Rx Only

cobas® SARS-CoV-2 Qualitative

Nucleic acid test for use on the cobas® 5800/6800/8800 Systems

For in vitro diagnostic use

cobas® SARS-CoV-2 Qualitative 192T	P/N: 09446109190
cobas® SARS-CoV-2 Qualitative 480T	P/N: 09448870190
cobas® SARS-CoV-2 Qualitative Control Kit	P/N: 09446117190
cobas® Buffer Negative Control Kit	P/N: 09051953190

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Intended use

cobas® SARS-CoV-2 Qualitative for use on the **cobas**® 5800/6800/8800 Systems is a real-time RT-PCR test intended for the qualitative detection of nucleic acids from SARS-CoV-2 in healthcare provider-instructed self-collected anterior nasal (nasal) swab and saliva specimens (collected on site), and healthcare provider-collected nasal, nasopharyngeal, and oropharyngeal swab specimens collected from any individuals, including those suspected of COVID-19 by their healthcare provider, and those without symptoms or other reasons to suspect COVID-19.

This test is also intended for the qualitative detection of nucleic acids from SARS-CoV-2 in pooled samples containing up to and including six individual samples from healthcare provider-instructed self-collected nasal swab specimens (collected on site), or healthcare provider-collected nasal, nasopharyngeal, and oropharyngeal swab specimens. Negative results from pooled samples should be treated as presumptive and, if inconsistent with clinical signs and symptoms or necessary for patient management, pooled samples should be tested individually. Specimens included in pools with a positive or presumptive positive result must be tested individually prior to reporting a result. Specimens with low SARS-CoV-2 RNA concentrations may not be detected in sample pools due to the decreased sensitivity of pooled testing.

Results are for the detection of SARS-CoV-2 RNA. The SARS-CoV-2 RNA is generally detectable in respiratory specimens during the acute phase of infection. Positive results are indicative of the presence of SARS-CoV-2 RNA but may not represent the presence of SARS-CoV-2 RNA; clinical correlation with patient history and other diagnostic information is necessary to determine patient infection status. Positive results do not rule out bacterial infection or co-infection with other viruses.

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

cobas® SARS-CoV-2 Qualitative is intended for use by trained clinical laboratory personnel specifically instructed and trained in the techniques of real-time PCR and in vitro diagnostic procedures.

Summary and explanation of the test

Explanation of the test

cobas® SARS-CoV-2 Qualitative is a qualitative nucleic acid test for use on the **cobas**® 5800 System, **cobas**® 6800 System or **cobas**® 8800 System for the detection of the 2019 novel coronavirus (SARS-CoV-2) RNA in individual saliva specimens collected in a sterile empty collection container and individual or pooled nasal, nasopharyngeal and oropharyngeal swab samples collected in Copan Universal Transport Medium System (UTM-RT), BD™ Universal Viral Transport System (UVT), **cobas**® PCR Media, or 0.9% physiological saline. The RNA Internal Control, used to monitor the entire sample preparation and PCR amplification process, is introduced into each specimen during sample processing. In addition, the test utilizes external controls (low titer positive control and a negative control).

Principles of the procedure

cobas® SARS-CoV-2 Qualitative is based on fully automated sample preparation (nucleic acid extraction and purification) followed by PCR amplification and detection. The **cobas**® 5800 System is designed as one integrated instrument. The **cobas**® 6800/8800 Systems consist of the sample supply module, the transfer module, the processing module, and the analytic module. Automated data management is performed by the **cobas**® 5800 System or **cobas**® 6800/8800 Systems software(s), which assigns test results for all tests. Results can be reviewed directly on the system screen, and printed as a report.

Nucleic acid from patient samples and added internal control RNA (RNA IC) molecules are simultaneously extracted. Nucleic acid is released by addition of proteinase and lysis reagent to the sample. The released nucleic acid binds to the silica surface of the added magnetic glass particles. Unbound substances and impurities, such as denatured protein, cellular debris and potential PCR inhibitors, are removed with subsequent wash steps and purified nucleic acid is eluted from the magnetic glass particles with elution buffer at elevated temperature. External controls (positive and negative) are processed in the same way.

Selective amplification of target nucleic acid from the sample is achieved by the use of target-specific forward and reverse primers for ORF1 a/b non-structural region that is unique to SARS-CoV-2. Additionally, a conserved region in the structural protein envelope E-gene were chosen for pan-Sarbecovirus detection. The pan-Sarbecovirus detection sets will also detect SARS-CoV-2 virus.

Selective amplification of RNA Internal Control is achieved by the use of non-competitive sequence specific forward and reverse primers which have no homology with the coronavirus genome. A thermostable DNA polymerase enzyme is used for amplification.

The **cobas**® SARS-CoV-2 Qualitative master mix contains detection probes which are specific for the coronavirus type SARS-CoV-2, members of the Sarbecovirus subgenus, and the RNA Internal Control nucleic acid. The coronavirus and RNA Internal Control detection probes are each labeled with unique fluorescent dyes that act as a reporter. Each probe also has a second dye which acts as a quencher. When not bound to the target sequence, the fluorescent signals of the intact probes are suppressed by the quencher dye. During the PCR amplification step, hybridization of the probes to the specific single-stranded DNA template results in cleavage of the probe by the 5' to 3' exonuclease activity of the DNA polymerase resulting in separation of the reporter and quencher dyes and the generation of a fluorescent signal. With each PCR cycle, increasing amounts of cleaved probes are generated and the cumulative signal of the reporter dye increases concomitantly. Each reporter dye is measured at defined wavelengths, which enables simultaneous detection and discrimination of the amplified coronavirus target and the RNA Internal Control. The master mix includes deoxyuridine triphosphate (dUTP), instead of deoxythimidine triphosphate (dTTP), which is incorporated into the newly synthesized DNA (amplicon). Any contaminating amplicons from previous PCR runs are destroyed by the AmpErase enzyme [uracil-N-glycosylase], which is included in the PCR mix, when heated in the first thermal cycling step. However, newly formed amplicons are not destroyed since the AmpErase enzyme is inactivated once exposed to temperatures above 55°C.

Reagents and materials

The materials provided for **cobas®** SARS-CoV-2 Qualitative can be found in Table 1. Materials required, but not provided can be found in Table 2, Table 3, Table 4, Table 8, Table 9, and Table 10.

Refer to the **Reagents and materials** section and **Precautions and handling requirements** section for the hazard information for the product.

cobas® SARS-CoV-2 Qualitative reagents and controls

All unopened reagents and controls shall be stored as recommended in Table 1 to Table 4.

Table 1 cobas® SARS-CoV-2 Qualitative

cobas® SARS-CoV-2 Qualitative Store at 2-8°C 192 test cassette (P/N 09446109190) 480 test cassette (P/N 09448870190)			
Kit components	**Reagent ingredients**	**Quantity per kit 192 tests**	**Quantity per kit 480 tests**
Proteinase Solution (PASE)	Tris buffer, < 0.05% EDTA, calcium chloride, calcium acetate, 8% proteinase, glycerol EUH210: Safety data sheet available on request. EUH208: Contains Subtilisin from *Bacillus subtilis*. May produce an allergic reaction.	22.3 mL	38 mL
RNA Internal Control (RNA IC)	Tris buffer, < 0.05% EDTA, < 0.001% non-Sarbecovirus related armored RNA construct containing primer and probe specific primer sequence regions (non-infectious RNA in MS2 bacteriophage), < 0.1% sodium azide	21.2 mL	38 mL
Elution Buffer (EB)	Tris buffer, 0.2% methyl-4 hydroxybenzoate	21.2 mL	38 mL
Master Mix Reagent 1 (MMX-R1)	Manganese acetate, potassium hydroxide, < 0.1% sodium azide	7.5 mL	14.5 mL
SARS-CoV-2 QL Master Mix Reagent 2 (SARS-CoV-2 QL MMX-R2)	Tricine buffer, potassium acetate, < 18% dimethyl sulfoxide, glycerol, < 0.1% Tween 20, EDTA, < 0.12% dATP, dCTP, dGTP, dUTPs, < 0.01% upstream and downstream SARS-CoV-2 and Sarbecovirus primers, < 0.01% Internal Control forward and reverse primers, < 0.01% fluorescent-labeled oligonucleotide probes specific for SARS-CoV-2, Sarbecovirus, and the RNA Internal Control, < 0.01% oligonucleotide aptamer, < 0.1% Z05D DNA polymerase, < 0.10% AmpErase (uracil-N-glycosylase) enzyme (microbial), < 0.1% sodium azide	9.7 mL	17.5 mL

Table 2 cobas® SARS-CoV-2 Qualitative Control Kit

cobas® SARS-CoV-2 Qualitative Control Kit Store at 2–8°C (P/N 09446117190)		
Kit components	**Reagent ingredients**	**Quantity per kit**
SARS-CoV-2 QL Positive Control (SARS-CoV-2 QL (+)C)	Tris buffer, < 0.05% Sodium azide, < 0.005% EDTA, 0.003% Poly rA, < 0.01% Non-infectious plasmid DNA (microbial) containing SARS-CoV-2 sequence, < 0.01% Non-infectious plasmid DNA (microbial) containing pan-Sarbecovirus sequence	16 mL (16 x 1 mL)

Table 3 cobas® Buffer Negative Control Kit

cobas® Buffer Negative Control Kit Store at 2-8°C (P/N 09051953190)		
Kit components	**Reagent ingredients**	**Quantity per kit**
cobas® Buffer Negative Control (BUF (–) C)	Tris buffer, < 0.1% sodium azide, EDTA, 0.002% Poly rA RNA (synthetic)	16 mL (16 x 1 mL)

cobas omni reagents for sample preparation

Table 4 cobas omni reagents for sample preparation*

Reagents	Reagent ingredients	Quantity per kit	Safety symbol and warning**
cobas omni MGP Reagent (MGP) Store at 2–8°C (P/N 06997546190)	Magnetic glass particles, Tris buffer, 0.1% methyl-4 hydroxybenzoate, < 0.1% sodium azide	480 tests	Not applicable
cobas omni Specimen Diluent (SPEC DIL) Store at 2–8°C (P/N 06997511190)	Tris buffer, 0.1% methyl-4 hydroxybenzoate, < 0.1% sodium azide	4 x 875 mL	Not applicable
cobas omni Lysis Reagent (LYS) Store at 2–8°C (P/N 06997538190)	43% (w/w) guanidine thiocyanate***, 5% (w/v) polydocanol***, 2% (w/v) dithiothreitol***, dihydro sodium citrate	4 x 875 mL	 **DANGER** H302 + H332: Harmful if swallowed or if inhaled. H314: Causes severe skin burns and eye damage. H411: Toxic to aquatic life with long lasting effects. EUH032: Contact with acids liberates very toxic gas. P273: Avoid release to the environment. P280: Wear protective gloves/ protective clothing/ eye protection/ face protection/ hearing protection. P303 + P361 + P353: IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water. P304 + P340 + P310: IF INHALED: Remove person to fresh air and keep comfortable for breathing. Immediately call a POISON CENTER/doctor. P305 + P351 + P338 + P310: IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. Immediately call a POISON CENTER/ doctor. P391: Collect spillage. 593-84-0 Guanidinium thiocyanate 9002-92-0 Polidocanol 3483-12-3 (R*,R*)-1,4-dimercaptobutane-2,3-diol
cobas omni Wash Reagent (WASH) Store at 15–30°C (P/N 06997503190)	Sodium citrate dihydrate, 0.1% methyl-4 hydroxybenzoate	4.2 L	Not applicable

* These reagents are not included in the **cobas**® SARS-CoV-2 Qualitative kits. See listing of additional materials required (Table 8 and Table 9).

** Product safety labeling primarily follows EU GHS guidance.

***Hazardous substance.

Reagent storage requirements

Reagents shall be stored and will be handled as specified in Table 5, Table 6 and Table 7.

When reagents are not loaded on the **cobas**® 5800 or **cobas**® 6800/8800 Systems, store them at the corresponding temperature specified in Table 5.

Table 5 Reagent storage (when reagent is not on the system)

Reagent	Storage temperature
cobas® SARS-CoV-2 Qualitative 192T	2–8°C
cobas® SARS-CoV-2 Qualitative 480T	2–8°C
cobas® SARS-CoV-2 Qualitative Control Kit	2–8°C
cobas® Buffer Negative Control Kit	2–8°C
cobas omni Lysis Reagent	2–8°C
cobas omni MGP Reagent	2–8°C
cobas omni Specimen Diluent	2–8°C
cobas omni Wash Reagent	15–30°C

Reagent handling requirements for cobas® 5800 System

Reagents loaded onto the **cobas**® 5800 System are stored at appropriate temperatures and their expiration is monitored by the system. The **s**ystem allows reagents to be used only if all of the conditions shown in Table 6 are met. The system automatically prevents use of expired reagents. Table 6 allows the user to understand the reagent handling conditions enforced by the **cobas**® 5800 System.

Table 6 Reagent expiry conditions enforced by the **cobas**® 5800 System

Reagent	Kit expiration date	Open-kit stability	Number of runs for which this kit can be used	On-board stability
cobas® SARS-CoV-2 Qualitative 192T	Date not passed	90 days from first usage	Max 40 runs	Max 36 days**
cobas® SARS-CoV-2 Qualitative 480T	Date not passed	90 days from first usage	Max 40 runs	Max 36 days**
cobas® SARS-CoV-2 Qualitative Control Kit	Date not passed	Not applicable*	Not applicable	Max 36 days**
cobas® Buffer Negative Control Kit	Date not passed	Not applicable*	Not applicable	Max 36 days**
cobas omni Lysis Reagent	Date not passed	30 days from loading**	Not applicable	Not applicable
cobas omni MGP Reagent	Date not passed	30 days from loading**	Not applicable	Not applicable
cobas omni Specimen Diluent	Date not passed	30 days from loading**	Not applicable	Not applicable
cobas omni Wash Reagent	Date not passed	30 days from loading**	Not applicable	Not applicable

* Single use reagents.

