GenMark Dx
A Member of the Roche Group

cobas® eplex

respiratory pathogen panel 2

package insert

For Use Under Emergency Use Authorization Only

EUA

For *in vitro* Diagnostic Use
For Prescription Use Only

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INTENDED USE

The **cobas® eplex** respiratory pathogen panel 2 (RP2 Panel) is a multiplexed nucleic acid *in vitro* diagnostic test intended for use on the **cobas eplex** Instrument for the simultaneous qualitative detection and differentiation of nucleic acids from multiple respiratory viral and bacterial organisms, including nucleic acid from Severe Acute Respiratory Syndrome coronavirus 2 (SARS-CoV-2), in nasopharyngeal swabs (NPS) eluted in viral transport media obtained from individuals suspected of respiratory viral infection consistent with COVID-19 by their healthcare provider. Clinical signs and symptoms of respiratory viral infection due to SARS-CoV-2 and the targeted respiratory viral and bacterial organisms can be similar. Testing is limited to laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. § 263a, that meet requirements to perform moderate or high complexity tests.

The **cobas eplex** RP2 panel is intended for the detection and differentiation of nucleic acid from SARS-CoV-2 and the following virus types, subtypes, and bacteria: adenovirus, coronavirus (229E, HKU1, NL63, OC43), SARS-CoV-2, human metapneumovirus, human rhinovirus/enterovirus, influenza A, influenza A H1, influenza A H1-2009, influenza A H3, influenza B, parainfluenza virus 1, parainfluenza virus 2, parainfluenza virus 3, parainfluenza virus 4, respiratory syncytial virus (RSV) A, respiratory syncytial virus (RSV) B, *Chlamydia pneumoniae*, and *Mycoplasma pneumoniae*.

SARS-CoV-2 RNA and nucleic acids from the other respiratory viral and bacterial organisms identified by this test are generally detectable in NPS specimens during the acute phase of infection. The detection and identification of specific viral and bacterial nucleic acids from individuals exhibiting signs and/or symptoms of respiratory infection aids in the diagnosis of respiratory infection when used in conjunction with other clinical and epidemiological information. The results of this test should not be used as the sole basis for diagnosis, treatment, or other patient management decisions. Positive results are indicative of active infection with the identified respiratory pathogen but do not rule out infection or co-infection with non-panel organisms. The agent detected by the **cobas eplex** RP2 panel may not be the definitive cause of disease.

Negative results for SARS-CoV-2 and other organisms on the **cobas eplex** RP2 panel may be due to infection with pathogens that are not detected by this test, or lower respiratory tract infection that may not be detected by a nasopharyngeal swab specimen. Negative results do not preclude infection with SARS-CoV-2 or other organisms on the **cobas eplex** RP2 panel 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.

Negative results for other organisms detected by the test may require additional laboratory testing (e.g., bacterial and viral culture, immunofluorescence and radiography) when evaluating a patient with possible respiratory tract infection.

The **cobas eplex** RP2 panel is intended for use by qualified clinical laboratory personnel specifically instructed and trained in performing testing on the **cobas eplex** system and in vitro diagnostic procedures. The **cobas eplex** RP2 panel is only for use under the Food and Drug Administration's Emergency Use Authorization.

SUMMARY AND EXPLANATION OF TEST

The **cobas eplex** RP2 panel is an automated qualitative nucleic acid multiplex *in vitro* diagnostic test for simultaneous detection and identification of multiple respiratory viral and bacterial nucleic acids in nasopharyngeal swabs (NPS) collected in viral transport media (VTM). The test is able to detect 16 respiratory viral targets and 2 bacterial targets as summarized in **Table 1**. This test is performed on the **cobas eplex** instrument.

Respiratory viruses and bacteria are responsible for a wide range of respiratory tract infections including the common cold, influenza, and croup, and represent the most common cause of acute illness. Disease severity can be especially high in the young, the immunocompromised, and elderly patients. Respiratory infections cause more doctor visits and absences from school and work than any other illness.[1] Influenza viruses have a peak season in the winter months in the northern hemisphere and the severity of the flu season varies each year based on the particular strain or strains that are in circulation and how effective the vaccine is for that year.[2] Globally, seasonal influenza results in about 3-5 million severe cases and 250,000 – 500,000 deaths annually.[3] In late 2019, a novel coronavirus was identified in Wuhan, China. The disease caused by this novel coronavirus was initially called "2019 novel coronavirus" or "2019-nCoV" and was later renamed Coronavirus Disease 2019, or COVID-19.[4] As of July 2020, cases have been identified in 188 countries around the world with over 16 million cases and 655,000 deaths.[5]

Influenza-like illness is a nonspecific respiratory illness characterized by fever, fatigue, cough, and other symptoms. The majority of influenza-like illnesses are not caused by influenza but by other viruses (*e.g.*, rhinovirus, respiratory syncytial virus, adenovirus, and parainfluenza virus).[6] Less common causes of influenza-like illness include bacteria such as *Chlamydia pneumoniae* and *Mycoplasma pneumoniae*.[6]

Table 1: Targets detected by the **cobas eplex** RP2 panel

Target	Classification (Genome Type)	Seasonal Prevalence*	Most Commonly Infected Demographic
Adenovirus (A-F)	Adenovirus (DNA)	Late winter to early summer[7]	All ages, immunocompromised[8]
Coronavirus (229E, HKU1, NL63, OC43)	Coronavirus (RNA)	Winter, spring[9]	All ages[9]
SARS-CoV-2	Coronavirus (RNA)	Unknown[4]	Not established[4]
Human Metapneumovirus	Paramyxovirus (RNA)	Winter[10]	Children, elderly, immunocompromised[11]
Human Rhinovirus/ Enterovirus	Picornavirus (RNA)	Fall, spring[12]/ Summer[13]	All ages, immunocompromised[12, 13, 14]
Influenza A	Orthomyxovirus (RNA)	Winter[3]	All ages[3]
Influenza A H1	Orthomyxovirus	Winter[3]	All ages[3]
Influenza A H1-2009	(RNA)	Winter[3]	All ages[3]
Influenza A H3	Orthomyxovirus	Winter[3]	All ages[3]
Influenza B	(RNA)	Winter[3]	All ages[3]
Parainfluenza Virus 1	Paramyxovirus (RNA)	Fall[15]	All ages[16]
Parainfluenza Virus 2	Paramyxovirus	Fall, early winter[15]	All ages[16]
Parainfluenza Virus 3	(RNA)	Spring, summer[15]	All ages[16]

Target	Classification (Genome Type)	Seasonal Prevalence*	Most Commonly Infected Demographic
Parainfluenza Virus 4	Paramyxovirus (RNA)	Fall, early winter[15]	All ages[16]
Respiratory Syncytial Virus A	Paramyxovirus (RNA)	Winter[17, 18]	Infants, children, older adults[17, 18]
Respiratory Syncytial Virus B	Paramyxovirus (RNA)	Winter[17, 18]	Infants, children, older adults[17]
Chlamydia pneumoniae	Bacterium (DNA)	No peak season[19]	All ages, most common in children[19]
Mycoplasma pneumoniae	Bacterium (DNA)	Late summer, fall[20]	Children, young adults[21]

* Based on northern hemisphere seasons

SUMMARY OF DETECTED ORGANISMS

Adenovirus: Adenoviruses are non-enveloped DNA viruses that include seven human species (A - G) and more than 60 serotypes.[22] Adenovirus species B, C, and E are frequently associated with upper respiratory infections; infections are common in children, and outbreaks often occur in crowded environments, such as military barracks.[8] There is no vaccine available to the general public, but the introduction of a live, oral vaccine to the US military in 2011 has reduced the incidence of adenovirus outbreaks in this population.[8, 23] Adenovirus infections generally cause mild illness but can result in severe disease in infants or in immunocompromised patients, particularly in hematopoietic stem cell transplant recipients.[8, 22] In addition to respiratory infections, adenovirus can also cause gastroenteritis, conjunctivitis, and cystitis.[8, 22] Adenovirus species A, D, and F are not typically associated with respiratory infections.

Coronavirus: Human coronaviruses usually cause mild to moderate upper respiratory infections but can cause significant disease in the elderly, young children, and immunocompromised individuals.[24, 25] Infection with coronaviruses 229E, HKU1, NL63, and OC43 is common worldwide.

SARS-CoV-2: In late 2019, a novel coronavirus was identified in Wuhan, China. The disease caused by this novel coronavirus was initially called “2019 novel coronavirus” or “2019-nCoV” and was later renamed Coronavirus Disease 2019, or COVID-19.[4] This novel coronavirus was named Severe Acute Respiratory Syndrome Coronavirus, or SARS-CoV-2 due to genetic similarity to the coronavirus responsible for an outbreak in 2003.[26] As of July 2020, cases have been identified in 188 countries around the world with over 16 million cases and 655,000 deaths.[5]

Human Metapneumovirus: Human metapneumovirus is a member of the paramyxovirus family and is closely related to RSV.[11] Metapneumovirus has been identified as an important respiratory pathogen in young children and is the second most common virus identified in pediatric respiratory tract infections.[11] Illness is more severe in children who are immunocompromised or have underlying conditions, such as HIV or cardiac disease; it can also cause more severe disease in immunocompromised adults, especially those with COPD (chronic obstructive pulmonary disease), asthma, cancer, or in transplant patients.[26]

Human Rhinovirus and Enterovirus: Rhinovirus and enterovirus are closely related RNA viruses in the *Picornaviridae* family.[13, 14] There are more than 100 different serotypes that all share high sequence homology.[25] Rhinovirus causes up to 80% of all cases of the common cold worldwide and is more common in children than adults. It is the cause of a significant number of mild upper respiratory tract

infections throughout the year, especially during the spring and fall seasons.[12, 29] Most infections are mild, but rhinovirus has been associated with severe infections in at-risk populations including young children, the elderly, immunocompromised patients, and those with asthma.[12, 13]

There are 62 non-polio enteroviruses that can cause disease in humans.[14] Enterovirus primarily infects the gastrointestinal tract but can also cause respiratory illness, which is generally mild, like the common cold, but can result in serious complications, especially in infants.[14] A 2014 outbreak of enterovirus D68 (EV-D68) resulted in severe respiratory infections, some of which were fatal.[30]

Influenza virus: There are three types of influenza viruses: A, B, and C.[3] In the northern hemisphere, influenza A and B circulate during the winter months causing seasonal epidemics most years; influenza C infections are less common and not believed to cause epidemics.[3, 31] Both influenza A and B mutate, and the impact of influenza varies from year to year depending on the severity of the changes and effectiveness of influenza vaccines.[32] The two most common influenza A subtypes infecting humans are H1N1 (including the 2009 Pandemic H1N1 variant) and H3N2, and prevalence varies annually.[33] Other rare influenza A subtypes also known to infect humans, such as H5N1 (avian influenza) and H3N2v, can cause severe illness and, in some cases, death.[33] Influenza is easily spread from person to person and those most at risk for complications from infection include infants and children, the elderly, and anyone who is immunocompromised or who has co-morbidities such as heart or lung disease.[34]

Influenza A 2009 H1N1: During the 2009 - 2010 influenza season, a new strain of influenza A, now known as 2009 H1N1 became the dominant circulating virus, accounting for approximately 95% of reported influenza infections.[31] This strain replaced the H1N1 virus that was previously circulating in humans and is common in both Europe and the U.S. [31, 33]

Parainfluenza Virus: The parainfluenza viruses are members of the paramyxovirus family that commonly cause respiratory infections in children.[35] Prevalence of parainfluenza viruses is seasonal and varies by type; most infections are mild and self-limited, but parainfluenza virus can cause life threatening pneumonia in immunocompromised patients, such as those with cystic fibrosis or transplant recipients.[36]

Respiratory Syncytial Virus: RSV is the most common cause of pediatric viral respiratory infections.[11] Infection with RSV can occur at any age, and those most at risk for complications and more severe disease are the very young, especially premature infants, the elderly, and anyone with a weakened immune system.[37] There are two types of respiratory syncytial virus, RSV A and B. Infections with RSV A are thought to be more severe than infections with RSV B.[17, 38]

Chlamydia pneumoniae (formerly known as Chlamydophila pneumoniae): *Chlamydia pneumoniae* is a common cause of upper respiratory infections including atypical pneumonia.[39] *C. pneumoniae* is transmitted person-to-person by respiratory secretions and outbreaks are common in close contact settings.[19] Infection severity can be mild or result in more severe disease, particularly in high risk populations such as people with heart or lung disease, diabetes, and the elderly.[19, 40] The true prevalence of *C. pneumoniae* infections is unknown, but the use of molecular diagnostics has improved detection of this organism, as it is difficult to identify using traditional laboratory methods.[40]

***Mycoplasma pneumoniae*:** *Mycoplasma pneumonia*e is a bacterium lacking a cell wall and is a major cause of respiratory disease.[21] *M. pneumonia*e is transmitted person-to-person by respiratory droplets and is a common cause of atypical, or walking pneumonia.[41] *M. pneumoniae* is frequently undiagnosed but is estimated to be involved in up to 30% of respiratory infections.[120] Infection often results in mild illness such as tracheobronchitis, or a chest cold, and is most prevalent in young adults and school-aged

children.[22, 41] Outbreaks of *M. pneumoniae* occur mostly in crowded environments, like schools, college dormitories, military barracks, and nursing homes.[41]

PRINCIPLES OF TECHNOLOGY

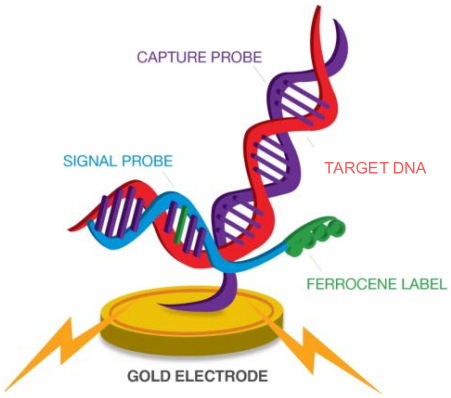
The **cobas eplex** system automates all aspects of nucleic acid testing including extraction, amplification, and detection, combining electrowetting and eSensor technology in a single-use cartridge. eSensor technology is based on the principles of competitive DNA hybridization and electrochemical detection, which is highly specific and is not based on fluorescent or optical detection.

Electrowetting, or digital microfluidics, uses electrical fields to directly manipulate discrete droplets on the surface of a hydrophobically coated printed circuit board (PCB). Sample and reagents are moved in a programmable fashion in the ePlex cartridge to complete all portions of the sample processing from nucleic acid extraction to detection.

A sample is loaded onto the **cobas eplex** cartridge and nucleic acids are extracted and purified from the specimen via magnetic solid phase extraction. For RNA targets, a reverse transcription step is performed to generate complementary DNA from the RNA, followed by PCR to amplify the targets. Exonuclease digestion creates single-stranded DNA in preparation for eSensor detection.

The target DNA is mixed with ferrocene-labeled signal probes that are complementary to the specific targets on the panel. Target DNA hybridizes to its complementary signal probe and capture probes, which are bound to gold-plated electrodes, as shown below in **Figure 1**. The presence of each target is determined by voltammetry which generates specific electrical signals from the ferrocene-labeled signal probe.

Figure 1: Hybridization complex. Target-specific capture probes are bound to the gold electrodes in the eSensor microarray on the **cobas eplex** cartridge. The amplified target DNA hybridizes to the capture probe and to a complementary ferrocene-labeled signal probe. Electrochemical analysis determines the presence or absence of targets using voltammetry.



MATERIALS PROVIDED

Table 2: cobas eplex respiratory pathogen panel 2 kit contents

Product	Item number	Components (quantity)	Storage
cobas eplex respiratory pathogen panel 2	GenMark: EA001222 Roche: 9555641001	**cobas eplex** respiratory pathogen panel 2 cartridge (12)	2 – 8 ºC (through printed expiration date) or 30 days at 25 °C (cartridges must be used within 30 days of 25 ºC storage start date)

Safety Data Sheets (SDS) for all reagents provided in this kit may be obtained at: https://dialog.roche.com. For paper copies, please contact GenMark Technical Support at cad.technical_support_us@roche.com.

REAGENT STORAGE, STABILITY, AND HANDLING

- Store the **cobas eplex** RP2 panel kit components at 2–8 ºC. Alternatively, cartridges can be stored at 25 ºC for up to 30 days. Cartridges must be used within 30 days from start of 25 ºC storage and should be considered expired once stored for 30 days at 25 ºC. Users should not return the kit to cold storage after storage at 25°C.
- Do not use RP2 panel kit components beyond the expiration date.
- Do not open a cartridge pouch until you are ready to perform testing.

MATERIALS NOT PROVIDED

Equipment

- **cobas eplex** system and Software
- Pipettes calibrated to deliver 200 µL
- Vortex mixer
- Printer (optional) - See **cobas eplex** User Assistance Manual for compatibility guidelines

Consumables

- Pipette tips, aerosol resistant, RNase/DNase-free
- Disposable, powder free gloves
- 10% bleach for decontamination of appropriate surfaces
- 70% ethanol or isopropyl alcohol

WARNINGS AND PRECAUTIONS

General

- For use under Emergency Use Authorization Only.
- For *in vitro* diagnostic use.
- For prescription use only.
- This product has not been FDA cleared or approved, but has been authorized for emergency use by FDA under an EUA for use by authorized laboratories.

- This product has been authorized only for the detection and differentiation of nucleic acids from multiple respiratory viral and bacterial organisms, including nucleic acid from Severe Acute Respiratory Syndrome coronavirus 2 (SARS-CoV-2).
- This emergency use of this product is only authorized for the duration of the declaration that circumstances exist justifying the authorization of emergency use of *in vitro* diagnostics for detection and/or diagnosis of COVID-19 under Section 564(b)(1) of the Federal Food, Drug and Cosmetic Act, 21 U.S.C. § 360bbb-3(b)(1), unless the declaration is terminated or authorization is revoked sooner.
- A trained healthcare professional should carefully interpret the results from the **cobas eplex** RP2 panel in conjunction with a patient's signs, symptoms, and results from other diagnostic tests.
- Positive results do not rule out co-infection with other viruses or bacteria. The agent detected may not be the definite cause of disease. The use of additional laboratory testing (e.g., bacterial and viral culture, immunofluorescence, and radiography) and clinical presentation must be taken into consideration in the diagnosis of respiratory infection.
- Do not reuse **cobas eplex** RP2 panel kit components.
- Do not use reagents beyond the expiration date printed on the labeling.
- Do not use a reagent that is damaged.
- Follow the procedure as described in this package insert. Read all instructions before starting the test. Any deviation from the procedures and guidelines may affect optimal test performance.
- All human-sourced materials should be considered potentially infectious and should be handled with universal precautions.
- The use of sterile, disposable, nuclease-free pipette tips is recommended. Use only supplied or specified required consumables to ensure optimal test performance.

Safety

- Handle all specimens and waste materials as if they were capable of transmitting infectious agents in accordance with Universal Precautions. Observe safety guidelines such as those outlined in CDC/NIH *Biosafety in Microbiological and Biomedical Laboratories,* CLSI Document M29 *Protection of Laboratory Workers from Occupationally Acquired Infections*, or other appropriate guidelines.
- Do not eat, smoke, drink, apply cosmetics, or handle contact lenses in areas where reagents or human specimens are handled.
- Follow national biological safety procedures for handling biological samples. (*e.g.*, do not pipette by mouth, wear appropriate protective clothing and eye protection).
- If infection with a novel influenza A virus is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions for novel virulent Influenza viruses and sent to state or local health department for testing. Viral culture should not be attempted in these cases unless a BSL-3+ facility is available to receive and culture specimens.
- Dispose materials used in this test, including reagents, specimens, and used vials, in accordance with all federal, state, and local regulations.
- Do not stick fingers or other objects inside the **cobas eplex** system bays.
- Wash hands thoroughly with soap and water after handling reagents. Launder contaminated clothing prior to re-use.
- Do not puncture or pierce reagent blisters on the **cobas eplex** cartridge. Reagents may cause irritation to skin, eyes, and respiratory tract. Harmful if swallowed or inhaled. Contains oxidizing liquids.
- The **cobas eplex** RP2 panel cartridge contains chemicals that are classified as hazardous. Review the Safety Data Sheet (SDS) before use, and in cases of exposure, refer to the SDS for more information.
- Observe safety guidelines such as wearing proper protective equipment including laboratory coats, gowns, gloves, eye protection, and a biological safety cabinet as outlined in Biosafety in Microbiological and Biomedical Laboratories (BMBL) 5th Edition https://www.cdc.gov/labs/BMBL.html.

- If infection with SARS-CoV-2 is suspected based on current clinical and epidemiological screening criteria recommended by public health authorities, specimens should be collected with appropriate infection control precautions.
- Thoroughly decontaminate the lab and all equipment with 10% bleach followed by 70% ethanol or isopropyl alcohol (or equivalent) prior to processing a specimen.
- Immediately clean up any spill containing potentially infectious material with a 0.5-1% (w/v) sodium hypochlorite (20% v/v bleach).
- Performance characteristics have been determined with nasopharyngeal swab samples from human patients with signs and symptoms of respiratory infection.
- Specimens should be processed in a Class II (or higher) biological safety cabinet.
- To mitigate the risk of sample-to-sample contamination, change gloves after dispensing sample into the cartridge.
- Contamination of the sample may occur if the sample is loaded in an area where PCR amplicons for respiratory pathogens are generated. Avoid loading sample in areas that are potentially contaminated with PCR amplicon.

Laboratory

- Contamination of the sample may occur if laboratory personnel processing the sample are infected with common respiratory pathogens. To avoid this, specimens should be processed in biosafety cabinets. If a biosafety cabinet is not used, a splash shield or face mask should be used when processing samples.
- A biosafety cabinet that is used for viral or bacterial culture should not be used for sample preparation.
- Samples and cartridges should be handled and/or tested one at a time. To mitigate the risk of sample-to-sample contamination, change gloves after dispensing sample into the cartridge.
- Thoroughly decontaminate the lab and all equipment with 10% bleach followed by 70% ethanol or isopropyl alcohol (or equivalent) prior to processing a specimen.
- Contamination of the sample may occur if the sample is loaded in an area where PCR amplicons for respiratory pathogens are generated. Avoid loading sample in areas that are potentially contaminated with PCR amplicon.

SPECIMEN COLLECTION, HANDLING, AND STORAGE

Nasopharyngeal Swab Collection – Nasopharyngeal swab specimen collection should be performed according to standard technique and placed in viral transport media.

Minimum Sample Volume – 200 µL nasopharyngeal swab specimen in viral transport media is required for testing.

Transport and Storage – Clinical specimens can be stored at room temperature (15–30 ºC) for up to 12 hours or refrigerated at 2-8 ºC for up to 10 days after collection in viral transport media. Specimens can also be stored at -20 ºC or -80 ºC for 12 months with up to 2 freeze/thaw cycles.

Inadequate or inappropriate specimen collection, storage, and transport are likely to yield false test results. Training in specimen collection is highly recommended due to the importance of specimen quality. CLSI MM13-A may be referenced as an appropriate resource.

