CLIA Waiver by Application Approval Determination

Decision Summary

A. **Document Number**

CW220014

B. Parent Document Number

K223591

C. CLIA Waiver Type:

Dual 510(k) and CLIA Waiver by Application (Dual Submission)

D. Applicant

Roche Molecular Systems, Inc.

E. Proprietary and Established Names

cobas SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System

F. Measurand (analyte)

The **cobas** SARS-CoV-2 & Influenza A/B test detects SARS-CoV-2, influenza A and influenza B virus RNA isolated from nasopharyngeal swab and nasal swab specimens from patients with signs and symptoms of respiratory viral infection.

G. Sample Type(s)

Nasopharyngeal Swabs (NPS) and Nasal swabs (NS)

H. Type of Test

This assay is a multiplex nucleic acid assay for the qualitative detection and differentiation of SARS-CoV-2, influenza A and influenza B RNA through nucleic acid extraction, amplification, and detection using real-time RT-PCR. All steps of the assay are automated within the **cobas** Liat System, after scanning the specimen ID barcode, scanning the assay tube barcode, and the manual addition of sample into the assay tube**.**

I. Test System Description

1. Overview

The **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System **(cobas** SARS-CoV-2 & Influenza A/B) is a rapid, automated in vitro diagnostic test for the qualitative detection of SARS-CoV-2, influenza A, and influenza B RNA in nasopharyngeal swab (NPS) and nasal swab (NS) specimens eluted in viral transport media.

The assay targets both the ORF1 a/b non-structural region and nucleocapsid protein gene that are unique to SARS-CoV-2, a well-conserved region of the matrix gene of influenza A, and the non-structural protein gene of influenza B. An Internal Process Control (IPC) is also included. The IPC is present to control for adequate processing of the target viruses and to monitor the presence of inhibitors in the sample preparation and RT-PCR.

The assay utilizes a single-use disposable **cobas** assay tube that holds the sample purification and PCR reagents and hosts the sample preparation and PCR processes. The **cobas** assay tube uses a flexible tube as a sample vessel. It contains all required unit dose reagents pre-packed in tube segments, separated by peelable seals, in the order of reagent use.

The **cobas** SARS-CoV-2 & Influenza A/B assay uses silica magnetic particle-based nucleic acid extraction and TaqMan probe-based real-time PCR amplification and detection. The **cobas** Liat Analyzer automates and integrates sample purification, nucleic acid amplification, and detection of the target sequence in biological samples. During the testing process, multiple sample processing actuators of the **cobas** Liat Analyzer compress the **cobas** Liat Tube to selectively release reagents from tube segments, move the sample from one segment to another, and control reaction volume, temperature, and incubation time. The **cobas** Liat Analyzer software controls and coordinates these actions to perform all required assay processes, including sample preparation, nucleic acid extraction, target enrichment, inhibitor removal, nucleic acid elution, and real-time PCR. All assay steps are performed within the closed and self-contained **cobas** SARS-CoV-2 & Influenza A/B assay tube.

The **cobas** Liat System was originally categorized as “waived” under K141338/CW140014 for the **cobas** Strep A Assay and two additional CLIA-waived assays have subsequently been implemented on the same instrument system (**Table 1**). The **cobas** SARS-CoV-2 & Influenza A/B assay is a modification of the cobas Influenza A/B & RSV Nucleic acid test for use on the cobas Liat System previously cleared under K153544/CLIA Waived under

CW150018. The modifications included the addition of the SARS-CoV-2 reagents. Therefore, the original analytical and clinical studies of the **cobas** Influenza A/B & RSV assay remain relevant for the performance of influenza A/B targets in the **cobas** SARS-CoV-2 & Influenza A/B assay.

Table 1. Previously CLIA Waived tests for use with the **cobas** Liat System

510(k) Number	CLIA Waiver	Device Name	Effective Date
K141338	CW140014	Liat Strep A Assay	05/15/2015
K111387	CW150013	Cobas Liat Influenza A/B Assay	09/18/2015
K153544	CW150018	Cobas Influenza A/B & RSV nucleic acid test for use on the cobas Liat System	07/25/2016

2. Test System Components

   a. The **cobas** SARS-CoV-2 & Influenza A/B kit includes:
      - 20 **cobas** SARS-CoV-2 & Influenza A/B test assay tubes (P/N 09211101190)
      - 2 **cobas** transfer pipette pack (12 pipettes/pack- P/N 9329676001)
      - 1 Package Insert Barcode card

   b. The **cobas** SARS-CoV-2 & Influenza A/B Quality Control Kit (P/N 09211128190) is provided separately, and includes:
      - **cobas** Influenza A/B Positive Control (3 X 10 µL)
      - **cobas** SARS-CoV-2 Positive Control (3 X 0.25 mL)
      - **cobas** Dilution UTM (negative control) (3 X 0.3 mL)
      - 11 transfer pipettes
      - 1 Control Kit Barcode

J. Demonstrating "Simple"

- The **cobas** Liat System automates all nucleic acid test (NAT) processes, including reagent preparation, target enrichment, inhibitor removal, nucleic acid extraction, amplification, real-time detection, and result interpretation in a rapid manner.
- The assay utilizes NPS and NS specimens collected in VTM/UTM or 0.9% saline, without the need for any specimen manipulation. When the sample is added to the sample segment of the assay tube, the tube is capped and remains closed for the entire test process. No further materials need to be added or removed from the tube. This approach avoids cross contamination, reduces biohazard risks, and helps preserve sample integrity.
- An untrained operator can conduct the test by performing four simple steps: transfer liquid sample to the assay tube with fixed volume pipette, 2) scan the barcodes on the assay tube and sample ID, 3) run the test on the **cobas** Liat System, and 4) read the results.

- Running the assay requires no reagent manipulation. The assay tube uses a flexible assay tube as a sample processing vessel that contains all assay reagents pre-packed in tube segments separated by seals. The assay tube can only be inserted in the **cobas** Liat Analyzer in one direction.
- The test does not require any operator intervention during the analysis step.
- Technical or specialized training is not required for troubleshooting or error code interpretation. If an error code is shown, simple on-screen instructions are provided to the operator.
- The system requires no electronic or mechanical maintenance tasks by the operator. The analyzer performs self-diagnostics during startup (initialization) and utilizes an advanced error diagnostics system to monitor the analyzer's performance during an assay. Under normal operation, the analyzer alerts the operator if a malfunction or error is detected. The analyzer requires no adjustment or calibration from the operator.
- The **cobas** Liat System performs automated analysis of test results and eliminates subjectivity associated with visual reading of results by the end-user.
- Results are reported on the **cobas** Liat System in English as "Detected", "Not Detected", "Invalid" for each target, or "Aborted", and requires no operator calibration, interpretation, or calculation.
- The Quick Reference instructions are written at a 7th grade comprehension level. In studies in which intended operators (i.e., test operator with limited or no training or hands-on experience in conducting clinical laboratory testing) performed the assay, the **cobas** SARS-CoV-2 & Influenza A/B demonstrated test performance comparable to the predicate device.