Time is measured from the first time that reagent is loaded onto the **cobas® 5800 System.

Reagent handling requirements for cobas® 6800/8800 Systems

Reagents loaded onto the **cobas**® 6800/8800 Systems are stored at appropriate temperatures and their expiration is monitored by the system. The **cobas**® 6800/8800 Systems allow reagents to be used only if all of the conditions shown in Table 7 are met. The system automatically prevents use of expired reagents. Table 7 allows the user to understand the reagent handling conditions enforced by the **cobas**® 6800/8800 Systems.

Table 7 Reagent expiry conditions enforced by the **cobas**® 6800/8800 Systems

Reagent	Kit expiration date	Open-kit stability	Number of runs for which this kit can be used	On-board stability (cumulative time on board outside refrigerator)
cobas® SARS-CoV-2 Qualitative 192T	Date not passed	90 days from first usage	Max 40 runs	Max 40 hours**
cobas® SARS-CoV-2 Qualitative 480T	Date not passed	90 days from first usage	Max 20 runs	Max 20 hours**
cobas® SARS-CoV-2 Qualitative Control Kit	Date not passed	Not applicable*	Not applicable	Max 8 hours**
cobas® Buffer Negative Control Kit	Date not passed	Not applicable*	Not applicable	Max 10 hours**
cobas omni Lysis Reagent	Date not passed	30 days from loading**	Not applicable	Not applicable
cobas omni MGP Reagent	Date not passed	30 days from loading**	Not applicable	Not applicable
cobas omni Specimen Diluent	Date not passed	30 days from loading**	Not applicable	Not applicable
cobas omni Wash Reagent	Date not passed	30 days from loading**	Not applicable	Not applicable

* Single use reagents.

** Time is measured from the first time that reagent is loaded onto the **cobas**® 6800/8800 Systems.

Additional materials required for cobas® 5800 System

Table 8 Materials and consumables for use on the **cobas**® 5800 System

Material	P/N
cobas omni Processing Plate 24	08413975001
cobas omni Amplification Plate 24	08499853001
cobas omni Liquid Waste Plate 24	08413983001
Tip CORE TIPS with Filter, 1mL	04639642001
Tip CORE TIPS with Filter, 300uL	07345607001
cobas omni Liquid Waste Container	07094388001
cobas omni Lysis Reagent	06997538190
cobas omni MGP Reagent	06997546190
cobas omni Specimen Diluent	06997511190
cobas omni Wash Reagent	06997503190
Solid Waste Bag or Solid Waste Bag With Insert	07435967001 or 08030073001
cobas omni Secondary Tubes 13x75 (optional)	06438776001
cobas® PCR Media Tube Replacement Cap Kit	07958056190
cobas® PCR Media Disposable Tube Stand (Optional)	07958064190
MPA RACK 16 MM LIGHT GREEN 7001-7050*,**	03143449001
RD5 RACK – RD Standard rack 0001-0050 LR*,**	11902997001
16-position tube carrier*	09224319001
5-position rack carrier*	09224475001

*Contact your local Roche representative for a detailed order list for sample racks.

*,** MPA 16mm rack or 16-position tube carrier are the preferred racks for use with samples collected in **cobas**® PCR Media tubes. If RD5 racks are used, make sure to fill in the sample tubes with not less than the recommended minimum sample input. The tubes sit higher in an RD5 rack because of the rubber gasket at the bottom of each tube position. Therefore, it is possible that when using RD5 racks, the system could accept tubes that are below the minimum sample input volume and cause pipetting errors later in the run.

Additional materials required for cobas® 6800/8800 Systems

Table 9 Materials and consumables for use on the **cobas**® 6800/8800 Systems

Material	P/N
cobas omni Processing Plate	05534917001
cobas omni Amplification Plate	05534941001
cobas omni Pipette Tips	05534925001
cobas omni Liquid Waste Container	07094388001
cobas omni Lysis Reagent	06997538190
cobas omni MGP Reagent	06997546190
cobas omni Specimen Diluent	06997511190
cobas omni Wash Reagent	06997503190
Solid Waste Bag and Solid Waste Container or Solid Waste Bag With Insert and Kit Drawer	07435967001 and 07094361001 or 08030073001 and 08387281001
cobas omni Secondary Tubes 13x75 (optional)	06438776001
cobas® PCR Media Tube Replacement Cap Kit	07958056190
cobas® PCR Media Disposable Tube Stand (Optional)	07958064190
MPA RACK 16 MM LIGHT GREEN 7001-7050*,**	03143449001
RD5 RACK – RD Standard rack 0001-0050 LR*,**	11902997001

* MPA 16 mm or RD5 racks are required to use **cobas**® SARS-COV-2 Qualitative. Contact your local Roche representative for a detailed order list for sample racks, racks for clotted tips and rack trays accepted on the instruments.

MPA 16 mm rack is the preferred rack for use with samples collected in **cobas® PCR Media tubes. If RD5 racks are used, make sure to fill in the sample tubes with not less than the recommended minimum sample input. The tubes sit higher in an RD5 rack because of the rubber gasket at the bottom of each tube position. Therefore, it is possible that when using RD5 racks, the system could accept tubes that are below the minimum sample input volume and cause pipetting errors later in the run.

Alternative collection kits for swab specimens for use on the cobas® 5800/6800/8800 Systems

Table 10 Alternative specimen collection kits used with **cobas**® SARS-CoV-2 Qualitative for swab specimens

Collection Kit	P/N
cobas® PCR Media Uni Swab Sample Kit	07958030190
cobas® PCR Media Dual Swab Sample Kit	07958021190
cobas® PCR Media 100 tube kit	06466281190
cobas® Uni Swab 100 Kit	09205098190

Instrumentation and software required

The **cobas**® 5800 software and **cobas**® SARS-CoV-2 Qualitative analysis package for the **cobas**® 5800 System must be installed on the **cobas**® 5800 instrument. The Data Manager software and PC for the **cobas**® 5800 System will be provided with the system.

The **cobas**® 6800/8800 Systems software and **cobas**® SARS-CoV-2 Qualitative analysis package for the **cobas**® 6800/8800 Systems must be installed on the instrument(s). The Instrument Gateway (IG) server will be provided with the system.

Table 11 Instrumentation

Equipment	P/N
cobas® 5800 System	08707464001
cobas® 6800 System (Moveable Platform)	05524245001 or 06379672001
cobas® 6800 System (Fixed Platform)	05524245001 or 06379664001
cobas® 8800 System	05412722001
Sample Supply Module (**cobas**® 6800/8800 Systems only)	06301037001

Refer to the **cobas**® 5800 System or **cobas**® 6800/8800 Systems – User Assistance and/or User Guide for additional information.

Note: Contact your local Roche representative for a detailed order list for sample racks, racks for clotted tips and rack trays accepted on the instruments.

Precautions and handling requirements

Warnings and precautions

As with any test procedure, good laboratory practice is essential to the proper performance of this assay. Due to the high sensitivity of this test, care should be taken to keep reagents and amplification mixtures free of contamination.

- For in vitro diagnostic use.
- Positive results are indicative of the presence of SARS-CoV-2 RNA.
- All patient samples should be handled as if infectious, using good laboratory procedures as outlined in Biosafety in Microbiological and Biomedical Laboratories and in the CLSI Document M29-A4.[1,2] Only personnel proficient in handling infectious materials and the use of **cobas**® SARS-CoV-2 Qualitative and the **cobas**® 5800/6800/8800 Systems should perform this procedure.
- All human-sourced materials should be considered potentially infectious and should be handled with universal precautions. If spillage occurs, immediately disinfect with a freshly prepared solution of 0.5% sodium hypochlorite in distilled or deionized water (dilute household bleach 1:10) or follow appropriate site procedures.
- The use of sterile disposable pipettes and nuclease-free pipette tips is recommended. Use only supplied or specified required consumables to ensure optimal test performance.
- Safety Data Sheets (SDS) are available on request from your local Roche representative.
- Closely follow procedures and guidelines provided to ensure that the test is performed correctly. Any deviation from the procedures and guidelines may affect optimal test performance.
- False positive results may occur if carryover of samples is not adequately controlled during sample handling and processing.
- **Some positive samples may not be detected when diluted and tested in pools**. SARS-CoV-2 RNA concentration is reduced when a positive sample is pooled with other samples, and the reduction corresponds inversely to the pool size. For example, if there is only one positive sample in a pool of 6, the concentration in the original sample would need to be 6 times the assay limit of detection in order for the concentration in the pool to be at the limit of detection.
- Inform your local competent authority about any serious incidents which may occur when using this assay.
- Reliable saliva results are dependent on proper specimen collection, handling, and storage. In case of visible particles or discoloration in the saliva sample, samples should not be further processed, but patient shall be asked to provide a new sample. Food particles and increased levels of mucin might cause failures in processing of the saliva specimen.

Reagent handling

- Handle all reagents, controls, and samples according to good laboratory practice in order to prevent carryover of samples or controls.
- Before use, visually inspect each reagent cassette, diluent, lysis reagent, and wash reagent to ensure that there are no signs of leakage. If there is any evidence of leakage, do not use that material for testing.
- **cobas omni** Lysis Reagent contains guanidine thiocyanate, a potentially hazardous chemical. Avoid contact of reagents with the skin, eyes, or mucous membranes. If contact does occur, immediately wash with generous amounts of water; otherwise, burns can occur.

- **cobas**® SARS-CoV-2 Qualitative, **cobas**® SARS-CoV-2 Qualitative Control Kit, **cobas**® Buffer Negative Control Kit, **cobas omni** MGP Reagent, and **cobas omni** Specimen Diluent contain sodium azide as a preservative. Avoid contact of reagents with the skin, eyes, or mucous membranes. If contact does occur, immediately wash with generous amounts of water; otherwise, burns can occur. If these reagents are spilled, dilute with water before wiping dry.
- Do not allow **cobas omni** Lysis Reagent, which contains guanidine thiocyanate, to contact sodium hypochlorite (bleach) solution. This mixture can produce a highly toxic gas.
- Dispose of all materials that have come in contact with samples and reagents in accordance with country, state, and local regulations.

Good laboratory practice

- Do not pipette by mouth.
- Do not eat, drink, or smoke in designated work areas.
- Wear laboratory gloves, laboratory coats, and eye protection when handling samples and reagents. Gloves must be changed between handling samples and **cobas**® SARS-CoV-2 Qualitative kits, **cobas**® SARS-CoV-2 Qualitative Control Kit, **cobas**® Buffer Negative Control Kit and **cobas omni** reagents to prevent contamination. Avoid contaminating gloves when handling samples and controls.
- Wash hands thoroughly after handling samples and kit reagents, and after removing the gloves.
- Thoroughly clean and disinfect all laboratory work surfaces with a freshly prepared solution of 0.5% sodium hypochlorite in distilled or deionized water (dilute household bleach 1:10). Follow by wiping the surface with 70% ethanol.
- If spills occur on the **cobas**® 5800 or **cobas**® 6800/8800 instrument, follow the instructions in the **cobas**® Systems – User Assistance and/or User Guide to properly clean and decontaminate the surface of the instrument(s).

Sample collection, transport, and storage

Note: Handle all samples and controls as if they are capable of transmitting infectious agents.

Sample collection – swab specimen types

Ensure that the correct collection device is used with the appropriate sample type by referring to the table below:

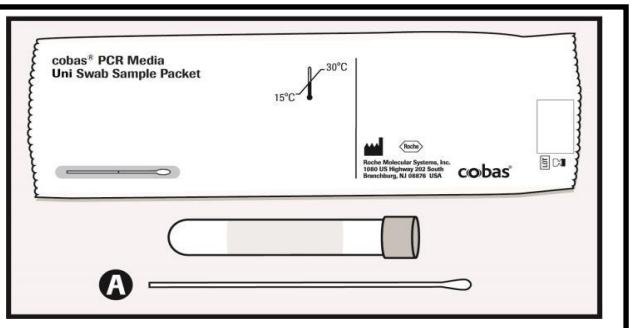
Table 12 Overview of collection devices and sample types

Collection Device	Sample Type		
	Nasopharyngeal	Oropharyngeal	Nasal
Copan Universal Transport Media (UTM-RT®)	√	√	√
BD™ Universal Viral Transport (UVT)	√	√	√
cobas® PCR Media Uni Swab Sample Kit			√
cobas® PCR Media Dual Swab Sample Kit			√
cobas® PCR Media Kit (and 100 tube PCR Media Kit)			√
0.9% Physiological saline			√

- Collect nasal, nasopharyngeal and oropharyngeal specimens according to standard collection technique using flocked or polyester-tipped swabs and immediately place in 3 mL of Copan Universal Transport Medium (UTM-RT) or BD™ Universal Viral Transport (UVT).
- Collect nasal specimens according to standard collection technique using flocked or polyester-tipped swabs and immediately place into **cobas**® PCR Media tube from **cobas**® PCR Media Kit (P/N 06466281190).
- Collect nasal specimens using the **cobas**® PCR Media Uni Swab Sample Kit (P/N 07958030190) or **cobas**® PCR Media Dual Swab Sample Kit (P/N 07958021190) according to instructions below.
- Refer to the Instructions for Use of the Collection Devices for hazard information.