PROCEDURE

Procedural Notes

- All frozen samples should be thawed completely before testing.
- Samples should be nasopharyngeal swabs in viral transport media.
- Cartridge can be used immediately upon removal from 2-8 °C storage. There is no need to equilibrate to room temperature before use.
- Alternatively, cartridges can be stored at 25 °C for up to 30 days. Cartridges must be used within 30 days from start of 25 ºC storage and should be considered expired once stored for 30 days at 25 ºC. Users should not return the kit to cold storage after storage at 25°C.
- Once cartridge is removed from foil pouch, it should be used within 2 hours. Do not open the cartridge pouch until the sample is ready to be tested.
- Once the sample is loaded into the **cobas eplex** RP2 panel cartridge, the sample should be tested as soon as possible or within 2 hours.
- Do not re-use cartridges.
- Use a new, sterile pipette tip for loading each sample.
- Do not insert a wet cartridge into the **cobas eplex** system. If the cartridge or sample has leaked, dispose of cartridge in accordance with all federal, state, and local regulations.
- Samples should be transferred into the **cobas eplex** RP2 panel cartridge in an amplicon-free, clean environment.
- Samples, consumables, and lab areas should be protected from aerosol or direct contamination with amplicon. Decontaminate laboratory areas and affected equipment with 10% bleach followed by 70% ethanol or isopropyl alcohol (or equivalent).
- Samples and cartridges should be handled and/or tested one at a time. To mitigate the risk of sample-to-sample contamination, change gloves after dispensing sample into the cartridge.
- Specimens should be processed in biosafety cabinets. If a biosafety cabinet is not used, a splash shield or face mask should be used when processing samples.
- Dispose materials used in this test, including reagents, specimens, and used vials, in accordance with all regulations.

Detailed Procedure

1. Decontaminate the clean area used for setting up the **cobas eplex** RP2 panel with 10% bleach followed by 70% ethanol or isopropyl alcohol (or equivalent).
2. Remove one **cobas eplex** RP2 panel cartridge pouch from kit packaging.
3. Open the RP2 panel cartridge pouch.
4. Write the accession ID or place a barcode label with accession ID on the RP2 panel cartridge.
5. Vortex the sample for 3-5 seconds.
6. Use a calibrated pipette to aspirate 200 µL of sample and dispense into the sample loading port of the **cobas eplex** RP2 panel cartridge.
7. Close the sample loading port by sliding the cap over the port and firmly pushing down on the cap to securely seal the sample delivery port.
   NOTE: Bubbles can be present when closing the cap.
8. Scan the RP2 panel cartridge using the barcode reader provided with the **cobas eplex** system.
   NOTE: If an accession ID barcode label is not used, manually enter accession ID with the on-screen keyboard and scan the cartridge barcode when prompted by the **cobas eplex** system.
   NOTE: The barcode scanner will read both the accession ID barcode (if placed on the cartridge by the operator) and the 2D barcode printed on the cartridge label; however, the barcode scanner will only beep once to indicate that both barcodes have been read.
9. Insert the RP2 panel cartridge into any available bay, indicated by a flashing, white LED light. The test will begin automatically when the cartridge has been inserted into the bay and the pre-run check (cartridge initialization) is completed, indicated by a blue LED light.

QUALITY CONTROL

Internal Controls

Each cartridge includes internal controls that monitor performance of each step of the testing process. A DNA control verifies extraction, amplification and detection of DNA targets, and RNA controls verify amplification and detection of RNA targets.

Each amplification reaction on the cartridge has at least one internal control and in each reaction either the internal control or a target must generate signal above the defined threshold for a valid test result. Internal control results are interpreted by the **cobas eplex** software and displayed on the RP2 panel Reports as Internal Control with a result of PASS, FAIL, N/A or INVALID. **Table 3** includes details on the interpretation of Internal Control results.

Table 3: Internal control results

Internal Control Result	Explanation	Action
PASS	The internal control or a target from each amplification reaction has generated signal above the threshold. The test was completed and internal controls were successful, indicating valid results were generated.	All results are displayed on the **cobas eplex** RP2 panel Detection Report. Test is valid, report results.
FAIL	Neither the internal control nor any target in at least one amplification reaction generates signal above the threshold. The test was completed but at least one internal control was not detected, indicating that results are not valid.	No results are displayed on the RP2 panel Detection Report. Test is not valid, repeat the test using a new cartridge.
N/A	The internal control in every amplification reaction does not generate signal above the threshold, but a target in every amplification reaction does generate signal above the threshold. The test was completed and internal controls were not successful, however detection of signal above the threshold for a target in every amplification reaction indicates valid results were generated.	All results are displayed on the RP2 panel Detection Report. Test is valid, report results.
INVALID	An error has occurred during processing that prevents analysis of signal data. The test has not successfully completed and results for this test are not valid. This is often due to an instrument or software error.	No results are displayed on the RP2 panel Detection Report. Test is not valid, repeat the test using a new cartridge.

External Controls

Positive and negative external controls should be tested with each new lot of reagents or monthly, whichever occurs first. Viral transport medium can be used as the negative control. Previously characterized positive samples or viral transport medium spiked with well characterized organisms can be used as the external positive control. External controls should be run in accordance with laboratory protocols and accrediting organizations, as applicable.

RESULTS

Table 4: Interpretation of results on the **cobas eplex** RP2 panel detection report

Target Result	Explanation	Action
Target Detected	The test was completed successfully, and the target has generated signal above its defined threshold, and the Internal Control was reported as PASS.	All results are displayed on the RP2 panel Detection Report. Test is valid, report results.
Multiple Targets Detected	The test was completed successfully, and multiple targets have generated signal above their defined threshold, and the Internal Control was reported as PASS.	All results are displayed on the RP2 panel Detection Report. Test is valid, report results. Detection of more than 3 pathogens may indicate contamination. Re-test of the sample is recommended to confirm results.
Target Not Detected	The test was completed successfully, and the target did not generate signal above its defined threshold, and the Internal Control was reported as PASS.	All results are displayed on the RP2 panel Detection Report. Test is valid, report results.
Invalid	The test has not successfully completed, and results for this test are not valid. This is often due to an instrument or software error or failure of an internal control.	No results are displayed on the RP2 panel Detection Report. Test is not valid, repeat test.

Influenza A Results

The **cobas eplex** RP2 panel detects Influenza A and the H1, H1-2009, and H3 subtypes using unique assays for each. Interpretation of results for Influenza A are described in **Table 5**.

Table 5: Results for influenza A

Results for Influenza A and Subtypes	Explanation	Results on Report	Recommended Action
Influenza A detected, at least one subtype (H1, H1-2009, or H3) reported as detected.	This is an expected result.	Result reported as influenza A and influenza A subtype detected.	None
Influenza A detected, all subtypes (H1, H1-2009, and H3) reported as not detected	Low virus titers can result in detection of influenza A matrix without a subtype. Detection of influenza A matrix without a subtype can also indicate the presence of a novel strain.	Result reported as influenza A detected. No Influenza A subtype detected.	Re-test to confirm result. If the original result is confirmed, contact the appropriate public health authorities for additional testing. If the re-test provides a different result, test the sample a third time to ensure the accuracy of the result.

Results for Influenza A and Subtypes	Explanation	Results on Report	Recommended Action
Influenza A detected and more than one subtype (H1, H1-2009, or H3) reported as detected.	Sample is co-infected with multiple influenza subtypes. Infection with multiple subtypes of influenza are possible but rare. A live intranasal multivalent influenza virus vaccine may cause false positive results for influenza A, A/H1, A/H3, A/H1-2009, and/or influenza B. Contamination has occurred.	Result reported as influenza A and multiple subtypes detected.	Re-test to confirm result. If the re-test result confirms the original result, it is recommended that the sample be further investigated using a different FDA-cleared influenza A subtyping assay.
Influenza A not detected, at least one subtype (H1, H1-2009, or H3) reported as detected.	Low virus titers can result in detection of influenza A subtype without the influenza A matrix. Detection of influenza A subtype without the influenza A matrix can also indicate the presence of a novel strain.	Influenza A (subtype) detected. Re-testing of this sample to confirm Influenza A (subtype) is recommended. Refer to package insert for additional information.	Re-test to confirm result. If the re-test result confirms the original result, the influenza A subtype is considered positive. It is recommended that the sample be further investigated using a different FDA-cleared influenza A subtyping assay and/or sending the residual sample to local public health laboratory for further testing.

TEST REPORTS

There are several different reports that are available on the **cobas eplex** system. Results are provided in a printable format, may be viewed electronically, or may be exported for additional analysis. Reports can be customized with account specific information such as the address, logo, and institution specific footers on each report. For more information on **cobas eplex** reports, refer to the **cobas eplex** User Assistance Manual.

Detection Report

The RP2 Panel detection report includes the results for each individual sample run on the **cobas eplex** system.

The summary section indicates the overall test result and lists all detected targets in that sample. The results section includes a list of all targets on the panel with an individual result for each. Results for each target are reported as Detected, Not Detected, or Invalid (displayed as a red **x**); results for the Internal Control are reported as PASS, FAIL, INVALID, or N/A.

External Control Report

The **cobas eplex** RP2 panel external control report is generated for an external control that has been pre-defined in the **cobas eplex** RP2 panel software. For more information on defining external controls on **cobas eplex** RP2 panel, refer to the **cobas eplex** User Assistance Manual.

The summary section indicates the overall result (Pass or Fail status) and lists all detected targets for that external control. The results section includes a list of all panel targets with the result, expected result, and Pass/Fail status for each. Results are reported as Detected, Not Detected, or Invalid (displayed as a red **x**). A target is reported as pass if the actual result matches the expected result (as defined for that control); a target is reported as fail if the actual result does not match the expected result. If the actual results for each target match the expected result for each target (all targets reported as pass), the overall result for the external control is reported as pass in the summary section. If the actual result for any target does not match the expected result, the overall result for the external control is reported as fail in the summary section.

Summary Report

The summary report allows the operator to use defined searchable criteria to create customized reports, using specified targets, dates, range of dates, sample, external control, test bay, or operator. For more information on creating summary reports, refer to the **cobas eplex** User Assistance Manual.

LIMITATIONS OF THE PROCEDURE

- This product can be used only with the **cobas eplex** system.
- At high titers, cross-reactivity with SARS-CoV-1 was observed with the **cobas eplex** RP2 panel.
- Due to the genetic similarity between human rhinovirus and enterovirus, this test cannot reliably differentiate them. A **cobas eplex** RP2 panel rhinovirus/enterovirus positive result should be followed up using an alternate method (e.g. cell culture or sequence analysis) if differentiation between the viruses is required.
- This test is a qualitative test and does not provide a quantitative value of detected organism present.
- The performance of the test has been evaluated for use with human sample material only.
- This test has not been validated for testing samples other than nasopharyngeal swab samples in viral transport media.
- The performance of this test has not been established for immunocompromised individuals.
- The performance of this test has not been established for patients without signs and symptoms of respiratory infection.
- Results from this test must be correlated with the clinical history, epidemiological data, and other data available to the clinician evaluating the patient.
- The effect of antibiotic treatment on test performance has not been evaluated.
- The performance of this test has not been established for screening of blood or blood products.
- Targets (viral and bacterial nucleic acids) may persist *in vivo*, independent of viral or bacterial viability. Detection of target(s) does not imply that the corresponding virus(es) or bacteria are infectious or are the causative agents for clinical symptoms.
- The detection of viral or bacterial nucleic acid is dependent upon proper specimen collection, handling, transportation, storage, and preparation. Failure to observe proper procedures in any one of these steps can lead to incorrect results. There is a risk of false positive or false negative values resulting from improperly collected, transported, or handled samples.
- There is a risk of false negative values due to the presence of sequence variants in the viral or bacterial targets of the test, the presence of inhibitors, technical error, sample mix-up, or an

infection caused by an organism not detected by the panel. Test results may be affected by concurrent antibacterial or antiviral therapy or levels of bacteria or virus in the sample that are below the limit of detection for the test. A result of No Targets Detected on the ePlex RP2 Panel should not be used as the sole basis for diagnosis, treatment, or other patient management decisions.

- A result of No Targets Detected on the **cobas eplex** RP2 panel in the setting of a respiratory illness may be due to infection with pathogens that are not detected by this test or lower respiratory tract infection that is not detected by a nasopharyngeal swab sample.
- There is a risk of false positive results due to contamination of the sample with target organisms, their nucleic acids, or amplicons. Particular attention should be given to the Laboratory precautions noted under the *Warnings and Precautions* section.
- There is a risk of false positive results due to non-specific amplification and cross-reactivity with organisms found in the respiratory tract. Erroneous results due to cross-reactivity with organisms that were not specifically evaluated or new variant sequences that emerge are possible.
- If four or more organisms are detected in a sample, retesting is recommended to confirm polymicrobial result.
- The **cobas eplex** RP2 panel influenza A subtyping reagents target the influenza A hemagglutinin gene only. The **cobas eplex** RP2 panel does not detect or differentiate the influenza A neuraminidase gene.
- The performance of this test has not been established for monitoring treatment of infection with any of the panel organisms.
- Positive and negative predictive values are highly dependent on prevalence. False negative test results are more likely during peak activity when prevalence of disease is high. False positive test results are more likely during periods when prevalence is moderate to low.
- Clinical performance was established when influenza A H3 and influenza A H1-2009 were the predominant influenza A viruses in circulation. When other influenza A viruses emerge, performance may vary.
- Due to the small number of positive samples collected for *Chlamydia pneumoniae* during the prospective and retrospective clinical studies, performance characteristics for *Chlamydia pneumoniae* were established primarily with contrived clinical specimens. Performance characteristics for Influenza A H1 were established using contrived clinical specimens only.
- Clinical evaluation indicates a lower sensitivity for the detection of coronavirus OC43. If infection with coronavirus OC43 is suspected, negative samples should be confirmed using an alternative method.
- The effect of interfering substances has only been evaluated for those listed in this package insert. Interference due to substances other than those described in the “Interfering Substances” section can lead to erroneous results.
- At concentrations greater than 1% weight/volume in the sample, tobramycin was found to inhibit assay performance.
- Minimum Essential Media (MEM) may be inhibitory and negatively impact the performance of the **cobas eplex** RP2 panel.
- Diluents from external quality controls or proficiency testing materials that include the following substances have been shown to interfere with the performance of the **cobas eplex** RP2 panel: human plasma proteins and 941 L media with methanol.
- The performance of this test has not been specifically evaluated for specimens collected from individuals who recently received influenza vaccine. Recent administration of a live intranasal influenza virus vaccine may cause false positive results for influenza A, H1, H3, H1-2009, and/or influenza B.
- The **cobas eplex** RP2 panel cannot differentiate variant viruses, such as H3N2v, from seasonal influenza A viruses. If variant virus infection is suspected, clinicians should contact their state or local health department to arrange specimen transport and request a timely diagnosis at a state public health laboratory.
- The performance of this test was established based on the evaluation of a limited number of clinical specimens. Clinical performance has not been established with all circulating variants but is anticipated to be reflective of the prevalent variants in circulation at the time and location of the

clinical evaluation. Performance at the time of testing may vary depending on the variants circulating, including newly emerging strains of SARS-CoV-2 and their prevalence, which change over time.

Conditions of Authorization for the Laboratory

The **cobas eplex** RP2 panel Letter of Authorization, along with the authorized Fact Sheet for Healthcare Providers, the authorized Fact Sheet for Patients, and authorized labeling are available on the FDA website: https://www.fda.gov/medical-devices/coronavirus-disease-2019-covid-19-emergency-use-authorizations-medical-devices/vitro-diagnostics-euas.

However, to assist clinical laboratories using the **cobas eplex** RP2 panel, the relevant Conditions of Authorization are listed below:

A. Authorized laboratories[1] using the **cobas eplex** RP2 panel must include with test result reports, all authorized Fact Sheets. Under exigent circumstances, other appropriate methods for disseminating these Fact Sheets may be used, which may include mass media.

B. Authorized laboratories using the **cobas eplex** RP2 panel must use the **cobas eplex** RP2 panel as outlined in the authorized labeling. Deviations from the authorized procedures, including the authorized instruments, authorized extraction methods, authorized clinical specimen types, authorized control materials, authorized other ancillary reagents and authorized materials required to use the **cobas eplex** RP2 panel are not permitted.

C. Authorized laboratories that receive the **cobas eplex** RP2 panel must notify the relevant public health authorities of their intent to run the **cobas eplex** RP2 panel prior to initiating testing.

D. Authorized laboratories using the **cobas eplex** RP2 panel must have a process in place for reporting test results to healthcare providers and relevant public health authorities, as appropriate.

E. Authorized laboratories must collect information on the performance of the **cobas eplex** RP2 panel and must report any significant deviations from the established performance characteristics of the **cobas eplex** RP2 panel test of which they become aware to DMD/OHT7/OPEQ/CDRH (via email: CDRH-EUA-Reporting@fda.hhs.gov) and GenMark Diagnostics, Inc. (via email: cad.technical_support_us@roche.com).

F. All laboratory personnel using the **cobas eplex** RP2 panel must be appropriately trained in RT-PCR techniques and use appropriate laboratory and personal protective equipment when handling this kit, and use the **cobas eplex** RP2 panel in accordance with the authorized labeling.

G. GenMark Diagnostics, Inc., authorized distributors and authorized laboratories using the **cobas eplex** RP2 panel must ensure that any records associated with this EUA are maintained until otherwise notified by FDA. Such records must be made available to FDA for inspection upon request.

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

PERFORMANCE CHARACTERISTICS

cobas eplex RP and RP2 panels

The **cobas eplex** RP2 panel was developed by incorporating the reagents required to detect the SARS-CoV-2 targets from the ePlex SARS-CoV-2 Test into the existing **cobas eplex** respiratory pathogen panel (RP panel). The assays for detection of SARS-CoV-2 were added into PCR pools that contain additional targets. The targets that are now co-amplified with SARS-CoV-2 are influenza A, influenza A H1, influenza A H1-2009, influenza A H3, influenza B, and adenovirus; assays for all other targets were unchanged. Studies were conducted to demonstrate that the performance characteristics of the RP Panel were not affected by the addition of the SARS-CoV-2 assays. Additional studies to support the addition of SARS-CoV-2 are included in the sections below. The original studies from the RP panel are still relevant for the **cobas eplex** RP2 panel.

CLINICAL PERFORMANCE

EXPECTED VALUES

A prospective, multicenter clinical study was conducted to evaluate the clinical performance of the **cobas eplex** RP panel in nasopharyngeal swab samples. 2462 nasopharyngeal swab samples were prospectively collected at 8 collection sites in 2 phases from patients of all ages and genders presenting with signs and/or symptoms of respiratory infection. In the first phase from March 2013 through August 2014, 1951 samples were prospectively collected and frozen; from September 2016 through October 2016, 511 samples were prospectively collected and tested fresh (never frozen). The expected values of individual analytes based on **cobas eplex** RP panel results in prospective samples for each phase are summarized in **Tables 6-9**. **NOTE:** Expected values for SARS-CoV-2 have not been determined.

Table 6: Expected value (as determined by **cobas eplex** RP panel) summary by age group in the prospective clinical evaluation (phase 1: March 2013 – August 2014)

Organism	All Ages (N=1951) n (%)	Age 0-1 (N=315) n (%)	Age >1-5 (N=250) n (%)	Age >5-21 (N=246) n (%)	Age >21-65 (N=745) n (%)	Age >65 (N=395) n (%)
Adenovirus	72 (3.7)	31 (9.8)	24 (9.6)	7 (2.8)	7 (0.9)	3 (0.8)
Coronavirus (229E, HKU1, NL63, OC43)	102 (5.2)	19 (6.0)	18 (7.2)	16 (6.5)	32 (4.3)	17 (4.3)
Human Metapneumovirus	113 (5.8)	22 (7.0)	28 (11.2)	6 (2.4)	31 (4.2)	26 (6.6)
Human Rhinovirus/Enterovirus	388 (19.9)	113 (35.9)	94 (37.6)	58 (23.6)	87 (11.7)	36 (9.1)
Influenza A	110 (5.6)	6 (1.9)	18 (7.2)	20 (8.1)	49 (6.6)	17 (4.3)
Influenza A H1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Influenza A H1-2009	76 (3.9)	4 (1.3)	13 (5.2)	14 (5.7)	37 (5.0)	8 (2.0)
Influenza A H3	34 (1.7)	1 (0.3)	5 (2.0)	6 (2.4)	12 (1.6)	10 (2.5)
Influenza B	62 (3.2)	4 (1.3)	9 (3.6)	10 (4.1)	24 (3.2)	15 (3.8)
Parainfluenza Virus 1	24 (1.2)	4 (1.3)	12 (4.8)	4 (1.6)	3 (0.4)	1 (0.3)
Parainfluenza Virus 2	10 (0.5)	4 (1.3)	4 (1.6)	0 (0.0)	2 (0.3)	0 (0.0)
Parainfluenza Virus 3	99 (5.1)	31 (9.8)	20 (8.0)	3 (1.2)	27 (3.6)	18 (4.6)
Parainfluenza Virus 4	7 (0.4)	3 (1.0)	2 (0.8)	1 (0.4)	1 (0.1)	0 (0.0)
RSV A	28 (1.4)	13 (4.1)	6 (2.4)	3 (1.2)	2 (0.3)	4 (1.0)
RSV B	83 (4.3)	33 (10.5)	19 (7.6)	6 (2.4)	15 (2.0)	10 (2.5)

Organism	All Ages (N=1951) n (%)	Age 0-1 (N=315) n (%)	Age >1-5 (N=250) n (%)	Age >5-21 (N=246) n (%)	Age >21-65 (N=745) n (%)	Age >65 (N=395) n (%)
Chlamydia pneumoniae	3 (0.2)	0 (0.0)	0 (0.0)	1 (0.4)	1 (0.1)	1 (0.3)
Mycoplasma pneumoniae	5 (0.3)	1 (0.3)	1 (0.4)	2 (0.8)	1 (0.1)	0 (0.0)

Table 7: Expected value (as determined by **cobas eplex** RP Panel) summary by age group in the prospective clinical evaluation (phase 2: September 2016 – October 2016)

Organism	All Ages (N=511) n (%)	Age 0-1 (N=73) n (%)	Age >1-5 (N=75) n (%)	Age >5-21 (N=75) n (%)	Age >21-65 (N=181) n (%)	Age >65 (N=107) n (%)
Adenovirus	10 (2.0)	3 (4.1)	4 (5.3)	1 (1.3)	1 (0.6)	1 (0.9)
Coronavirus (229E, HKU1, NL63, OC43)	8 (1.6)	2 (2.7)	0 (0.0)	1 (1.3)	4 (2.2)	1 (0.9)
Human Metapneumovirus	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Human Rhinovirus/Enterovirus	188 (36.8)	37 (50.7)	40 (53.3)	33 (44.0)	58 (32.0)	20 (18.7)
Influenza A	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Influenza A H1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Influenza A H1-2009	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Influenza A H3	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Influenza B	2 (0.4)	0 (0.0)	0 (0.0)	1 (1.3)	1 (0.6)	0 (0.0)
Parainfluenza Virus 1	1 (0.2)	0 (0.0)	1 (1.3)	0 (0.0)	0 (0.0)	0 (0.0)
Parainfluenza Virus 2	13 (2.5)	3 (4.1)	4 (5.3)	3 (4.0)	2 (1.1)	1 (0.9)
Parainfluenza Virus 3	5 (1.0)	2 (2.7)	1 (1.3)	1 (1.3)	1 (0.6)	0 (0.0)
Parainfluenza Virus 4	8 (1.6)	1 (1.4)	4 (5.3)	2 (2.7)	1 (0.6)	0 (0.0)
RSV A	8 (1.6)	5 (6.8)	3 (4.0)	0 (0.0)	0 (0.0)	0 (0.0)
RSV B	9 (1.8)	3 (4.1)	4 (5.3)	0 (0.0)	2 (1.1)	0 (0.0)
Chlamydia pneumoniae	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Mycoplasma pneumoniae	4 (0.8)	0 (0.0)	1 (1.3)	2 (2.7)	1 (0.6)	0 (0.0)