K. Demonstrating "Insignificant Risk of an Erroneous Result"- Failure Alerts and Fail-safe Mechanisms

1. Risk Analysis

   Risk analysis was performed by the firm according to the principles of risk minimization as found in the standard EN ISO 14971 *Medical Devices – Application of risk management to medical devices*. The detailed analysis was provided during Interactive Review. Potential sources of errors that could adversely affect system performance were identified and mitigated first through system design and then through additional cautions in the labeling. All risks of harm to the patient or operator were mitigated to an acceptable level and were supported by flex studies and/or operator instructions.

2. Fail-Safe and Failure Alert Mechanisms

   The **cobas** Liat System is designed with a variety of fail-safe and/or failure alert mechanisms to prevent operator and instrument error. These mechanisms were originally validated as being effective in reducing the likelihood of error to broadly acceptable levels under CW140014 for the **cobas** Strep A assay. Subsequently, two additional CLIA-waived assays have been implemented on the **cobas** Liat System (**Table 1** above) and the **cobas** SARS-CoV-2 & Influenza A/B assay has also been authorized for emergency use in a point-of-care setting. Under the current submission (K223591/CW220014), the

sponsor did not describe any changes to the fail-safe and lock-out features associated with the **cobas** Liat System (**Table 2**).

Table 2. Fail-safe and failure alert mechanisms the **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System cited by the manufacturer

#	Description
1	Operator permissions restrict access to specific instrument functions/test capabilities
2	On-screen instructions
3	Use of barcodes to automate assay selection and provide traceability
4	Lock-out feature to prevent use of expired or previously used reagents
5	Controls to ensure correct tube insertion
6	Sample volume detection
7	Error diagnostic and recovery to prevent out-of-specification operation
8	System self-checks for instrument subsystems
9	Auto-calibration and monitoring
10	Automated data acquisition, analysis, and result interpretation
11	Internal Process Control to monitor the test procedure for each individual sample
12	Mandatory "Add Lot" procedure for new reagent lots

3. Flex Studies

Due to the similarities between the **cobas** SARS-CoV-2 & Influenza A/B test and the cleared and CLIA waived **cobas** Influenza A/B & RSV test, FDA has agreed to leverage data from flex studies originally performed for **cobas** Influenza A/B & RSV Nucleic acid test for use on the **cobas** Liat System (originally cleared under K153544 and CLIA Waived under CW150018). For both assays, the **cobas** Liat Assay Tube and **cobas** Liat System are the same with regard to operational robustness, the assays utilize the same specimen type (swabs collected in "liquids"), sample volumes and sample processing. The flex studies that were previously performed with **cobas** Influenza A/B & RSV, that are applicable to **cobas** SARS-CoV-2 & Influenza A/B are described in **Table 3.**

Table 3. Flex Studies Demonstrating Insignificant Risk of an Erroneous Result – Failure Alerts and Fail-safe Mechanisms

Flex Study	Demonstrated for cobas Influenza A/B & RSV	Studies leveraged from existing tests on the cobas Liat System and not conducted for cobas SARS-CoV-2 & Influenza A/B
Operating Temperature	Yes	These studies were not performed since it has been already established that the system is robust under the variable range of these environmental conditions (CW140014, CW150003, and CW150018). These control measurements are system specific and not related to assay chemistry
Humidity	Yes	
Altitude/Pressure	Yes	
Tilt Testing	Yes	
Movement of Liat Analyzers	Yes	
Bubbles with Sample	Yes	cobas Influenza A/B & RSV and cobas SARS-CoV-2 & Influenza A/B use the same sample types, therefore the impact of bubbles in sample established in cobas Influenza A/B & RSV should apply to cobas SARS-CoV-2 & Influenza A/B
Assay Tube orientation post sample addition	Yes	The assay tube configuration, workflow, and chemistry are the same as cobas Influenza A/B & RSV, therefore same outcome is expected
Equilibrate temperature of reagent and sample prior to run	Yes	The assay workflow, reagent formulation and assay chemistry are the same as cobas Influenza A/B & RSV, therefore the same outcome is expected.
Incorrect sample input volume	Yes	The formulation and workflow are the same as cobas Influenza A/B & RSV, especially sample input volume, sample preparation reagents, nucleic acid extraction and capture steps, therefore the same outcome is expected
Tube Seal Break	Yes	The assay tube configuration, fill volume, reagent formulation and assay chemistry are the same as cobas Influenza A/B & RSV, therefore the same outcome is expected.
Improper Tube storage	Yes	The assay tube configuration/packaging, reagent formulation and assay chemistry are the same as for cobas Influenza A/B & RSV, therefore the same outcome is expected.

The above analytical flex studies results were applied to validate the insensitivity of the test system to variation under stress conditions (Tier 1) and verify and/or validate the effectiveness of control measures at operational limits (Tier 2). Stress conditions tested include operating temperature, humidity, altitude / barometric pressure, operating on non-level surface, specimen storage, **cobas** Liat Tube storage, **cobas** Liat Tube seal breakage, input sample volume, presence of bubbles within the specimen, **cobas** Liat Tube hold time between sample addition and initiation of the test, and movement of the **cobas** Liat System during an assay run. The study results demonstrated that the test is insensitive to the stresses of environmental conditions and potential user errors. Flex studies for previously FDA cleared and CLIA Waived tests for use on the **cobas** Liat System are described in CW Decision Summaries CW140014, CW150013, and CW150018, which support CLIA Waiver for the **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System.

Additional flex studies, deemed as assay specific, were performed to evaluate the robustness of the **cobas** Liat System and **cobas** SARS-CoV-2 & Influenza A/B reagents to variations in workflow and control effectiveness that may reasonably be expected to occur with untrained operators in the intended use CLIA Waived setting. Test conditions were designed based on a risk analysis of the complete test system and included conditions intended to verify the effectiveness of built in controls.

Flex studies performed for the **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System included: (1) Evaluation of Assay tube hold time, (2) Internal Processing Control (IPC) Effectiveness, and (3) External Control Effectiveness. These studies and the results are described below.

a. Evaluation of Assay Tube Hold Time/ On-Board Stability

The insensitivity of the **cobas** SARS-CoV-2 & Influenza A/B test to variation of hold time between addition of the sample to the **cobas** SARS-CoV-2 & Influenza A/B Assay tube and initiation of the run on the **cobas** Liat Analyzer was evaluated. To perform these studies, co-formulated panel consisting of ~3x LoD concentrations of inactivated SARS-CoV-2 (USA-WA1/2020, catalog number 0810587CFHI, ZeptoMetrix, NY), influenza A (Brisbane/59/07, catalog number 0810244CF, ZeptoMetrix, NY), and influenza B (Florida/04/06, catalog number 0810255CF; ZeptoMetrix, NY) viral cultures in pooled negative clinical nasopharyngeal swab specimens (NPS) collected in Universal Transport Media was used. The samples were added to the assay tube and run initiated immediately, and after being stored at room temperature (25ºC) for 2, 4, and 6 hours. For each condition, five (5) replicates of negative nasopharyngeal swab specimens (NPS) and five (5) replicates of negative nasal swab specimens (NS) collected in Universal Transport Media (UTM) and five (5) replicates of co-formulated panel consisting of ~3x LoD of influenza A, influenza B and SARS-CoV-2 viral cultures spiked in negative NPS and NS each were tested.