Nasal (anterior nares) swab specimen collection - clinician or self-collected on site

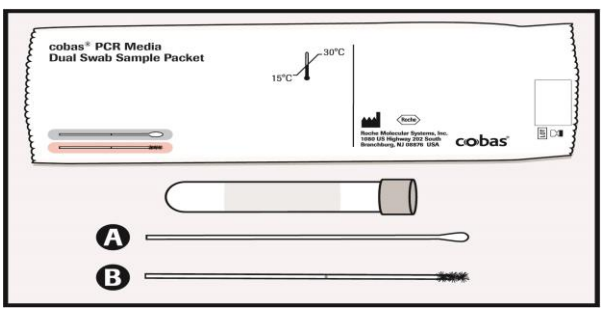
WARNING: DO NOT PRE-WET SWAB IN cobas® PCR MEDIA BEFORE COLLECTION!



The **cobas**® PCR Media Uni Swab Sample kit contains:

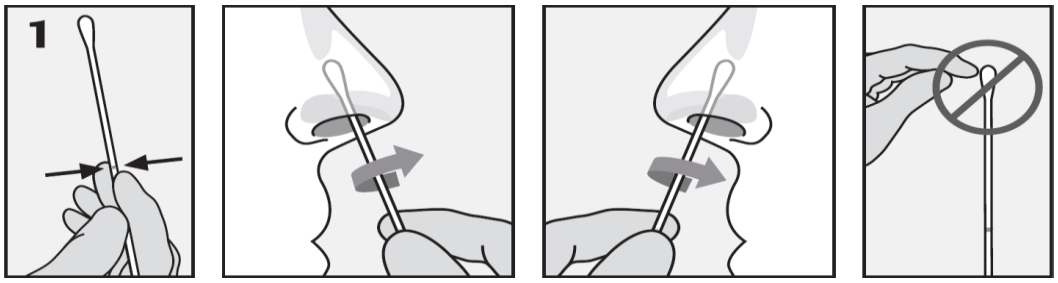
cobas® PCR Media tube
Woven Swab: A

OR



The **cobas**® PCR Media Dual Swab Sample kit contains:

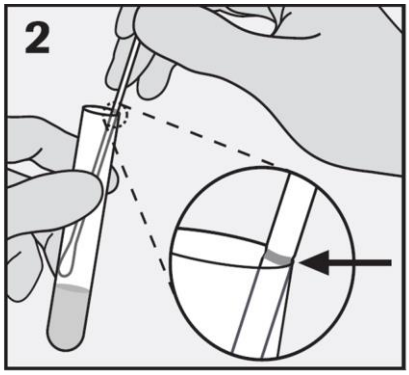
cobas® PCR Media Tube
Woven Swab: A
Flocked Swab: B



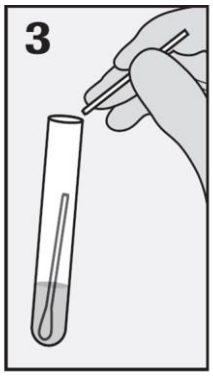
DO NOT PRE-WET SWAB IN cobas® PCR MEDIA BEFORE COLLECTION!

1. **COLLECT:** Hold the woven swab (Swab A) or the flocked swab (Swab B) with the scoreline above your hand. Insert the swab 1-2 cm into one of the anterior nares. Rotate the swab against the nasal mucosa for about 3 seconds and withdraw. Repeat with the other anterior nare using the same swab.

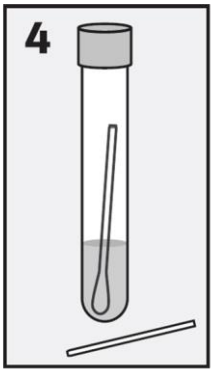
   Do not let the swab touch any surface before placing it into the collection tube.



2. **ALIGN:** Remove the cap from the **cobas**® PCR Media Tube and lower the swab specimen into the tube until the visible scoreline on the swab is aligned with the tube rim.



3. **BREAK:** Carefully leverage the swab against the tube rim to break the swab shaft at the scoreline.



4. **CLOSE: Tightly** re-cap the **cobas**® PCR Media Tube. The specimen is now ready for transport. Discard the top portion of the swab.

- Collect nasal specimens according to standard collection technique using flocked or polyester-tipped swabs and immediately place in 3 mL of 0.9% physiological saline.

Sample collection – saliva

- Do NOT eat or brush teeth 60 minutes before collecting the saliva sample.
- Collect saliva on the floor of your mouth and allow to pool without swallowing. Do not cough.
- Spit passively collected saliva into a sterile collection container.
- Repeat the procedure above until at least 1 mL, but no more than 5 mL of saliva are collected in the container.
- Replace cap on the saliva collection container.
- Return the saliva collection container.

Raw saliva needs to be liquified within 9 days of collection (48 hours at 2-25°C followed by 7 days below -18°C) by estimating the volume of saliva and adding double the amount of 0.9% physiological saline solution to the saliva collection container. After addition of saline, the sample needs to be mixed (e.g., vortex 10 – 20 seconds) prior to further storage or processing.

Transport and storage – swab specimen types

- Transportation of collected specimens must comply with all applicable regulations for the transport of etiologic agents.
- Samples collected in UTM-RT®,
    - After collection, specimens can be stored for up to 48 hours at 2-25°C followed by up to 3 days at 2-8°C and at ≤ -70°C for up to 30 days.
- Samples collected in **cobas**® PCR Media,
    - After collection, specimens can be stored for up to 24 hours at 2-25°C followed by up to 3 days at 2-8°C and at ≤ -70°C for up to 30 days.
- Samples collected in 0.9% physiological saline,
    - After collection, specimens can be stored for up to 48 hours at 2-25°C followed by up to 3 days at 2-8°C and at ≤ -70°C for up to 30 days.
- Specimen are stable for up to two freeze/thaw cycles when frozen at ≤ -70°C.

Transport and storage – saliva

- Transportation of collected specimens must comply with all applicable regulations for the transport of etiologic agents.
- Raw saliva samples collected in a sterile polypropylene collection device,
    - After collection, specimens can be stored for up to 48 hours at 2-25°C followed by up to 7 days ≤ -18°C.
- Liquified saliva,
    - After addition of 2 parts of 0.9% physiological saline solution and intensive mixing, the liquefied saliva specimens can be stored for up to 48 hours at 2-25°C followed by up to 7 days at 2-8°C.

Instructions for use

Procedural notes

- Do not use **cobas**® SARS-CoV-2 Qualitative, **cobas**® SARS-CoV-2 Qualitative Control Kit, **cobas**® Buffer Negative Control Kit, or **cobas omni** reagents after their expiry dates.
- Do not reuse consumables. They are for one-time use only.
- Ensure that specimen barcode labels on sample tubes are visible through the openings on the side of the sample racks. Refer to the **cobas**® 5800 System or **cobas**® 6800/8800 Systems User Guide for proper barcode specifications and additional information on loading sample tubes.
- Refer to the **cobas**® 5800 System or **cobas**® 6800/8800 Systems – User Assistance and/or User Guides for proper maintenance of instruments.

Running cobas® SARS-CoV-2 Qualitative utilizing swab specimens

For testing swab specimens, **cobas**® SARS-CoV-2 Qualitative can be run with a minimum required sample volume of 0.6 mL in the **cobas omni** Secondary Tube for specimens collected in Copan Universal Transport Medium (UTM-RT), BD™ Universal Viral Transport (UVT), **cobas**® PCR Media or 0.9% physiological saline. Specimens collected using **cobas**® PCR Media Uni Swab Sample Kit or **cobas**® PCR Media Dual Swab Sample Kit can be run in their primary collection tube with a minimum required sample volume of 1.0 mL.

Specimens collected in cobas® PCR Media, 0.9% physiological saline, UTM-RT or UVT

Specimens collected in tubes compatible with the **cobas**® 5800 and **cobas**® 6800/8800 Systems may be loaded directly onto the **cobas**® 5800 and **cobas**® 6800/8800 Systems. Specimens collected in Copan Universal Transport Medium (UTM-RT), BD™ Universal Viral Transport (UVT), **cobas**® PCR Media or 0.9% physiological saline tubes which are not compatible with the **cobas**® 5800 and **cobas**® 6800/8800 Systems must be transferred into a secondary tube prior to processing on the **cobas**® 5800 and **cobas**® 6800/8800 Systems. The **cobas omni** Secondary Tube is the preferred option. Samples should be processed using the sample type selection in the user interface (UI) as described in Table 13. Additional tubes for testing **cobas**® SARS-CoV-2 Qualitative are available. Contact your local Roche representative for detailed testing instructions and an order list of primary tubes and secondary tubes compatible with the instruments.

Always use caution when transferring specimens from a primary collection tube to a secondary tube.

Use pipettes with aerosol-barrier or positive-displacement tips to handle specimens.

Always use a new pipette tip for each specimen.

Ensure samples are equilibrated to room temperature prior to transfer into a cobas omni Secondary Tube.

Follow the steps below to transfer patient sample from a primary collection tube into a **cobas omni** Secondary Tube:

- Unscrew the primary sample tube cap.
- Lift the cap and any attached swab to allow a pipette to be inserted into the sample tube. ·
- Transfer 0.6 mL into the prepared barcoded secondary tube. ·
- Transfer secondary tube to a rack. Close the primary sample tube cap.

Specimens collected using cobas® PCR Media Uni or Dual Swab Sample Kit or cobas® PCR Media together with cobas® Uni Swab 100 Kit

Samples collected using **cobas**® PCR Media Uni Swab Sample Kit or **cobas**® PCR Media Dual Swab Sample Kit or **cobas**® PCR Media together with the **cobas**® Uni Swab 100 Kit must be uncapped and can be loaded directly onto racks for processing on the **cobas**® 5800/6800/8800 Systems. Transfer into a secondary tube is not necessary. **cobas**® PCR Media tubes fit on to the MPA RACK 16 MM LIGHT GREEN 7001-7050 (P/N 03143449001) or the 16-position tube carrier (P/N 09224319001) and can be processed with the swab remaining in the tube. Samples collected using **cobas**® PCR Media Uni Swab Sample Kit or **cobas**® PCR Media Dual Swab Sample Kits or **cobas**® PCR Media together with the **cobas**® Uni Swab 100 Kit should be processed using the '**cobas**® PCR media swab' sample type selection in the user interface (UI) of the **cobas**® SARS-CoV-2 Qualitative as described in Table 13.

A properly collected swab specimen should have a single swab with the shaft broken at the scoreline. Swab shafts which are broken above the score line will appear longer than normal and may also be bent over to fit into the **cobas**® PCR Media tube. This may create an obstruction to the pipetting system which might cause the loss of sample, test results and/or mechanical damage to the instrument. In the event that a swab specimen has an improperly broken shaft, remove the swab prior to sample processing on the **cobas**® 5800/6800/8800 Systems. Use caution when disposing of specimen swabs; avoid splashing or touching swabs to other surfaces during disposal to prevent contamination.

Incoming **cobas**® PCR Media primary swab specimen tubes with no swabs or with two swabs have not been collected according to the instructions in their respective collection kit instruction for use and should not be tested. If the sample containing two swabs in the **cobas**® PCR Media primary tubes must be tested, transfer 0.6 mL into the prepared barcoded secondary tube.

Occasionally, incoming swab specimens contain excessive mucus which may induce a pipetting error (e.g., clot or other obstruction) on the **cobas**® 5800/6800/8800 Systems. Prior to retesting of specimens that exhibited clots during initial processing, remove and discard the swab, then re-cap and vortex these specimens for 30 seconds to disperse the excess mucus.

Swab specimens can be processed twice on the **cobas**® 5800/6800/8800 Systems while the swab is in the collection tube. If additional testing is required, or if the first test fails due to specimen pipetting error (e.g., clot or other obstruction), the swab must be removed and the remaining fluid must have a minimum volume of 1.0 mL.

Running cobas® SARS-CoV-2 Qualitative utilizing saliva specimens

Raw saliva specimens collected in a sterile polypropylene container need to be liquified prior to processing. For liquifaction, the volume of raw saliva is estimated and the double amount of 0.9% physiological saline is added. The collection device must be recapped and the solution mixed (e.g., vortex 10 – 20 seconds) prior to the required transfer into a secondary tube and processing on the **cobas**® 5800/6800/8800 Systems. The **cobas omni** Secondary Tube is the preferred option. Liquified saliva samples transferred to **s**econdary tubes should be processed using the 'Saliva' sample type selection in the user interface (UI) of the **cobas**® SARS-CoV-2 Qualitative.

Always use caution when transferring specimens from a primary collection container to the secondary tube.

Use pipettes with aerosol-barrier or positive-displacement tips to handle specimens.

Always use a new pipette tip for each specimen.

Ensure samples are equilibrated to room temperature prior to liquifaction and transfer into a secondary tube.

Follow the steps below to transfer saliva samples from a primary collection container into a secondary tube:

- Unscrew the primary sample container cap and lift the cap.
- Estimate the volume of raw saliva and add the double amount of 0.9% physiological saline.
- Recap the container and mix (e.g., vortex 10-20 seconds) until a homogenous solution results.
- Unscrew the primary sample container cap and lift the cap.
- Transfer 1.2 mL into the prepared barcoded secondary tube.
- Close the primary sample container cap.
- Transfer secondary tube to the rack.