Table 8: Expected value (as determined by **cobas eplex** RP panel) summary by sample collection site in the prospective clinical evaluation (phase 1: March 2013 – August 2014)

Organism	All Sites (N=1951) n (%)	Site 1 (N=165) n (%)	Site 2 (N=248) n (%)	Site 3 (N=350) n (%)	Site 4 (N=892) n (%)	Site 5 (N=296) n (%)
Adenovirus	72 (3.7)	4 (2.4)	8 (3.2)	28 (8.0)	23 (2.6)	9 (3.0)
Coronavirus (229E, HKU1, NL63, OC43)	102 (5.2)	8 (4.8)	11 (4.4)	32 (9.1)	29 (3.3)	22 (7.4)
Human Metapneumovirus	113 (5.8)	10 (6.1)	23 (9.3)	27 (7.7)	30 (3.4)	23 (7.8)
Human Rhinovirus/Enterovirus	388 (19.9)	27 (16.4)	33 (13.3)	61 (17.4)	185 (20.7)	82 (27.7)
Influenza A	110 (5.6)	5 (3.0)	21 (8.5)	48 (13.7)	19 (2.1)	17 (5.7)
Influenza A H1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Influenza A H1-2009	76 (3.9)	3 (1.8)	22 (8.9)	31 (8.9)	5 (0.6)	15 (5.1)
Influenza A H3	34 (1.7)	2 (1.2)	0 (0.0)	18 (5.1)	12 (1.3)	2 (0.7)
Influenza B	62 (3.2)	9 (5.5)	9 (3.6)	9 (2.6)	19 (2.1)	16 (5.4)
Parainfluenza Virus 1	24 (1.2)	0 (0.0)	0 (0.0)	5 (1.4)	2 (0.2)	17 (5.7)
Parainfluenza Virus 2	10 (0.5)	0 (0.0)	0 (0.0)	0 (0.0)	10 (1.1)	0 (0.0)
Parainfluenza Virus 3	99 (5.1)	13 (7.9)	3 (1.2)	28 (8.0)	41 (4.6)	14 (4.7)
Parainfluenza Virus 4	7 (0.4)	0 (0.0)	0 (0.0)	1 (0.3)	4 (0.4)	2 (0.7)

Organism	All Sites (N=1951) n (%)	Site 1 (N=165) n (%)	Site 2 (N=248) n (%)	Site 3 (N=350) n (%)	Site 4 (N=892) n (%)	Site 5 (N=296) n (%)
RSV A	28 (1.4)	4 (2.4)	6 (2.4)	7 (2.0)	4 (0.4)	7 (2.4)
RSV B	83 (4.3)	6 (3.6)	15 (6.0)	24 (6.9)	15 (1.7)	23 (7.8)
Chlamydia pneumoniae	3 (0.2)	0 (0.0)	0 (0.0)	1 (0.3)	2 (0.2)	0 (0.0)
Mycoplasma pneumoniae	5 (0.3)	1 (0.6)	0 (0.0)	3 (0.9)	0 (0.0)	1 (0.3)

Table 9: Expected value (as determined by **cobas eplex** RP panel) summary by sample collection site in the prospective clinical evaluation (phase 2: September 2016 – October 2016)

Organism	All Sites (N=511) n (%)	Site 5 (N=49) n (%)	Site 6 (N=101) n (%)	Site 7 (N=161) n (%)	Site 8 (N=200) n (%)
Adenovirus	10 (2.0)	2 (4.1)	3 (3.0)	3 (1.9)	2 (1.0)
Coronavirus (229E, HKU1, NL63, OC43)	8 (1.6)	0 (0.0)	2 (2.0)	4 (2.5)	2 (1.0)
Human Metapneumovirus	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Human Rhinovirus/Enterovirus	188 (36.8)	24 (49.0)	49 (48.5)	62 (38.5)	53 (26.5)
Influenza A	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Influenza A H1	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Influenza A H1-2009	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Influenza A H3	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Influenza B	2 (0.4)	1 (2.0)	0 (0.0)	0 (0.0)	1 (0.5)
Parainfluenza Virus 1	1 (0.2)	0 (0.0)	0 (0.0)	1 (0.6)	0 (0.0)
Parainfluenza Virus 2	13 (2.5)	2 (4.1)	4 (4.0)	3 (1.9)	4 (2.0)
Parainfluenza Virus 3	5 (1.0)	2 (4.1)	2 (2.0)	0 (0.0)	1 (0.5)
Parainfluenza Virus 4	8 (1.6)	1 (2.0)	1 (1.0)	4 (2.5)	2 (1.0)
RSV A	8 (1.6)	0 (0.0)	8 (7.9)	0 (0.0)	0 (0.0)
RSV B	9 (1.8)	1 (2.0)	4 (4.0)	0 (0.0)	4 (2.0)
Chlamydia pneumoniae	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Mycoplasma pneumoniae	4 (0.8)	0 (0.0)	3 (3.0)	0 (0.0)	1 (0.5)

CLINICAL PERFORMANCE

Clinical performance of the cobas eplex RP2 panel and SARS-CoV-2

Performance characteristics of the **cobas eplex** RP2 panel for SARS-CoV-2 detection were established using previously frozen clinical specimens (nasopharyngeal swab (NPS) samples) collected from U.S. patients.

In the first arm of the study, a total of 189 samples, 174 NPS samples (60 known SARS-CoV-2 positive and 114 from the initial RP panel clinical study) and 15 contrived samples were tested with the **cobas eplex** RP2 panel in the clinical evaluation study. Samples with final, valid results and a comparator result were considered evaluable. Four samples (1 known SARS-CoV-2 positive, 3 from the initial RP panel clinical study) were not evaluable because they did not have final, valid **cobas eplex** RP2 panel results and were excluded from analysis.

The comparator methods for the SARS-CoV-2 target were COVID-19 molecular diagnostic tests that received FDA Emergency Use Authorization (EUA). Only the 60 SARS-CoV-2 known positive NPS samples were tested with these methods. There was no comparator method for the SARS-CoV-2 target in the remaining 114 NPS samples from the initial clinical study. These samples were presumed SARS-

CoV-2 negative based on their collection prior to 2017. The comparator method for the other RP2 panel targets was the **cobas eplex** RP panel. Only the 114 NPS samples from the initial RP panel clinical study were tested with this method.

Positive percent agreement (PPA) was calculated by dividing the number of true positive (TP) results by the sum of TP and false negative (FN) results, while negative percent agreement (NPA) was calculated by dividing the number of true negative (TN) results by the sum of TN and false positive (FP) results. A TP result was one where the detected **cobas eplex** RP2 panel result matched the detected comparator method result, while a TN result was one where a negative **cobas eplex** RP2 panel result matched a presumed negative result. Since archived negative samples were not tested by the comparator method, false positives are calculated based on a presumed negative clinical truth. The two-sided 95% confidence interval was also calculated. Results are shown in **Table 10** below.

Table 10. Positive percent agreement (PPA) and negative percent agreement (NPA) for SARS-CoV-2 in the **cobas eplex** RP2 panel clinical study

cobas eplex RP2 panel	Comparator result Positive	Comparator result Negative	Comparator result Total
Positive	59	0	59
Negative	0	111	111
Total	59	111	170

PPA: 59/59 100% (95% CI: 93.9-100)
NPA: 111/111 100% (95% CI: 96.7-100

In the second arm of the study, testing was done to evaluate the performance of **cobas eplex** RP2 panel targets co-amplified with the SARS-CoV-2 assays (assays for SARS-CoV-2 were incorporated into PCR pools that also include influenza A, influenza A H1, influenza A H1-2009, influenza A H3, influenza B, and adenovirus; assays for all other targets were unchanged). Samples were tested with the **cobas eplex** RP2 panel and the original **cobas eplex** respiratory pathogen panel, or to the known contrived organism. For the Influenza A H1 target only, contrived samples were evaluated. Results are shown in **Tables 11-12** below.

Table 11: Positive percent agreement (PPA) and negative percent agreement (NPA) of the **cobas eplex** RP2 panel with the **cobas eplex** RP panel in nasopharyngeal swab samples

Organism	Positive % Agreement TP/TP+FN	Positive % Agreement PPA (95% CI)	Negative % Agreement TN/TN+FP	Negative % Agreement NPA (95% CI)
Adenovirus	25/25	100 (86.7-100)	83/86[a]	96.5 (90.2-98.8)
Influenza A	56/57[b]	98.2 (90.7-99.7)	54/54	100 (93.4-100)
Influenza A H1	0/0	---	111/111	100 (96.7-100)
Influenza A H1-2009	26/27[c]	96.3 (81.7-99.3)	84/84	100 (95.6-100)
Influenza A H3	29/30[b]	96.7 (83.3-99.4)	81/81	100 (95.5-100)
Influenza B	28/29[d]	96.6 (82.8-99.4)	82/82	100 (95.5-100)

[a] 3 FP Adenovirus results were detected by the **cobas eplex** RP panel (IUO) in the original clinical study.
[b] 1 FN Influenza A H3 result was not detected by the RP panel (IUO) in the original clinical study.
[c] 1 FN Influenza A 2009 H1N1 result was detected by the RP panel (IUO) in the original clinical study.
[d] 1 FN Influenza B result was detected by the RP panel (IUO) in the original clinical study.

Table 12: Positive percent agreement (PPA) for the **cobas eplex** RP2 panel with the **cobas eplex** RP panel with contrived samples

Organism	Positive % Agreement TP/TP+FN	Positive % Agreement PPA (95% CI)
Influenza A	15/15	100 (79.6-100)
Influenza A H1	15/15	100 (79.6-100)

RP2 clinical study cobas eplex instrument performance

A total of 189 samples (174 NPS and 15 contrived) were initially tested with the **cobas eplex** RP2 panel and 183/189 = 96.8% (95% CI: 93.2% - 98.5%) generated valid results on the first attempt. Two samples were re-tested and both had valid results. The remaining invalid samples did not have sufficient volume for repeat testing.

Clinical performance of the cobas eplex RP panel

Comparator method

The performance of the **cobas eplex** RP panel was compared to an FDA-cleared multiplexed molecular respiratory pathogen panel and analytically validated PCR tests with bi-directional sequencing for confirmation of RSV subtypes. Details of the comparator method are described in **Table 13**.

Table 13: Comparator methods used to assess **cobas eplex** RP panel clinical performance

Target	Comparator Method
Adenovirus	FDA-cleared multiplexed molecular respiratory pathogen panel
Coronavirus (229E, HKU1, NL63, OC43)	FDA-cleared multiplexed molecular respiratory pathogen panel
Human Metapneumovirus	FDA-cleared multiplexed molecular respiratory pathogen panel
Human Rhinovirus/Enterovirus	FDA-cleared multiplexed molecular respiratory pathogen panel
Influenza A	FDA-cleared multiplexed molecular respiratory pathogen panel
Influenza A H1	FDA-cleared multiplexed molecular respiratory pathogen panel
Influenza A H1-2009	FDA-cleared multiplexed molecular respiratory pathogen panel
Influenza A H3	FDA-cleared multiplexed molecular respiratory pathogen panel
Influenza B	FDA-cleared multiplexed molecular respiratory pathogen panel
Parainfluenza Virus 1	FDA-cleared multiplexed molecular respiratory pathogen panel
Parainfluenza Virus 2	FDA-cleared multiplexed molecular respiratory pathogen panel
Parainfluenza Virus 3	FDA-cleared multiplexed molecular respiratory pathogen panel
Parainfluenza Virus 4	FDA-cleared multiplexed molecular respiratory pathogen panel
Respiratory Syncytial Virus A	FDA-cleared multiplexed molecular respiratory pathogen panel followed by a PCR test with bi-directional sequencing confirmation
Respiratory Syncytial Virus B	FDA-cleared multiplexed molecular respiratory pathogen panel followed by a PCR test with bi-directional sequencing confirmation
Chlamydia pneumoniae	FDA-cleared multiplexed molecular respiratory pathogen panel
Mycoplasma pneumoniae	FDA-cleared multiplexed molecular respiratory pathogen panel

Prospective clinical samples

Clinical performance was evaluated in nasopharyngeal swab samples in VTM prospectively collected at 8 clinical sites in 2 phases. From March 2013 through August 2014, 2218, samples were prospectively collected and frozen; from September 2016 through October 2016, 514 samples were prospectively collected and tested fresh (never frozen). A total of 2732 samples were collected across the 2 phases.

Prior to the start of investigational testing, 263 samples were withdrawn (251 had sample handling deviations, 9 were tested outside of protocol timelines, 2 had insufficient volume, and 1 had incomplete documentation). Of the 2469 prospectively collected samples eligible for testing, 2462 were evaluable. Samples with final, valid results and a valid comparator result were considered evaluable. Seven prospectively collected samples were not evaluable because they did not have final, valid **cobas eplex** RP panel results and were excluded from performance evaluations. Demographic information for prospectively collected samples is described in **Table 14**. Subjects enrolled in this study were from a diverse demographic distribution and represent the intended patient population.

Table 14: Subject demographic data for prospectively collected samples by collection site (N=2462)

-	All sites N=2462 n (%)	Site 1 N=165 n (%)	Site 2 N=248 n (%)	Site 3 N=350 n (%)	Site 4 N=892 n (%)	Site 5 N=345 n (%)	Site 6 N=101 n (%)	Site 7 N=161 n (%)	Site 8 N=200 n (%)
Sex: Male	1247 (50.6)	96 (58.2)	118 (47.6)	186 (53.1)	450 (50.4)	188 (54.5)	43 (42.6)	84 (52.2)	82 (41.0)
Sex: Female	1215 (49.4)	69 (41.8)	130 (52.4)	164 (46.9)	442 (49.6)	157 (45.5)	58 (57.4)	77 (47.8)	118 (59.0)
Age: 0–1 years	388 (15.8)	17 (10.3)	21 (8.5)	74 (21.1)	164 (18.4)	45 (13.0)	28 (27.7)	3 (1.9)	36 (18.0)
Age: > 1–5 years	325 (13.2)	12 (7.3)	22 (8.9)	62 (17.7)	64 (7.2)	100 (29.0)	39 (38.6)	16 (9.9)	10 (5.0)
Age: > 5–21 years	321 (13.0)	15 (9.1)	6 (2.4)	38 (10.9)	82 (9.2)	116 (33.6)	34 (33.7)	18 (11.2)	12 (6.0)
Age: > 21–65 years	926 (37.6)	87 (52.7)	131 (52.8)	98 (28.0)	385 (43.2)	55 (15.9)	0 (0.0)	92 (57.1)	78 (39.0)
Age: > 65 years	502 (20.4)	34 (20.6)	68 (27.4)	78 (22.3)	197 (22.1)	29 (8.4)	0 (0.0)	32 (19.9)	64 (32.0)

Prospective clinical performance

Positive percent agreement (PPA) was calculated by dividing the number of true positive (TP) results by the sum of TP and false negative (FN) results, while negative percent agreement (NPA) was calculated by dividing the number of true negative (TN) results by the sum of TN and false positive (FP) results. A TP result was one where the detected **cobas eplex** RP Panel result matched the detected comparator method result, while a TN result was one where a negative **cobas eplex** RP panel result matched a negative comparator method result. The two-sided 95% confidence interval was also calculated.

A total of 2462 prospectively collected samples (511 tested fresh and 1951 tested after previously frozen) were evaluated for 17 ePlex RP Panel organisms. PPA and NPA results are summarized by target in **Tables 15 and 16** below.

Table 15: Positive percent agreement (PPA) and negative percent agreement (NPA) in the **cobas eplex** RP panel clinical study (fresh)

Organism	Prevalence	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Adenovirus	1.6%	6/8[a]	75.0 (40.9-92.9)	499/503[a]	99.2 (98.0-99.7)
Coronavirus (229E, HKU1, NL63, OC43)	1.4%	7/7	100 (64.6-100)	503/504	99.8 (98.9-100)

Organism	Prevalence	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Human Metapneumovirus	0.0%	0/0	---	511/511	100 (99.3-100)
Human Rhinovirus/Enterovirus	35.8%	176/183[b]	96.2 (92.3-98.1)	316/328[b]	96.3 (93.7-97.9)
Influenza A	0.0%	0/0	---	511/511	100 (99.3-100)
Influenza A H1	0.0%	0/0	---	511/511	100 (99.3-100)
Influenza A H1-2009	0.0%	0/0	---	511/511	100 (99.3-100)
Influenza A H3	0.0%	0/0	---	511/511	100 (99.3-100)
Influenza B	0.2%	1/1	100 (20.7-100)	509/510	99.8 (98.9-100)
Parainfluenza Virus 1	0.2%	1/1	100 (20.7-100)	510/510	100 (99.3-100)
Parainfluenza Virus 2	2.5%	12/13	92.3 (66.7-98.6)	497/498	99.8 (98.9-100)
Parainfluenza Virus 3	1.0%	5/5	100 (56.6-100)	506/506	100 (99.2-100)
Parainfluenza Virus 4	0.6%	3/3	100 (43.9-100)	503/508[c]	99.0 (97.7-99.6)
RSV A	1.8%	8/9	88.9 (56.5-98.0)	501/501	100 (99.2-100)
RSV B	2.0%	9/10	90.0 (59.6-98.2)	500/500	100 (99.2-100)
Chlamydia pneumoniae	0.0%	0/0	---	511/511	100 (99.3-100)
Mycoplasma pneumoniae	0.6%	3/3	100 (43.9-100)	507/508[d]	99.8 (98.9-100)

[a] Adenovirus was not detected in 2 of 2 FN samples and detected in 4 of 4 FP samples using PCR/sequencing.
[b] Human rhinovirus/enterovirus was not detected in 1 of 7 FN samples and detected in 9 of 12 FP samples using PCR/sequencing.
[c] Parainfluenza virus 4 was detected in 3 of 5 FP samples using PCR/sequencing.
[d] *M. pneumoniae* was detected in the 1 FP sample using PCR/sequencing.

Table 16: Positive percent agreement (PPA) and negative percent agreement (NPA) in the **cobas eplex** RP panel clinical study (after previously frozen)

Organism	Prevalence	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Adenovirus	2.7%	48/53[a]	90.6 (79.7-95.9)	1874/1898[a]	98.7 (98.1-99.1)
Coronavirus (229E, HKU1, NL63, OC43)	5.6%	89/110[b]	80.9 (72.6-87.2)	1828/1841[b]	99.3 (98.8-99.6)
Human Metapneumovirus	5.8%	107/113[c]	94.7 (88.9-97.5)	1832/1838[c]	99.7 (99.3-99.9)
Human Rhinovirus/Enterovirus	17.2%	317/336[d]	94.3 (91.3-96.4)	1544/1615[d]	95.6 (94.5-96.5)
Influenza A[e]	5.7%	106/111[f]	95.5 (89.9-98.1)	1836/1840[f]	99.8 (99.4-99.9)
Influenza A H1	0.0%	0/0	---	1951/1951	100 (99.8-100)
Influenza A H1-2009	3.6%	70/71	98.6 (92.4-99.8)	1874/1880[g]	99.7 (99.3-99.9)
Influenza A H3	1.9%	34/37[h]	91.9 (78.7-97.2)	1914/1914	100 (99.8-100)
Influenza B	3.3%	58/65[i]	89.2 (79.4-94.7)	1882/1886[i]	99.8 (99.5-99.9)
Parainfluenza Virus 1	1.2%	23/24	95.8 (79.8-99.3)	1926/1927	99.9 (99.7-100)
Parainfluenza Virus 2	0.5%	9/9	100 (70.1-100)	1941/1942	99.9 (99.7-100)
Parainfluenza Virus 3	5.3%	94/104[j]	90.4 (83.2-94.7)	1842/1847[j]	99.7 (99.4-99.9)
Parainfluenza Virus 4	0.3%	5/5	100 (56.6-100)	1944/1946	99.9 (99.6-100)
RSV A	1.6%	27/31	87.1 (71.1-94.9)	1917/1918	99.9 (99.7-100)
RSV B	4.4%	81/86	94.2 (87.1-97.5)	1861/1863[k]	99.9 (99.6-100)
Chlamydia pneumoniae	0.3%	2/5[l]	40.0 (11.8-76.9)	1945/1946[l]	99.9 (99.7-100)
Mycoplasma pneumoniae	0.3%	4/5[m]	80.0 (37.6-96.4)	1945/1946	99.9 (99.7-100)

[a] Adenovirus was not detected in 1 of 5 FN samples and detected in 9 of 24 FP samples using PCR/sequencing.

[b] Coronavirus was not detected in 2 of 21 FN samples and detected in 3 of 13 FP samples using PCR/sequencing.
[c] Human Metapneumovirus was not detected in 1 of 6 FN samples and detected in 4 of 6 FP samples using PCR/sequencing.
[d] Human rhinovirus/enterovirus was not detected in 6 of 19 FN samples and detected in 33 of 71 FP samples using PCR/sequencing.
[e] Influenza A comparator results contain 71 samples with A H1-2009, 37 samples with A H3, and 3 samples with no subtype detected.
[f] Influenza A was not detected in 1 of 3 FN samples (2 samples were not tested by PCR/sequencing) and detected in 1 of 4 FP samples using PCR/sequencing.
[g] Influenza A H1-2009 was detected in 4 of 6 FP samples using PCR/sequencing.
[h] Influenza A H3 was not detected in 1 of 3 FN samples using PCR/sequencing.
[i] Influenza B was not detected in 3 of 7 FN samples and detected in 2 of 4 FP samples using PCR/sequencing.
[j] Parainfluenza virus 3 was not detected in 3 of 10 FN samples and detected in 4 of 5 FP samples using PCR/sequencing.
[k] RSV B was detected in 1 of 2 FP samples using PCR/sequencing.
[l] *C. pneumoniae* was not detected in 1 of 3 FN samples and detected in the 1 FP sample using PCR/sequencing.
[m] *M. pneumoniae* was not detected in the 1 FN sample using PCR/sequencing.

Retrospective clinical samples

To supplement the number of positives for targets that were not sufficiently represented in the prospective collection, additional nasopharyngeal swab in VTM samples were retrospectively collected from 6 sites. A total of 535 nasopharyngeal swab samples that had previously tested positive for one or more of the target organisms during standard-of-care (SOC) testing were collected and stored frozen. Prior to the start of investigational testing, 11 samples were withdrawn due to noncompliance with the study protocol, and 52 samples were withdrawn because the organisms present had sufficient representation in other samples. In addition, the composition and integrity of the retrospective samples were confirmed with the same comparator method employed in the prospective clinical study (i.e., an FDA-cleared multiplexed respiratory pathogen panel). As the result of this confirmation testing using the comparator method, 26 additional samples were withdrawn because the original SOC testing positive results for the intended organisms were not confirmed when tested with the comparator method. Of the remaining 446 retrospectively collected samples eligible for testing, all 446 were evaluable. Demographic information for retrospectively collected samples is described in **Table 17**. Subjects enrolled in this study were from a diverse demographic distribution and represent the intended patient population.