At ~3x LoD, all five (5) replicates of positive samples for each condition, each sample type (NPS & NS) were valid and were positive for influenza A, influenza B and SARS-CoV-2. All five (5) replicates of samples for each condition, each sample type (NPS & NS) containing no target were valid and negative. The results are summarized in **Table 4**.

Table 4. Assay Tube Hold Results

Hold Time (Hour)	Test Condition	Positive Sample Hit Rate (~3x LoD)		Negative Sample Hit Rate	
		NPS	NS	NPS	NS
0	Add sample to Liat tube and run immediately (Control)	100% (5/5)	100% (5/5)	0% (0/5)	0% (0/5)
2	Run after 120 minutes	100% (5/5)	100% (5/5)	0% (0/5)	0% (0/5)
4	Run after 240 minutes	100% (5/5)	100% (5/5)	0% (0/5)	0% (0/5)
6	Run after 360 minutes	100% (5/5)	100% (5/5)	0% (0/5)	0% (0/5)

NPS= Pooled negative nasopharyngeal swab matrix, NS= Pooled negative nasal swab matrix
* At time point 0 hour; samples were added to all Liat tubes for all time points/test conditions (0, 2, 4 and 6 hours). Hit rate refers to the number of positive replicates divided by the total number of replicates tested. 100% Hit rate observed for influenza A, influenza B & SARS-COV-2 channels for ~3x spiked positive samples & 0% hit rate in all three channels for negative samples.

These results demonstrate **cobas** Liat SARS-CoV-2 & Influenza A/B Test can tolerate hold time between addition of the sample to the assay tube and initiation of the run on the **cobas** Liat Analyzer for up to 6 hours at room temperature (25°C).

b. Internal Processing Control (IPC) Effectiveness

This study evaluated the effectiveness of the Internal Process Control (IPC) to monitor the performance of the **cobas** SARS-CoV-2 & Influenza A/B test sample processing and PCR amplification/detection under simulated process and reagent failures. To perform these studies, co-formulated panel consisting of ~3x LoD concentrations of inactivated SARS-CoV-2 (USA-WA1/2020, catalog number 0810587CFHI, ZeptoMetrix, NY), influenza A (Brisbane/59/07, catalog number 0810244CF, ZeptoMetrix, NY), and influenza B (Florida/04/06, catalog number 0810255CF; ZeptoMetrix, NY) viral cultures in pooled negative clinical nasopharyngeal swab specimens (NPS) collected in Universal Transport Media was used. For each condition, five (5) replicates of negative nasopharyngeal swab specimens (NPS) collected in Universal Transport Media (UTM) and five (5) replicates of co-formulated panel consisting of ~3x LoD of influenza A, influenza B and SARS-CoV-2 viral cultures spiked in negative NPS were tested for each condition (Table 5).

Table 5. Test Condition for IPC Effectiveness

Test Samples	Performance of	Failure Conditions	Replicates
SARS-CoV-2 and Influenza A/B Negative Sample (Negative NPS) and Contrived Positive Sample (3x LoD targets in Negative NPS)	Control	Normal Conditions	5
	Systematic Error in Sample Preparation	Process Failure: Failure to capture magnetic beads during nucleic acid extraction	5
	Assay Tube Lot in Sample Preparation	Reagent Failure: Break off frangible seal between the assay tube Sample Preparation segments	5
	Systematic Error in PCR amplification and detection	Process Failure: Deviation in PCR temperature	5
	Assay Tube Lot in PCR amplification and detection	Reagent Failure: Break off frangible seal between the assay tube segments PCR	5

Results indicate that under failure mode testing conditions, both positive and negative test samples yielded invalid results when tested using **cobas** SARS-CoV-2 & Influenza A/B. Therefore, the IPC appears to be effective in monitoring the performance of sample processing and PCR amplification/detection failure conditions. The results are summarized in **Table 6**.

Table 6. IPC Effectiveness Results

Test Sample	Testing Condition	Hit Rate for Target Results	Flu A Result	Flu B Result	SARS-CoV-2 Result	IPC Result
Negative Sample (Negative NPS)	Control	100% (5/5)	Not Detected	Not Detected	Not Detected	Detected
	Sample Prep Process Failure	100% (5/5)	Invalid	Invalid	Invalid	Invalid
	Sample Prep Reagent Failure	100% (5/5)	Invalid	Invalid	Invalid	Invalid
	PCR Process Failure	100% (5/5)	Invalid	Invalid	Invalid	Invalid
	PCR Reagent Failure	100% (5/5)	Invalid	Invalid	Invalid	Invalid
Contrived Positive Sample (3x LoD targets in Negative NPS)	Control	100% (5/5)	Detected	Detected	Detected	Detected
	Sample Prep Process Failure	100% (5/5)	Invalid	Invalid	Invalid	Invalid
	Sample Prep Reagent Failure	100% (5/5)	Invalid	Invalid	Invalid	Invalid
	PCR Process Failure	100% (5/5)	Invalid	Invalid	Invalid	Invalid
	PCR Reagent Failure	100% (5/5)	Invalid	Invalid	Invalid	Invalid

c. External Control Effectiveness

This study evaluated the external positive and negative control results under the cobas SARS-CoV-2 & Influenza A/B assay process and reagent/assay tube failure conditions in order to substantiate the ability of Positive Controls (PC) and Negative Controls (NC) to monitor such processes. To demonstrate the ability of PC and NC to monitor the performance of sample processing and PCR amplification/detection, the conditions and replicates shown in **Table 7** were tested.

Table 7. Test Conditions for External Control Effectiveness

Test Samples	Performance of	Failure Conditions	Replicates
SARS-CoV-2 and Influenza A/B Positive and Negative Control	Control	Normal Conditions	5
	Systematic Error in Sample Preparation	Process Failure: Failure to capture magnetic beads during nucleic acid extraction	5
	Assay Tube Lot in Sample Preparation	Reagent Failure: Break off frangible seal between the assay tube Sample Preparation segments	5
	Systematic Error in PCR amplification and detection	Process Failure: Deviation in PCR temperature	5
	Assay Tube Lot in PCR amplification and detection	Reagent Failure: Break off frangible seal between the assay tube PCR segments	5
	NA	Run NC to simulate low level contamination	5

The NC & PC both failed under the reagent and process failure modes tested (**Table 8**). All runs with the simulated failure conditions yielded invalid results for the targets and Internal Control (IC) channels for all five replicates, thus demonstrating that the Positive and Negative control are effective in monitoring the process and reagent/assay tube failure conditions.

