608814-1 | 23.4 | 23.6 | Detected | Positive | 1312500 |
| 608814-10 | 22.9 | 21.8 | Detected | Positive | 1312500 |
| 608814-2 | 22.9 | 21.6 | Detected | Positive | 1312500 |
| 608814-3 | 22.4 | 21.5 | Detected | Positive | 1312500 |
| 608814-4 | 22.7 | 22.9 | Detected | Positive | 1312500 |
| 608814-5 | 22.8 | 21.7 | Detected | Positive | 1312500 |
| 608814-6 | 23.1 | 21.6 | Detected | Positive | 1312500 |
| 608814-7 | 22.8 | 22.8 | Detected | Positive | 1312500 |
| 608814-8 | 25.3 | 26.8 | Detected | Positive | 1312500 |
| 608814-9 | 22.9 | 22.7 | Detected | Positive | 1312500 |
| 608822-1 | 22.6 | 22.1 | Detected | Positive | 1312500 |
| 608822-10 | 23.4 | 22.6 | Detected | Positive | 1312500 |
| 608822-2 | 22.8 | 22.7 | Detected | Positive | 1312500 |
| 608822-3 | 22.5 | 22.4 | Detected | Positive | 1312500 |
| 608822-4 | 22.3 | 21.8 | Detected | Positive | 1312500 |
| 608822-5 | 22.4 | 21.6 | Detected | Positive | 1312500 |
| 608822-6 | 22.4 | 21.4 | Detected | Positive | 1312500 |
| 608822-7 | 23.2 | 21.6 | Detected | Positive | 1312500 |
| 608822-8 | 22.0 | 20.7 | Detected | Positive | 1312500 |
| 608822-9 | 23.0 | 22.2 | Detected | Positive | 1312500 |
| 608816-1 | 23.5 | Undetermined | Not Detected | Negative | 0 |
| 608816-10 | 23.3 | Undetermined | Not Detected | Negative | 0 |
| 608816-2 | 23.6 | Undetermined | Not Detected | Negative | 0 |
| 608816-3 | 23.7 | Undetermined | Not Detected | Negative | 0 |
| 608816-4 | 22.8 | Undetermined | Not Detected | Negative | 0 |
| 608816-5 | 23.4 | Undetermined | Not Detected | Negative | 0 |
| 608816-6 | 23.5 | Undetermined | Not Detected | Negative | 0 |
| 608816-7 | 23.2 | Undetermined | Not Detected | Negative | 0 |
| 608816-8 | 22.8 | Undetermined | Not Detected | Negative | 0 |

| | | | | | |
|---|---|---|---|---|---|
| 608816-9 | 22.7 | Undetermined | Not Detected | Negative | 0 |
| 608812-1 | 22.6 | Undetermined | Not Detected | Negative | 0 |
| 608812-10 | 22.4 | Undetermined | Not Detected | Negative | 0 |
| 608812-2 | 22.7 | Undetermined | Not Detected | Negative | 0 |
| 608812-3 | 22.4 | Undetermined | Not Detected | Negative | 0 |
| 608812-4 | 22.6 | Undetermined | Not Detected | Negative | 0 |
| 608812-5 | 22.2 | Undetermined | Not Detected | Negative | 0 |
| 608812-6 | 22.7 | Undetermined | Not Detected | Negative | 0 |
| 608812-7 | 22.8 | Undetermined | Not Detected | Negative | 0 |
| 608812-8 | 22.9 | Undetermined | Not Detected | Negative | 0 |
| 608812-9 | 22.6 | Undetermined | Not Detected | Negative | 0 |
| 608817-1 | 22.4 | Undetermined | Not Detected | Negative | 0 |
| 608817-10 | 22.3 | Undetermined | Not Detected | Negative | 0 |
| 608817-2 | 22.6 | Undetermined | Not Detected | Negative | 0 |
| 608817-3 | 22.1 | Undetermined | Not Detected | Negative | 0 |
| 608817-4 | 22.7 | Undetermined | Not Detected | Negative | 0 |
| 608817-5 | 21.5 | Undetermined | Not Detected | Negative | 0 |
| 608817-6 | 25.0 | Undetermined | Not Detected | Negative | 0 |
| 608817-7 | 22.5 | Undetermined | Not Detected | Negative | 0 |
| 608817-8 | 23.7 | Undetermined | Not Detected | Negative | 0 |
| 608817-9 | 22.0 | Undetermined | Not Detected | Negative | 0 |

## REVISION HISTORY

**Version 01 Effective on 19-Feb-2021**
New Document

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Minor update to language

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Document category changed

## DOCUMENT ELECTRONIC SIGNATURES

### DOCUMENT APPROVAL WORKFLOW

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**Required Workflow Steps for this Category**

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