Table 17: Subject demographic data for retrospectively collected samples by collection site (N=446)

-	**All sites N=446 n (%)**	**Site 1 N=1 n (%)**	**Site 2 N=1 n (%)**	**Site 3 N=129 n (%)**	**Site 4 N=18 n (%)**	**Site 5 N=131 n (%)**	**Site 6 N=166 n (%)**
Sex: Male	232 (52.0)	0 (0.0)	1 (100)	76 (58.9)	11 (61.1)	68 (51.9)	76 (45.8)
Sex: Female	214 (48.0)	1 (100)	0 (0.0)	53 (41.1)	7 (38.9)	63 (48.1)	90 (54.2)
Age: 0 – 1 years	122 (27.4)	0 (0.0)	0 (0.0)	24 (18.6)	5 (27.8)	56 (42.7)	37 (22.3)
Age: > 1 – 5 years	107 (24.0)	0 (0.0)	1 (100)	51 (39.5)	3 (16.7)	16 (12.2)	36 (21.7)
Age: > 5 – 21 years	59 (13.2)	0 (0.0)	0 (0.0)	9 (7.0)	2 (11.1)	19 (14.5)	29 (17.5)
Age: > 21 – 65 years	99 (22.2)	1 (100)	0 (0.0)	11 (8.5)	8 (44.4)	31 (23.7)	48 (28.9)
Age: > 65 years	59 (13.2)	0 (0.0)	0 (0.0)	34 (26.4)	0 (0.0)	9 (6.9)	16 (9.6)

Retrospective clinical performance

A total of 446 retrospectively collected samples were evaluated for 17 **cobas eplex** RP panel organisms. The following specimens with the original positive SOC results for the unintended organisms that were not confirmed by the comparator method were excluded from the performance calculation for the respective organism: 1 coronavirus positive specimen, 3 human rhinovirus/enterovirus positive

specimens, 1 influenza A positive specimen, 1 influenza A H3 positive specimen, 1 parainfluenza virus positive specimen. In addition, 5 unintended RSV positive specimens by the comparator method were not confirmed by PCR/sequencing with regard to determining RSV subtypes and therefore were excluded from the performance calculations for RSV A and RSV B. PPA and NPA results are summarized by target in **Table 18** below.

Table 18: Positive percent agreement (PPA) and negative percent agreement (NPA) of the **cobas eplex** RP panel with comparator methods (retrospective collection)

Organism	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Adenovirus	55/56[a]	98.2 (90.6-99.7)	386/390[a]	99.0 (97.4-99.6)
Coronavirus (229E, HKU1, NL63, OC43)	121/138[b]	87.7 (81.2-92.2)	307/307	100 (98.8-100)
Human Metapneumovirus	5/7	71.4 (35.9-91.8)	439/439	100 (99.1-100)
Human Rhinovirus/Enterovirus	37/41	90.2 (77.5-96.1)	384/402	95.5 (93.0-97.1)
Influenza A[c]	75/82[d]	91.5 (83.4-95.8)	363/363	100 (99.0-100)
Influenza A H1	0/0	---	446/446	100 (99.1-100)
Influenza A H1-2009	27/31[e]	87.1 (71.1-94.9)	415/415	100 (99.1-100)
Influenza A H3	45/51[f]	88.2 (76.6-94.5)	394/394	100 (99.0-100)
Influenza B	1/1	100 (20.7-100)	445/445	100 (99.1-100)
Parainfluenza Virus 1	43/48[g]	89.6 (77.8-95.5)	396/397	99.7 (98.6-100)
Parainfluenza Virus 2	46/51	90.2 (79.0-95.7)	395/395	100 (99.0-100)
Parainfluenza Virus 3	2/2	100 (34.2-100)	444/444	100 (99.1-100)
Parainfluenza Virus 4	18/20	90.0 (69.9-97.2)	426/426	100 (99.1-100)
RSV A	25/27	92.6 (76.6-97.9)	414/414	100 (99.1-100)
RSV B	21/22	95.5 (78.2-99.2)	419/419	100 (99.1-100)
Chlamydia pneumoniae	1/1	100 (20.7-100)	445/445	100 (99.1-100)
Mycoplasma pneumoniae	7/7	100 (64.6-100)	439/439	100 (99.1-100)

[a] Adenovirus was not detected in the 1 FN sample and detected in 2 of 4 FP samples using PCR/sequencing.
[b] Coronavirus was not detected in 2 of 16 FN samples using PCR/sequencing (1 sample was not tested by PCR/sequencing).
[c] Influenza A comparator results contain 31 samples with A H1-2009 and 51 samples with A H3 detected.
[d] Influenza A was not detected in 3 of 7 FN samples using PCR/sequencing.
[e] Influenza A H1-2009 was not detected in 2 of 4 FN samples using PCR/sequencing.
[f] Influenza A H3 was not detected in 1 of 6 FN samples using PCR/sequencing.
[g] Parainfluenza virus 1 was not detected in 2 of 5 FN samples using PCR/sequencing.

Contrived sample performance

There were 327 contrived samples created and tested to supplement the low prevalence targets on the RP panel; 104 contained one or more low prevalence organisms and 223 were negative for the contrived organisms. All 327 contrived samples were tested with the **cobas eplex** RP panel and 326 were evaluable. PPA and NPA results are summarized for these low prevalence organisms in **Table 19** below.

Table 19: Positive percent agreement (PPA) and negative percent agreement (NPA) of the **cobas eplex** RP panel with comparator method (contrived samples)

Organism	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Chlamydia pneumoniae	52/52	100 (93.1-100)	274/274	100 (98.6-100)
Influenza A H1	51/51	100 (93.0-100)	275/275	100 (98.6-100)

Clinical and contrived sample performance by target

Tables 20-36 below include the clinical performance by pathogen of prospective samples tested fresh (shown in **Table 15**), prospective samples tested after previously freezing (shown in **Table 16**), retrospective samples (shown in **Table 18**), and contrived samples (shown in **Table 1**).

Table 20: Positive percent agreement (PPA) and negative percent agreement (NPA) for adenovirus in the **cobas eplex** RP panel clinical study

Adenovirus	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	6/8[a]	75.0 (40.9-92.9)	499/503[a]	99.2 (98.0-99.7)
Prospectively collected samples	Frozen	48/53[b]	90.6 (79.7-95.9)	1874/1898[b]	98.7 (98.1-99.1)
Prospectively collected samples	Total	54/61	88.5 (78.2-94.3)	2373/2401	98.8 (98.3-99.2)
Retrospectively collected samples	-	55/56[c]	98.2 (90.6-99.7)	386/390[c]	99.0 (97.4-99.6)

[a] Adenovirus was not detected in 2 of 2 FN samples and detected in 4 of 4 FP samples using PCR/sequencing.
[b] Adenovirus was not detected in 1 of 5 FN samples and detected in 9 of 24 FP samples using PCR/sequencing.
[c] Adenovirus was not detected in the 1 FN sample and detected in 2 of 4 FP samples using PCR/sequencing.

Table 21: Positive percent agreement (PPA) and negative percent agreement (NPA) for coronavirus (229E, HKU1, NL63, OC43) in the **cobas eplex** RP panel clinical study

Coronavirus (229E, HKU1, NL63, OC43)	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples[a]	Fresh	7/7	100 (64.6-100)	503/504	99.8 (98.9-100)
Prospectively collected samples[a]	Frozen	89/110[b]	80.9 (72.6-87.2)	1828/1841[b]	99.3 (98.8-99.6)
Prospectively collected samples[a]	Total	96/117	82.1 (74.1-88.0)	2331/2345	99.4 (99.0-99.6)
Retrospectively collected samples[c]	-	121/138[d]	87.7 (81.2-92.2)	307/307	100 (98.8-100)

[a] 20 FN prospectively collected frozen samples were repeat tested with the comparator method and 12 had coronavirus detected. Of these 12 samples, 11 were repeat tested with the **cobas eplex** RP panel and 3 had coronavirus detected.
[b] Coronavirus was not detected in 2 of 21 FN samples and detected in 3 of 13 FP samples using PCR/sequencing.
[c] 10 FN retrospectively collected samples were repeat tested with the comparator method and all 10 had coronavirus detected. Of these 10 samples, 9 were repeat tested with the **cobas eplex** RP panel and 5 had coronavirus detected.
[d] Coronavirus was not detected in 2 of 16 FN samples using PCR/sequencing (1 sample was not tested by PCR/sequencing).

Table 22: Positive percent agreement (PPA) and negative percent agreement (NPA) for human metapneumovirus in the **cobas eplex** RP panel clinical study

Human metapneumovirus	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	0/0	---	511/511	100 (99.3-100)
Prospectively collected samples	Frozen	107/113[a]	94.7 (88.9-97.5)	1832/1838[a]	99.7 (99.3-99.9)
Prospectively collected samples	Total	107/113	94.7 (88.9-97.5)	2343/2349	99.7 (99.4-99.9)
Retrospectively collected samples	-	5/7	71.4 (35.9-91.8)	439/439	100 (99.1-100)

[a] Human metapneumovirus was not detected in 1 of 6 FN samples and detected in 4 of 6 FP samples using PCR/sequencing.

Table 23: Positive percent agreement (PPA) and negative percent agreement (NPA) for human rhinovirus/enterovirus in the **cobas eplex** RP panel clinical study

Human rhinovirus/enterovirus	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	176/183[a]	96.2 (92.3-98.1)	316/328[a]	96.3 (93.7-97.9)
Prospectively collected samples	Frozen	317/336[b]	94.3 (91.3-96.4)	1544/1615[b]	95.6 (94.5-96.5)
Prospectively collected samples	Total	493/519	95.0 (92.8-96.6)	1860/1943	95.7 (94.7-96.5)
Retrospectively collected samples	-	37/41	90.2 (77.5-96.1)	384/402	95.5 (93.0-97.1)

[a] Human rhinovirus/enterovirus was not detected in 1 of 7 FN samples and detected in 9 of 12 FP samples using PCR/sequencing.
[b] Human rhinovirus/enterovirus was not detected in 6 of 19 FN samples and detected in 33 of 71 FP samples using PCR/sequencing.

Table 24: Positive percent agreement (PPA) and negative percent agreement (NPA) for influenza A in the **cobas eplex** RP panel clinical study

Influenza A	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples[a]	Fresh	0/0	---	511/511	100 (99.3-100)
Prospectively collected samples[a]	Frozen	106/111[b]	95.5 (89.9-98.1)	1836/1840[b]	99.8 (99.4-99.9)
Prospectively collected samples[a]	Total	106/111	95.5 (89.9-98.1)	2347/2351	99.8 (99.6-99.9)
Retrospectively collected samples[c]	-	75/82[d]	91.5 (83.4-95.8)	363/363	100 (99.0-100)

[a] Influenza A comparator results contain 71 samples with A H1-2009, 37 samples with A H3, and 3 samples with no subtype detected.
[b] Influenza A was not detected in 1 of 3 FN samples (2 samples were not tested by PCR/sequencing) and detected in 1 of 4 FP samples using PCR/sequencing.
[c] Influenza A comparator results contain 31 samples with A H1-2009 and 51 samples with A H3 detected.
[d] Influenza A was not detected in 3 of 7 FN samples using PCR/sequencing.

Table 25: Positive percent agreement (PPA) and negative percent agreement (NPA) for influenza A H1 in the **cobas eplex** RP panel clinical study

Influenza A H1	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	0/0	---	511/511	100 (99.3-100)
Prospectively collected samples	Frozen	0/0	---	1951/1951	100 (99.8-100)
Prospectively collected samples	Total	0/0	---	2462/2462	100 (99.8-100)
Retrospectively collected samples	-	0/0	---	446/446	100 (99.1-100)
Contrived samples	-	51/51	100 (93.0-100)	275/275	100 (98.6-100)

Table 26: Positive percent agreement (PPA) and negative percent agreement (NPA) for influenza A H1-2009 in the **cobas eplex** RP panel clinical study

Influenza A H1-2009	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	0/0	---	511/511	100 (99.3-100)
Prospectively collected samples	Frozen	70/71	98.6 (92.4-99.8)	1874/1880[a]	99.7 (99.3-99.9)
Prospectively collected samples	Total	70/71	98.6 (92.4-99.8)	2385/2391	99.7 (99.5-99.9)
Retrospectively collected samples	-	27/31[b]	87.1 (71.1-94.9)	415/415	100 (99.1-100)

[a] Influenza A H1-2009 was detected in 4 of 6 FP samples using PCR/sequencing.
[b] Influenza A H1-2009 was not detected in 2 of 4 FN samples using PCR/sequencing.

Table 27: Positive percent agreement (PPA) and negative percent agreement (NPA) for influenza A H3 in the **cobas eplex** RP panel clinical study

Influenza A H3	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	0/0	---	511/511	100 (99.3-100)
Prospectively collected samples	Frozen	34/37[a]	91.9 (78.7-97.2)	1914/1914	100 (99.8-100)
Prospectively collected samples	Total	34/37	91.9 (78.7-97.2)	2425/2425	100 (99.8-100)
Retrospectively collected samples	-	45/51[b]	88.2 (76.6-94.5)	394/394	100 (99.0-100)

[a] Influenza A H3 was not detected in 1 of 3 FN samples using PCR/sequencing.
[b] Influenza A H3 was not detected in 1 of 6 FN samples using PCR/sequencing.

Table 28: Positive percent agreement (PPA) and negative percent agreement (NPA) for influenza B in the **cobas eplex** RP panel clinical study

Influenza B	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	1/1	100 (20.7-100)	509/510	99.8 (98.9-100)
Prospectively collected samples	Frozen	58/65[a]	89.2 (79.4-94.7)	1882/1886[a]	99.8 (99.5-99.9)
Prospectively collected samples	Total	59/66	89.4 (79.7-94.8)	2391/2396	99.8 (99.5-99.9)
Retrospectively collected samples	-	1/1	100 (20.7-100)	445/445	100 (99.1-100)

[a] Influenza B was not detected in 3 of 7 FN samples and detected in 2 of 4 FP samples using PCR/sequencing.

Table 29: Positive percent agreement (PPA) and negative percent agreement (NPA) for parainfluenza virus 1 in the **cobas eplex** RP panel clinical study

Parainfluenza virus 1	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	1/1	100 (20.7-100)	510/510	100 (99.3-100)
Prospectively collected samples	Frozen	23/24	95.8 (79.8-99.3)	1926/1927	99.9 (99.7-100)
Prospectively collected samples	Total	24/25	96.0 (80.5-99.3)	2436/2437	100 (99.8-100)
Retrospectively collected samples	-	43/48[a]	89.6 (77.8-95.5)	396/397	99.7 (98.6-100)

[a] Parainfluenza virus 1 was not detected in 2 of 5 FN samples using PCR/sequencing.

Table 30: Positive percent agreement (PPA) and negative percent agreement (NPA) for parainfluenza virus 2 in the **cobas eplex** RP panel clinical study

Parainfluenza virus 2	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	12/13	92.3 (66.7-98.6)	497/498	99.8 (98.9-100)
Prospectively collected samples	Frozen	9/9	100 (70.1-100)	1941/1942	99.9 (99.7-100)
Prospectively collected samples	Total	21/22	95.5 (78.2-99.2)	2438/2440	99.9 (99.7-100)
Retrospectively collected samples	-	46/51	90.2 (79.0-95.7)	395/395	100 (99.0-100)

Table 31: Positive percent agreement (PPA) and negative percent agreement (NPA) for parainfluenza virus 3 in the **cobas eplex** RP panel clinical study

Parainfluenza virus 3	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	5/5	100 (56.6-100)	506/506	100 (99.2-100)
Prospectively collected samples	Frozen	94/104[a]	90.4 (83.2-94.7)	1842/1847[a]	99.7 (99.4-99.9)
Prospectively collected samples	Total	99/109	90.8 (83.9-94.9)	2348/2353	99.8 (99.5-99.9)
Retrospectively collected samples	-	2/2	100 (34.2-100)	444/444	100 (99.1-100)

[a] Parainfluenza virus 3 was not detected in 3 of 10 FN samples and detected in 4 of 5 FP samples using PCR/sequencing.

Table 32: Positive percent agreement (PPA) and negative percent agreement (NPA) for parainfluenza virus 4 in the **cobas eplex** RP panel clinical study

Parainfluenza virus 4	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	3/3	100 (43.9-100)	503/508[a]	99.0 (97.7-99.6)
Prospectively collected samples	Frozen	5/5	100 (56.6-100)	1944/1946	99.9 (99.6-100)
Prospectively collected samples	Total	8/8	100 (67.6-100)	2447/2454	99.7 (99.4-99.9)
Retrospectively collected samples	-	18/20	90.0 (69.9-97.2)	426/426	100 (99.1-100)

[a] Parainfluenza virus 4 was detected in 3 of 5 FP samples using PCR/sequencing.

Table 33: Positive percent agreement (PPA) and negative percent agreement (NPA) for respiratory syncytial virus A in the **cobas eplex** RP panel clinical study

Respiratory syncytial virus A	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	8/9	88.9 (56.5-98.0)	501/501	100 (99.2-100)
Prospectively collected samples	Frozen	27/31	87.1 (71.1-94.9)	1917/1918	99.9 (99.7-100)
Prospectively collected samples	Total	35/40	87.5 (73.9-94.5)	2418/2419	100 (99.8-100)
Retrospectively collected samples	-	25/27	92.6 (76.6-97.9)	414/414	100 (99.1-100)

Table 34: Positive percent agreement (PPA) and negative percent agreement (NPA) for respiratory syncytial virus B in the **cobas eplex** RP panel clinical study

Respiratory syncytial virus B	Sample type	Positive % agreement TP/TP+FN	Positive % agreement PPA (95% CI)	Negative % agreement TN/TN+FP	Negative % agreement NPA (95% CI)
Prospectively collected samples	Fresh	9/10	90.0 (59.6-98.2)	500/500	100 (99.2-100)

Respiratory syncytial virus B	**Sample type**	**Positive % agreement TP/TP+FN**	**Positive % agreement PPA (95% CI)**	**Negative % agreement TN/TN+FP**	**Negative % agreement NPA (95% CI)**
Prospectively collected samples	Frozen	81/86	94.2 (87.1-97.5)	1861/1863[a]	99.9 (99.6-100)
Prospectively collected samples	Total	90/96	93.8 (87.0-97.1)	2361/2363	99.9 (99.7-100)
Retrospectively collected samples	-	21/22	95.5 (78.2-99.2)	419/419	100 (99.1-100)

[a] RSV B was detected in 1 of 2 FP samples using PCR/sequencing.

Table 35: Positive percent agreement (PPA) and negative percent agreement (NPA) for *Chlamydia pneumoniae* in the **cobas eplex** RP panel clinical study

Chlamydia pneumoniae	**Sample type**	**Positive % agreement TP/TP+FN**	**Positive % agreement PPA (95% CI)**	**Negative % agreement TN/TN+FP**	**Negative % agreement NPA (95% CI)**
Prospectively collected samples	Fresh	0/0	---	511/511	100 (99.3-100)
Prospectively collected samples	Frozen	2/5[a]	40.0 (11.8-76.9)	1945/1946[a]	99.9 (99.7-100)
Prospectively collected samples	Total	2/5	40.0 (11.8-76.9)	2456/2457	100 (99.8-100)
Retrospectively collected samples	-	1/1	100 (20.7-100)	445/445	100 (99.1-100)
Contrived samples	-	52/52	100 (93.1-100)	274/274	100 (98.6-100)

[a] *C. pneumoniae* was not detected in 1 of 3 FN samples and detected in the 1 FP sample using PCR/sequencing.

Table 36: Positive percent agreement (PPA) and negative percent agreement (NPA) for *Mycoplasma pneumoniae* in the **cobas eplex** RP panel clinical study

Mycoplasma pneumoniae	**Sample type**	**Positive % agreement TP/TP+FN**	**Positive % agreement PPA (95% CI)**	**Negative % agreement TN/TN+FP**	**Negative % agreement NPA (95% CI)**
Prospectively collected samples	Fresh	3/3	100 (43.9-100)	507/508[a]	99.8 (98.9-100)
Prospectively collected samples	Frozen	4/5[b]	80.0 (37.6-96.4)	1945/1946	99.9 (99.7-100)
Prospectively collected samples	Total	7/8	87.5 (52.9-97.8)	2452/2454	99.9 (99.7-100)
Retrospectively collected samples	-	7/7	100 (64.6-100)	439/439	100 (99.1-100)

[a] *M. pneumoniae* was detected in the 1 FP sample using PCR/sequencing.

[b] *M. pneumoniae* was not detected in the 1 FN sample using PCR/sequencing.

Co-detections in prospective clinical samples

The **cobas eplex** RP panel identified a total of 135 prospective samples with multiple organisms detected, or 5.5% of all prospectively collected samples. Of these, 118 (4.8%) had two organisms, 14 (0.6%) had three organisms, and 3 (0.1%) had four organisms detected. Of the 135 co-detected samples, 58 included 1 or more organisms that had not been detected by the comparator method(s). Results are summarized in **Table 37, Table 37a and Table 38**.

Table 37: Distinct co-detection combinations detected by the **cobas eplex** RP panel in the prospective clinical samples

Distinct co-detection combinations detected by the cobas eplex RP Panel Organism 1	Distinct co-detection combinations detected by the cobas eplex RP Panel Organism 2	Distinct co-detection combinations detected by the cobas eplex RP Panel Organism 3	Distinct co-detection combinations detected by the cobas eplex RP Panel Organism 4	Total number Of co-detections (% of samples)	Number of discrepant co-detections	Discrepant organism(s) [a]
ADV	CoV	-	-	2 (0.08%)	0	-
ADV	CoV	HRV/EV	-	2 (0.08%)	1	ADV (1)
ADV	Flu A (unk)	Flu B	HRV/EV	1 (0.04%)	1	ADV (1), Flu A (unk) (1), Flu B (1), HRV/EV (1)
ADV	Flu AH3	-	-	1 (0.04%)	0	-
ADV	Flu B	HRV/EV	RSV B	1 (0.04%)	1	ADV (1), Flu B (1)
ADV	FluA09H1	-	-	1 (0.04%)	1	ADV (1), FluA09H1 (1)
ADV	FluA09H1	HRV/EV	-	1 (0.04%)	0	-
ADV	FluA09H1	PIV 3	-	1 (0.04%)	1	PIV 3 (1)
ADV	HMPV	-	-	3 (0.12%)	2	ADV (2)
ADV	HMPV	HRV/EV	RSV A	1 (0.04%)	1	RSV A (1)
ADV	HRV/EV	-	-	18 (0.73%)	7	ADV (6), HRV/EV (1)
ADV	HRV/EV	Mpneum	-	1 (0.04%)	0	-
ADV	HRV/EV	PIV 1	-	1 (0.04%)	1	PIV 1 (1)
ADV	HRV/EV	PIV 4	-	1 (0.04%)	1	ADV (1), PIV 4 (1)
ADV	HRV/EV	RSV B	-	1 (0.04%)	0	-
ADV	PIV 2	-	-	2 (0.08%)	1	ADV (1)
ADV	PIV 3	-	-	2 (0.08%)	1	ADV (1)
ADV	PIV 4	-	-	1 (0.04%)	1	ADV (1)
ADV	RSV B	-	-	2 (0.08%)	2	ADV (2)
CPneum	HRV/EV	-	-	1 (0.04%)	0	-
CoV	FluA09H1	-	-	1 (0.04%)	0	-
CoV	HMPV	-	-	4 (0.16%)	0	-
CoV	HMPV	HRV/EV	-	2 (0.08%)	0	-
CoV	HRV/EV	-	-	12 (0.49%)	4	CoV (1), HRV/EV (4)
CoV	HRV/EV	RSV B	-	1 (0.04%)	1	CoV (1)
CoV	PIV 1	-	-	1 (0.04%)	0	-
CoV	RSV A	-	-	3 (0.12%)	0	-
CoV	RSV B	-	-	3 (0.12%)	2	CoV (2)
Flu A (unk)	HRV/EV	-	-	1 (0.04%)	1	Flu A (unk) (1)
Flu AH3	HRV/EV	-	-	2 (0.08%)	1	HRV/EV (1)
Flu AH3	RSV B	-	-	1 (0.04%)	0	-
Flu B	HRV/EV	-	-	4 (0.16%)	2	HRV/EV (2)
Flu B	HRV/EV	RSV B	-	1 (0.04%)	0	-
Flu B	PIV 3	-	-	1 (0.04%)	0	-
FluA09H1	HMPV	HRV/EV	-	1 (0.04%)	1	HRV/EV (1)
FluA09H1	HRV/EV	-	-	2 (0.08%)	1	HRV/EV (1)
HMPV	HRV/EV	-	-	5 (0.20%)	1	HRV/EV (1)

Distinct co-detection combinations detected by the cobas eplex RP Panel Organism 1	Distinct co-detection combinations detected by the cobas eplex RP Panel Organism 2	Distinct co-detection combinations detected by the cobas eplex RP Panel Organism 3	Distinct co-detection combinations detected by the cobas eplex RP Panel Organism 4	Total number Of co-detections (% of samples)	Number of discrepant co-detections	Discrepant organism(s) [a]
HMPV	HRV/EV	RSV B	-	1 (0.04%)	1	HRV/EV (1)
HMPV	PIV 3	-	-	1 (0.04%)	0	-
HRV/EV	PIV 1	-	-	3 (0.12%)	0	-
HRV/EV	PIV 2	-	-	7 (0.28%)	3	HRV/EV (1), PIV 2 (2)
HRV/EV	PIV 3	-	-	11 (0.45%)	5	HRV/EV (5)
HRV/EV	PIV 4	-	-	4 (0.16%)	4	PIV 4 (4)
HRV/EV	RSV A	-	-	5 (0.20%)	0	-
HRV/EV	RSV B	-	-	11 (0.45%)	6	HRV/EV (6)
PIV 1	PIV 4	-	-	1 (0.04%)	1	PIV 4 (1)
PIV 3	RSV B	-	-	1 (0.04%)	0	-
RSV A	RSV B	-	-	1 (0.04%)	1	RSV B (1)

Note: ADV= adenovirus, CoV= coronavirus (229E, HKU1, NL63, OC43), HMPV= human metapneumovirus, HRV/EV= human rhinovirus/enterovirus, Flu= Influenza, (unk)= unknown subtype, PIV= parainfluenza, RSV= respiratory syncytial virus, Cpneum= *C. pneumoniae*, Mpneum= *M. pneumoniae*

[a] A discrepant organism is defined as one that was detected by the **cobas eplex** RP panel but not by the comparator method(s).