Table 13 Sample type selection in the user interface of the **cobas**® SARS-CoV-2 Qualitative

Collection kit/Matrix type	Minimum volume (mL) Processing tube	Process as Sample Type
Copan Universal Transport Medium BD™ Universal Viral Transport 0.9% physiological saline **cobas**® PCR Media Kit	0.6 mL **cobas omni** Secondary Tube	**VTM**
cobas® PCR Media Uni or Dual Swab Sample Kit **cobas**® PCR Media Kit together with the **cobas**® Uni Swab 100 Kit	1.0 mL Primary tube	**cobas**® **PCR media swab**
Liquified Saliva in a sterile polypropylene container	1.2 mL **cobas omni** Secondary Tube	**Saliva**

Sample pooling for SARS-CoV-2 testing

Pools of up to and including 6 samples may be tested using **cobas**® SARS-CoV-2 Qualitative. The pool size implemented by the laboratory should be based on the required efficiency gains, the positivity rate of SARS-CoV-2 in the testing population, and the potential risks of testing in pools. Combination of multiple sample types in a pool has not been validated.

When resource availability is sufficient to meet testing demand, it is recommended that laboratories consider whether the risks of reduced test sensitivity with pooling outweigh the benefits of resource conservation.

- Use a process that ensures traceability between individual sample IDs and pool IDs.
- To reduce potential contamination of the **cobas**® 5800/6800/8800 Systems, do not transfer samples into the secondary tubes while the samples are in the Roche 5 position racks (RD5 and/or MPA and /or 16 position tube carrier).
- Ensure appropriate sample handling techniques to reduce the risk of cross-contamination of pools and original patient samples.

Note: Sample pooling does not apply to saliva specimen.

Pooling methods

1. Identify a uniquely labeled secondary tube for pooling.
2. Associate the samples to be pooled with the pool tube identification using either a pooling worksheet or validated sample tracking system.
3. Roche suggests using Biological Safety Cabinet or other approved safety measures during sample handling (i.e., sample transfer to secondary tube).
4. For manual pooling, it is recommended to work with only the samples for one pool at a time.
5. Ensure each sample has sufficient volume for pool construction and any possible resolution testing (pool deconvolution) that may be required. Example: for pools of 6, a minimum volume of 100 µL (for pool) plus 600 µL (for resolution) are required for a minimum sample volume of 700 µL available prior to pooling (Table 14).

Table 14 Minimum sample volumes for pooling

Pool Size	Volume required for pool (mL)	Volume required for resolution testing (mL)	Minimum volume required prior to pooling (mL)
6	0.100	0.600	0.700
5	0.120	0.600	0.720
4	0.150	0.600	0.750
3	0.200	0.600	0.800
2	0.300	0.600	0.900

6. Using a calibrated micropipettor with a fresh pipette tip for each sample, carefully pipette each individual sample associated with that pool into the appropriate secondary tube to prepare the pool.
7. Ensure complete mixing after addition of all samples to the secondary tube (i.e., through pipetting up and down). Use caution to avoid creating bubbles, foam or aerosols while mixing.
8. For manual pooling, it is recommended to visually compare the pooled sample volume in the secondary tube to a secondary tube containing the target pool volume. If the pooling tube level is less or more than the standard pool volume, then the manually prepared pool should be discarded and prepared again.
9. Process pooled samples as described in Figure 1 and Figure 2.

Pool result reporting and follow-up testing

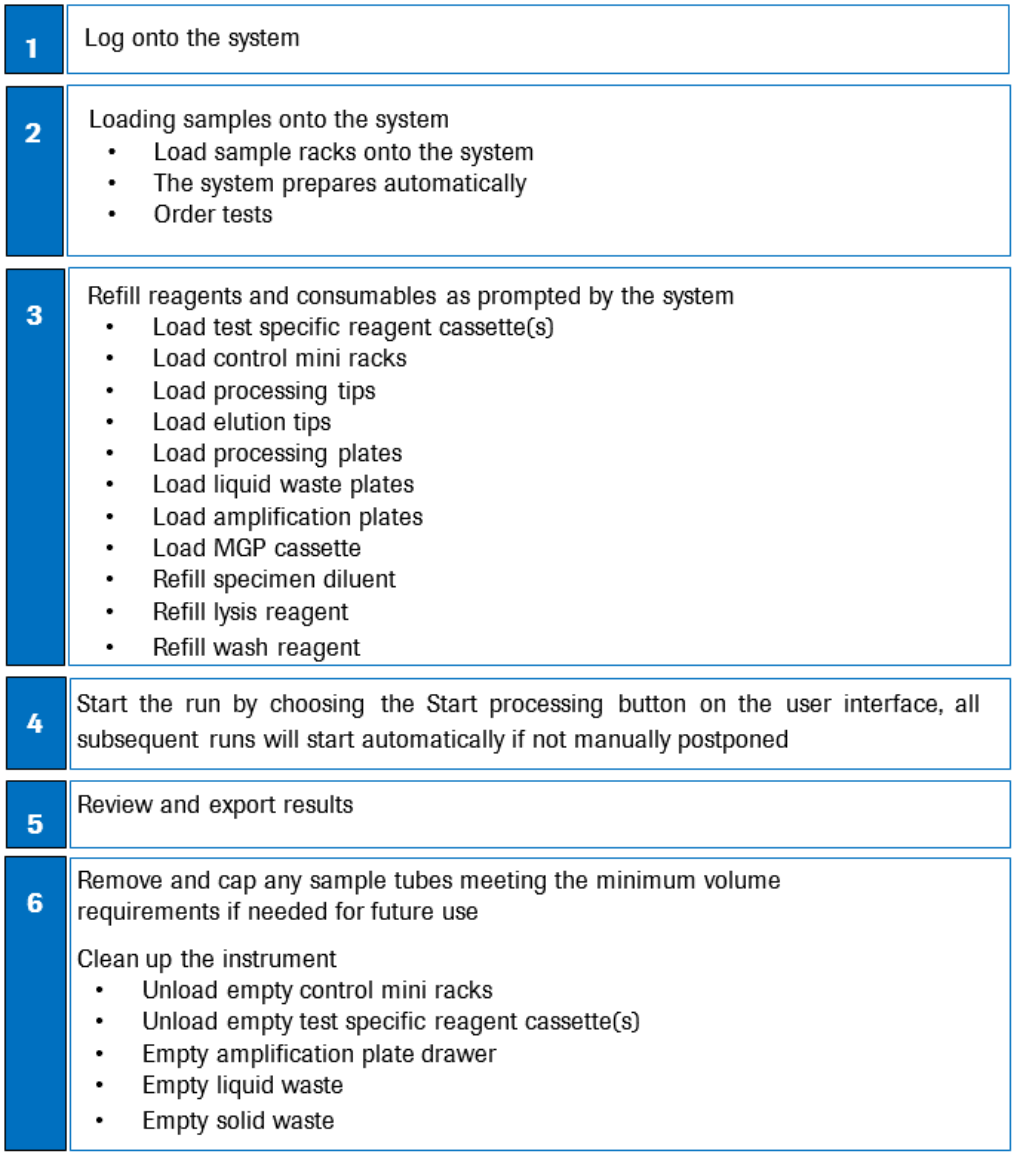
Interpretation of pool results is the same as for individual results as described in the **Interpretation of results** section.

- If the result of the pool is negative, then each constituent sample can be reported as negative. The result report should include a comment that pooling was used during testing. Refer to the **Warnings and precautions** section for additional information regarding decreased sensitivity of pool testing.
- If the result of the pool is positive or presumptive positive, then each of the constituent samples must be re-tested as a separate individual sample. Use the laboratory defined tracking system to ensure the correct individual samples are tested. Individual test results supersede the pool result.

Running cobas® SARS-CoV-2 Qualitative on cobas® 5800 System

The test procedure is described in detail in the **cobas**® 5800 Systems User Assistance and/or User Guide. Figure 1 below summarizes the procedure.

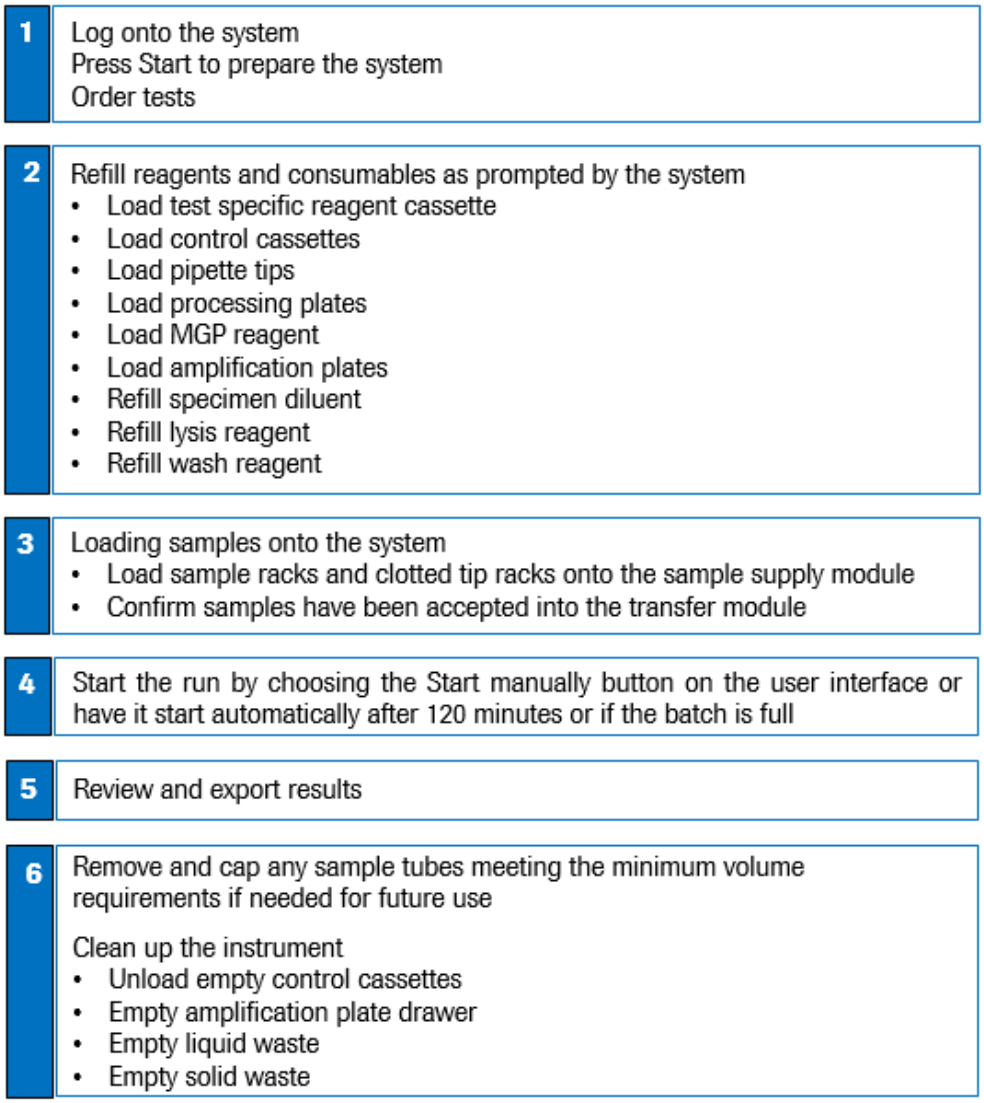
Figure 1 **cobas**® SARS-CoV-2 Qualitative test procedure on the **cobas**® 5800 System



Running cobas® SARS-CoV-2 Qualitative on cobas® 6800/8800 Systems

The test procedure is described in detail in the **cobas**® 6800/8800 Systems – User Assistance and/or User Guide. Figure 2 below summarizes the procedure.

Figure 2 **cobas**® SARS-CoV-2 Qualitative procedure on the **cobas**® 6800/8800 Systems



Results

The **cobas**® 5800/6800/8800 Systems automatically detect the SARS-CoV-2, for each individually processed sample and control, displaying individual target results for samples as well as test validity and overall results for controls.

Quality control and validity of results on the cobas® 5800 System

- One **cobas**® Buffer Negative Control [BUF (-) C] and one positive control [SARS-CoV-2 QL (+) C] are processed at least every 72 hours and with every new kit lot. Positive and/or negative controls can be scheduled more frequently based on laboratory procedures and/or local regulations.
- In the **cobas**® 5800 System software and/or report, check for flags and their associated results to ensure the result validity.

Invalidation of results is performed automatically by the **cobas**® 5800 software based on negative or positive control failures.

NOTE: The **cobas**® 5800 System will be delivered with the standard setting of running a set of controls (positive and negative) with every run, but can be configured to a less frequent scheduling up to every 72 hours based on laboratory procedures and/or local regulations. Please contact your Roche service engineer and/or Roche customer technical support for more information.

Control results on cobas® 5800 System

The results of the controls are shown in the **cobas**® 5800 software in the "Controls" app.

- Controls are marked with "Valid" in the column "Control result" if all Targets of the control are reported valid. Controls are marked with "Invalid" in the column "Control result" if all or one Target of the control are reported invalid.
- Controls marked with "Invalid" show a flag in the "Flags" column. More information on why the control is reported invalid including flag information is shown in the detail view
- If one of the controls is invalid, repeat testing of all controls and all associated samples is required.