Table 8. External Control Effectiveness Results

Test Sample	Testing Condition	Report Target Results	Hit Rate for Target Results			
			Flu A Result	Flu B Result	SARS-CoV-2 Result	IPC Result
Negative Control (NC)	Control	Valid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)
	Sample Prep Process Failure	Invalid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)
	Sample Prep Reagent Failure	Invalid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)
	PCR Process Failure	Invalid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)
	PCR Reagent Failure	Invalid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)
	Contaminated NC	Invalid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)
Positive Control	Control	Valid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)
	Sample Prep Process Failure	Invalid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)
	Sample Prep Reagent Failure	Invalid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)
	PCR Process Failure	Invalid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)
	PCR Reagent Failure	Invalid	100% (5/5)	100% (5/5)	100% (5/5)	100% (5/5)

Overall, based on flex studies previously performed for the **cobas** Liat System and currently performed for the **cobas** SARS-CoV-2 & Influenza A/B test specifically, the **cobas** SARS-CoV-2 & Influenza A/B test is robust to foreseeable user-dependent variations in the assay workflow and that built-in assay controls and fail-safe and/or failure alert mechanisms are effective in preventing the generation of erroneous results due to operator error and/or use of the **cobas** Liat System outside the specified operating environmental conditions.

L. Demonstrating "Insignificant Risk of an Erroneous Result" –Accuracy

1. Comparison Study

a. Study Design

i. Study Sites and Duration

Clinical performance characteristics of the **cobas** SARS-CoV-2 & Influenza A/B test were evaluated in a multi-site prospective study during the 2021-2022 influenza season in the U.S. Ten (10) sites throughout the U.S. participated in the clinical study. The sites consisted of emergency rooms/ urgent care clinics, outpatient clinics, physician's offices and drive through COVID-19 testing sites. All the sites qualified as representative of CLIA waived intended use sites for this device.

The prospective clinical study was supplemented by additional testing at four of the U.S. sites that was performed on frozen, archived specimens obtained from various clinical laboratories distributed worldwide. These specimens were selected for inclusion in the study based on their known microbial content as determined by the source laboratory, and were confirmed using the same comparator method used for the prospectively collected specimens.

ii. Operators

There were a total of 30 operators representative of intended CLIA waived users across the ten clinical testing sites, with 2 to 5 operators per site. The participants consisted of research/medical assistants or managers, nurses, research/study coordinators, phlebotomists, and other patient care providers. The test operators who participated in the study were untrained in the use of the **cobas** SARS-CoV-2 & Influenza A/B test and none were trained laboratory technicians.

Of the 30 operators who participated in the prospective clinical study, 17 (56.7%) processed at least 5 nasopharyngeal or nasal samples that were positive and 5 that were negative, as determined by the applicable and relevant comparator methods.

iii. Instructions for Use

The operators were provided the **cobas** SARS-CoV-2 & Influenza A/B Instructions for Use (IFU), the Quick Reference Instructions, and the **cobas** Liat System User Guide. No other materials or instructions were provided and the operators received no training in the use of the test.

iv. Subjects (Patients)

Specimens analyzed for the prospective clinical study were collected under informed consent (if required by the local Institutional Review Board), or, if the subject was < 18 years of age, with parental permission and assent. The Inclusion Criteria for the study analysis population were as follows:

Subjects who:

- Presented with signs/symptoms of a respiratory infection including but not limited to fever, cough, sore throat, runny nose, myalgia, headache, chills, or fatigue.
- Were willing and able to provide a nasopharyngeal swab and a nasal swab specimen.

The Exclusion Criteria for the study were as follows:

- Prior enrollment in this study.
- Not exhibiting signs/ symptoms of respiratory tract infection.
- Nasal or nasopharyngeal cavity sampling other than study sample sampling occurring on the same day as study sample collection, except for SOC sampling (if collected).
- Receipt of any formulation of antiviral medication in the preceding 7 days, including but not limited to rimantadine (Flumadine), amantadine (Gocovri), oseltamivir (Tamiflu), zanamivir (Relenza), peramivir (Rapivab), baloxavir marboxil (Xofluza), ribavirin (Copegus, Rebetol, Ribasphere, and Virazole), and remdesivir (GS-5734).
- Receipt of topical mupirocin in the preceding 7 days.
- Contraindication to nasopharyngeal cavity sampling as performed according to the clinical site policies and procedures.
- Receipt of influenza vaccination that was administered through the nasopharynx (e.g., FluMist) within the preceding 6 weeks.
- If collected, SOC specimen collection performed same day with both nostrils (either nasopharyngeal or nasal sampling).
- Same-day nasopharyngeal or nasal aspirate and nasopharyngeal or nasal wash performed.

v. Samples

The clinical performance of the **cobas** SARS-CoV-2 & Influenza A/B Nucleic Acid test for use on the **cobas** Liat System was evaluated using a combination of prospectively collected and archived specimens as described below.

Prospectively Collected Specimens
One nasopharyngeal swab (NPS) and one nasal swab (NS) were collected from each subject using standard collection methods and each eluted in UTM. Prospectively collected NPS and NS specimens were tested fresh at the clinical site with **cobas** SARS-CoV-2 & Influenza A/B by the intended use operators following the product IFU and/or Quick Reference Instructions. After investigational assay testing, the samples were shipped to the reference testing laboratory, where they were tested on the comparator methods following the products' IFUs.

Prospective clinical specimens were collected and tested from February–June 2022. In total, prospectively collected paired NPS and NS specimens from 640 evaluable individuals were included in the analysis population for the evaluation of **cobas** SARS-CoV-2 & Influenza A/B. Of these, 24 NPS and 24 NS samples were excluded or non-evaluable. For NPS, 13 NPS specimens had no comparator results due to incidents (11) or were missing or not tested (2), and 11 NPS specimen results from **cobas** SARS-CoV-2 & Influenza A/B were non-evaluable due to protocol deviation (8), not tested (1), or invalid results (2). For NS, 11 NS specimens had no comparator results due to incidents (9) or were missing/not tested (2), and 13 NS specimen results from **cobas** SARS-CoV-2 & Influenza A/B were non-evaluable due to protocol deviation (8) or invalid results (5). The remaining 616 prospective NPS and NS samples were included for analysis.

No coinfections with SARS-CoV-2 and influenza A/B were detected by the comparator method. No prospective specimens tested in this performance evaluation were influenza B positive by the comparator method.

Retrospective Specimens
To supplement the prospective data for influenza A and influenza B, retrospective frozen positive and negative NPS (n=178) and NS (n=190) specimens prospectively obtained during the 2013-2014, 2014-2015, and 2019-2020 flu seasons and during the COVID-19 pandemic (March–June 2021) were distributed to 4 of the 10 sites and worked into the daily workflow of sites for testing. Subject demographic data was not available for the retrospective samples.