Table 37a: Total Distinct co-detection combinations detected by the **cobas eplex** RP panel in the prospective clinical samples

Distinct co-detection combinations detected by the cobas eplex RP Panel	Total number Of co-detections (% of samples)	Number of discrepant co-detections	Discrepant organism(s) [a]
Total Number of Co-Detections	135 (5.5%)	57	64/290[b]
Total Number with 2 Organisms Detected	118 (4.8%)	47	49/236
Total Number with 3 Organisms Detected	14 (0.6%)	7	8/42
Total Number with 4 Organisms Detected	3 (0.1%)	3	7/12

[a] A discrepant organism is defined as one that was detected by the **cobas eplex** RP panel but not by the comparator method(s).

[b] 64/64 discrepant organisms were investigated using PCR/sequencing; the discrepant organism was detected in 20/64 cases:

-In 8/18 samples, adenovirus was detected by PCR/sequencing.
-In 1/4 samples, coronavirus was detected by PCR/sequencing.
-In 7/25 samples, human rhinovirus/enterovirus was detected by PCR/sequencing.
-In 1/1 sample, influenza A H1-2009 was detected by PCR/sequencing.
-In 1/1 sample, parainfluenza virus 3 was detected by PCR/sequencing.
-In 2/6 samples, parainfluenza virus 4 was detected by PCR/sequencing.

Table 38: Additional co-detection combinations detected by the comparator method in the prospective clinical samples

Distinct co-detection combinations detected by the comparator method Organism 1	Distinct co-detection combinations detected by the comparator method Organism 2	Distinct co-detection combinations detected by the comparator method Organism 3	Total number Of co-detections (% of samples)	Number of discrepant co-detections	Discrepant organism(s) [a,b]
ADV	CoV	-	1 (0.04%)	1	ADV (1), CoV (1)
ADV	HRV/EV	-	4 (0.16%)	4	ADV (4)
ADV	HRV/EV	PIV 3	1 (0.04%)	1	HRV/EV (1), PIV 3 (1)
ADV	HRV/EV	RSV A	1 (0.04%)	1	ADV (1)
CPneum	HRV/EV	-	1 (0.04%)	1	CPneum (1)
CPneum	PIV 3	-	1 (0.04%)	1	CPneum (1)
CoV	FluA09H1	-	2 (0.08%)	2	CoV (2)
CoV	HMPV	-	1 (0.04%)	1	CoV (1)
CoV	HRV/EV	-	6 (0.24%)	6	CoV (4), HRV/EV (2)
CoV	PIV 3	-	1 (0.04%)	1	CoV (1)
CoV	RSV B	-	3 (0.12%)	3	CoV (2), RSV B (1)
Flu AH3	HRV/EV	PIV 3	1 (0.04%)	1	Flu AH3 (1), PIV 3 (1)
Flu AH3	PIV 3	-	1 (0.04%)	1	PIV 3 (1)
FluA09H1	HMPV	HRV/EV	1 (0.04%)	1	HMPV (1), HRV/EV (1)
HMPV	HRV/EV	-	1 (0.04%)	1	HRV/EV (1)
HRV/EV	PIV 1	-	1 (0.04%)	1	HRV/EV (1)
HRV/EV	PIV 3	-	2 (0.08%)	2	HRV/EV (2)
HRV/EV	PIV 3	RSV B	1 (0.04%)	1	PIV 3 (1)
HRV/EV	RSV A	-	2 (0.08%)	2	RSV A (2)

[a] A discrepant organism is defined as one that was detected by the comparator method(s) but not by the **cobas eplex** RP panel.
[b] 36/36 discrepant organisms were investigated using PCR/sequencing; the discrepant organism was not detected in 10/36 cases:
-In 2/6 samples, adenovirus was not detected by PCR/sequencing.
-In 1/2 samples, *Chlamydia pneumoniae* was not detected by PCR/sequencing.
-In 1/11 samples, coronavirus was not detected by PCR/sequencing.
-In 5/8 samples, human rhinovirus/enterovirus was not detected by PCR/sequencing.
-In 1/1 sample, influenza A H3 was not detected by PCR/sequencing.

Clinical study for cobas eplex RP panel system performance

A total of 3281 samples (including prospective, retrospective, and contrived samples) were initially tested in the clinical evaluations and 3127/3281 = 95.3% (95% CI: 94.5%-96.0%) generated valid results on the first attempt. After re-test, 8 samples had invalid results; final validity rate was 3273/3281 = 99.8% (95% CI: 99.5%-99.9%).

ANALYTICAL PERFORMANCE CHARACTERISTICS

Limit of detection for SARS-CoV-2

The limit of detection (LoD), or analytical sensitivity was identified and verified for SARS-CoV-2 using commercially available heat inactivated quantified virus. Serial dilutions were prepared in a natural clinical matrix (pooled, negative nasopharyngeal swab in VTM) and at least 20 replicates per concentration were tested in the study. The limit of detection was defined as the lowest concentration at which SARS-CoV-2 is detected at least 95% of the time. The confirmed LoD for detection of SARS-CoV-2 is shown in **Table 39**.

Table 39: SARS-CoV-2 LoD results summary

Target	Strain	LoD concentration
SARS-CoV-2	USA-WA1/2020	1 x 10^{-2} $TCID_{50}$/mL[a]

[a] The LoD concentration for detection of SARS-CoV-2 was determined to be 0.01 $TCID_{50}$/mL, which corresponds to 250 genomic copies per milliliter, as determined by digital droplet PCR.

FDA SARS-CoV-2 reference panel testing

The evaluation of sensitivity and MERS-CoV cross-reactivity was performed using reference material (T1), blinded samples and a standard protocol provided by the FDA. The study included a range finding study and a confirmatory study for LoD. Blinded sample testing was used to establish specificity and to corroborate the LoD. The **cobas eplex** RP2 panel showed no cross-reactivity with MERS-CoV at the highest concentration supplied. The LoD of the **cobas eplex** RP2 panel using the FDA SARS-CoV-2 Reference Panel was observed to be 1.8x10^5 NDU/mL. The results of the FDA SARS-CoV-2 Reference Panel testing are summarized in **Table 40**.

Table 40: FDA SARS-CoV-2 reference panel testing summary

FDA reference material	Specimen type	cobas eplex RP2 panel LoD	Cross-reactivity
SARS-CoV-2	NPS	1.8 x 10^5 NDU/mL[a]	N/A
MERS-CoV	NPS	N/A	ND

[a] The sample matrix used for the FDA reference panel, Minimum Essential Media (MEM), is not a media typically used for collection of respiratory specimens; this material was not tested as part of the interfering substances study (shown in Table 67). Additional characterization testing indicated that the diluent used for the FDA SARS-CoV-2 Reference Panel (Minimal Essential Media; MEM) may interfere with the performance of the **cobas eplex** RP2 Panel.
NDU/mL: RNA NAAT detectable units/mL
N/A: Not Applicable
ND: Not Detected

Limit of detection for all other RP2 panel targets

The limit of detection (LoD), or analytical sensitivity was identified and verified for each viral and bacterial target on the **cobas eplex** RP2 panel using quantified reference strains/isolates. Serial dilutions were prepared in a natural clinical matrix (pooled, negative nasopharyngeal swab in VTM samples) with one or more organisms per series, and at least 20 replicates per target were tested in the study. The limit of detection was defined as the lowest concentration at which each target is detected at least 95% of the time. The confirmed LoD for each **cobas eplex** RP2 panel organism is shown in **Table 41.**

Table 41: LoD results summary

Target	Strain	LoD concentration
Adenovirus	Type 1 (C)	1 x 10^3 $TCID_{50}$/mL
Adenovirus	Type 4 (E)	2 x 10^0 $TCID_{50}$/mL
Adenovirus	Type 7 (B)	2 x 10^0 $TCID_{50}$/mL
Coronavirus 229E	229E	1 x 10^0 $TCID_{50}$/mL
Coronavirus HKU1	HKU1[a]	5 x 10^4 copies/mL
Coronavirus NL63	NL63	7.5 x 10^0 $TCID_{50}$/mL
Coronavirus OC43	OC43	5 x 10^2 $TCID_{50}$/mL

Target	Strain	LoD concentration
Human Metapneumovirus	A1 IA3-2002	2 x 10^{-1} $TCID_{50}$/mL
Human Metapneumovirus	Type B[b]	2 x 10^{3} $TCID_{50}$/mL
Human Metapneumovirus	B1 Peru2-2002	2 x 10^{2} $TCID_{50}$/mL
Human Metapneumovirus	B2 Peru1-2002	2.25 x 10^{2} $TCID_{50}$/mL
Human Rhinovirus/Enterovirus	Enterovirus Type 68 (2007)	1 x 10^{0} $TCID_{50}$/mL
Human Rhinovirus/Enterovirus	Rhinovirus 1A	1.5 x 10^{0} $TCID_{50}$/mL
Human Rhinovirus/Enterovirus	Rhinovirus B14	1 x 10^{0} $TCID_{50}$/mL
Human Rhinovirus/Enterovirus	Rhinovirus C[a]	1 x 10^{5} copies/mL
Influenza A	H1N1 Brisbane/59/07	3 x 10^{-1} $TCID_{50}$/mL
Influenza A H1	H1N1 Brisbane/59/07	3 x 10^{-1} $TCID_{50}$/mL
Influenza A H1-2009	NY/01/2009	1 x 10^{-1} $TCID_{50}$/mL
Influenza A H3	A/Perth/16/2009	1 x 10^{1} $TCID_{50}$/mL
Influenza A H3	A/Texas/50/2012	1 x 10^{0} $TCID_{50}$/mL
Influenza A H3	A/Victoria/361/2011	5 x 10^{-1} $TCID_{50}$/mL
Influenza A H3	H3N2 Brisbane/10/07	5 x 10^{1} $TCID_{50}$/mL
Influenza B (Victoria Lineage)	B/Brisbane/60/2008	1 x 10^{0} $TCID_{50}$/mL
Influenza B (Victoria Lineage)	B/Montana/5/2012	1 x 10^{0} $TCID_{50}$/mL
Influenza B (Victoria Lineage)	B/Nevada/03/2011	1 x 10^{0} $TCID_{50}$/mL
Influenza B (Yamagata Lineage)	Florida/02/06	1 x 10^{-1} $TCID_{50}$/mL
Influenza B (Yamagata Lineage)	B/Massachusetts/02/2012	1 x 10^{2} $TCID_{50}$/mL
Influenza B (Yamagata Lineage)	B/Texas/06/2011	1 x 10^{-1} $TCID_{50}$/mL
Influenza B (Yamagata Lineage)	B/Wisconsin/01/2010	1 x 10^{0} $TCID_{50}$/mL
Parainfluenza Virus 1	Clinical Isolate	4 x 10^{-1} $TCID_{50}$/mL
Parainfluenza Virus 2	Clinical Isolate	5 x 10^{1} $TCID_{50}$/mL
Parainfluenza Virus 3	Clinical Isolate	5 x 10^{0} $TCID_{50}$/mL
Parainfluenza Virus 4	4a	3 x 10^{1} $TCID_{50}$/mL
Respiratory Syncytial Virus A	2006 Isolate	1.5 x 10^{0} $TCID_{50}$/mL
Respiratory Syncytial Virus B	CH93(18)-18	2 x 10^{-1} $TCID_{50}$/mL
Chlamydia pneumoniae	AR-39	3 x 10^{2} $TCID_{50}$/mL
Mycoplasma pneumoniae	FH strain of Eaton Agent [NCTC 10119]	3 x 10^{2} CCU/mL

[a] Clinical samples confirmed positive for coronavirus HKU1 and human rhinovirus C by bi-directional sequencing and quantified by real-time RT-PCR were used for determination of LoD.
[b] The human metapneumovirus strain tested was originally identified as A2 IA14-2003. Subsequent vendor communications identified an error and corrected the strain identification as type B.

Analytical reactivity (inclusivity)

Reactivity of SARS-CoV-2 assays

Inclusivity was evaluated using RNA for SARS-CoV-2 (Hong Kong/VM20001061/2020) at 750 copies/mL. Three replicates were tested and all replicates were detected as expected as shown in **Table 42.**

Table 42: Analytical reactivity (inclusivity) results for SARS-CoV-2

Target	Test material	concentration	Percent detected (positive replicates / total)
SARS-CoV-2	Hong Kong/VM20001061/2020 (BEI Resource – Isolated RNA)	750 copies/mL	100% (3/3)

Predicted (*in silico*) reactivity (inclusivity) results for SARS-CoV-2

In silico analysis of sequences from GISAID are conducted routinely to assess the ability of the **cobas eplex** RP2 panel to detect the most recent COVID-19 strains. The results of these analyses, as of January 2024, show that the RP2 panel will detect all variants in circulation. For the most up to date information on detection of SARS-CoV-2 strains currently in circulation, please contact your local affiliate: https://www.roche.com/about/business/roche_worldwide.htm.

Inclusivity of all other RP2 targets

Inclusivity of all other RP2 panel targets was evaluated using a panel of strains/isolates representing the genetic, temporal, and geographic diversity of each target on the panel to demonstrate analytical reactivity. Each strain/isolate was tested in triplicate at 3x LoD in natural clinical matrix (pooled, negative nasopharyngeal swab in VTM samples); if the organism was not detected at this concentration, testing of higher concentrations was performed. Additional *in silico* analysis was also performed on a subset of **cobas eplex** RP2 panel organisms.

All of the strains/isolates tested for inclusivity were detected by the **cobas eplex** RP2 panel. Results of analytical reactivity are shown in **Tables 43-53**.

Table 43: Analytical reactivity (inclusivity) results for adenovirus

Adenovirus species	Serotype	Concentration	Multiple of LoD detected
A	Type 31	3×10^3 $TCID_{50}$/mL	3x
B	Type 3	6×10^0 $TCID_{50}$/mL	3x
B	Type 11	6×10^0 $TCID_{50}$/mL	3x
B	De Wit Type 14	6×10^0 $TCID_{50}$/mL	3x
B	Ch.79 Type 16	2×10^2 $TCID_{50}$/mL	100x[a]
B	Type 21	6×10^0 $TCID_{50}$/mL	3x
B	Compton Type 34	6×10^0 $TCID_{50}$/mL	3x
B	Holden Type 35	6×10^0 $TCID_{50}$/mL	3x
B	Wan Type 50	2×10^1 $TCID_{50}$/mL	10x[b]
C	Type 2	3×10^3 $TCID_{50}$/mL	3x
C	Type 5	3×10^3 $TCID_{50}$/mL	3x
C	Type 6	3×10^3 $TCID_{50}$/mL	3x
D	Type 26	3×10^3 $TCID_{50}$/mL	3x
D	Type 37	3×10^3 $TCID_{50}$/mL	3x
F	Type 40 Dugan	3×10^3 $TCID_{50}$/mL	3x
F	Type 41/ Strain Tak	3×10^3 $TCID_{50}$/mL	3x

[a] *In silico* analysis revealed good homology to primers and probes. Lower sensitivity is likely the result of incorrect estimation of genetic material present in the culture of this or the reference strain ($TCID_{50}$ value is based only on infectious virus particles).
[b] *In silico* analysis revealed that lower sensitivity may be a result of mismatches in the assay primers and/or probes.

Table 44: Analytical reactivity (inclusivity) results for human metapneumovirus

Metapneumovirus subtype	Strain	Concentration	Multiple of LoD detected
Human metapneumovirus	Peru6-2003 G, B2	6.75 x 10^2 $TCID_{50}$/mL	3x

Table 45: Analytical reactivity (inclusivity) results for human rhinovirus/enterovirus

Rhinovirus/enterovirus	Strain	Concentration	Multiple of LoD detected
Human rhinovirus	Type A2	4.5 x 10^0 $TCID_{50}$/mL	3x
Human rhinovirus	Type A7	1.5 x 10^1 $TCID_{50}$/mL	10x[a]
Human rhinovirus	Type A16	4.5 x 10^0 $TCID_{50}$/mL	3x
Human rhinovirus	Type A18	1.5 x 10^2 $TCID_{50}$/mL	100x[a]
Human rhinovirus	Type A34	4.5 x 10^0 $TCID_{50}$/mL	3x
Human rhinovirus	Type A57	4.5 x 10^0 $TCID_{50}$/mL	3x
Human rhinovirus	Type A77	4.5 x 10^0 $TCID_{50}$/mL	3x
Human rhinovirus	277G	4.5 x 10^0 $TCID_{50}$/mL	3x
Human rhinovirus	Type B3	1.5 x 10^1 $TCID_{50}$/mL	10x[a]
Human rhinovirus	Type B17	1.5 x 10^1 $TCID_{50}$/mL	10x[a]
Human rhinovirus	Type B42	4.5 x 10^0 $TCID_{50}$/mL	3x
Human rhinovirus	Type B83	4.5 x 10^0 $TCID_{50}$/mL	3x
Human rhinovirus	Type B84	4.5 x 10^0 $TCID_{50}$/mL	3x
Human rhinovirus	FO2-2547	4.5 x 10^0 $TCID_{50}$/mL	3x
Enterovirus	Type 71	3 x 10^0 $TCID_{50}$/mL	3x
Coxsackievirus	A9	3 x 10^0 $TCID_{50}$/mL	3x
Coxsackievirus	A10	3 x 10^0 $TCID_{50}$/mL	3x
Coxsackievirus	A21	3 x 10^0 $TCID_{50}$/mL	3x
Coxsackievirus	A24	3 x 10^0 $TCID_{50}$/mL	3x
Coxsackievirus	B2	1 x 10^2 $TCID_{50}$/mL	100x[a]
Coxsackievirus	B3	3 x 10^0 $TCID_{50}$/mL	3x
Coxsackievirus	B4	3 x 10^0 $TCID_{50}$/mL	3x
Coxsackievirus	B5	1 x 10^1 $TCID_{50}$/mL	10x[a]
Echovirus	9	3 x 10^0 $TCID_{50}$/mL	3x
Echovirus	E6	1 x 10^1 $TCID_{50}$/mL	10x[b]
Echovirus	25	1 x 10^1 $TCID_{50}$/mL	10x[a]
Echovirus	30	3 x 10^0 $TCID_{50}$/mL	3x
Poliovirus	1	1 x 10^2 $TCID_{50}$/mL	100x[a]

[a] *In silico* analysis revealed that lower sensitivity may be a result of mismatches in the assay primers and/or probes.
[b] *In silico* analysis revealed good homology to primers and probes. Lower sensitivity is likely the result of incorrect estimation of genetic material present in the culture of this or the reference strain ($TCID_{50}$ value is based only on infectious virus particles).

Table 46: Analytical reactivity (inclusivity) results for influenza A

Note: Due to different assays for influenza A matrix and influenza A subtypes on the **cobas eplex** RP panel, if different LoDs are observed for inclusivity for an Influenza A matrix vs. HA subtype, the differences are noted in the Multiple of LoD Detected column.

Influenza A subtype	Strain	Concentration	Multiple of LoD detected
Influenza A H1	A/New Caledonia/20/1999	3 x 10^1 $TCID_{50}$/mL	100x[b]

Influenza A subtype	Strain	Concentration	Multiple of LoD detected
Influenza A H1	A/PR/8/34	9 x 10^{-1} $TCID_{50}$/mL	3x (Influenza A matrix) H1 subtype not detected[a]
Influenza A H1	A/Solomon Islands/3/2006	3 x 10^{0} $TCID_{50}$/mL	10x[b]
Influenza A H1	A/Taiwan/42/06	3 x 10^{1} $TCID_{50}$/mL	100x[b]
Influenza A H3	A/Port Chalmers/1/73	1.5 x 10^{2} $TCID_{50}$/mL	3x
Influenza A H3	A/Nanchang/933/95	1.5 x 10^{2} $TCID_{50}$/mL	3x
Influenza A H3	A/Victoria/3/75	1.5 x 10^{2} $TCID_{50}$/mL	3x
Influenza A H3	A/Wisconsin/67/05	1.5 x 10^{2} $TCID_{50}$/mL	3x
Influenza A 2009 H1N1	A/California/7/2009	3 x 10^{-1} $TCID_{50}$/mL	3x
Influenza A 2009 H1N1	A/Mexico/4108/09	3 x 10^{-1} $TCID_{50}$/mL	3x
Influenza A 2009 H1N1	A/NY/02/2009	3 x 10^{-1} $TCID_{50}$/mL	3x
Influenza A 2009 H1N1	A/Swine NY/03/2009	3 x 10^{-1} $TCID_{50}$/mL	3x

[a] *In silico* analysis revealed little homology between this non-contemporary influenza strain sequence and the H1 primer sequences.
[b] For Influenza A matrix, *in silico* analysis revealed good homology to primers and probes. Lower sensitivity is likely the result of incorrect estimation of genetic material present in the culture of this or the reference strain ($TCID_{50}$ value is based only on infectious virus particles). For H1 subtype, *in silico* analysis revealed that lower sensitivity may be a result of mismatches in the assay primers and/or probes.