Interpretation of results on the cobas® 5800 System

The results of the samples are shown in the **cobas**® 5800 System software in the "Results" app.

For a valid control batch, check each individual sample for flags in the **cobas**® 5800 System software and/or report. The result interpretation should be as follows:

Table 15 Example of **cobas**® SARS-CoV-2 Qualitative results display **cobas**® 5800 System

Sample ID*	Test	Control Result	Flags**	Status	Result		Creation date/time
Sample_01	SCoV2-QL	Valid		Released	SCoV2 Negative	PanSarb Negative	7/7/2021 8:27:39 AM
Sample _C1	SCoV2-QL	Invalid		Released	Invalid	Invalid	7/7/2021 8:27:39 AM
Sample _B1	SCoV2-QL	Valid		Released	SCoV2 Negative	PanSarb Positive	7/7/2021 8:27:39 AM
Sample _B2	SCoV2-QL	Valid		Released	SCoV2 Positive	PanSarb Positive	7/7/2021 8:27:39 AM

Sample _D1	SCoV2-QL	Valid		Released	SCoV2 Negative	PanSarb Negative	7/7/2021 8:27:39 AM
Sample _A6	SCoV2-QL	Valid		Released	SCoV2 Positive	PanSarb Negative	7/7/2021 8:27:39 AM
Sample _E1	SCoV2-QL	Valid		Released	SCoV2 Positive	Invalid	7/7/2021 8:27:39 AM
Sample _A2	SCoV2-QL	Valid		Released	Invalid	PanSarb Positive	7/7/2021 8:27:39 AM

*Table applies for all sample types used.

** The result overview shows a flag symbol in case of invalid results. Detailed flag descriptions are available in the result details.

- Samples associated with a valid control batch are shown as "Valid" in the "Control result" column if all Control Target Results reported valid. Samples associated with a failed control batch are shown as 'Invalid' in the "Control result" column if Control Results are reported invalid.
- If the associated controls of a sample result are invalid, a specific flag will be added to the sample result as follows:
  - Q05D: Result validation failure because of an invalid positive control.
  - Q06D: Result validation failure because of an invalid negative control.
- The values in "Result" column for individual sample target result should be interpreted as show in Table 15 above.
- If one or more sample targets are marked with "Invalid" the **cobas**® 5800 software shows a flag in the "Flags" column. More information on why the sample target(s) is reported invalid including flag information is shown in the detail view.

Quality control and validity of results on the cobas® 6800/8800 Systems

- One **cobas**® Buffer Negative Control [BUF (-) C] and one Positive Control [SARS-CoV-2 QL (+)C] are processed with each batch.
- In the **cobas**® 6800/8800 Systems software and/or report, check for flags and their associated results to ensure the batch validity.
- All flags are described in the **cobas**® 6800/8800 Systems User Guide.
- The batch is valid if no flags appear for any controls. If the batch is invalid, repeat testing of the entire batch.

Validation of results is performed automatically by the **cobas**® 6800/8800 Systems software based on negative and positive control performance.

Interpretation of results on the cobas® 6800/8800 Systems

For a valid batch, check each individual sample for flags in the **cobas**® 6800/8800 Systems software and/or report. The result interpretation should be as follows:

- A valid batch may include both valid and invalid sample results.
- Results display examples for **cobas**® SARS-CoV-2 Qualitative are shown in Table 16.
- The "Valid" and "Overall Result" columns are not applicable to sample results for the **cobas**® SARS-CoV-2 Qualitative.
- Invalid results for one or more target combinations are possible and are reported out specifically for each target. If any individual target result is invalid, the presence or absence of that individual target cannot be determined.
- Other initial valid target results can be interpreted as described in Table 17.

Table 16 Example of **cobas**® SARS-CoV-2 Qualitative results display on **cobas**® 6800/8800 Systems

Test	Sample ID	Valid*	Flags	Sample type	Overall result*	Target 1	Target 2
SCoV2- QL	Sample _01	NA		VTM	NA	SCoV2 Negative	PanSarb Negative
SCoV2- QL	Sample _C1	NA	Y40T	VTM	NA	Invalid	Invalid
SCoV2- QL	Sample _B1	NA		VTM	NA	SCoV2 Negative	**PanSarb Positive**
SCoV2- QL	Sample _B2	NA		VTM	NA	**SCoV2 Positive**	**PanSarb Positive**
SCoV2- QL L	Sample _D1	NA		VTM	NA	SCoV2 Negative	PanSarb Negative
SCoV2- QL	Sample _A6	NA		VTM	NA	**SCoV2 Positive**	PanSarb Negative
SCoV2- QL	Sample _E1	NA	C01H2	VTM	NA	**SCoV2 Positive**	Invalid
SCoV2- QL	Sample _A2	NA	C01H1	VTM	NA	Invalid	**PanSarb Positive**
SCoV2- QL	C161420284090428828404	Yes		(-) Ctrl	Valid	Valid	Valid
SCoV2- QL	C161420284093009580264	Yes		SCoV2-QL (+) C	Valid	Valid	Valid

The "Valid" and "Overall Result" columns are not applicable to sample results for the **cobas® SARS-CoV-2 Qualitative. Refer to Table 17, **cobas**® SARS-CoV-2 Qualitative results interpretation, for specific instructions on test results interpretation.

Table 17 cobas® SARS-CoV-2 Qualitative results interpretation

Target 1	Target 2	Interpretation
SCoV2 Positive	**PanSarb Positive**	All Target Results were valid. Result for SARS-CoV-2 RNA is Detected.
SCoV2 Positive	**PanSarb Negative**	All Target Results were valid. Result for SARS-CoV-2 RNA is Detected. A positive Target 1 result and a negative Target 2 result is suggestive of 1) a sample at concentrations near or below the limit of detection of the test, 2) a mutation in the Target 2, target region, or 3) other factors.
SCoV2 Negative	**PanSarb Positive**	All Target Results were valid. Result for SARS-CoV-2 RNA is Presumptive Positive. A negative Target 1 result and a positive Target 2 result is suggestive of 1) a sample at concentrations near or below the limit of detection of the test, 2) a mutation in the Target 1 target region in the oligo binding sites, or 3) infection with some other Sarbecovirus (e.g., SARS-CoV or some other Sarbecovirus previously unknown to infect humans), or 4) other factors. For samples with a Presumptive Positive result, additional confirmatory testing may be conducted, if it is necessary to differentiate between SARS-CoV-2 and SARS-CoV-1 or other Sarbecovirus currently unknown to infect humans, for epidemiological purposes or clinical management.
SCoV2 Negative	**PanSarb Negative**	All Target Results were valid. Result for SARS-CoV-2 RNA is Not Detected.
SCoV2	**Invalid**	Not all Target Results were valid. Result for SARS-CoV-2 RNA is Detected.
Invalid	**PanSarb Positive**	Not all Target Results were valid. Result for SARS-CoV-2 RNA is Presumptive Positive. For samples with a Presumptive Positive result, additional confirmatory testing may be conducted, if it is necessary to differentiate between SARS-CoV-2 and SARS-CoV-1 or other Sarbecovirus currently unknown to infect humans, for epidemiological purposes or clinical management.
SCoV2 Negative	**Invalid**	Not all Target Results were valid. Sample should be retested. If the result is still invalid, a new specimen should be obtained.
Invalid	**PanSarb Negative**	Not all Target Results were valid. Sample should be retested. If the result is still invalid, a new specimen should be obtained.
Invalid	**Invalid**	All Target Results were invalid.* Sample should be retested. If the result is still invalid, a new specimen should be obtained.

*For further details also refer to section **Procedural limitations**

Procedural limitations

- **cobas**® SARS-CoV-2 Qualitative has been evaluated only for use in combination with the **cobas**® SARS-CoV-2 Qualitative Control Kit, **cobas**® Buffer Negative Control Kit, **cobas omni** MGP Reagent, **cobas omni** Lysis Reagent, **cobas omni** Specimen Diluent, and **cobas omni** Wash Reagent for use on the **cobas**® 5800/6800/8800 Systems.
- Detection of SARS-CoV-2 RNA may be affected by sample collection methods, patient factors (e.g., presence of symptoms), and/or stage of infection.
- Reliable results depend on proper sample collection, storage and handling procedures.
- Due to the nature of the saliva sample type and the individual patient sample variability, an increased invalid and clot rate may occur. Additionally, food particles and increased levels of mucin indicated by potentially discolored samples might cause failures in processing of the saliva specimen. Refer to the **Sample collection – saliva** section for the appropriate precautions to be taken during collection to assure optimal performance.
- Occasionally, incoming saliva specimens may induce a pipetting error (e.g. clot or other obstruction) on the **cobas**® 5800/6800/8800 Systems. A potential additional processing step prior to retesting the specimens that exhibited clots during initial processing is to centrifuge the samples at 2000g for 1 minute and reload the samples on the instrument. Aerosols might occur during centrifugation. To avoid any contamination or transmission of the virus, please handle centrifuged samples carefully .
- This test is intended to be used for the detection of SARS-CoV-2 RNA in nasal, nasopharyngeal and oropharyngeal swab samples collected in a Copan UTM-RT System (UTM-RT) or BD™ Universal Viral Transport System (UVT) and nasal swab samples collected in **cobas**® PCR Media and 0.9% physiological saline. Additionally the test is intended to be used for the detection of SARS-CoV-2 RNA in saliva specimens liquified with 0.9% physiological saline. Testing of other sample types with **cobas**® SARS-CoV-2 Qualitative may result in inaccurate results.
- As with any molecular test, mutations within the target regions of **cobas**® SARS-CoV-2 Qualitative could affect primer and/or probe binding resulting in failure to detect the presence of virus.
- Due to inherent differences between technologies, it is recommended that, prior to switching from one technology to the next, users perform method correlation studies in their laboratory to qualify technology differences. One hundred percent agreement between the results should not be expected due to aforementioned differences between technologies. Users should follow their own specific policies/procedures.
- False negative or invalid results may occur due to interference. The Internal Control is included in **cobas**® SARS-CoV-2 Qualitative to help identify the specimens containing substances that may interfere with nucleic acid isolation and PCR amplification.
- The addition of AmpErase enzyme into the **cobas**® SARS-CoV-2 Qualitative Master Mix reagent enables selective amplification of target RNA; however, good laboratory practices and careful adherence to the procedures specified in this Instructions For Use document are necessary to avoid contamination of reagents.

Use of pooling based on prevalence

Pooling may increase throughput in laboratories testing samples from populations with low prevalence of SARS-CoV-2. In populations with higher prevalence, smaller pool sizes or individual sample testing may be indicated.

When considering pooling strategies, laboratories should consider the appropriateness of the pooling strategy based on the positivity rate in the testing population and efficiency of the pooling workflow. Laboratories may also consider the sensitivity of pooled testing based on the assay's limit of detection.

Table 18 provides estimated maximal efficiency gained based on N-sample pooling and on the percent of SARS-CoV-2 positive samples in a population.

Table 18 Efficiency of pooling based on prevalence

P, percent of positive subjects in the tested population	$n_{maxefficiency}$ (n corresponding to the maximal efficiency)	Efficiency (F) of n-sample pooling (a maximum increase in the number of tested patients when Dorfman n-pooling strategy used)
1% - 4%	6	4.44 - 2.60
5% - 6%	6	2.32 - 2.10
7% - 12%	6	1.92 - 1.42
13% - 25%	6	1.36 - 1.01
1% - 4%	5	4.02 - 2.60
5% - 6%	5	2.35 - 2.15
7% - 12%	5	1.98 - 1.49
13% - 25%	5	1.43 - 1.04
1% - 4%	4	3.46 - 2.50
5% - 6%	4	2.30 - 2.13
7% - 12%	4	1.99 - 1.54
13% - 25%	4	1.48 - 1.07
1% - 4%	3	2.75 - 2.23
5% - 6%	3	2.10 - 1.99
7% - 12%	3	1.89 - 1.53
13% - 25%	3	1.48 - 1.10
1% - 4%	2	1.92 - 1.73
5% - 6%	2	1.67 - 1.62
7% - 12%	2	1.57 - 1.38
13% - 25%	2	1.35 - 1.07

Because a positive pool requires individual retesting of each sample in the pool, the efficiency of any pooling strategy depends on the positivity rate. The efficiency (F) of n-sample pooling for positivity rate (P) can be calculated using the following formula F=1/(1+1/n-(1-P)n). The efficiency (F) indicates how many more samples can be tested with n-sample pools compared to individual testing. For example, a 6-sample pooling strategy increases the number of tested samples by 2.10 times for positivity rate P of 6% (F = 2.10). At F = 2.10, 1,000 tests can cover testing of 2,100 samples on average.

Non-clinical performance evaluation

Analytical sensitivity (Limit of Detection) – swab specimen types

Limit of detection (LoD) studies determine the lowest detectable concentration of SARS-CoV-2 at which greater or equal to 95% of all (true positive) replicates tested positive.

To determine the LoD, the WHO International Standard for SARS-CoV-2 RNA (NIBSC code: 20/146) was serially diluted in simulated clinical matrix. A total of 5 concentration levels, and 3 independent dilution series, were tested with a total of 24 replicates per concentration and lot, with an additional 60 replicates of a blank sample (i.e., clinical sample pools).

As shown in Table 19 and Table 20, the concentration level with observed hit rates greater than or equal to 95% were 250 and 125 IU/mL for SARS-CoV-2 (Target 1) and pan-Sarbecovirus (Target 2), respectively and the Probit predicted 95% hit rates were 200 and 102 IU/mL for SARS-CoV-2 (Target 1) and pan-Sarbecovirus (Target 2), respectively.