Of the 178 retrospective NPS specimens (44 influenza A positive, 22 influenza B-positive, and 112 negative) that were tested at sites, two retrospective NPS samples were non-evaluable due to obtaining invalid results from the comparator device, and three obtained invalid results for influenza B with the candidate device, leaving 176 evaluable retrospective NPS samples for influenza A and 173 for influenza B. Of the 190 retrospective NS specimens (37 influenza A-positive, 35 influenza B-positive, and 118 negative) that were tested at sites, three retrospective NS samples were non-evaluable due to obtaining invalid results from the comparator device, and one was aborted by the candidate device, leaving 186 evaluable retrospective NS samples for influenza A and influenza B.

vi. Comparative Method (CM)

A composite comparator method (CCM) consisting of three highly sensitive EUA authorized molecular RT-PCR assays was used as the comparator method for demonstrating the performance accuracy of SARS-CoV-2. The composite comparator results were defined as concordant results from two of the three comparator assays.

An acceptable molecular assay for influenza was used as the comparator method for demonstrating the performance accuracy of influenza A and influenza B targets in support of the CLIA waiver.

b. Results and Analysis

i. Statistical Analysis of Clinical Performance for NPS Specimens

As shown in **Table 9** for prospective symptomatic subjects, 101 NPS specimens tested positive for SARS-CoV-2 with both the **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System and the composite comparator; five SARS-CoV-2-composite comparator positive specimens tested negative for SARS-CoV-2 with the **cobas** SARS-CoV-2 & Influenza A/B test. A total of 507 NPS specimens tested negative for SARS-CoV-2 with both the **cobas** SARS-CoV-2 & Influenza A/B test and the composite comparator; three SARS-CoV-2 composite comparator negative specimens tested positive for SARS-CoV-2 with the **cobas** SARS-CoV-2 & Influenza A/B test. All discordant SARS-CoV-2 results showed late Ct values, which are indicative of NPS specimens from individuals with viral loads near or below the limit of detection of both **cobas** SARS-CoV-2 & Influenza A/B and the composite comparator methods.

For SARS-CoV-2, the results of the clinical performance evaluation using NPS specimens from prospective symptomatic subjects demonstrated 95.3% positive percent agreement (PPA) (101/106; 95% score CI: 89.4% - 98.0%) and 99.4% negative percent agreement (NPA) (507/510; 95% score CI: 98.3% - 99.8%) as compared to the composite comparator method.

Table 9. Clinical performance comparison – SARS-CoV-2 for prospective NPS specimens

		Composite Comparator Method SARS-CoV-2 Result	
		Positive	Negative
cobas SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System **Nasopharyngeal Swab (NPS)**	Positive	101	3
	Negative	5	507

PPA	95.3% (95% CI: 89.4% - 98.0%)
NPA	99.4% (95% CI: 98.3% - 99.8%)

As shown in **Table 10** for prospective symptomatic subjects, 18 NPS specimens tested positive for influenza A with both the **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System and the comparator assay; one influenza A comparator positive specimen tested negative for influenza A with the **cobas** SARS-CoV-2 & Influenza A/B test. A total of 595 NPS specimens tested negative for influenza A with both the **cobas** SARS-CoV-2 & Influenza A/B test and the comparator assay; two influenza A comparator negative specimens tested positive for influenza A with the **cobas** SARS-CoV-2 & Influenza A/B test.

For influenza A, the results of the clinical performance evaluation using NPS specimens from prospective symptomatic subjects demonstrated 94.7% PPA (18/19; 95% score CI: 75.4% - 99.1%) and 99.7% NPA (595/597; 95% score CI: 98.8% – 99.9%) as compared to the comparator method.

Table 10. Clinical performance comparison – Influenza A for prospective NPS specimens

		Comparator Method Influenza A Result	
		Positive	Negative
cobas SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System **Nasopharyngeal Swab (NPS)**	Positive	18	2
	Negative	1	595

PPA	94.7% (95% CI: 75.4% - 99.1%)
NPA	99.7% (95% CI: 98.8% - 99.9%)

As shown in **Table 11** for retrospective NPS specimens, the results of the clinical performance evaluation for influenza A demonstrated 97.7% PPA (43/44; 95% score CI: 88.2% - 99.6%) and 99.2% NPA (131/132; 95% score CI: 95.8% – 99.9%) as compared to the comparator method.

Table 11: Clinical performance comparison – Influenza A for retrospective NPS specimens

		Comparator Method Influenza A Result	
		Positive	Negative
	Positive	43	1

cobas SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System **Nasopharyngeal Swab (NPS)**	Negative	1	131

PPA	97.7% (95% CI: 88.2% - 99.6%)
NPA	99.2% (95% CI: 95.8% - 99.9%)

As shown in **Table** 12 for retrospective NPS specimens, the results of the clinical performance evaluation for influenza B demonstrated 100.0% PPA (22/22; 95% score CI: 85.1% - 100.0%) and 100.0% NPA (151/151; 95% score CI: 97.5% - 100.0%) as compared to the comparator method.

For prospective symptomatic subjects, PPA was not calculable because no fresh specimens were influenza B-positive by the comparator method. For influenza B, the results of the clinical performance evaluation using NPS specimens from prospective symptomatic subjects demonstrated 100.0% NPA (616/616; 95% score CI: 99.4% – 100.0%) as compared to the comparator method.

Table 12. Clinical performance comparison – Influenza B for retrospective NPS specimens

		Comparator Method Influenza B Result	
		Positive	Negative
cobas SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System **Nasopharyngeal Swab (NPS)**	Positive	22	0
	Negative	0	151

PPA	100.0% (95% CI: 85.1% - 100.0%)
NPA	100.0% (95% CI: 97.5% - 100.0%)

ii. Statistical Analysis of Comparison Study Results for NS Specimens

As shown in **Table 13** for prospective symptomatic subjects, 105 NS specimens tested positive for SARS-CoV-2 with both the **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System and the composite comparator; four SARS-CoV-2 comparator positive specimens tested negative for SARS-CoV-2 with the **cobas** SARS-CoV-2 & Influenza A/B test. A total of 503 NS specimens tested negative for SARS-CoV-2 with both the **cobas** SARS-CoV-2 & Influenza A/B test and the composite comparator; four SARS-CoV-2 comparator negative specimens tested positive for SARS-CoV-2 with the **cobas** SARS-CoV-2 & Influenza A/B test. All eight of the discordant SARS-CoV-2 results showed late Ct values, which are indicative of NS specimens from individuals potentially with viral loads near or below the limit of detection of both **cobas** SARS-CoV-2 & Influenza A/B and the composite comparator methods.

For SARS-CoV-2, the results of the clinical performance evaluation using NS specimens from prospective symptomatic subjects demonstrated 96.3% PPA (105/109; 95% score CI: 90.9% - 98.6%) and 99.2% NPA (503/507; 95% score CI: 98.0% - 99.7%) as compared to the composite comparator method.