Table 47: Analytical reactivity (inclusivity) results for influenza A strains titered with methods different from the reference strain

Influenza A subtype	Strain	Concentration detected
Influenza A H1	A/FM/1/47	2.81 x 10^{4} $CEID_{50}$/mL
Influenza A H1	A/NWS/33	7.40 x 10^{2} $CEID_{50}$/mL (Influenza A matrix) H1 subtype not detected[c]
Influenza A H3	A/Hong Kong/8/68	1.58 x 10^{2} $CEID_{50}$/mL
Influenza A H1N1	A/Virginia/ATCC1/2009	2.90 x 10^{0} PFU/mL
Influenza A H1N1	A/Virginia/ATCC2/2009	6.10 x 10^{2} PFU/mL
Influenza A H1N1	A/Virginia/ATCC3/2009	1.80 x 10^{3} PFU/mL
Influenza A H5N8	A/Gyrfalcon/Washington/41088-6/2014 BPL	1.58 x 10^{3} EID_{50}/mL (Influenza A matrix) No subtype detected[a]
Influenza A H5N2	A/Northern Pintail/Washington/40964/2014 BPL	2.51 x 10^{3} EID_{50}/mL (Influenza A matrix) No subtype detected[a]
Influenza A H7N9	A/ANHUI/1/2013	7.94 x 10^{3} EID_{50}/mL (Influenza A matrix) No subtype detected[b]
Influenza A H3N2v	A/Indiana/21/2012	2.51 x 10^{4} EID_{50}/mL (Influenza A matrix and H3 subtype)

[a] Detection of the H5 Subtype not expected
[b] Detection of the H7 Subtype not expected
[c] *In silico* analysis revealed little homology between this non-contemporary strain sequence and the H1 signal probe/capture probe sequences.
NOTE: $CEID_{50}$/mL= Chick Embryo Infectious Dose; EID_{50}/mL= Egg Infectious Dose; PFU/mL = Plaque Forming Units Quantitation

Supplemental analytical reactivity (inclusivity) for influenza A

For human, avian, and swine influenza strains not available for testing on the **cobas eplex** RP panel, *in silico* analysis was performed. Bioinformatics analysis was used to predict a result based on the number and location of mismatches in the primers, capture probes, and signal probes found in the **cobas eplex** RP panel relative to an alignment of GenBank sequences.

Table 48: Predicted (*in silico*) reactivity (inclusivity) results for influenza A

Influenza A subtype	Host	Strain	GenBank ID	Predicted cobas eplex result
H2N2	Human	A/Albany/20/1957(H2N2)	CY022014	Influenza A
H2N2	Human	Kilbourne F38: A/Korea/426/68 (HA, NA) x A/Puerto Rico/8/34	CY037296	Influenza A
H2N2	Avian	A/chicken/New York/13828-3/1995(H2N2)	CY014822	Influenza A
H2N2	Avian	A/Japan/305/1957(H2N2)	CY014977	Influenza A
H2N2	Avian	A/Korea/426/1968(H2N2)	CY031596	Influenza A
H4N6	Avian	A/Blue-winged teal/Minnesota/Sg-00043/2007(H4N6)	CY063978	Influenza A
H5N1	Avian	A/Peregrine falcon/Aomori/7/2011	AB629716	Influenza A
H5N1	Avian	A/Chicken/West Bengal/239022/2010	CY061305	Influenza A
H5N1	Avian	A/Chicken/West Bengal/193936/2009	GU272009	Influenza A
H5N1	Avian	A/Chicken/Hunan/1/2009	HM172150	Influenza A
H5N1	Avian	A/Chicken/Hunan/8/2008	GU182162	Influenza A
H5N1	Avian	A/Chicken/West Bengal/106181/2008	GU083632	Influenza A
H5N1	Avian	A/Chicken/Primorsky/85/2008	FJ654298	Influenza A
H5N1	Avian	A/Chicken/West Bengal/82613/2008	GU083648	Influenza A
H5N1	Avian	A/Duck/France/080036/2008	CY046185	Influenza A
H5N1	Avian	A/Duck/Vietnam/G12/2008	AB593450	Influenza A
H5N1	Avian	A/Chicken/Thailand/PC-340/2008	EU620664	Influenza A
H5N1	Avian	A/Great egret/Hong Kong/807/2008	CY036240	Influenza A
H5N1	Avian	A/Rook/Rostov-on-Don/26/2007(H5N1)	EU814504	Influenza A
H5N1	Avian	A/Turkey/VA/505477-18/2007(H5N1)	GU186510	Influenza A
H5N1	Avian	A/Chicken/Bangladesh/1151-10/2010(H5N1)	HQ156766	Influenza A
H5N1	Human	A/Bangladesh/3233/2011	CY088772	Influenza A
H5N1	Human	A/Cambodia/R0405050/2007(H5N1)	HQ200572	Influenza A
H5N1	Human	A/Cambodia/S1211394/2008	HQ200597	Influenza A
H5N1	Human	A/Hong Kong/486/97(H5N1)	AF255368	Influenza A
H5N1	Swine	A/Swine/East Java/UT6010/2007(H5N1)	HM440124	Influenza A
H5N2	Avian	A/Duck/Pennsylvania/10218/1984(H5N2)	AB286120	Influenza A
H5N2	Avian	A/American black duck/Illinois/08OS2688/2008	CY079453	Influenza A
H5N2	Avian	A/American green-winged teal/California/HKWF609/2007	CY033447	Influenza A
H5N2	Avian	A/Canada goose/New York/475813-2/2007	GQ923358	Influenza A
H5N2	Avian	A/Blue-winged teal/Saskatchewan/22542/2007	CY047705	Influenza A
H5N2	Avian	A/Chicken/Taiwan/A703-1/2008	AB507267	Influenza A
H5N2	Avian	A/Duck/France/080032/2008	CY046177	Influenza A
H5N2	Avian	A/Duck/New York/481172/2007	GQ117202	Influenza A
H5N2	Avian	A/Gadwall/Altai/1202/2007	CY049759	Influenza A
H5N2	Avian	A/Mallard/Louisiana/476670-4/2007	GQ923390	Influenza A
H5N2	Avian	A/Waterfowl/Colorado/476466-2/2007	GQ923374	Influenza A
H5N3	Avian	A/Duck/Singapore/F119/3/1997(H5N3)	GU052803	Influenza A

Influenza A subtype	Host	Strain	GenBank ID	Predicted cobas eplex result
H6N1	Avian	A/Duck/PA/486/1969(H6N1)	EU743287	Influenza A
H6N2	Avian	A/Mallard/Czech Republic/15902-17K/2009(H6N2)	HQ244433	Influenza A
H7N2	Avian	A/Chicken/Hebei/1/2002	AY724263	Influenza A
H7N2	Avian	A/Chicken/PA/149092-1/02	AY241609	Influenza A
H7N2	Avian	A/Chicken/NJ/294508-12/2004	EU743254	Influenza A
H7N2	Avian	A/Chicken/New York/23165-6/2005	CY031077	Influenza A
H7N2	Avian	A/Muscovy duck/New York/23165-13/2005	CY033226	Influenza A
H7N2	Avian	A/Muscovy duck/New York/87493-3/2005	CY034791	Influenza A
H7N2	Avian	A/Mallard/Netherlands/29/2006	CY043833	Influenza A
H7N2	Avian	A/Northern shoveler/California/JN1447/2007	CY076873	Influenza A
H7N2	Human	A/New York/107/2003(H7N2)	EU587373	Influenza A
H7N3	Human	A/Canada/rv504/2004(H7N3)	CY015007	Influenza A
H7N7	Avian	A/American green-winged teal/Mississippi/09OS046/2009	CY079309	Influenza A
H7N7	Avian	A/Chicken/Germany/R28/03	AJ619676	Influenza A
H7N7	Avian	A/Chicken/Netherlands/1/03	AY340091	Influenza A
H7N7	Avian	A/Mallard/California/HKWF1971/2007	CY033383	Influenza A
H7N7	Avian	A/Mallard/Korea/GH171/2007	FJ959087	Influenza A
H7N7	Avian	A/Mute swan/Hungary/5973/2007	GQ240816	Influenza A
H7N7	Avian	A/Northern shoveler/Mississippi/ 09OS643/2009	CY079413	Influenza A
H7N7	Human	A/Netherlands/219/03(H7N7)	AY340089	Influenza A
H7N9	Human	A/Shanghai/1/2013(H7N9)	EPI439493	Influenza A
H7N9	Avian	A/Northern shoveler/Mississippi/11OS145/2011(H7N9)	CY133650	Influenza A
H7N9	Avian	A/Ruddy turnstone/Delaware Bay/220/1995(H7N9)	CY127254	Influenza A
H7N9	Avian	A/Turkey/Minnesota/1/1988(H7N9)	CY014787	Influenza A
H7N9	Avian	A/Blue-winged teal/Ohio/566/2006(H7N9)	CY024819	Influenza A
H9N2	Human	A/Hong Kong/1073/99(H9N2)	AJ278647	Influenza A
H9N2	Avian	A/Turkey/Wisconsin/1/1966(H9N2)	CY014664	Influenza A
H10N7	Avian	A/chicken/Germany/N/1949(H10N7)	GQ176135	Influenza A
H11N9	Avian	A/Duck/Memphis/546/1974(H11N9)	GQ257441	Influenza A
H1N1	Swine	A/Swine/Wisconsin/1/1971(H1N1)	CY022414	Influenza A
H1N1	Human	A/California/UR06-0393/2007(H1N1)	CY026540	Influenza A H1
H1N1	Human	A/California/UR06-0393/2007(H1N1)	CY026539	Influenza A H1
H1N2	Human	A/New York/297/2003(H1N2)	CY002664	Influenza A H1
H1N2	Human	A/New York/297/2003(H1N2)	CY002665	Influenza A H1
H1N1 (2009)	Human	A/Aalborg/INS133/2009(H1N1)	CY063606	Influenza A H1-2009
H1N1 (2009)	Human	A/Aalborg/INS133/2009(H1N1)	CY063607	Influenza A H1-2009
H1N1 (2009)	Human	A/South Carolina/02/2010(H1N1)	KC781370	Influenza A H1-2009

Influenza A subtype	Host	Strain	GenBank ID	Predicted cobas eplex result
H1N1 (2009)	Human	A/South Carolina/02/2010(H1N1)	KC781372	Influenza A H1-2009
H1N2	Swine	A/Swine/Hong Kong/NS857/2001(H1N2)	GQ229350	Influenza A
H1N2	Swine	A/Swine/Sweden/1021/2009(H1N2)	GQ495135	Influenza A
H3N1	Avian	A/Blue-winged teal/ALB/452/1983(H3N1)	CY004635	Influenza A
H3N2v	Human	A/Iowa/07/2011(H3N2)	JQ070760	Influenza A H3
H3N2v	Human	A/Iowa/07/2011(H3N2)	JQ290177	Influenza A H3
H3N2v	Human	A/Iowa/08/2011(H3N2)	JQ070768	Influenza A H3
H3N2v	Human	A/Iowa/08/2011(H3N2)	JQ290167	Influenza A H3
H3N2v	Human	A/Iowa/09/2011(H3N2)	JQ070776	Influenza A H3
H3N2v	Human	A/Iowa/09/2011(H3N2)	JQ290183	Influenza A H3
H3N2v	Human	A/Indiana/08/2011(H3N2)	JQ070800	Influenza A H3
H3N2v	Human	A/Indiana/08/2011(H3N2)	JQ070795	Influenza A H3
H3N2v	Human	A/Maine/06/2011(H3N2)	JN866181	Influenza A H3
H3N2v	Human	A/Maine/06/2011(H3N2)	JN866186	Influenza A H3
H3N2v	Human	A/Maine/07/2011(H3N2)	JN992746	Influenza A
H3N2v	Human	A/Pennsylvania/09/2011(H3N2)	JN655534	Influenza A
H3N2v	Human	A/Pennsylvania/11/2011(H3N2)	JN655540	Influenza A
H3N2v	Human	A/Pennsylvania/10/2011(H3N2)	JN655550	Influenza A
H3N2v	Human	A/West Virginia/06/2011(H3N2)	JQ290159	Influenza A H3
H3N2v	Human	A/West Virginia/06/2011(H3N2)	JQ290164	Influenza A H3
H3N2v	Human	A/West Virginia/07/2011(H3N2)	JQ348839	Influenza A
H3N2v	Human	A/Indiana/10/2011(H3N2)	KJ942592	Influenza A H3
H3N2v	Human	A/Indiana/10/2011(H3N2)	JQ070787	Influenza A H3
H3N2v	Human	A/Boston/38/2008(H3N2)	CY044580	Influenza A H3
H3N2v	Human	A/Boston/38/2008(H3N2)	CY044581	Influenza A H3
H3N2v	Swine	A/swine/NY/A01104005/2011(H3N2v)	JN940422	Influenza A H3
H3N2v	Swine	A/Maine/06/2011(H3N2)	JN866181	Influenza A H3
H3N2v	Swine	A/Maine/06/2011(H3N2)	JN866186	Influenza A H3
H3N2v	Swine	A/Indiana/08/2011(H3N2)	JN655558	Influenza A H3
H3N2v	Swine	A/Indiana/08/2011(H3N2)	JN638733	Influenza A H3
H3N2v	Avian	A/American black duck/North Carolina/675-075/2004(H3N2)	GU051135	Influenza A
H3N2v	Avian	A/American black duck/North Carolina/675-075/2004(H3N2)	GU051136	Influenza A
H3N5	Avian	A/Mallard/Netherlands/2/1999(H3N5)	CY060261	Influenza A
H3N5	Avian	A/Mallard/Netherlands/2/1999(H3N5)	CY060264	Influenza A
H3N6	Avian	A/American black duck/New Brunswick/25182/2007(H3N6)	CY047696	Influenza A
H3N6	Avian	A/American black duck/New Brunswick/25182/2007(H3N6)	CY047697	Influenza A
H3N7	Avian	A/Northern shoveler/California/HKWF1367/2007(H3N7)	CY033372	Influenza A
H3N7	Avian	A/Northern shoveler/California/HKWF1367/2007(H3N7)	CY033375	Influenza A

Influenza A subtype	Host	Strain	GenBank ID	Predicted cobas eplex result
H3N8	Avian	A/American black duck/Washington/699/1978(H3N8)	GU052300	Influenza A H3
H3N8	Avian	A/American black duck/Washington/699/1978(H3N8)	GU052299	Influenza A H3

Table 49: Analytical reactivity (inclusivity) results for influenza B

Influenza B subtype	Strain	Concentration	Multiple of LoD detected
Influenza B (Yamagata lineage)	B/Lee/40	3 x 10^{-1} $TCID_{50}$/mL	3x
Influenza B (Yamagata lineage)	B/Allen/45	1 x 10^{0} $TCID_{50}$/mL	10x[a]
Influenza B (Yamagata lineage)	B/Maryland/1/59	3.38 x 10^{1} $CEID_{50}$/mL	N/A (Strain titered differently from reference strain)
Influenza B (Yamagata lineage)	B/Taiwan/2/62	1 x 10^{2} $TCID_{50}$/mL	1000x[a]
Influenza B (Victoria lineage)	B/Hong Kong/5/72	1 x 10^{1} $TCID_{50}$/mL	100x[b]
Influenza B (Victoria lineage)	B/Malaysia/2506/04	3 x 10^{-1} $TCID_{50}$/mL	3x
Influenza B (Lineage unknown)	B/GL/1739/54	3 x 10^{-1} $TCID_{50}$/mL	3x

[a] No sequence data available. Lower sensitivity may be a result of mismatches in the assay primers and/or probes. In addition, the reduced sensitivity may be the result of incorrect estimation of genetic material present in the culture of this or the reference strain ($TCID_{50}$/mL value is based only on infectious virus particles).
[b] *In silico* analysis revealed that lower sensitivity may be a result of mismatches in the assay primers and/or probes.

Table 50: Analytical reactivity (inclusivity) results for parainfluenza virus

Parainfluenza subtype	Strain	Concentration	Multiple of LoD detected
Parainfluenza virus 1	C35	1.2 x 10^{0} $TCID_{50}$/mL	3x
Parainfluenza virus 2	Greer	1.5 x 10^{2} $TCID_{50}$/mL	3x
Parainfluenza virus 3	C-243	5 x 10^{1} $TCID_{50}$/mL	10x[a]
Parainfluenza virus 4	4b	9 x 10^{1} $TCID_{50}$/mL	3x

[a] *In silico* analysis revealed that lower sensitivity may be a result of mismatches in the assay primers and/or probes.

Table 51: Analytical reactivity (inclusivity) results for respiratory syncytial virus

RSV Subtype	Strain	Concentration	Multiple of LoD detected
Respiratory syncytial virus A	A2	4.5 x 10^{0} $TCID_{50}$/mL	3x
Respiratory syncytial virus A	Long	4.5 x 10^{0} $TCID_{50}$/mL	3x
Respiratory syncytial virus B	9320	6 x 10^{-1} $TCID_{50}$/mL	3x
Respiratory syncytial virus B	Wash/18537/62	6 x 10^{-1} $TCID_{50}$/mL	3x
Respiratory syncytial virus B	WV/14617/85	6 x 10^{-1} $TCID_{50}$/mL	3x

Table 52: Analytical reactivity (inclusivity) results for *Chlamydia pneumoniae*

-	Strain	Concentration	Multiple of LoD detected
Chlamydia pneumoniae	CWL-029	9 x 10^{2} CFU/mL	3x
Chlamydia pneumoniae	TWAR strain 2043	9 x 10^{2} CFU/mL	3x

Table 53: Analytical reactivity (inclusivity) results for *Mycoplasma pneumoniae*

-	Strain	Concentration	Multiple of LoD detected
Mycoplasma pneumoniae	[Bru]	9 x 10^2 CCU/mL	3x
Mycoplasma pneumoniae	M129-B170	9 x 10^2 CCU/mL	3x
Mycoplasma pneumoniae	M129-B7	9 x 10^2 CCU/mL	3x
Mycoplasma pneumoniae	[M52]	9 x 10^2 CCU/mL	3x
Mycoplasma pneumoniae	[Mac]	9 x 10^2 CCU/mL	3x
Mycoplasma pneumoniae	Mutant 22	3 x 10^4 CCU/mL	100x[a]
Mycoplasma pneumoniae	PI 1428	3 x 10^4 CCU/mL	100x[b]

[a] No sequence data available. Lower sensitivity may be a result of mismatches in the assay primers and/or probes. In addition, the reduced sensitivity may be the result of incorrect estimation of genetic material present in the culture of this or the reference strain (CCU/ml value is based only on live bacteria).
[b] *In silico* analysis revealed good homology to primers and probes.The reduced sensitivity is likely the result of incorrect estimation of genetic material present in the culture of this or the reference strain (CCU/ml value is based only on live bacteria).

Analytical specificity (Cross-reactivity and exclusivity)

Cross-reactivity of the SARS-CoV-2 assays

Cross-reactivity of the SARS-CoV-2 assays was evaluated using both *in silico* analysis and by testing quantified analytes for organisms likely to be found in circulation and other pathogens in the same genetic family. Synthetic constructs were used for analytes where high-titer cultures were not available (SARS-CoV-1, MERS-CoV, human bocavirus, and coronavirus HKU1). A pool of two to four analytes were tested in triplicate. Viral analytes were diluted to testing concentrations ranging from 1x10^4 - 1x10^6 $TCID_{50}$/mL. Bacterial and fungal analytes were diluted to a testing concentration of 1x10^7 - 1x10^8 CFU/mL. Synthetic constructs were tested at a concentration of 1x10^5 - 1x10^6 copies/mL. The parainfluenza virus 3 (PIV3) analyte is a clinical sample that was used as a diluent to generate a viral pool and therefore a viral concentration is not provided. A summary of the results of cross-reactivity testing are shown in **Table 54** below. At high titers, cross-reactivity with SARS-CoV-1 was observed with the **cobas eplex** RP2 panel.

Table 54: Cross-reactivity of SARS-CoV-2 assays with on and off-panel organisms

Virus/bacteria	Strain	Concentration	Cross-reactivity
Adenovirus C	1	1 x 10^3 $TCID_{50}$/mL	Not observed
Coronavirus	229E	1 x 10^4 $TCID_{50}$/mL	Not observed
Coronavirus	HKU1[a]	1 x 10^5 copies/mL	Not observed
Coronavirus	NL63	1 x 10^5 $TCID_{50}$/mL	Not observed
Coronavirus	OC43	1 x 10^6 $TCID_{50}$/mL	Not observed
Coronavirus	MERS-CoV[b]	1 x 10^5 copies/mL	Not observed
Coronavirus	SARS-CoV-1[a]	1 x 10^6 copies/mL	Observed
Human bocavirus	HBoV1[b]	1 x 10^6 copies/mL	Not observed
Echovirus	30	1 x 10^5 $TCID_{50}$/mL	Not observed
Enterovirus	68	1 x 10^5 $TCID_{50}$/mL	Not observed
Influenza A	H1N1/NY01/2009	1 x 10^{-1} $TCID_{50}$/mL	Not observed
Influenza B	Yamagata B/Florida/02/06	1 x 10^{-1} $TCID_{50}$/mL	Not observed
Human metapneumovirus	B2 Peru1-2002	1 x 10^5 $TCID_{50}$/mL	Not observed
Parainfluenza	1	1 x 10^5 $TCID_{50}$/mL	Not observed
Parainfluenza	2	1 x 10^5 $TCID_{50}$/mL	Not observed

Virus/bacteria	Strain	Concentration	Cross-reactivity
Parainfluenza	3	N/A	Not observed
Parainfluenza	4a	1 x 10^5 $TCID_{50}$/mL	Not observed
Respiratory syncytial virus A	2006	1 x 10^5 $TCID_{50}$/mL	Not observed
Human rhinovirus	B14	1 x 10^6 $TCID_{50}$/mL	Not observed
Bordetella pertussis	ATCC53894	1 x 10^8 CFU/mL	Not observed
Candida albicans	ATCC24433	1 x 10^7 CFU/mL	Not observed
Corynebacterium diphtheriae	ATCC53281	1 x 10^7 CFU/mL	Not observed
Haemophilus influenzae	ATCC43065	1 x 10^8 CFU/mL	Not observed
Legionella pneumophila	ATCC35096	1 x 10^8 CFU/mL	Not observed
Mycobacterium tuberculosis	ATCC25177	1 x 10^8 CFU/mL	Not observed
Moraxella catarrhalis	ATCC23246	1 x 10^7 CFU/mL	Not observed
Mycoplasma pneumoniae	ATCC29085	1 x 10^8 CFU/mL	Not observed
Neisseria meningitidis	NCTC10026	1 x 10^8 CFU/mL	Not observed
Pseudomonas aeruginosa	ATCC BAA-1744	1 x 10^8 CFU/mL	Not observed
Staphylococcus aureus	ATCC25923	1 x 10^8 CFU/mL	Not observed
Staphylococcus epidermidis	ATCC700567	1 x 10^8 CFU/mL	Not observed
Staphylococcus salivarius	ATCC25975	1 x 10^7 CFU/mL	Not observed
Streptococcus pneumoniae	ATCC49136	1 x 10^8 CFU/mL	Not observed
Streptococcus pyogenes	ATCC49399	1 x 10^8 CFU/mL	Not observed
Pooled nasal swab	Human clinical sample	N/A	Not observed

[a] in vitro transcript

[b] plasmid

In silico analysis of the cobas eplex RP2 panel SARS-CoV-2 assays

In silico analysis was performed for the gene regions targeted by the **cobas eplex** RP2 panel to evaluate cross-reactivity. GenMark conducted a primer BLAST® search of the NCBI database against all bacteria, negative-stranded RNA viruses (negarnaviricota), picornaviruses, adenoviruses, common human coronaviruses, MERS, *Candida albicans*, and *Pneumocystis*. The BLAST searches did not identify any cross-reactivity with the exception of SARS coronavirus, which is in the same subgenus (Sarbecovirus) as SARS-CoV-2.