Table 19 Summary of LoD for SARS-CoV-2 using WHO International Standard (NIBSC code: 20/146)

Viral Strain	Kit lot	95% Probit [IU/mL]	95% CI of Probit [IU/mL]	Hit rate ≥ 95% [IU/mL]	Mean Ct at ≥ 95% Hit rate
WHO International Standard for SARS-CoV-2 RNA (NIBSC code: 20/146)	Lot 1	**202**	157-319	**250**	33.2
	Lot 2	**121**	97-183	**125**	34.1
	Lot 3	**259**	196-413	**250**	33.2
	Lot 1-3	**200**	170-252	**250**	33.4

Table 20 Summary of LoD for pan-Sarbecovirus using WHO International Standard (NIBSC code: 20/146)

Viral Strain	Kit lot	95% Probit [IU/mL]	95% CI of Probit [IU/mL]	Hit rate ≥ 95% [IU/mL]	Mean Ct at ≥ 95% Hit rate
WHO International Standard for SARS-CoV-2 RNA (NIBSC code: 20/146)	Lot 1	**83**	64-127	**125**	35.2
	Lot 2	**67**	46-454	**125**	36.0
	Lot 3	**132**	99-233	**125**	34.8
	Lot 1-3	**102**	83-140	**125**	35.3

Further, the sensitivity of the assay was determined using a cultured virus of an isolate from a US patient (USA-WA1/2020, catalog number NR-52281, lot number 70033175, 2.8E+05 $TCID_{50}$/mL[§]) was serially diluted in simulated clinical matrix. A total of 7 concentration levels, with 3-fold serial dilutions between the levels, were tested with a total of 21 replicates per concentration, with an additional 10 replicates of a blank sample (i.e., simulated clinical matrix).

As shown in Table 21 the concentration level with observed hit rates greater than or equal to 95% were 0.009 and 0.003 $TCID_{50}$/mL for SARS-CoV-2 (Target 1) and pan-Sarbecovirus (Target 2), respectively. As shown in Table 22, the Probit predicted 95% hit rates were 0.007 and 0.004 $TCID_{50}$/mL for SARS-CoV-2 (Target 1) and pan-Sarbecovirus (Target 2), respectively.

Table 21 LoD determination using USA-WA1/2020 strain

Strain	Concentration [$TCID_{50}$/mL]	Total valid results	Hit rate [%]**		Mean Ct*	
			Target 1	Target 2	Target 1	Target 2
USA-WA1/2020* (stock concentration 2.8E+05 $TCID_{50}$/mL) Lot 70033175***	0.084	21	100	100	31.0	33.0
	0.028	21	100	100	31.8	34.1
	0.009	21	100	100	32.7	35.2
	0.003	21	38.1	100	33.5	36.4
	0.001	21	0	52.4	n/a	37.9
	0.0003	21	0	14.3	n/a	37.2
	0.0001	21	0	9.5	n/a	38.5
	0 (blank)	10	0	0	n/a	n/a

* The following reagent was deposited by the Centers for Disease Control and Prevention and obtained through BEI Resources, NIAID, NIH: SARS-Related Coronavirus 2, Isolate USA-WA1/2020, NR-52281

** All replicates where Target 1 was positive were also positive for Target 2.

*** Based on the information provided in the Certificate of Analysis from the vendor, 1 TCID50/mL is equal to 7,393 genome equivalents by ddPCR

Table 22 Probit Predicted 95% Hit Rates Using USA-WA1/2020 Strain

Strain	Probit Predicted 95% Hit Rate [$TCID_{50}$/mL]	
	Target 1	Target 2
USA-WA1/2020 (stock concentration 2.8E+05 $TCID_{50}$/mL)	0.007 (95% CI: 0.005 – 0.036)	0.004 (95% CI: 0.002 – 0.009)

Analytical sensitivity (Limit of Detection) – saliva specimen types

Limit of detection (LoD) studies determine the lowest detectable concentration of SARS-CoV-2 at which greater or equal to 95% of all (true positive) replicates tested positive.

To determine the LoD, the WHO International Standard for SARS-CoV-2 RNA (NIBSC code: 20/146) was serially diluted in pools of negative Saliva clinical specimens. A total of 8 concentration levels and 3 independent dilution series/saliva pools, were tested with a total of 32 replicates per concentration and lot, with an additional 96 replicates of a blank sample (i.e., clinical sample pools).

As shown in Table 23 and Table 24, the concentration levels with observed hit rates greater than or equal to 95% were 150 IU/mL and 75 IU/mL for SARS-CoV-2 (Target 1) and pan-Sarbecovirus (Target 2), respectively and the Probit predicted 95% hit rates were 92 and 72 IU/mL for SARS-CoV-2 (Target 1) and pan-Sarbecovirus (Target 2), respectively.

Table 23 Summary of LoD for SARS-CoV-2 using WHO International Standard (NIBSC code: 20/146)

Viral Strain	Kit lot	95% Probit [IU/mL]	95% CI of Probit [IU/mL]	Hit rate ≥ 95% [IU/mL]	Mean Ct at ≥ 95% Hit rate
WHO International Standard for SARS-CoV-2 RNA (NIBSC code: 20/146)	Lot 1	**102**	76-156	**150**	34.0
	Lot 2	**92**	71-140	**150**	33.9
	Lot 3	**82**	64-121	**150**	33.8
	Lot 1-3	**92**	78-114	**150**	33.9

Table 24 Summary of LoD for pan-Sarbecovirus using WHO International Standard (NIBSC code: 20/146)

Viral Strain	Kit lot	95% Probit [IU/mL]	95% CI of Probit [IU/mL]	Hit rate ≥ 95% [IU/mL]	Mean Ct at ≥ 95% Hit rate
WHO International Standard for SARS-CoV-2 RNA (NIBSC code: 20/146)	Lot 1	**62**	48-94	**75**	36.6
	Lot 2	**75**	54-128	**150**	35.6
	Lot 3	**79**	58-130	**75**	36.5
	Lot 1-3	**72**	60-92	**75**	36.5

Inclusivity

The inclusivity of **cobas**® SARS-CoV-2 Qualitative for the detection of SARS-CoV-2 was confirmed by testing nine SARS-CoV-2 strains, including six variant strains. The lowest target analyte at which all four tested replicates were positive are reported in Table 25.

Table 25 Summary of inclusivity

Strain	Catalog Number	Lot Number	Test Concentration with 100% Positivity
Hong Kong/VM20001061/2020	0810590CFHI	325659	1.06E+02 cp/mL
Italy-INMI1	0810589CFHI	325658	1.00E+02 cp/mL
USA-WA1/2020	0810587CFHI	325656	5.03E+01 cp/mL
UK (B.1.1.7)	0810614CFHI	326230	2.4E+01 cp/mL
Japan / Brazil (P.1)	NR-54982	70042875	1.9E+02 cp/mL
South Africa (B.1.351)	0810613CFHI	326229	2.4E+01 cp/mL
US NY (B.1.526)	NR-55359	70043342	1.9E+02 cp/mL
India (B.1.617.1)	NR-55486	70044706	2.5E+02 cp/mL
India (B.1.617.2)	NR-55611	70045238	7.0E+01 cp/mL

Precision

Within-laboratory precision was examined using a panel of SARS-CoV-2 (USA-WA1/2020, heat-inactivated) cultures diluted in simulated clinical matrix in universal transport media. Sources of variability were examined with a panel consisting of three concentration levels, using three lots of **cobas**® SARS-CoV-2 Qualitative reagents and three instruments over a course of 15 instrument days (2 runs/day x 3 instruments x 5 days/instrument) for a total of 30 runs. A description of the precision panel and the observed positivity rates are shown in Table 26. All negative panel members tested negative throughout the study. Analysis of standard deviation and percent coefficient of variation (CV) of the Ct values from tests performed on positive panel members (see Table 27) yielded overall CV percentage ranging from 1.1% to 2.2% for **cobas**® SARS-CoV-2 Qualitative.

Table 26 Summary of within laboratory precision

Target	Panel Member	Level (x LoD)	Positive Results	Total Results	Positivity %	Two-sided 95% CI Lower Bound	Two-sided 95% CI Upper Bound
Target 1 (SARS-CoV-2)	Weak positive	~0.3x	9	90	10%	5%	18%
	Low positive	~1.0x	82	90	91%	83%	96%
	Moderate positive	~3.0x	90	90	100%	96%	100%
Target 2 (pan-Sarbecovirus)	Weak positive	~0.3x	31	90	34%	25%	45%
	Low positive	~1.0x	84	90	93%	86%	97%
	Moderate positive	~3.0x	90	90	100%	96%	100%
N/A	Negative	Blank	0	90	0%	0%	4%

Table 27 Overall mean, standard deviation, and percent coefficient of variation for Ct values by positive panel member

Target	Level (x LoD)	Hit rate	Mean Ct	Instrument-to-Instrument		Lot-to-Lot		Day-to-Day		Run-to-Run		Within Run		Total	
				SD	CV %	SD	CV %	SD	CV %	SD	CV %	SD	CV %	SD	CV %
Target 1 (SARS-CoV-2)	~0.3x	10.0%	32.51	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.5	1.4	0.5	1.4
	~1.0x	91.1%	32.1	0.0	0.0	0.2	0.6	0.1	0.3	0.0	0.0	0.6	1.8	0.6	1.9
	~3.0x	100.0%	31.18	0.0	0.0	0.2	0.7	0.0	0.0	0.0	0.0	0.3	0.9	0.4	1.1
Target 2 (pan-Sarbecovirus)	~0.3x	34.4%	35.36	0.0	0.0	0.5	1.3	0.3	0.8	0.1	0.2	0.5	1.5	0.8	2.2
	~1.0x	93.3%	34.21	0.0	0.0	0.1	0.3	0.2	0.6	0.0	0.0	0.7	2	0.7	2.2
	~3.0x	100.0%	32.9	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.4	1.1	0.4	1.1

Analytical specificity / cross-reactivity

A panel of 48 viruses, bacteria, and fungi (including those commonly found in respiratory tract) were tested with **cobas**® SARS-CoV-2 Qualitative to assess analytical specificity. The organisms listed in Table 28 were spiked at concentrations of 1 x 10^5 units/mL for viruses and 1 x 10^6 units/mL for other organisms, unless otherwise noted.

Testing was performed with each potential interfering organism in the absence and presence of SARS-CoV-2 target (spiked at ~3x LoD). None of the organisms interfered with the test performance. Testing of SARS-CoV-1 generated an expected pan-Sarbecovirus positive result.

Table 28 Cross-reactivity test results

Microorganism	Concentration
Human coronavirus 229E	1.0E+05 $TCID_{50}$/mL
Human coronavirus OC43	1.0E+05 $TCID_{50}$/mL
Human coronavirus HKU1	1.0E+05 $TCID_{50}$/mL
Human coronavirus NL63	1.0E+05 $TCID_{50}$/mL
MERS coronavirus	1.0E+05 genomic equivalent/mL
SARS coronavirus	1.0E+05 PFU/mL
Adenovirus B (Type 34)	1.0E+05 $TCID_{50}$/mL
Bocavirus	1.0E+05 cp/mL
Cytomegalovirus	1.0E+05 $TCID_{50}$/mL
Epstein Barr virus	1.0E+05 cp/mL
Human Metapneumovirus (hMPV)	1.0E+05 $TCID_{50}$/mL
Measles virus	1.0E+05 $TCID_{50}$/mL
Mumps virus	1.0E+05 $TCID_{50}$/mL
Parainfluenza virus Type 1	1.0E+05 $TCID_{50}$/mL
Parainfluenza virus Type 2	1.0E+05 $TCID_{50}$/mL
Parainfluenza virus Type 3	1.0E+05 $TCID_{50}$/mL

Microorganism	Concentration
Parainfluenza virus Type 4	1.0E+05 $TCID_{50}$/mL
Influenza A (H1N1)	1.0E+05 $TCID_{50}$/mL
Influenza A virus (H1N1-2009, H1N3, H3N2)	1.0E+05 $TCID_{50}$/mL
Influenza B	1.0E+05 $TCID_{50}$/mL
Enterovirus E (Type 1)	1.0E+05 $TCID_{50}$/mL
Parechovirus	1.0E+05 $TCID_{50}$/mL
Respiratory syncytial virus	1.0E+05 PFU/mL
Rhinovirus	1.0E+05 $TCID_{50}$/mL
Candida albicans	1.0E+06 CFU/mL
Chlamydia pneumoniae	1.0E+06 $TCID_{50}$/mL
Corynebacterium diphtheriae	1.0E+06 CFU/mL
Escherichia coli	1.0E+06 CFU/mL
Haemophilus influenzae	1.0E+06 CFU/mL
Lactobacillus gasseri	1.0E+06 CFU/mL
Legionella pneumophila	1.0E+06 CFU/mL
Legionella jordanis (non-pneumophila)	1.0E+06 CFU/mL
Moraxella catarrhalis	1.0E+06 CFU/mL
Mycobacterium tuberculosis	1.0E+06 cells/mL
Neisseria elongata	1.0E+06 CFU/mL
Neisseria meningitidis	1.0E+06 CFU/mL
Pseudomonas aeruginosa	1.0E+06 CFU/mL
Pneumocystis jirovecii	1:20 of Patient Sample
Staphylococcus aureus	1.0E+06 CFU/mL
Staphylococcus epidermidis	1.0E+06 CFU/mL
Streptococcus pneumonia	1.0E+06 CFU/mL
Streptococcus pyrogenes	1.0E+06 CFU/mL
Streptococcus salivarius	1.0E+06 CFU/mL
Bordetella pertussis	1.0E+06 CFU/mL
Mycoplasma pneumoniae	1.0E+06 CFU/mL
Pooled Nasal Wash	1:20 of Patient Sample

Interference

The effect of exogenous substances potentially secreted into respiratory specimens was evaluated (Table 29). Each potentially interfering substance was tested at or above clinically relevant levels in negative simulated clinical matrix stabilized in universal transport media in absence and presence of SARS-CoV-2 target (spiked at ~3x LoD).