Table 13. Clinical performance comparison – SARS-CoV-2 for prospective NS specimens

		Composite Comparator Method SARS-CoV-2 Result	
		Positive	Negative
cobas SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System **Nasal Swab (NS)**	Positive	105	4
	Negative	4	503

PPA	96.3% (95% CI: 90.9% - 98.6%)
NPA	99.2% (95% CI: 98.0% - 99.7%)

Note : The nasal swabs were comprised of healthcare provider-collected nasal swab specimens and nasal swab specimens self-collected on-site with healthcare provider instructions.

As shown in **Table 14** for prospective symptomatic subjects, all 20 NS specimens tested positive for influenza A with both the **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System and the comparator assay. A total of 595 NS specimens tested negative for influenza A with both the **cobas** SARS-CoV-2 & Influenza A/B test and the comparator assay; one influenza A comparator negative specimens tested positive for influenza A with the **cobas** SARS-CoV-2 & Influenza A/B test.

For influenza A, the results of the clinical performance evaluation using NS specimens from prospective symptomatic subjects demonstrated 100.0% PPA (20/20; 95% score CI: 83.9% - 100.0%) and 99.8% NPA (595/596; 95% score CI: 99.1% - 100.0%) as

compared to the comparator method.

Table 14. Clinical performance comparison – Influenza A for prospective NS specimens

		Comparator Method Influenza A Result	
		Positive	Negative
cobas SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System **Nasal Swab (NS)**	Positive	20	1
	Negative	0	595

PPA	100.0% (95% CI: 83.9% - 100.0%)
NPA	99.8% (95% CI: 99.1% - 100.0%)

Note : The nasal swabs were comprised of healthcare provider-collected nasal swab specimens and nasal swab specimens self-collected on-site with healthcare provider instructions.

As shown in **Table 15** for retrospective NS specimens, the results of the clinical performance evaluation for influenza A demonstrated 97.2% PPA (35/36; 95% score CI: 85.8% - 99.5%) and 100.0% NPA (150/150; 95% score CI: 97.5% - 100.0%) as compared to the comparator method.

Table 15. Clinical performance comparison – Influenza A for retrospective NS specimens

		Comparator Method Influenza A Result	
		Positive	Negative
cobas SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System **Nasal Swab (NS)**	Positive	35	0
	Negative	1	150

PPA	97.2% (95% CI: 85.8% - 99.5%)
NPA	100.0% (95% CI: 97.5% - 100.0%)

As shown in **Table 16** for retrospective NS specimens, the results of the clinical performance evaluation for influenza B demonstrated 100.0% PPA (32/32; 95% score CI: 89.3% - 100.0%) and 100.0% NPA (154/154; 95% score CI: 97.6% - 100.0%) as compared to the comparator method.

For prospective symptomatic subjects, PPA was not calculable because no fresh specimens were influenza B-positive by the comparator method. For influenza B, the

results of the clinical performance evaluation using NS specimens from prospective symptomatic subjects demonstrated 100.0% NPA (616/616; 95% score CI: 99.4% - 100.0%) as compared to the comparator method.

Table 16. Clinical performance comparison – Influenza B for retrospective NS specimens

		Comparator Method Influenza B Result	
		Positive	Negative
cobas SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System **Nasal Swab (NS)**	Positive	32	0
	Negative	0	154

PPA	100.0% (95% CI: 89.3% - 100.0%)
NPA	100.0% (95% CI: 97.6% - 100.0%)

iii. Invalid Rate for Clinical Evaluation Samples

In total, 828 test results from NPS samples were obtained with **cobas** SARS-CoV-2 & Influenza A/B during the clinical evaluation, including samples that required repeat testing in accordance with this IFU. Of these, a total of 6 (0.7%) failed tests and 6 (0.7%) invalid results were obtained. An additional 10 tests experienced protocol deviations, leaving a total of 806 (97.3%) valid NPS results obtained with **cobas** SARS-CoV-2 & Influenza A/B during the clinical evaluation.

In total, 834 test results from NS samples were obtained with **cobas** SARS-CoV-2 & Influenza A/B during the clinical evaluation, including samples that required repeat testing in accordance with this IFU. Of these, a total of 1 (0.1%) failed test and 7 (0.8%) invalid results were obtained. An additional 11 tests experienced protocol deviations, leaving a total of 815 (97.7%) valid NS results obtained with **cobas** SARS-CoV-2 & Influenza A/B during the clinical evaluation.

Tables 17 and 18 describe the number of samples from the enrolled prospective and retrospective populations combined that obtained invalid or failed results with the **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System.

Table 17. Numbers of Valid and Failed/Invalid Test Results From Specimens From Valid QC Batches by Lot, Site/Instrument and Operator - Nasopharyngeal swab (NPS)

				Invalid Test Results				
					Incidents			
	Factor Number	Total Tests From Valid QC Batches N	Failed Tests{a} n (%)	Invalid Results n (%)	Instrument Errors n (%)	Other Incidents {b} n (%)	Protocol Deviations n (%)	Tests With Valid Results n (%)
Site	1	144	2 (1.4)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	142 (98.6)
	2	248	1 (0.4)	2 (0.8)	0 (0.0)	0 (0.0)	0 (0.0)	245 (98.8)
	3	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	6 (100.0)
	4	80	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	6 (7.5)	74 (92.5)
	5	54	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	54 (100.0)
	6	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	9 (100.0)
	7	26	0 (0.0)	2 (7.7)	0 (0.0)	0 (0.0)	2 (7.7)	22 (84.6)
	8	125	3 (2.4)	1 (0.8)	0 (0.0)	0 (0.0)	0 (0.0)	121 (96.8)
	9	30	0 (0.0)	1 (3.3)	0 (0.0)	0 (0.0)	1 (3.3)	28 (93.3)
	10	106	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.9)	105 (99.1)
	Total	828	6 (0.7)	6 (0.7)	0 (0.0)	0 (0.0)	10 (1.2)	806 (97.3)

Note: This table includes both prospective and retrospective sample results.
Note: A quality control (QC) batch is defined as the set of external control runs (typically 1 SARS-CoV-2 & Influenza A/B–Positive and 1 SARS-CoV-2 Influenza A/B- Negative).
Note: An invalid QC batch is one where at least one external control is invalid or was not performed or there is an incident or protocol deviation that invalidates the entire QC batch.
{a} A failed QC batch is one where a failed test occurred for at least one of the external controls (SARS-CoV-2 & Influenza A/B-Positive or SARS-CoV-2 & Influenza A/B-Negative external control) and the external control was not retested.
{b} Other incidents include: reagent, operational and other errors.