Cross-reactivity and exclusivity of other RP2 panel targets

The design of the **cobas eplex** RP2 panel incorporates assays for the detection of SARS-CoV-2 without affecting the original designs of the **cobas eplex** RP panel assays. The original RP panel targets impacted by the addition of the SARS-CoV-2 assays (influenza A, influenza A H1, influenza A H1-2009, influenza A H3, influenza B, and adenovirus) were tested and no cross-reactivity was observed. Therefore, the established cross-reactivity claims of the **cobas eplex** RP panel are applicable to the **cobas eplex** RP2 panel.

Cross-reactivity of each viral and bacterial target on the **cobas eplex** RP panel was evaluated at high concentrations (1 x 10^5 $TCID_{50}$/mL for viruses, 1 x 10^6 CFU/mL or CCU/mL for bacterial isolates, or 1 x 10^6 copies/mL for *in vitro* transcripts) of quantified strains/isolates diluted in viral transport media. *In vitro* transcript for coronavirus HKU1 was diluted in PBS. Additional Influenza A strains were tested at the following concentrations: Influenza A H7N9 at 7.94 x 10^5 EID_{50}/mL, Influenza A H3N2v at 2.51 x 10^5

EID_{50}/mL, Influenza A H5N2 at 2.51 x 10^5 EID_{50}/mL, Influenza A H5N8 at 1.58 x 10^5 EID_{50}/mL. **Table 55** summarizes the results of the on-panel viral and bacterial strains/isolates tested. No cross-reactivity was observed between any of the on-panel viruses or bacteria.

Table 55: Cross-reactivity with **cobas eplex** RP panel target organisms

Target	Strain	Concentration	Cross-reactivity results
Adenovirus A	Type 31	1 x 10^5 $TCID_{50}$/mL	Not observed
Adenovirus B	Type 7A	1 x 10^5 $TCID_{50}$/mL	Not observed
Adenovirus C	Type 1	1 x 10^5 $TCID_{50}$/mL	Not observed
Adenovirus D	Type 9	1 x 10^5 $TCID_{50}$/mL	Not observed
Adenovirus E	Type 4	1 x 10^5 $TCID_{50}$/mL	Not observed
Adenovirus F	Type 41	1 x 10^5 $TCID_{50}$/mL	Not observed
Coronavirus	229E	1 x 10^5 $TCID_{50}$/mL	Not observed
Coronavirus	HKU1 *in vitro* transcript	1 x 10^6 copies/mL	Not observed
Coronavirus	NL63	1 x 10^5 $TCID_{50}$/mL	Not observed
Coronavirus	OC43	1 x 10^5 $TCID_{50}$/mL	Not observed
Enterovirus	Type 68 2007 isolate	1 x 10^5 $TCID_{50}$/mL	Not observed
Human metapneumovirus	B1	1 x 10^5 $TCID_{50}$/mL	Not observed
Human rhinovirus	1A	1 x 10^5 $TCID_{50}$/mL	Not observed
Influenza A	A/Brisbane/59/07	1 x 10^5 $TCID_{50}$/mL	Not observed
Influenza A H1	A/Brisbane/59/07	1 x 10^5 $TCID_{50}$/mL	Not observed
Influenza A H1-2009	A/NY/01/2009	1 x 10^5 $TCID_{50}$/mL	Not observed
Influenza A H3	A/Brisbane/10/07	1 x 10^5 $TCID_{50}$/mL	Not observed
Influenza A H3N2v[a]	A/Indiana/21/2012	2.51 x 10^5 EID_{50}/mL	Not observed
Influenza A H5N2[b]	A/Northern Pintail Washington/40964/14BPL	2.51 x 10^5 EID_{50}/mL	Not observed
Influenza A H5N8[c]	A/Gyrfalcon/Washington /410886/2014 BPL	1.58 x 10^5 EID_{50}/mL	Not observed
Influenza A H7N9[d]	A/ANHUI/1/2013	7.94 x 10^5 EID_{50}/mL	Not observed
Influenza B	B/Florida/02/06	1 x 10^5 $TCID_{50}$/mL	Not observed
Parainfluenza Virus 1	C35	1 x 10^5 $TCID_{50}$/mL	Not observed
Parainfluenza Virus 2	Type 2	1 x 10^5 $TCID_{50}$/mL	Not observed
Parainfluenza Virus 3	Type 3	1 x 10^5 $TCID_{50}$/mL	Not observed
Parainfluenza Virus 4	Type 4a	1 x 10^5 $TCID_{50}$/mL	Not observed
RSV A	2006 Isolate	1 x 10^5 $TCID_{50}$/mL	Not observed
RSV B	CH93(18)-18	1 x 10^5 $TCID_{50}$/mL	Not observed
Chlamydia pneumoniae	AR-39	1 x 10^6 CFU/mL	Not observed
Mycoplasma pneumoniae	FH strain of Eaton Agent [NCTC 10119]	1 x 10^6 CCU/mL	Not observed

[a] Influenza A H3N2v detected as influenza A, influenza A H3

[b] Influenza A H5N2 detected as influenza A

[c] Influenza A H5N8 detected as influenza A

[d] Influenza A H7N9 detected as influenza A

Cross-reactivity of viruses, bacteria, and fungi that are not targets on the **cobas eplex** RP panel was evaluated at high concentrations (1 x 10^5 $TCID_{50}$/mL for viruses, 1 x 10^6 CFU/mL for bacterial and yeast

isolates, or 1 x 10^5 - 1 x 10^6 copies/mL for plasmid DNA or genomic RNA) by diluting quantified strains in viral transport media. Plasmid for bocavirus and genomic RNA for MERS coronavirus (MERS-CoV) were diluted in PBS. Varicella zoster virus was not diluted and was tested at the stock titer of 8.9 x 10^3 $TCID_{50}$/mL. **Table 56** summarizes the results of the strains tested. No cross-reactivity was observed between any of the off-panel viruses, bacteria or fungi with the **cobas eplex** RP panel targets.

Table 56: Cross-reactivity with organisms not detected by the **cobas eplex** RP panel (exclusivity)

Target	Strain	Concentration	Cross-reactivity results
Acinetobacter baumanii	ATCC® 19606	1 x 10^6 CFU/mL	Not observed
Bordetella pertussis	18323 [NCTC 10739]	1 x 10^6 CFU/mL	Not observed
Bordetella parapertussis	ATCC 15311	1 x 10^6 CFU/mL	Not observed
Burkholderia cepacia	ATCC 25416	1 x 10^6 CFU/mL	Not observed
Candida albicans	ATCC 10231	1 x 10^6 CFU/mL	Not observed
Candida glabrata	ATCC 15126	1 x 10^6 CFU/mL	Not observed
MERS coronavirus (MERS-CoV)	EMC/2012[a]	1 x 10^5 copies/mL	Not observed
Corynebacterium diphtheriae	ATCC 13812	1 x 10^6 CFU/mL	Not observed
Cytomegalovirus	AD 169	1 x 10^5 $TCID_{50}$/mL	Not observed
Epstein barr virus	Strain B95-8	1 x 10^5 $TCID_{50}$/mL	Not observed
Escherichia coli	ATCC 10279	1 x 10^6 CFU/mL	Not observed
Haemophilus influenzae	ATCC 43065	1 x 10^6 CFU/mL	Not observed
Herpes simplex virus	Isolate 2	1 x 10^5 $TCID_{50}$/mL	Not observed
Human bocavirus	Bocavirus plasmid[b]	1 x 10^6 copies/mL	Not observed
Klebsiella pneumoniae	ATCC 51504	1 x 10^6 CFU/mL	Not observed
Lactobacillus acidophilus	ATCC 314	1 x 10^6 CFU/mL	Not observed
Lactobacillus plantarum	ATCC 8014	1 x 10^6 CFU/mL	Not observed
Legionella pneumophila	Philadelphia-1	1 x 10^6 CFU/mL	Not observed
Measles	N/A	1 x 10^5 $TCID_{50}$/mL	Not observed
Moraxella catarrhalis	ATCC 23246	1 x 10^6 CFU/mL	Not observed
Mumps	Isolate 2	1 x 10^5 $TCID_{50}$/mL	Not observed
Mycobacterium tuberculosis	ATCC 25177	1 x 10^6 CFU/mL	Not observed
Neisseria meningiditis	ATCC 13077	1 x 10^6 CFU/mL	Not observed
Neisseria sicca	ATCC 29193	1 x 10^6 CFU/mL	Not observed
Porphyromonas gingivalis	ATCC 33277	1 x 10^6 CFU/mL	Not observed
Proteus vulgaris	ATCC 33420	1 x 10^6 CFU/mL	Not observed
Pseudomonas aeruginosa	ATCC 15442	1 x 10^6 CFU/mL	Not observed
Serratia marcescens	ATCC 13880	1 x 10^6 CFU/mL	Not observed
Staphylococcus aureus (MRSA)	NRS384	1 x 10^6 CFU/mL	Not observed
Staphylococcus aureus (MSSA)	ATCC 25923	1 x 10^6 CFU/mL	Not observed
Staphylococcus epidermidis (MRSE)	ATCC 35983	1 x 10^6 CFU/mL	Not observed
Staphylococcus epidermidis (MSSE)	ATCC 49134	1 x 10^6 CFU/mL	Not observed
Staphylococcus haemolyticus	ATCC 29970	1 x 10^6 CFU/mL	Not observed
Streptococcus agalactiae	ATCC 12401	1 x 10^6 CFU/mL	Not observed
Streptococcus dysgalactiae	ATCC 35666	1 x 10^6 CFU/mL	Not observed
Streptococcus mitis	ATCC 15914	1 x 10^6 CFU/mL	Not observed

Target	Strain	Concentration	Cross-reactivity results
Streptococcus pneumoniae	ATCC 49619	1 x 10^6 CFU/mL	Not observed
Streptococcus pyogenes	ATCC 12384	1 x 10^6 CFU/mL	Not observed
Streptococcus salivarius	ATCC 13419	1 x 10^6 CFU/mL	Not observed
Varicella Zoster Virus	82	8.9 x 10^3 $TCID_{50}$/mL	Not observed

[a] Extracted genomic RNA

[b] Plasmid does not contain full length viral genome.

Reproducibility

A multisite reproducibility study of the **cobas eplex** RP panel was performed to evaluate agreement with expected results across major sources of variability, such as site-to-site, lot-to-lot, day-to-day, and operator-to-operator. Testing occurred at 3 sites (2 external, 1 internal) on one **cobas eplex** instrument per site with either 3 or 4 towers. Two operators performed testing at each site on 6 days (5 nonconsecutive days) with 3 unique lots of RP panel cartridges. A reproducibility panel consisting of 3 panel members with 6 organisms (representing 7 RP Panel targets) at 3 concentrations (moderate positive- 3x LoD, low positive- 1x LoD, and negative) was tested in triplicate. The 6 organisms tested included adenovirus, coronavirus (229E, HKU1, NL63, OC43), human metapneumovirus, influenza A H3, parainfluenza virus 1, and RSV A; organisms were diluted in natural clinical matrix (pooled, negative nasopharyngeal swab samples). Negative samples consisted of natural clinical matrix only. Each simulated sample was divided into aliquots and stored frozen (-70 °C) prior to testing. Each operator tested 9 samples (3 member reproducibility panel in triplicate) each day; each panel member was tested 108 times (3 replicates x 3 sites x 2 operators x 3 lots x 2 days of testing/operator/lot) for a maximum of 324 tests. After completion of initial and repeat testing for invalid results, 1 low positive sample tested at Site 3 had an invalid result and was excluded from reproducibility performance analyses.

Percent agreement (95% CI) with expected results was 100% for all 7 targets for the moderate positive and negative panel, and 100% for 6 of 7 low positive panel targets (coronavirus, human metapneumovirus, influenza A, influenza A H3, parainfluenza 1, and RSV A); percent agreement was 91.6% for adenovirus. Summary results for the 7 **cobas eplex** RP panel targets that correspond to the 6 organisms in the reproducibility panel are provided in **Tables 57-63**. Summary results for the 10 **cobas eplex** RP panel targets that did not have organisms included in the reproducibility panel are provided in **Table 64**.

Table 57: Percent agreement for adenovirus

Adenovirus concentration	Site	Agreement with expected results Agreed / N	Agreement with expected results %	Agreement with expected results 95% CI
Moderate positive 3x LoD 6 x 10^0 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Moderate positive 3x LoD 6 x 10^0 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Moderate positive 3x LoD 6 x 10^0 $TCID_{50}$/mL	3	36/36	100	(90.4-100)
Moderate positive 3x LoD 6 x 10^0 $TCID_{50}$/mL	All	108/108	100	(96.6-100)

Adenovirus concentration	Site	Agreement with expected results Agreed / N	Agreement with expected results %	Agreement with expected results 95% CI
Low positive 1x LoD 2 x 10^0 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Low positive 1x LoD 2 x 10^0 $TCID_{50}$/mL	2	34/36	94.4	(81.9-98.5)
Low positive 1x LoD 2 x 10^0 $TCID_{50}$/mL	3	28/35	80.0	(64.1-90.0)
Low positive 1x LoD 2 x 10^0 $TCID_{50}$/mL	All	98/107	91.6	(84.8-95.5)
Negative	1	36/36	100	(90.4-100)
Negative	2	36/36	100	(90.4-100)
Negative	3	36/36	100	(90.4-100)
Negative	All	108/108	100	(96.6-100)

CI=Confidence Interval

Table 58: Percent agreement for coronavirus (229E, HKU1, NL63, OC43)

Coronavirus (229E, HKU1, NL63, OC43) concentration	Site	Agreement with expected results Agreed / N	Agreement with expected results %	Agreement with expected results 95% CI
Moderate positive 3x LoD 1.5 x 10^3 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.5 x 10^3 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.5 x 10^3 $TCID_{50}$/mL	3	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.5 x 10^3 $TCID_{50}$/mL	All	108/108	100	(96.6-100)
Low positive 1x LoD 5 x 10^2 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Low positive 1x LoD 5 x 10^2 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Low positive 1x LoD 5 x 10^2 $TCID_{50}$/mL	3	35/35	100	(90.1-100)
Low positive 1x LoD 5 x 10^2 $TCID_{50}$/mL	All	107/107	100	(96.5-100)
Negative	1	36/36	100	(90.4-100)
Negative	2	36/36	100	(90.4-100)
Negative	3	36/36	100	(90.4-100)
Negative	All	108/108	100	(96.6-100)

Table 59: Percent agreement for human metapneumovirus

Human metapneumovirus concentration	Site	Agreement with expected results Agreed / N	Agreement with expected results %	Agreement with expected results 95% CI
Moderate positive 3x LoD 6.75×10^2 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Moderate positive 3x LoD 6.75×10^2 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Moderate positive 3x LoD 6.75×10^2 $TCID_{50}$/mL	3	36/36	100	(90.4-100)
Moderate positive 3x LoD 6.75×10^2 $TCID_{50}$/mL	All	108/108	100	(96.6-100)
Low positive 1x LoD 2.25×10^2 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Low positive 1x LoD 2.25×10^2 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Low positive 1x LoD 2.25×10^2 $TCID_{50}$/mL	3	35/35	100	(90.1-100)
Low positive 1x LoD 2.25×10^2 $TCID_{50}$/mL	All	107/107	100	(96.5-100)
Negative	1	36/36	100	(90.4-100)
Negative	2	36/36	100	(90.4-100)
Negative	3	36/36	100	(90.4-100)
Negative	All	108/108	100	(96.6-100)

Table 60: Percent agreement for influenza A

Influenza A concentration	Site	Agreement with expected results Agreed / N	Agreement with expected results %	Agreement with expected results 95% CI
Moderate positive 3x LoD 1.5×10^2 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.5×10^2 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.5×10^2 $TCID_{50}$/mL	3	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.5×10^2 $TCID_{50}$/mL	All	108/108	100	(96.6-100)
Low positive 1x LoD 5×10^1 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Low positive 1x LoD 5×10^1 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Low positive 1x LoD 5×10^1 $TCID_{50}$/mL	3	35/35	100	(90.1-100)

Influenza A concentration	Site	Agreement with expected results Agreed / N	Agreement with expected results %	Agreement with expected results 95% CI
Low positive 1x LoD 5 x 10^1 $TCID_{50}$/mL	All	107/107	100	(96.5-100)
Negative	1	36/36	100	(90.4-100)
Negative	2	36/36	100	(90.4-100)
Negative	3	36/36	100	(90.4-100)
Negative	All	108/108	100	(96.6-100)

Table 61: Percent agreement for influenza A H3

Influenza A H3 concentration	Site	Agreement with expected results Agreed / N	Agreement with expected results %	Agreement with expected results 95% CI
Moderate positive 3x LoD 1.5 x 10^2 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.5 x 10^2 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.5 x 10^2 $TCID_{50}$/mL	3	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.5 x 10^2 $TCID_{50}$/mL	All	108/108	100	(96.6-100)
Low positive 1x LoD 5 x 10^1 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Low positive 1x LoD 5 x 10^1 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Low positive 1x LoD 5 x 10^1 $TCID_{50}$/mL	3	35/35	100	(90.1-100)
Low positive 1x LoD 5 x 10^1 $TCID_{50}$/mL	All	107/107	100	(96.5-100)
Negative	1	36/36	100	(90.4-100)
Negative	2	36/36	100	(90.4-100)
Negative	3	36/36	100	(90.4-100)
Negative	All	108/108	100	(96.6-100)

Table 62: Percent agreement for parainfluenza virus

Parainfluenza virus 1 concentration	Site	Agreement with expected results Agreed / N	Agreement with expected results %	Agreement with expected results 95% CI
Moderate positive 3x LoD 1.2 x 10^0 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.2 x 10^0 $TCID_{50}$/mL	2	36/36	100	(90.4-100)

Parainfluenza virus 1 concentration	Site	Agreement with expected results Agreed / N	Agreement with expected results %	Agreement with expected results 95% CI
Moderate positive 3x LoD 1.2 x 10^0 $TCID_{50}$/mL	3	36/36	100	(90.4-100)
Moderate positive 3x LoD 1.2 x 10^0 $TCID_{50}$/mL	All	108/108	100	(96.6-100)
Low positive 1x LoD 4 x 10^{-1} $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Low positive 1x LoD 4 x 10^{-1} $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Low positive 1x LoD 4 x 10^{-1} $TCID_{50}$/mL	3	35/35	100	(90.1-100)
Low positive 1x LoD 4 x 10^{-1} $TCID_{50}$/mL	All	107/107	100	(96.5-100)
Negative	1	36/36	100	(90.4-100)
Negative	2	36/36	100	(90.4-100)
Negative	3	36/36	100	(90.4-100)
Negative	All	108/108	100	(96.6-100)

Table 63: Percent agreement for respiratory syncytial virus

Respiratory syncytial virus A Concentration	Site	Agreement with expected results Agreed / N	Agreement with expected results %	Agreement with expected results 95% CI
Moderate positive 3x LoD 4.5 x 10^0 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Moderate positive 3x LoD 4.5 x 10^0 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Moderate positive 3x LoD 4.5 x 10^0 $TCID_{50}$/mL	3	36/36	100	(90.4-100)
Moderate positive 3x LoD 4.5 x 10^0 $TCID_{50}$/mL	All	108/108	100	(96.6-100)
Low positive 1x LoD 1.5 x 10^0 $TCID_{50}$/mL	1	36/36	100	(90.4-100)
Low positive 1x LoD 1.5 x 10^0 $TCID_{50}$/mL	2	36/36	100	(90.4-100)
Low positive 1x LoD 1.5 x 10^0 $TCID_{50}$/mL	3	35/35	100	(90.1-100)
Low positive 1x LoD 1.5 x 10^0 $TCID_{50}$/mL	All	107/107	100	(96.5-100)
Negative	1	36/36	100	(90.4-100)
Negative	2	36/36	100	(90.4-100)
Negative	3	36/36	100	(90.4-100)
Negative	All	108/108	100	(96.6-100)

Table 64: Negative percent agreement with organisms not included in the reproducibility panel

Target	Site	Agreement with expected negative results Agreed / N	Agreement with expected negative results %	Agreement with expected negative results 95% CI
Human rhinovirus/enterovirus	1	108/108	100	(96.6-100)
Human rhinovirus/enterovirus	2	108/108	100	(96.6-100)
Human rhinovirus/enterovirus	3	104/107	97.2	(92.1-99.0)
Human rhinovirus/enterovirus	All	320/323	99.1	(97.3-99.7)
Influenza A H1	1	108/108	100	(96.6-100)
Influenza A H1	2	108/108	100	(96.6-100)
Influenza A H1	3	107/107	100	(96.5-100)
Influenza A H1	All	323/323	100	(98.8-100)
Influenza A H1-2009	1	108/108	100	(96.6-100)
Influenza A H1-2009	2	108/108	100	(96.6-100)
Influenza A H1-2009	3	107/107	100	(96.5-100)
Influenza A H1-2009	All	323/323	100	(98.8-100)
Influenza B	1	108/108	100	(96.6-100)
Influenza B	2	108/108	100	(96.6-100)
Influenza B	3	107/107	100	(96.5-100)
Influenza B	All	323/323	100	(98.8-100)
Parainfluenza virus 2	1	108/108	100	(96.6-100)
Parainfluenza virus 2	2	108/108	100	(96.6-100)
Parainfluenza virus 2	3	107/107	100	(96.5-100)
Parainfluenza virus 2	All	323/323	100	(98.8-100)
Parainfluenza virus 3	1	108/108	100	(96.6-100)
Parainfluenza virus 3	2	108/108	100	(96.6-100)
Parainfluenza virus 3	3	106/107	99.1	(94.9-99.8)
Parainfluenza virus 3	All	322/323	99.7	(98.3-99.9)
Parainfluenza virus 4	1	108/108	100	(96.6-100)
Parainfluenza virus 4	2	108/108	100	(96.6-100)
Parainfluenza virus 4	3	107/107	100	(96.5-100)
Parainfluenza virus 4	All	323/323	100	(98.8-100)
Respiratory syncytial virus B	1	108/108	100	(96.6-100)
Respiratory syncytial virus B	2	108/108	100	(96.6-100)
Respiratory syncytial virus B	3	107/107	100	(96.5-100)
Respiratory syncytial virus B	All	323/323	100	(98.8-100)
Chlamydia pneumoniae	1	108/108	100	(96.6-100)
Chlamydia pneumoniae	2	108/108	100	(96.6-100)
Chlamydia pneumoniae	3	107/107	100	(96.5-100)
Chlamydia pneumoniae	All	323/323	100	(98.8-100)
Mycoplasma pneumoniae	1	108/108	100	(96.6-100)
Mycoplasma pneumoniae	2	107/108	99.1	(94.9-99.8)
Mycoplasma pneumoniae	3	106/107	99.1	(94.9-99.8)

Target	Site	Agreement with expected negative results Agreed / N	Agreement with expected negative results %	Agreement with expected negative results 95% CI
Mycoplasma pneumoniae	All	321/323	99.4	(97.8-99.8)

Samples with co-detected organisms

Co-detection of SARS-CoV-2 with other organisms

Detection of SARS-CoV-2 in the presence of another clinically relevant organism was evaluated using a natural clinical matrix (pooled, negative nasopharyngeal swab samples) spiked with SARS-CoV-2 and a second organism co-amplified in the same PCR reaction. In this study, SARS-CoV-2 was tested at a low concentration (3x LoD) in combination with the second organism at a high concentration (1×10^3 - 1×10^6 $TCID_{50}$/mL). SARS-CoV-2 was also tested at a high concentration (2.5×10^6 copies/mL) in combination with the second organism at low concentration (3x LoD). **Table 65** contains the results of co-detection testing which demonstrated that there is no competitive inhibition when SARS-CoV-2 is co-amplified at low concentrations in the presence of the indicated organisms at high concentrations or when SARS-CoV-2 at high concentration is co-amplified with the indicated organism at low concentration.