None of the substances interfered with the test performance by generating false-negative, false-positive or invalid results.

Table 29 List of exogenous substances tested for interference

Substance	Concentration
Oxymetazoline	0.011 mg/mL
Galphimia glauca, Luffa operculata, Sabadilla	0.023 mg/mL
Lidocaine and Phenylephrine	2.68 mg/mL
Budesonide	0.039 mg/mL
Phenol	0.47 mg/mL
Fluticasone propionate	166.67 µg/mL
Mupirocin	0.20 mg/mL
Zanamivir	0.0015 mg/mL
Oseltamivir	0.0073 mg/mL
Benzocaine and Menthol	5.00 mg/mL
Tobramycin	0.018 mg/mL
Petroleum Jelly	1% (w/v)
Nicotine	1% (w/v)
Camphor-synthetic eucalyptus oil and menthol ointment	1% (w/v)
0.65% NaCl, Phenylcarbinol, Benzalkonium chloride	1% (w/v)

Endogenous substances that may be present in respiratory specimens were tested for interference (Table 30). Each potentially interfering substance was tested at or above clinically relevant levels in negative simulated clinical matrix stabilized in universal transport media in absence and presence of SARS-CoV-2 target (spiked at ~3x LoD).

None of the substances interfered with the test performance by generating false-negative, false-positive or invalid results.

Table 30 List of endogenous substances tested for interference

Substance	Concentration
Human Genomic DNA	20 ng/µL
Mucus	One sputum swab/mL
Human Peripheral Blood Mononuclear Cells (PBMC)	1.0E+03 cells/µL
Human Whole Blood	1% (v/v)
Human Whole Blood	2% (v/v)
Human Whole Blood	5% (v/v)

Matrix equivalency – UTM-RT/UVT, cobas® PCR Media and 0.9% physiological saline

Equivalence between different collection media (UTM-RT/UVT, **cobas**® PCR Media, and saline) was evaluated. Equivalence between UTM-RT/UVT and **cobas**® PCR Media was evaluated the WHO International Standard for SARS-CoV-2 RNA (NIBSC code: 20/146). The WHO International Standard was used to formulate to a target concentration of approximately 2x LoD (low positive) and 4x LoD (moderate positive) into paired individual negative clinical samples, stabilized either in Universal Transport Media (UTM-RT/UVT) or, **cobas**® PCR Media (CPM).

Equivalence between UTM-RT/UVT and 0.9% physiological saline was evaluated using cultured virus (USA-WA1/2020 strain). The cultured virus was used to formulate to a target concentration of approximately 2X LoD (low positive) and 4X LoD (moderate positive) into paired individual negative clinical samples, stabilized either in Universal Transport Media (UTM-RT/UVT) or 0.9% physiological saline (NaCl).

At least 20 replicates per low positive sample and 10 replicates per moderate positive sample were tested for each collection media type. All replicates tested were positive for SARS-CoV-2 in all the three collection media types. UTM-RT/UVT, **cobas**® PCR Media, and 0.9% physiological saline are acceptable for use with **cobas**® SARS-CoV-2 Qualitative.

Whole system failure

The whole system failure rate was assessed by testing 100 specimens of simulated clinical matrix spiked with WHO International Standard for SARS-CoV-2 RNA (acid-heat inactivated England/02/2020 isolate, NIBSC code: 20/146) to a concentration of approximately 3x LoD. The results of this study determined that all replicates were valid and positive for SARS-CoV-2, resulting in a whole system failure rate of 0% with an upper one-sided 95% confidence interval of 2.95%.

Cross contamination

Studies were performed to evaluate potential cross contamination on the **cobas**® 6800/8800 Systems using **cobas**® SARS-CoV-2 Qualitative. Cross-contamination can cause false positive results. In this performance study, the sample-to-sample cross-contamination rate of **cobas**® SARS-CoV-2 Qualitative was 0.0% (0/239; with an upper one-sided 95% CI of 1.25%) in UTM samples and 0.6% (3/480; with a 95% CI from 0.1% to 1.8%) in Saliva samples when alternating high viral level positive and negative samples were tested over multiple runs. High viral level positive samples in the study were prepared to generate a Ct value that exceeds the 95th percentile of all positive samples observed via real world monitoring of **cobas**® SARS-CoV-2 Qualitative (>10 million results). The likelihood of encountering such specimens in the routine use of **cobas**® SARS-CoV-2 Qualitative is proportional to SARS-CoV-2 prevalence in the testing population. Therefore, the sample-to-sample cross-contamination rate for saliva samples in routine use of **cobas**® SARS-CoV-2 Qualitative will likely be less than 0.6% x 5% x SARS-CoV-2 prevalence (in percent) in the testing population. With an assumed prevalence of 10%, the estimated cross-contamination rate would be 0.6% x 5% x 10% = 0.003%.

Performance in sample pools

The performance of **cobas**® SARS-CoV-2 Qualitative when testing nasopharyngeal samples collected in UTM or UVT was evaluated using one **cobas**® 6800 System and one **cobas**® 8800 System. Thirty positive samples were tested individually and in pools of 6 containing 1 positive and 5 negative samples, and in pools of 3 containing 1 positive and 2 negative samples. Additionally, negative samples were tested individually, in 20 negative pools of 6, and in 20 negative pools of 2.

The 30 individual positive specimens had pan-Sarbecovirus Target 2 Ct values between 15.1 – 35.3, including a subset of 8 Low Positive samples (~27% of the samples) with Target 2 Ct values between 33.4 and 35.3. The Low Positive subset of samples targeted within 2-3 Ct (actual 1.1 – 3) of the mean Ct for Target 2 at the Limit of Detection.

The performance of testing sample pools of 6 and pools of 3 containing one positive sample each, compared to testing individual samples, is shown inTable 31 and Table 32, respectively. Positive and presumptive positive results (as defined in Table 17) were used for the Positive Percent Agreement (pools vs. individual) calculations, as all the constituent samples would require re-testing as separate individual samples. Results are summarized for all samples, and separately summarized for the subset of Low Positive samples, for each tested pool size.

Table 31 Reactivity in positive sample pools of 6

Samples in Pools of 6	Negative Pool Results	Invalid Pool Results	Positive or Presumptive Positive Pool Results	Total N valid Pool Results	Positive Percent Agreement (pools vs, individual)
Positive (Including Low Positive)	0	0	30*	30	100% (30/30) (95% CI: 88.6 – 100%)
Low Positive	0	0	8*	8	100% (8/8) (95% CI: 67.6 – 100%)

*Note: One low positive sample was presumptive positive when tested in a pool of 6.

Table 32 Reactivity in positive sample pools of 3

Samples in Pools of 3	Negative Pool Results	Invalid Pool Results	Positive or Presumptive Positive Pool Results	Total N valid Pool Results	Positive Percent Agreement (pools vs, individual)
Positive (Including Low Positive)	0	0	30	30	100% (30/30) (95% CI: 88.6 – 100%)
Low Positive	0	0	8	8	100% (8/8) (95% CI: 67.6 – 100%)

The performance of testing sample pools of 6 and pools of 2 containing only negative samples compared to testing individual samples, is shown in Table 33.

Table 33 Specificity in negative sample pools of 6 and pools of 2

Pool Size	Negative Pool Results	Invalid Pool Results	Positive or Presumptive Positive Pool Results	Total N valid Pool Results	Observed Negative Rate
Pools of 6	20	0	0	20	100% (20/20) (95% CI: 83.9 – 100%)
Pools of 2	20	0	0	20	100% (20/20) (95% CI: 83.9 – 100%)

Note: Some positive samples may not be detected when diluted and tested in pools. Performance estimations above may underestimate the loss of detection from testing in pools. Laboratories should also consider the assay's limit of detection when evaluating testing in pools (see **Warnings and precautions**).

Clinical performance evaluation

Performance with clinical specimens – swab specimen types

The performance of **cobas**® SARS-CoV-2 Qualitative was evaluated across three studies with archived or fresh, prospectively collected specimens. Combined, all three studies compared the performance of **cobas**® SARS-CoV-2 Qualitative across four external testing sites (one site in the EU, three in the US) using a common highly sensitive CE-IVD SARS-CoV-2 assay as the comparator method. The specimens in all studies were collected into VTM.

The first study consisted of archived nasopharyngeal swab (NPS) specimens from individuals with signs and symptoms of a respiratory infection evaluated at one external site. The second study consisted of one external site evaluating archived specimens from individuals without symptoms or other reasons to suspect COVID-19. The final study was a large multi-center study with three external testing sites evaluating prospectively collected fresh clinical specimens from individuals with signs and symptoms of a respiratory infection. Participants from 12 geographically distributed enrollment centers provided NPS and NS (nasal swab) specimens as part of a dual collection procedure where (a) the collection order was varied such that the first specimen collected will be ~50% NPS and ~50% NS, and (b) the collection method for NS specimens was also varied to yield ~50% self-collected and ~50% healthcare worker–collected.

Across the three studies, a total of 1,500 SARS-CoV-2 nasopharyngeal swab specimen results were evaluable and included in the data analysis. The accuracy (method correlation) of the **cobas**® SARS-CoV-2 Qualitative in comparison to a highly sensitive CE-IVD SARS-CoV-2 assay is shown in Table 34. Overall, the **cobas**® SARS-CoV-2 Qualitative Positive Percent Agreement (PPA) was 97.2% (140/144) and Negative Percent Agreement (NPA) was 99.9% (1,354/1,356).

Table 34 Summary of NPS clinical performance of **cobas**® SARS-CoV-2 Qualitative

Specimen Type	Target	Total (N)	PPA	PPA LCL 95% Score CI	PPA UCL 95% Score CI	NPA	NPA LCL 95% Score CI	NPA UCL 95% Score CI
Nasopharyngeal	SARS-CoV-2	1500	97.2% (140/144)	93.1%	98.9%	99.9% (1,354/1,356)	99.5%	100%

CI= confidence interval, LCL= lower confidence limit, NPA= negative percent agreement, PPA= positive percent agreement, UCL= upper confidence limit.

Additionally, the aforementioned prospective multi-center evaluation study was designed to evaluate the performance of the **cobas**® SARS-CoV-2 Qualitative test using NPS and NS specimens from subjects suspected of respiratory infection. This study used a composite comparator method wherein laboratory sites used up to 3 highly-sensitive CE-IVD SARS-CoV-2 assays to determine the infective status by majority rule. The composite comparator result was defined as the concordant results from 2 comparator assays (test A and test B). In case of discordance between the initial 2 comparator assays, the sample was tested by a third assay (test C) and the result of that test determined the composite comparator status.

When compared with the composite comparator result, **cobas**® SARS-CoV-2 Qualitative yielded a Positive Percent Agreement (PPA) of 98.7% for NPS and 96.2% for NS specimens. The Negative Percent Agreement (NPA) was 99.7% for NPS and 100% for NS specimens. **cobas**® SARS-CoV-2 Qualitative also demonstrated similar performance when using self-collected and healthcare worker collected nasal swab specimens as shown in Table 35.

Table 35 Summary of NPS/NS clinical performance of **cobas**® SARS-CoV-2 Qualitative - prospective evaluation

Specimen Type	Target	Total (N)	PPA	PPA LCL 95% Score CI	PPA UCL 95% Score CI	NPA	NPA LCL 95% Score CI	NPA UCL 95% Score CI
Nasopharyngeal*	SARS-CoV-2	938	98.7% (77/78)	93.1 %	99.8 %	99.7% (857/860)	99.0 %	99.9 %
Nasal Swab	SARS-CoV-2	941	96.2% (76/79)	89.4 %	98.7 %	100.0% (862/862)	99.6 %	100.0 %
Nasal Swab - Self Collected	SARS-CoV-2	481	100.0% (40/40)	91.2 %	100.0 %	100.0% (441/441)	99.1 %	100.0 %
Nasal Swab - HCW Collected	SARS-CoV-2	460	92.3% (36/39)	79.7 %	97.3 %	100.0% (421/421)	99.1 %	100.0 %

CI= confidence interval, LCL= lower confidence limit, NPA= negative percent agreement, PPA= positive percent agreement, UCL= upper confidence limit, HCW= healthcare worker.

* Nasopharyngeal specimen data from the prospective study are included in both Table 34 and Table 35. Test A of the SARS-CoV-2 composite comparator was the same method used as the single comparator in the summary analysis of all three studies.

Performance with clinical specimens – saliva specimen

The performance of **cobas**® SARS-CoV-2 Qualitative was evaluated with prospectively collected specimens. The study compared the performance of **cobas**® SARS-CoV-2 Qualitative at one external testing site within the EU against the paired nasopharyngeal swab result of **cobas**® SARS-CoV-2 Qualitative as the comparator. Nasopharyngeal specimens were collected into RT-UTM and saliva specimens were collected as raw saliva into a sterile device.