Table 18. Numbers of Valid and Failed/Invalid Test Results From Specimens From Valid QC Batches by Lot, Site/Instrument and Operator - Nasal swab (NS)

				Invalid Test Results				
					Incidents			
	Factor Number	Total Tests From Valid QC Batches N	Failed Tests{a} n (%)	Invalid Results n (%)	Instrument Errors n (%)	Other Incidents {b} n (%)	Protocol Deviations n (%)	Tests With Valid Results n (%)
Site	1	89	0 (0.0)	2 (2.2)	0 (0.0)	0 (0.0)	0 (0.0)	87 (97.8)
	2	286	0 (0.0)	4 (1.4)	0 (0.0)	0 (0.0)	1 (0.3)	281 (98.3)
	3	6	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	6 (100.0)
	4	102	0 (0.0)	1 (1.0)	0 (0.0)	0 (0.0)	7 (6.9)	94 (92.2)
	5	54	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	54 (100.0)
	6	9	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	9 (100.0)
	7	22	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (4.5)	21 (95.5)

				Invalid Test Results				
					Incidents			
	Factor Number	Total Tests From Valid QC Batches N	Failed Tests{a} n (%)	Invalid Results n (%)	Instrument Errors n (%)	Other Incidents {b} n (%)	Protocol Deviations n (%)	Tests With Valid Results n (%)
	8	131	1 (0.8)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	130 (99.2)
	9	29	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (3.4)	28 (96.6)
	10	106	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.9)	105 (99.1)
	Total	834	1 (0.1)	7 (0.8)	0 (0.0)	0 (0.0)	11 (1.3)	815 (97.7)

Note: This table includes both prospective and retrospective sample results.
Note: A quality control (QC) batch is defined as the set of external control runs (typically 1 SARS-CoV-2 & Influenza A/B–Positive and 1 SARS-CoV-2 Influenza A/B- Negative).
Note: An invalid QC batch is one where at least one external control is invalid or was not performed or there is an incident or protocol deviation that invalidates the entire QC batch.
{a} A failed QC batch is one where a failed test occurred for at least one of the external controls (SARS-CoV-2 & Influenza A/B-Positive or SARS-CoV-2 & Influenza A/B-Negative external control) and the external control was not retested.
{b} Other incidents include: reagent, operational and other errors.

iv. Device Performance with Analyte Concentrations Near the Cutoff (Reproducibility)

A reproducibility study was conducted to evaluate the performance of the **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System including weak positive samples when testing was performed by untrained users. The study assessed the total variability of the **cobas** Liat SARS-CoV-2 & Influenza A/B assay across operators, study sites, testing days, **cobas** Liat Analyzers, and **cobas** Liat assay tube lots. The **cobas** Liat SARS-CoV-2 & Influenza A/B assay was evaluated at three CLIA waived sites. Two (2) operators at each of the three sites tested a 3-member reproducibility panel in triplicate on five different days across 3 reagent lots, for a total of ~810 tests, ~270 tests/panel member (3 sites × 3 lots/site × 5 day/lot × 2 operators/day × 3 panel members/operator × 3 replicates/panel member). Each site utilized a minimum of three **cobas** Liat Analyzers, and all replicates for each panel member were tested on the same analyzer. The reproducibility panel contained a true negative; a low positive and a moderate positive member co-formulated with SARS-CoV-2, influenza A, and influenza B.

The reproducibility panel samples were prepared by spiking SARS-CoV-2 (USA-WA1/2020, catalog number 0810587CFHI, ZeptoMetrix, NY), influenza A virus (Brisbane/59/07-catalog number 0810244CF; ZeptoMetrix, NY) and influenza B virus (Florida/04/06-catalog number 0810255CF; ZeptoMetrix, NY) of known titer into negative simulated clinical matrix. (Refer to the FDA Decision Summary for K223591, section VII.B.2. Matrix Equivalency, for analytical study data

supporting the use of simulated clinical matrix). The moderate positive and low positive concentrations used for each of the strains corresponded to 3x-5x LoD and 1x-2x LoD, respectively. The true negative sample was comprised of negative simulated clinical matrix.

Three (3) CLIA waived sites and six operators (two operators per site) participated in this reproducibility study. The six operators consisted of two Medical Assistants, a Point-of-Care Coordinator, a Research assistant, an Administrative Assistant (billing), and a Lab Technician (institutional title only, no laboratory training) with no formal medical laboratory training. All operators had limited or no training or hands-on experience in conducting laboratory testing when the study was initiated.

The six operators at the three sites tested the members of the reproducibility panel in triplicate on five non-consecutive days. Three (3) **cobas** Liat Analyzers were used at each site for a total of nine **cobas** Liat Analyzers. Each site also used approximately equal amounts of three different lots of **cobas**SARS-CoV-2 & Influenza A/B assay tubes.

The qualitative and quantitative results are shown in **Table 19** and **Table 20**, respectively.

Table 19. Reproducibility Study- Qualitative Results

Target	Panel Conc.	% Agreement with Expected Results/ (n Agreement/N Tested) (95% CI)			
		Site 1	Site 2	Site 3	Overall
SARS-CoV-2	Low Positive (1x-2x LoD)	100% (90/90) (95.9-100)	98.9% (89/90) (93.4-99.8)	97.6% (81/83) (91.6-99.3)	98.9% (260/263) (96.7-99.6)
	Mod. Positive (3x-5x LoD)	98.9% (88/89) (93.9-99.8)	100% (89/89) (95.9-100)	100% (90/90) (95.9-100)	99.6% (267/268) (97.9-99.9)
	Negative	100% (89/89) (95.9-100)	100% (90/90) (95.9-100)	100% (87/87) (95.8-100)	100% (266/266) (98.6-100)
Flu A	Low positive (1x-2x LoD)	100% (90/90) (95.9-100)	95.6% (86/90) (89.1-98.3)	100% (83/83) (95.6-100)	98.5% (259/263) (96.2-99.4)
	Mod. Positive (3x-5x LoD)	100% (89/89) (95.9-100)	100% (89/89) (95.9-100)	100% (90/90) (95.9-100)	100% (268/268) (98.6-100)
	Negative	100% (89/89) (95.9-100)	100% (90/90) (95.9-100)	100% (87/87) (95.8-100)	100% (266/266) (98.6-100)
Flu B	Low positive (1x-2x LoD)	100% (90/90) (95.9-100)	100% (90/90) (95.9-100)	100% (83/83) (95.6-100)	100% (263/263) (98.6-100)
	Mod. Positive (3x- 5x LoD)	98.9% (88/89) (93.4-99.8)	100% (89/89) (95.9-100)	100% (90/90) (95.9-100)	99.6% (267/268) (97.9-100)
	Negative	100% (89/89)	100% (90/90)	100% (87/87)	100% (266/266)

		(95.9-100)	(95.9-100)	(95.8-100)	(98.6-100)

Mod = moderate, Conc= Concentration; Note: Results are shown only for the intended targets. Panel members were all co-spiked with all targets, so results are presented three times

The **cobas** SARS-CoV-2 & Influenza A/B assay demonstrated 100% agreement for Flu A, and 99.6% agreement for SARS-CoV-2 and Flu B moderate positive panel members. For low positive panel members, the assay yielded 100% agreement for Flu B, and 98.9% and 98.5% for SARS-CoV-2 and Flu A, respectively (**Table 19** above). A lower agreement for low positive panel members was expected, since the analyte concentration of the panel member ranged between 1x and 2x the limit of detection, which is expected to yield >95% detection rate. This performance is acceptable and demonstrates acceptable assay reproducibility when performed by untrained users in the Intended Use setting.