Table 65: Detection of co-detections

Organism 1	High titer	Organism 2	Multiple of LoD	% positive of organism 2
SARS-CoV-2	2.5×10^6 copies/mL	Influenza A H1-2009	3x	100%
SARS-CoV-2	2.5×10^6 copies/mL	Adenovirus	3x	100%
SARS-CoV-2	2.5×10^6 copies /mL	Influenza B	3x	100%
Influenza A H1-2009	1×10^3 $TCID_{50}$/mL	SARS-CoV-2	3x	100%
Adenovirus	1×10^6 $TCID_{50}$/mL	SARS-CoV-2	3x	100%
Influenza B	1×10^3 $TCID_{50}$/mL	SARS-CoV-2	3x	100%

Samples with co-detected organisms on the cobas eplex RP2 panel

Detection of more than one clinically relevant viral organism in a sample was evaluated with the **cobas eplex** RP panel using a natural clinical matrix (pooled, negative nasopharyngeal swab samples) spiked with two RP panel organisms: one organism at a low concentration (1-3x LoD) and the second organism at a high concentration (1×10^5 $TCID_{50}$/mL). **Table 66** contains the results of co-detection testing which demonstrated the ability of the **cobas eplex** RP panel to detect 2 organisms in a sample at both high and low concentrations as indicated in the table.

Table 66: Detection of co-detections

Organism 1	High titer	Organism 2	Low titer	Multiple of LoD
Influenza A H3	1×10^5 $TCID_{50}$/mL	Adenovirus B	2×10^0 $TCID_{50}$/mL	1x
Adenovirus	1×10^5 $TCID_{50}$/mL	Influenza A H3	5×10^1 $TCID_{50}$/mL	1x
Influenza A H3	1×10^5 $TCID_{50}$/mL	RSV A	1.5×10^0 $TCID_{50}$/mL	1x
RSV A	1×10^5 $TCID_{50}$/mL	Influenza A H3	5×10^1 $TCID_{50}$/mL	1x
Influenza A H1-2009	1×10^5 $TCID_{50}$/mL	RSV B	6×10^{-1} $TCID_{50}$/mL	3x
RSV B	1×10^5 $TCID_{50}$/mL	Influenza A H1-2009	$1x\ 10^{-1}$ $TCID_{50}$/mL	1x

Influenza A H1-2009	1 x 10^5 $TCID_{50}$/mL	Rhinovirus	1.5 x 10^0 $TCID_{50}$/mL	1x
Rhinovirus	1 x 10^5 $TCID_{50}$/mL	Influenza A H1-2009	3 x 10^{-1} $TCID_{50}$/mL	3x
Influenza A H1-2009	1 x 10^5 $TCID_{50}$/mL	Parainfluenza virus 3	5 x 10^0 $TCID_{50}$/mL	1x
Parainfluenza virus 3	1 x 10^5 $TCID_{50}$/mL	Influenza A H1-2009	1 x 10^{-1} $TCID_{50}$/mL	1x
Rhinovirus	1 x 10^5 $TCID_{50}$/mL	RSV A	1.5 x 10^0 $TCID_{50}$/mL	1x
RSV A	1 x 10^5 $TCID_{50}$/mL	Rhinovirus	1.5 x 10^0 $TCID_{50}$/mL	1x
Coronavirus (229E, HKU1, NL63, OC43)	1 x 10^5 $TCID_{50}$/mL	RSV A	1.5 x 10^0 $TCID_{50}$/mL	1x
RSV A	1 x 10^5 $TCID_{50}$/mL	Coronavirus (229E, HKU1, NL63, OC43)	7.5 x 10^0 $TCID_{50}$/mL	1x
Human metapneumovirus	1 x 10^5 $TCID_{50}$/mL	Adenovirus	2 x 10^0 $TCID_{50}$/mL	1x
Adenovirus	1 x 10^5 $TCID_{50}$/mL	Human metapneumovirus	2.25 x 10^2 $TCID_{50}$/mL	1x
Adenovirus	1 x 10^5 $TCID_{50}$/mL	RSV A	1.5 x 10^0 $TCID_{50}$/mL	1x
RSV A	1 x 10^5 $TCID_{50}$/mL	Adenovirus	2 x 10^0 $TCID_{50}$/mL	1x

Sample matrix equivalency

All analytical studies that utilized viral and bacterial cultures close to LoD were performed by spiking the viral and bacterial cultures into a pool of natural negative NPS in VTM as sample matrix. For analytical studies that used viral and bacterial cultures at a concentration which was at least 10x LoD or higher, the viral and bacterial cultures were spiked into MicroTest™ M5® transport media from Remel instead of negative pooled NPS for ease of use. A sample matrix equivalency study was performed to demonstrate equivalency between natural clinical matrix (pooled, negative nasopharyngeal swab in VTM samples) and viral transport media when spiked with targets at a concentration of approximately 10x LoD. Quantified, representative viral and bacterial strains were diluted in a natural clinical matrix (pooled, negative nasopharyngeal swab in VTM samples) and in viral transport media. All samples were tested in duplicate. There was no difference observed in detection of targets in natural clinical matrix vs. viral transport media.

Interfering substances

Substances commonly found in respiratory samples, substances that could be introduced during specimen collection, or medications commonly used to treat congestion, allergies, or asthma symptoms that could potentially interfere with the **cobas eplex** RP panel were individually evaluated. To simulate clinical samples, quantified representative viral and bacterial strains were diluted to 1x LoD in a natural clinical matrix (pooled, negative nasopharyngeal swab specimens) and tested in triplicate for negative and positive interference. Natural clinical matrix (pooled, negative nasopharyngeal swab samples) with no organisms added was used as a control. All substances and organisms tested for interference were shown to be compatible with the **cobas eplex** RP panel. No potentially interfering substances were found to inhibit the **cobas eplex** RP panel at the concentrations tested in **Table 67**.

Table 67: List of substances for testing

Potentially interfering substance	Active ingredient	Testing concentration
Control sample matrix[a]	Becton Dickinson UVT	N/A
Transport medium[a]	Copan eSwab (Liquid Amies media)	N/A
Viral Transport Medium[a]	MicroTest M4	N/A
Viral Transport Medium[a]	MicroTest M4-RT	N/A
Viral Transport Medium[a]	MicroTest M5	N/A

Potentially interfering substance	Active ingredient	Testing concentration
Viral Transport Medium[a]	MicroTest M6	N/A
Flocked swabs	Copan Mmnitip in UVT	N/A
Flocked swabs	Copan regular Tip in UVT	N/A
Blood (human)	Blood	2% v/v
Blood (human)	Human gDNA	50 ng/rxn
Throat lozenges, oral anesthetic and analgesic	Benzocaine, menthol	26% w/v
Mucin	Purified mucin protein	1% w/v
Nasal sprays or drops	Phenylephrine HCl (Neo-Synephrine®)	1.5% v/v
Nasal sprays or drops	Oxymetazoline HCl (Afrin®)	1% v/v
Nasal sprays or drops	Sodium chloride	0.8% w/v
Antibacterial, systemic	Tobramycin[b]	1% w/v
Antibiotic, nasal ointment	Mupirocin	2% w/v
Nasal corticosteroids	Beclomethasone	1.5% w/v
Nasal corticosteroids	Dexamethasone	1.5% w/v
Nasal corticosteroids	Flunisolide	1.5% w/v
Nasal corticosteroids	Budesonide (Rhinocort®)	0.9% v/v
Nasal corticosteroids	Triamcinolone (Nasacort®)	1.5% v/v
Nasal corticosteroids	Fluticasone (Flonase®)	1.5% v/v
ZICAM® allergy relief nasal gel	Luffa opperculata	1% v/v
ZICAM® allergy relief nasal gel	Sulfur	1% v/v
ZICAM® allergy relief nasal gel	Galphimia glauca	1% v/v
ZICAM® allergy relief nasal gel	Histaminum hydrochloricum	1% v/v
Anti-viral drugs	Zanamivir	550 ng/mL
Anti-viral drugs	Oseltamivir	142 ng/mL
Virus	Cytomegalovirus	1 x 10^5 $TCID_{50}$/mL
Bacteria	*Streptococcus pneumoniae*	1 x 10^6 CFU/mL
Bacteria	*Bordetella parapertussis*	1 x 10^6 CFU/mL
Bacteria	*Haemophilus influenza*	1 x 10^6 CFU/mL
Bacteria	*Staphylococcus aureus*	1 x 10^6 CFU/mL
Bacteria	*Neisseria meningitides*	1 x 10^6 CFU/mL
Bacteria	*Corynebacterium diptheriae*	1 x 10^6 CFU/mL

[a] Testing of media was done by adding a negative NPS collected in the specified media and diluting in the natural clinical matrix.
[b] At concentrations greater than 1% weight/volume in the sample, tobramycin was found to inhibit assay performance.

Carryover and cross-contamination

The carryover/cross-contamination rate of the **cobas eplex** RP panel and **cobas eplex** instrument was tested in a checkerboard approach by running high positive and negative samples interspersed in all bays of a four-tower **cobas eplex** system (24 bays total) over 5 separate runs on 5 separate days. Quantified parainfluenza virus 3 was prepared in viral transport media at a high concentration (1 x 10^5 $TCID_{50}$/mL, 20,000x LoD) to simulate a clinically relevant high positive and was tested as a representative target organism. Transport media was used to represent negative samples. On each round of testing, 24 **cobas eplex** RP panel cartridges were evaluated. 100% of parainfluenza 3-positive samples generated a result of Detected and 100% of parainfluenza 3-negative samples generated a parainfluenza 3 result of No

Target Detected, indicating no carryover or cross-contamination was observed between bays or within bays with the **cobas eplex** RP panel when testing consecutively or in adjacent bays.

TROUBLESHOOTING

Table 68: Troubleshooting table

For a complete list of all **cobas eplex** error messages and a description of the messages, please refer to the **cobas eplex** User Assistance Manual.

Error	Error messages	Description	Re-test recommendations
Test did not start	Cartridge failure The cartridge initialization test failed Cartridge not present Bay heater failure Unknown error Bay main / fluid motor failure Bay over pressured Bay temperature out of range The system was unable to read the cartridge Cartridge inserted doesn't match the serial number of the cartridge scanned The system is not ready to accept the cartridge The system failed to prepare the cartridge for processing	An error that occurs during pre-run checks (cartridge initialization) of the cartridge upon insertion into the bay. Cartridge initialization occurs when the cartridge is first inserted into the bay and takes approximately 90 seconds. Upon completion of cartridge initialization, the cartridge cannot be restarted, but prior to this point, the cartridge can be restarted. To verify cartridge initialization has completed, examine the cartridge label upon removal from the bay. If the cartridge label has been pierced, the test has already started, and cartridge cannot be reused. If the label has not been pierced, follow the recommendation as stated.	1. Remove cartridge from bay. a. Reset bay to clear the error b. Restart cartridge in any available bay 2. If the cartridge is not able to be run on the second try and again generates an error during cartridge initialization, this indicates an issue with the cartridge. This cartridge should be discarded following laboratory procedures and the sample should be repeated using a new cartridge. Bay(s) should be reset to clear the errors. Please contact Technical Support to alert them of the issue. If the bay remains in an error state (flashing red) after the cartridge has been removed, then the bay must be reset through the Bay Configuration menu before it can be used to run cartridges.
Test did not finish	Bay heater failure Bay main / fluid motor failure Bay voltage failure Bay sub-system communication timeout Cartridge failure Bay over pressured Bay auto-calibration failure Bay temperature out of range The system was unable to eject the cartridge from the bay	This type of error occurs during the run, after pre-run checks (cartridge initialization) have finished, and prevents the cartridge from being processed to completion.	Reagents have been consumed and the cartridge cannot be reused. Contact Technical Support and proceed with repeat testing of the sample using a new cartridge. If the bay remains in an error state (flashing red) after the cartridge has been removed, then the bay must be reset through the Bay Configuration menu before it can be used to run cartridges.
Invalid		This is an error that results in no valid results being generated. A test report will be generated, but all targets and the internal control will be invalid.	Reagents have been consumed and the cartridge cannot be reused. Contact Technical Support and proceed with repeat testing of the sample using a new cartridge.

Technical Support

GenMark Technical support is available 24 hours a day, 7 days a week to provide the highest level of customer support and satisfaction.

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In the US, please contact:
Technical support: 833.943.6627 (833.9GENMAR) or cad.technical_support_us@roche.com.
Customer service: 1-800-428-5076

GLOSSARY OF SYMBOLS

Symbol	Description	Symbol	Description
	Batch code		Catalog number
	Caution		Biological risks
	Contains sufficient for <n> tests		Upper limit of temperature
	Consult instructions for use		Lower limit of temperature
	Manufacturer		Temperature range
	Cartridge lot		Irritant, dermal sensitizer, acute toxicity (harmful), narcotic effects, respiratory tract irritation
	Use by date YYYY-MM-DD		Oxidizers
	Serial number		For Use Under the Emergency Use Authorization Only

REFERENCES

1. Upper Respiratory Infection (URI or Common Cold). Johns Hopkins Medicine. Retrieved from http://www.hopkinsmedicine.org/healthlibrary/conditions/pediatrics/upper_respiratory_infection_uri_or_common_cold_90,P02966/ (Date accessed: 3/22/2016).
2. Seasonal influenza, More Information. Centers for Disease Control and Prevention. Retrieved from http://www.cdc.gov/flu/about/qa/disease.htm (Date accessed: 6/10/2016).

3. Influenza (Seasonal). (2014). World Health Organization. Retrieved from http://www.who.int/mediacentre/factsheets/fs211/en/ (Date accessed: 3/22/2016).
4. Coronavirus Disease 2019 (COVID-19) Frequently Asked Questions. Centers for Disease Control and Prevention Retrieved from https://www.cdc.gov/coronavirus/2019-ncov/faq.html#Basics (Date accessed 6/18/2020)
5. COVID-19 Dashboard by the Center for Systems Science and Engineering (CSSE) at Johns Hopkins University (JHU). Retrieved from: https://gisanddata.maps.arcgis.com/apps/opsdashboard/index.html#/bda7594740fd40299423467b48e9ecf6 (Date accessed: 7/27/2020).
6. Mossad, S., Upper Respiratory Tract Infections. Cleveland Clinic Center for Continuing Education. Retrieved from http://www.clevelandclinicmeded.com/medicalpubs/diseasemanagement/infectious-disease/upper-respiratory-tract-infection/ (Date Published 8/2013).
7. Adenovirus Infections. University of Rochester Medical Center. Retrieved from https://www.urmc.rochester.edu/Encyclopedia/Content.aspx?ContentTypeID=90&ContentID=P02508 (Date accessed: 3/22/2016).
8. Adenoviruses Clinical Overview. Centers for Disease Control and Prevention. Retrieved from http://www.cdc.gov/adenovirus/hcp/clinical-overview.html (Date accessed: 3/22/2016).
9. Gaunt, E.R. et al. (2010). Epidemiology and Clinical Presentations of the Four Human Coronaviruses 229E, HKU1, NL63, and OC43 Detected over 3 Years Using a Novel Multiplex Real-Time PCR Method. J. Clin. Microbiol. 48(8) 2940-2947.
10. Human Metapneumovirus Clinical Features. Centers for Disease Control and Prevention. http://www.cdc.gov/surveillance/nrevss/hmpv/clinical.html (Date accessed: 3/22/2016).
11. Auwaerter, P., Metapneumovirus. Johns Hopkins Antibiotics (ABX) Guide. Retrieved from http://www.hopkinsguides.com/hopkins/view/Johns_Hopkins_ABX_Guide/540614/all/Metapneumovirus (Updated March 2013).
12. Anzueto, A., et al. (2003) Diagnosis and Treatment of Rhinovirus Respiratory Infections. Chest. 123(5) 1664-1672.
13. Auwaerter, P., Rhinovirus. Johns Hopkins Antibiotics (ABX) Guide. Retrieved from http://www.hopkinsguides.com/hopkins/view/Johns_Hopkins_ABX_Guide/540476/all/Rhinovirus?q=rhinvorirus&ti=0#0 (Updated February 2016).
14. Auwaerter, P., Enterovirus. Johns Hopkins Antibiotics (ABX) Guide. Retrieved from http://www.hopkinsguides.com/hopkins/view/Johns_Hopkins_ABX_Guide/540204/all/Enterovirus?q=enterovirus&ti=0#0 (Updated November 2014).
15. Henrickson, K.J. (2003). Parainfluenza viruses. Clin. Microbiol. Rev. 16(2):242-264.
16. Schomacker, H. et al., (2012) Pathogenesis of acute respiratory illness caused by human parainfluenza viruses. Curr Opin Virol. 2(3) 294-299.
17. Mahony, J.B. Detection of Respiratory Viruses by Molecular Methods. Clin. Microbiol. Rev. (2008). 21(4) 716-747.
18. Resch, B., et al., (2011). Epidemiology of Respiratory Syncytial Virus Infection in Preterm Infants. Open Microbiol J. 5(Suppl 2-M3) 135-143.
19. Atypical Pneumonia. *Chlamydia pneumoniae* Infection. Centers for Disease Control and Prevention. Retrieved from https://www.cdc.gov/pneumonia/atypical/cpneumoniae/index.html (Updated February 7, 2014).
20. Auwaerter, P., Mycoplasma Pneumoniae. Johns Hopkins Antibiotics (ABX) Guide. Retrieved from http://www.hopkinsguides.com/hopkins/view/Johns_Hopkins_ABX_Guide/540373/all/Mycoplasma%20pneumoniae (Date accessed: 3/28/2016).
21. Epidemiological update: *Mycoplasma pneumoniae* infections- recent increases reported in EU countries. (2012). European Centre for Disease Prevention and Control. Retreived from http://ecdc.europa.eu/en/press/news/_layouts/forms/News_DispForm.aspx?ID=340&List=8db7286c-fe2d-476c-9133-18ff4cb1b568 (Date accessed: 3/28/2016).
22. Spacek, L. and Auwaerter, P., Adenovirus. Johns Hopkins Antibiotics (ABX) Guide. Retrieved from

http://www.hopkinsguides.com/hopkins/view/Johns_Hopkins_ABX_Guide/540009/all/Adenovirus?q=adenovirus&ti=0#0 (Updated December 2014).
23. Technical Guidance, Naming the coronavirus disease (COVID-19) and the virus that causes it. World Health Organization. Retrieved from: https://www.who.int/emergencies/diseases/novel-coronavirus-2019/technical-guidance/naming-the-coronavirus-disease-(covid-2019)-and-the-virus-that-causes-it#:~:text=ICTV%20announced%20%E2%80%9Csevere%20acute,two%20viruses%20are%20different (Date accessed: 6/18/2020)
24. Friedman, J., (2012). Vaccination Program Appears to Reduce Respiratory Infections Among Recruits. Story number NNS120131-22. Retrieved from http://www.navy.mil/submit/display.asp?story_id=65070.
25. Coronavirus, About Coronavirus. Centers for Disease Control and Prevention. Retrieved from http://www.cdc.gov/coronavirus/about/index.html (Date accessed: 3/22/2016).
26. Coronavirus Infections. European Centre for Disease Prevention and Control. Retrieved from http://ecdc.europa.eu/en/healthtopics/coronavirus-infections/Pages/index.aspx (Date accessed: 3/24/2106).
27. Human Metapneumovirus Clinical Features, National Respiratory and Enteric Virus Surveillance System (NREVSS). Centers for Disease Control and Prevention. Retrieved from http://www.cdc.gov/surveillance/nrevss/hmpv/clinical.html (Date accessed: 3/22/2016).
28. Tapparel, C., et al., (2009). New Respiratory Enterovirus and Recombinant Rhinoviruses among Circulating Picornaviruses. Emerg Infect Dis Retrieved from http://wwwnc.cdc.gov/eid/article/15/5/08-1286 (Date accessed: 3/24/2016).
29. Jacobs, S., et al., (2013). Human Rhinoviruses. Clin Microbiol Rev 26(1) 135-162.
30. Enterovirus D68 detections in the USA, Canada and Europe, Second update 25 November 2014. European Centre for Disease Prevention and Control. Stockholm: ECDC; 2014. Retrieved from http://ecdc.europa.eu/en/publications/Publications/Enterovirus-68-detected-in-the-USA-Canada-Europe-second-update-25-November-2014.pdf (Date accessed: 3/22/2016).
31. Influenza, Types of Influenza Viruses. Centers for Disease Control and Prevention. Retrieved from http://www.cdc.gov/flu/about/viruses/types.htm (Date accessed: 3/22/2016).
32. Seasonal Influenza Factsheet for the General Public. European Centre for Disease Prevention and Control. Retrieved from http://ecdc.europa.eu/en/healthtopics/seasonal_influenza/basic_facts/Pages/factsheet_general_public.aspx (Date accessed: 3/24/2016).
33. Seasonal Influenza Factsheet for Health Professionals. European Centre for Disease Prevention and Control. Retrieved from http://ecdc.europa.eu/en/healthtopics/seasonal_influenza/basic_facts/Pages/factsheet_professionals_seasonal_influenza.aspx (Date accessed: 3/24/2016).
34. Auwaerter, P., and Bartlett, J., Influenza. Johns Hopkins Antibiotics (ABX) Guide. Retrieved from http://www.hopkinsguides.com/hopkins/view/Johns_Hopkins_ABX_Guide/540285/all/Influenza?q=influenza&ti=0#0 (Date accessed: 3/24/2016).
35. Human Parainfluenza Viruses, Symptoms and Illnesses. Centers for Disease Control and Prevention. Retrieved from http://www.cdc.gov/parainfluenza/about/symptoms.html (Date accessed 3/28/2016).
36. Bartlett, J., Parainfluenza Virus. Johns Hopkins Antibiotics (ABX) Guide. Retrieved from http://www.hopkinsguides.com/hopkins/view/Johns_Hopkins_ABX_Guide/540415/all/Parainfluenza_virus?q=parainfluenza&ti=0#0 (Date accessed: 3/24/2016).
37. Respiratory Syncytial Virus Infection, Infection and Incidence. Centers for Disease Control and Prevention. Retrieved from http://www.cdc.gov/rsv/about/infection.html (Date accessed: 3/24/2016).
38. Walsh, E., et al., (1997). Severity of Respiratory Syncytial Virus Infection Is Related to Virus Strain. J Infect Dis 175(4) 814-820.
39. Bartlett, J., *Chlamydophila pneumoniae*. Johns Hopkins Antibiotics (ABX) Guide. Retrieved from http://www.hopkinsguides.com/hopkins/view/Johns_Hopkins_ABX_Guide/540117/all/Chlamydophila%20pneumoniae (Updated October 4, 2015).

40. Pneumonia. Centers for Disease Control and Prevention. Retrieved from http://www.cdc.gov/pneumonia/index.html (Updated February 25, 2015).
41. *Mycoplasma pneumoniae* Infection. Centers for Disease Control and Prevention. Retrieved from http://www.cdc.gov/pneumonia/atypical/mycoplasma/index.html (Date accessed: 3/28/2016).

TRADEMARKS

PATENT INFORMATION

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