The study was evaluating prospectively collected clinical specimens from individuals with signs and symptoms of a respiratory infection, as well from individuals without signs and symptoms of a respiratory infection. Participants provided nasopharyngeal swab and saliva specimens as part of a dual collection procedure.

Paired specimens in a total of 652 subjects were evaluable and included in the data analysis, which included 298 (45.7%) who were symptomatic and 354 (54.3%) who were asymptomatic at the time of sample collection. The accuracy (method correlation) of the **cobas**® SARS-CoV-2 Qualitative using saliva in comparison to the **cobas**® SARS-CoV-2 Qualitative using nasopharyngeal swab specimen is shown in Table 36. Overall, the **cobas**® SARS-CoV-2 Qualitative Positive Percent Agreement (PPA) between the saliva and nasopharyngeal swab swab specimen types was 82.2% (120/146) and Negative Percent Agreement (NPA) was 97.2% (492/506).

Table 36 Summary of clinical performance of **cobas**® SARS-CoV-2 Qualitative using saliva in comparison to NPS

Specimen Type	Target	Total (N)	PPA	PPA LCL 95% Score CI	PPA UCL 95% Score CI	NPA	NPA LCL 95% Score CI	NPA UCL 95% Score CI
Saliva	SARS-CoV-2	652	82.2% (120/146)	75.2%	87.5%	97.2% (492/506)	95.4%	98.3%

CI= confidence interval, LCL= lower confidence limit, NPA= negative percent agreement, PPA= positive percent agreement, UCL= upper confidence limit.

The Positive Percent Agreement of the **cobas**® SARS-CoV-2 Qualitative using saliva in comparison to the **cobas**® SARS-CoV-2 Qualitative using nasopharyngeal swab specimen split into arbitrary viral level groups is shown in Table 37. The **cobas**® SARS-CoV-2 Qualitative Positive Percent Agreement (PPA) between the specimen types saliva and nasopharyngeal swab was 97.9.% (47/48) for NPS samples with a high viral level (Ct of target 1 (SARS-CoV-2) ≤ 23), 100.0% (50/50) for NPS samples with a moderate viral level (Ct of target 1 (SARS-CoV-2) > 23 to 30), and 47.9% (23/48) for NPS samples with a low viral level at and below the Limit of Detection of the NPS sample type (Ct of target 1 (SARS-CoV-2) > 30).

Table 37 Positive Percent Agreement **cobas**® SARS-CoV-2 Qualitative using saliva in comparison to the viral level detected in the paired NPS sample

Viral level based on Ct* of paired NPS specimen	Target	Total (N)	PPA	PPA LCL 95% Score CI	PPA UCL 95% Score CI
High (NPS Ct ≤ 23)	SARS-CoV-2	48	97.9% (47/48)	89.1%	99.6%
Moderate (NPS > Ct 23 to ≤ Ct 30)	SARS-CoV-2	50	100.0% (50/50)	92.9%	100.0%
Low (NPS Ct > 30, at and below LoD of NPS sample type)	SARS-CoV-2	48	47.9% (23/48)	34.5%	61.7%

CI= confidence interval, LCL= lower confidence limit, NPA= negative percent agreement, PPA= positive percent agreement, UCL= upper confidence limit.

* Ct of target 1 (SARS-CoV-2)

Testing of the 40 saliva specimens where results were discrepant between the paired nasopharyngeal swab and saliva specimen by an alternative highly sensitive CE-IVD NAT test resulted in a 100% agreement with the **cobas**® SARS-CoV-2 Qualitative saliva result. All 14 **cobas**® SARS-CoV-2 Qualitative NPS negative, but saliva positive results were confirmed as saliva positive by the alternative test, and all 26 **cobas**® SARS-CoV-2 Qualitative NPS positive, but saliva negative results were confirmed as saliva negative by the alternative test. This indicates that discrepant results when using saliva as a sample type are dependent on the differences between the two specimen types rather than on the assay performance.

Additionally, head-to-head comparison between saliva samples tested with the cobas® SARS-CoV-2 Qualitative and the Hologic Aptima™ SARS-CoV-2 tests was performed (Table 38). The two tests comparably detected SARS-CoV-2 RNA in saliva specimens with an overall Positive Percent Agreement (PPA) of 97.8% (131/134) and a Negative Percent Agreement (NPA) of 99.4% (514/517). The 95% confidence limits ranged from 93.6% to 99.2% for the PPA and from 98.3% to 99.8% for the NPA, respectively. All cobas-/Aptima+ saliva specimens generated negative results for the paired NPS specimens. For the cobas+/Aptima- saliva specimens, two of three results were positive for the paired NPS specimens. The third cobas+/Aptima- saliva specimen tested positive for target 2 only (pan-Sarbecovirus) with a late Ct value indicating a low SARS-CoV-2 RNA level near the limit of detection.

Table 38 Correlation between **cobas**® SARS-CoV-2 Qualitative and Aptima™ SARS-CoV-2 Test

Specimen Type	SARS–CoV-2				PPA [95% Score CI]	NPA [95% Score CI]
	Con +	Con -	cobas+ Aptima -	cobas - Aptima +		
Saliva	131	515	3	3	**97.8%** (131/134) [93.6–99.2%]	**99.4%** (515/518) [98.3–99.8%]

Con = Concordant; + = Positive; - = Negative, CI= confidence interval, NPA= negative percent agreement, PPA= positive percent agreement

Reproducibility

The reproducibility of **cobas**® SARS-CoV-2 Qualitative was evaluated across multiple factors that theoretically could affect reported results, including: reagent lot, testing site/instrument, day, and run. The evaluation was conducted at 3 testing sites, using 3 reagent lots, with a 4-member panel of positive and negative samples resulting in a total number of 216 tests per concentration (not including controls). The positive panel members contained SARS-CoV-2 viral culture material [WHO International Standard for SARS-CoV-2 RNA (NIBSC code: 20/146)] at 3 different concentrations in universal transport medium (UTM) based simulated clinical matrix. Each site tested two reagent lots for 6 days. Two runs were performed each day and 3 replicates of each panel member were performed for each run. An overall SARS-CoV-2 positive result was determined by a positive detection in either or both of the SARS-CoV-2 or/and pan-Sarbecovirus channels. The evaluation results are summarized in Table 39.

The test results showed good lot-to-lot, instrument-to-instrument (site), day-to-day, and between batch variability for the ~0.3x LoD, ~1x LoD, and ~3x LoD panel members (Table 39). Regardless of viral targets and viral concentrations, most of the variability was within batches, ranging from 79.5% to 100%. Site-to-site variability ranged from 0.0% to 10.1%, and between-batch variability ranged from 0.0% to 16.0%.

Table 39 Overall mean estimate, standard deviations, and coefficients of variation (%) for cycle threshold values by viral target and expected viral concentration (positive panel members)

Viral Target	Panel Member Concen-tration	N*/N	Mean Ct**	Site SD	Site CV(%)	Lot SD	Lot CV(%)	Day SD	Day CV(%)	Batch SD	Batch CV(%)	Within Batch SD	Within Batch CV(%)	Total SD**	Total CV(%)***
SARS-CoV-2	~0.3x LoD	45/216	33.6	0.00	0.0	0.00	0.0	0.11	0.3	0.00	0.0	0.35	1.1	0.37	1.1
SARS-CoV-2	~1x LoD	196/216	33.2	0.00	0.0	0.09	0.3	0.00	0.0	0.17	0.5	0.37	1.1	0.42	1.3
SARS-CoV-2	~3x LoD	216/216	32.2	0.05	0.2	0.02	0.1	0.00	0.0	0.03	0.1	0.24	0.8	0.25	0.8
pan-Sarbecovirus	~0.3x LoD	158/216	36.5	0.18	0.5	0.00	0.0	0.00	0.0	0.00	0.0	0.71	2.0	0.74	2.0
pan-Sarbecovirus	~1x LoD	214/216	35.4	0.00	0.0	0.00	0.0	0.00	0.0	0.00	0.0	0.67	1.9	0.67	1.9
pan-Sarbecovirus	~3x LoD	216/216	34.1	0.11	0.3	0.05	0.2	0.00	0.0	0.00	0.0	0.32	0.9	0.34	1.0

Ct = cycle threshold; LoD = limit of detection; SD = standard deviation; CV(%) = percent coefficient of variation; SARS-CoV-2 = severe acute respiratory syndrome coronavirus 2.

Note: SARS-CoV-2 is a dual target assay. Inactivated viral culture material was diluted to ~0.3/1/3x LoD based on the target 2 (SARS-CoV-2) LoD.

* 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.

** The mean and total standard deviation (SD) estimates were calculated from the PROC MIXED procedure.

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

The system showed a 99.1% negative percent agreement with a 95% CI of 96.7 - 99.9%. Of the 216 valid tests, 2 tests were positive (1 each for SARS-CoV-2 and pan-Sarbecovirus). Post-amplification DNA sequencing confirmed the presence of an amplification product in 1 sample (pan-Sarbecovirus positive, Ct 36.7) and did not detect amplification product for either target in the other (SARS-CoV-2 positive, Ct 34.4). The Ct values and the curve analysis of the reactive negative panel member may suggest a low level of contamination during specimen handling.

System equivalency / system comparison

System equivalency of the **cobas**® 5800, **cobas**® 6800 and **cobas**® 8800 Systems was demonstrated via performance studies. The results presented in the Instructions for Use support equivalent performance for all systems.

Additional information

Key test features

Sample type	Nasopharyngeal and oropharyngeal swab samples collected in the Copan UTM-RT System or the BD™ UVT System Nasal swab samples collected in the Copan UTM-RT System, the BD™ UVT System, the **cobas**® PCR Media, and 0.9% physiological saline Saliva samples
Minimum amount of sample required	Swab specimen types: 0.6 or 1.0 mL*,** Liquified Saliva: 1.2 mL
Sample processing volume	Swab specimen types: 0.4 mL Liquified Saliva: 0.85 mL

*Dead volume of 0.2 mL is identified for the **cobas omni** Secondary Tubes. Dead volume of 0.6 mL is identified for the **cobas**® PCR Media primary tubes. Other tubes compatible with **cobas**® 5800/6800/8800 Systems (consult User Assistance Guides) may have different dead volume and require more or less minimum volume.

**Additional volume is required if pooling.

Symbols

The following symbols are used in labeling for Roche PCR diagnostic products.

Table 40 Symbols used in labeling for Roche PCR diagnostics products

Age/DOB	Age or Date of Birth		Device not for near-patient testing	QS IU/PCR	QS IU per PCR reaction, use the QS International Units (IU) per PCR reaction in calculation of the results.
SW	Ancillary Software		Device not for self-testing		
Assigned Range [copies/mL]	Assigned Range (copies/mL)		Distributor *(Note: The applicable country/region may be designated beneath the symbol)*	SN	Serial number
Assigned Range [IU/mL]	Assigned Range (IU/mL)		Do not re-use	Site	Site
EC REP	Authorized representative in the European Community		Female	Procedure Standard	Standard Procedure
BARCODE	Barcode Data Sheet		For IVD performance evaluation only	STERILE EO	Sterilized using ethylene oxide
LOT	Batch code	GTIN	Global Trade Item Number		Store in dark
	Biological risks		Importer		Temperature limit
REF	Catalogue number	IVD	In vitro diagnostic medical device	TDF	Test Definition File
CE	CE marking of conformity; this device is in conformity with the applicable requirements for CE marking of an in vitro diagnostic medical device	LLR	Lower Limit of Assigned Range		This way up
			Male	Procedure UltraSensitive	Ultrasensitive Procedure
Collect Date	Collect date		Manufacturer	UDI	Unique Device Identifier
	Consult instructions for use	CONTROL −	Negative control	ULR	Upper Limit of Assigned Range
Σ	Contains sufficient for <*n*> tests	NON STERILE	Non-sterile	Urine Fill Line	Urine Fill Line
CONTENT	Content of kit		Patient Name	Rx Only	US Only: Federal law restricts this device to sale by or on the order of a physician.
CONTROL	Control		Patient number		Use-by date
	Date of manufacture		Peel here		
	Device for near-patient testing	CONTROL +	Positive control		
	Device for self-testing	QS copies / PCR	QS copies per PCR reaction, use the QS copies per PCR reaction in calculation of the results.		

Technical support

For technical support (assistance) please reach out to your local affiliate:
https://www.roche.com/about/business/roche_worldwide.htm

Manufacturer and importer

Table 41 Manufacturer and importer

Roche Molecular Systems, Inc.
1080 US Highway 202 South
Branchburg, NJ 08876 USA
www.roche.com

Made in USA

Roche Diagnostics GmbH
Sandhofer Strasse 116
68305 Mannheim, Germany

Trademarks and patents

See http://www.roche-diagnostics.us/patents

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EC REP Roche Diagnostics GmbH
Sandhofer Str. 116
68305 Mannheim
Germany

CE IVD

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2. Clinical and Laboratory Standards Institute (CLSI). Protection of laboratory workers from occupationally acquired infections. Approved Guideline-Fourth Edition. CLSI Document M29-A4:Wayne, PA;CLSI, 2014.

Document revision

Document Revision Information	
Doc Rev. 1.0 11/2021	First Publishing.
Doc Rev. 2.0 01/2022	The claim was extended to run additionally on the **cobas**® 5800 System, and with that all information required were added to the whole IFU. Please contact your local Roche Representative if you have any questions.