Table 20. Reproducibility Study- Ct Signal Variability Analysis Results

Viral Target	Panel Member Conc	n/N[a]	Mean Ct[b]	Between Sites		Between Lot		Between Day		Between Operator		Within-Run (Residual)		Total	
				SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD	CV%	SD[b]	CV%[c]
SARS-CoV-2	1x-2x LOD	260/263	33.3	0.00	0.0	0.36	1.1	0.29	0.9	0.00	0.0	1.08	3.3	1.18	3.5
SARS-CoV-2	3x-5x LOD	267/268	32.1	0.31	1.0	0.46	1.4	0.29	0.9	0.07	0.2	0.74	2.3	0.97	3.0
Influenza A	1x-2x LOD	259/263	33.0	0.07	0.2	0.49	1.5	0.19	0.6	0.00	0.0	0.81	2.5	0.97	2.9
Influenza A	3x-5x LOD	268/268	31.9	0.26	0.8	0.44	1.4	0.23	0.7	0.00	0.0	0.56	1.7	0.79	2.5
Influenza B	1x-2x LOD	263/263	30.2	0.15	0.5	0.38	1.3	0.50	1.6	0.00	0.0	0.66	2.2	0.92	3.1
Influenza B	3x-5x LOD	267/268	29.3	0.09	0.3	0.29	1.0	0.29	1.0	0.00	0.0	0.96	3.3	1.05	3.6

Ct: cycle threshold, CV%: percent coefficient of variation, LOD: limit of detection, SARS-CoV-2: severe acute respiratory syndrome coronavirus 2, SD: standard deviation.

[a]n is the number of positive tests, which contribute Ct values to the analysis. N is the total number of valid tests for the panel member.

[b]The Mean and total SD were estimated using the PROC MIXED procedure.

[c]Total CV (%) = (SD/Mean)*100.

The total Ct signal variability, as measured by the standard deviation, was less than or equal to 1.08 across all target viruses and concentrations. For all positive panel members, the within-run factor (i.e., random error) was the largest contributor to total variability. These results indicate that the reproducibility of the **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat system is robust in NPS samples when tested by untrained users in the Intended Use setting.

2. Operator Questionnaire

Upon completion of the Clinical and Near the Cutoff Studies (Reproducibility), the operators at each site were asked to complete an Operator Questionnaire that asked them to rate the ease of use of the test procedure and answer proficiency questions related to **cobas** SARS-CoV-2 & Influenza A/B result interpretation. The proficiency potion of the questionnaire included 15 images of a **cobas** Liat System display and asked if the results for SARS-CoV-2, influenza A, and influenza B were positive, negative, or could not be assessed (45 total points possible). The ease of use questionnaire asked the operators to reply to a series of 8 statements using an agreement scale (1= strongly disagree to 5= strongly agree).

Of the 30 operators who participated in the clinical study, there were three individuals who were no longer employed by the clinical sites at the time the questionnaire was given, so only 27 operators provided responses. **Table 21** shows the results of the proficiency questions by operator. The combined score for the proficiency portion of the questionnaire was 99.3% (1206/1215 correct responses).

Table 21. Study Operator Proficiency Test Results

Site ID	Operator	Overall Score	Overall Score Points
1	1	100.0%	45/45
	2	100.0%	45/45
	3	100.0%	45/45
	4	100.0%	45/45
2	2	100.0%	45/45
	4	100.0%	45/45
3	1	100.0%	45/45
	2	95.6%	43/45
4	1	100.0%	45/45
	2	100.0%	45/45
	4	95.6%	43/45
5	1	100.0%	45/45
	2	100.0%	45/45
	4	100.0%	45/45
6	1	100.0%	45/45
	2	95.6%	43/45
7	1	100.0%	45/45
	2	95.6%	43/45
8	1	100.0%	45/45
	2	100.0%	45/45
	3	97.8%	44/45
9	2	100.0%	45/45

	4	100.0%	45/45
10	1	100.0%	45/45
	2	100.0%	45/45
	3	100.0%	45/45
	5	100.0%	45/45
Total		99.3%	1206/1215

Note: Three operators were no longer at the institution to complete the post ease-of-use questionnaire.

Note: One operator completed the post ease-of-use questionnaire and test but did not test any samples and is not included in this summary table.

The operators' average scores indicating their agreement with the statements in the ease of use questionnaire are shown in **Table 22**. The average agreement with the statement ranged from 4.1 (4 being Agree) to 4.8 (5 being Strongly Agree). The overall score for the ease of use questions was 4.5 out of 5, indicating the operators agreed the device was easy to use overall.

Table 22. Operators Ease of Use Questionnaire Results

Statement	Average Agreement with Statement Score[a] (1 = Strongly Disagree, 5 = Strongly Agree)
The instructions to add lot and perform controls were easy to follow.	4.1
The instructions to test specimens were easy to follow.	4.5
It was easy to load the sample into the Liat assay tube.	4.6
It was easy to start the assay on the Liat analyzer.	4.7
It was easy to read the test results.	4.8
It was easy to understand the test results.	4.8
The Instructions For Use and Quick Reference Instructions clearly explain what to do if a test result is invalid.	4.2
I did not need help when I tested samples using the Liat assay.	4.4
Overall Score	4.5

[a]Statements were scored as follows: 1 = Strongly Disagree, 2 = Disagree, 3 = Neutral, 4 = Agree, 5 = Strongly Agree.

Note: Three operators were no longer at the institution to complete the post ease of use questionnaire.

Note: One operator completed the post ease of use questionnaire and test but did not test any samples and is not included in this summary table.

Labeling for Waived Devices

The labeling consists of:

1. **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System Instructions for Use

2. **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System Quick Reference Instructions

3. **cobas** Liat System User Guide

2. The following elements are appropriately present:

- The **cobas** Liat System User Guide specifies the environmental operating conditions under which testing may be performed.
- The **cobas** Liat System User Guide and **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System Instructions for Use are clear and easy to understand.
- The **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System Instructions for Use and Quick Reference Instructions identify the test as CLIA Waived.
- The **cobas** SARS-CoV-2 & Influenza A/B Nucleic acid test for use on the **cobas** Liat System Instructions for Use:
    - Indicate that laboratories with a Certificate of Waiver must follow the manufacturer's instructions for performing the test.
    - Include step-by-step instructions for performing the test.
    - Include safety considerations applicable for untrained users.
    - Specify the actions to be taken if an invalid test result is obtained.
    - Include a summary of the studies performed to support CLIA Waiver.
    - Include appropriate warnings and/or limitations pertaining to clinical interpretation of test results.
    - Include recommendations for Quality Control testing including the source of appropriate control materials and the frequency of testing.

- The labeling is sufficient and satisfies the requirements of 21 CFR Part 809.10.

M. Benefit-Risk Considerations

Not Applicable

N. Conclusion:

The submitted information in this CLIA waiver application supports a CLIA waiver approval decision.