Sansure Biotech

Six Respiratory Pathogens Nucleic Acid Diagnostic Kit (PCR-Fluorescence Probing)

Product identification

Product name: Six Respiratory Pathogens Nucleic Acid Diagnostic Kit (PCR-Fluorescence Probing)
Reference number: S3066E
Package specification: 24 tests/kit, Pre-packaged 12 tests/kit

Intended use

The Six Respiratory Pathogens Nucleic Acid Diagnostic Kit (PCR-Fluorescence Probing) is intended for the simultaneous qualitative detection of influenza A virus, influenza B virus, respiratory syncytial virus, adenovirus, human rhinovirus and mycoplasma pneumonia in human nasopharyngeal swabs(NPS), oropharyngeal swabs, alveolar lavage fluid. The detection results can be used for the auxiliary diagnosis of the patients with respiratory tract pathogen infection and providing a molecular diagnostic basis for the respiratory tract pathogen infection. This kit can distinguish the above pathogens, but for the same pathogen, can not distinguish the specific subtypes.

The detection results of this diagnostic kit can only be used for clinical reference, and not as the only criterion of clinical diagnosis.

For in vitro diagnostic use only. For professional use only.

Test principle summary and explanation

Summary

Respiratory infections are classified into the upper respiratory tract infections and lower respiratory tract infections, which means that pathogens infect the nose, throat, trachea, bronchi or lungs of the human body, characterized by fever, sore throat, cough, headache and other symptoms. The respiratory tract pathogen has the characteristics of strong infectivity, rapid spread, short incubation period and acute onset. which seriously harm human health.

Influenza Virus is a RNA virus of the Orthomyxoviridae family which lead to human and animal influenza. It causes acute upper respiratory tract infection, spreads rapidly through the air, has periodic pandemics around the world. Human influenza virus are in classified into three types, namely A, B and C. Influenza A is the most harmful, influenza B and influenza C have weak pathogenicity and do not easily mutate. Influenza A Virus (Inf. A) has many subtypes. So far, 18 subtypes of HA and 11 subtypes of NA are known. Influenza B Virus (Inf. B) can be divided into two phylogenetic lineages: Yamagata family and Victoria family.

Respiratory Syncytial Virus (RSV) is a member of the RNA viruses of the Paramyxoviridae family. Clinical research shows that the respiratory syncytial virus is the most common pathogen that causes virus pneumonia, which is more common in infants under the age of three, mainly with the symptoms of hyperpyrexia, nasitis, pharyngitis and laryngitis, and even bronchiolitis and pneumonia. Very few sick infants will have the complications of tympanitis, pleuritis and myocarditis, etc. When older children and adults get infected, the main symptom is upper respiratory tract infection.

Adenoviruses(Adv) belongs to Adenoviridae, with seven species (A to G) based on different immunology, biology and biochemical characterization, approximately 52 serotypes. Different serotype have different organ affinity and will cause corresponding clinical manifestation, which infection is mainly characterized by respiratory symptoms, including high fever, cough, sore throat, and a few can cause pneumonia.

Human Rhinovirus (HRV) belongs to the genus Picornaviridae, and is a group of single positive-stranded small RNA viruses. It is an important pathogenic factor in respiratory tract infection viruses. In infants and children, in addition to upper respiratory tract infection, it can also cause bronchitis and bronchopneumonia, acute asthma and other diseases.

Mycoplasma Pneumoniae **(MP)** is a common pathogen of community-acquired pneumonia. After infection with MP, it can cause mycoplasma pneumonia. The pathological changes of mycoplasma pneumonia are mainly interstitial pneumonia, sometimes complicated by bronchial pneumonia. It is mainly transmitted by droplets, with an incubation period of 2 to 3 weeks, and the incidence is highest in adolescents. The clinical symptoms are mild, and most patients only have general respiratory symptoms such as headache, sore throat, fever, and cough; a few will have persistent high fever, severe cough, rapid disease progression, respiratory failure, and multiple organ dysfunction in a short period of time. Critical illness manifested.

At present, the main methods of laboratory detection of the above-mentioned respiratory pathogens include: virus isolation and identification, RT-PCR, colloidal gold, etc.

Test principle

This kit uses specific primers and specific fluorescent probes designed for the conserved region of the nucleic acid of the pathogen to be detected, together with components such as PCR solution, and applies multiple real-time fluorescent quantitative PCR detection technology on the fluorescence quantitative PCR instrument. The changes of fluorescence signal enable rapid detection of respiratory pathogen nucleic acids in samples.

The PCR detection system contains a positive internal control (internal standard), which monitors the extraction process and PCR amplification process of the sample to be tested by detecting whether the human housekeeping gene encoding glyceraldehyde-3-phosphate dehydrogenase (GAPDH) in the sample is normally amplified. Avoid false negative results.

Materials provided

This kit is an amplification reaction reagent and contains the following components:

No.	Reagent Name	Specification & Qty.		Main Ingredients
		24T	Pre-packaged 12T	
1	Six RP-PCR Mix A	1044 μL/tube × 1 tube	43.5 μL/tube × 12 tubes	Primers, probes, dNTPs, PCR buffer, DEPC water, $MgCl_2$
2	Six RP-PCR Mix B	1044 μL/tube × 1 tube	43.5 μL/tube × 12 tubes	Primers, probes, dNTPs, PCR buffer, DEPC water, $MgCl_2$
3	Six RP-Enzyme Mix	72 μL/tube × 1 tube	1.5 μL/tube × 24 tubes	DNA polymerase, mMLV enzyme
4	Six RP-Positive Control	1000 μL/tube × 1 tube	1000 μL/tube × 1 tube	cloned plasmids (7.0×10^3copies/mL) and Lentivirus particles (7.0×10^3TU/mL) containing each pathogen gene sequence diluted with normal saline
5	Six RP-Negative Control	1000 μL/tube × 1 tube	1000 μL/tube × 1 tube	Normal saline
6	Six RP-Internal Control	50 μL/tube × 1 tube	50 μL/tube × 1 tube	Lentivirus particles (2.0×10^3TU/mL) containing GAPDH gene sequence diluted with normal saline

Materials required but not provided

1. Not included in the kit reagent: Multi-type Sample DNA/RNA Extraction-Purification Kit (Magnetic beads method) (Reference number: S1006E Series) and Nucleic Acid Extraction-Purification Kit (Reference number: S10016E / S50016E Series) manufactured by Sansure Biotech Inc
2. Materials required but not provided: 1.5 mL DNase-free and RNase-free centrifuge tubes, 0.2 mL PCR tubes, pipette tips (10 μL, 200 μL and 1000 μL tips with filters are preferred), desktop centrifuge, desktop vortex mixer, magnetic-bead separator, various models of pipettes.

Warnings and precautions

Warnings

1. Do not mix or exchange components from different kits.
2. All biological materials in the kit have been inactivated.

Precautions

1. For in vitro diagnostic use only. Please read the product instructions for use carefully before operation.
2. Please learn and be familiar with the operation procedures and precautions for each instrument before test. Please make sure quality control is performed for each test.
3. Laboratory management shall strictly follow management practices of PCR gene amplification laboratories. Laboratory personnel must receive professional training, test processes must be performed in separated rooms, all consumables should be for single use only after sterilization, special instruments and devices should be used for every process, all lab devices required in different processes and rooms should not be cross-used.
4. All samples for detection should be handled as potentially infectious. Wear laboratory coats, protective disposable gloves and change the gloves often to avoid cross-contamination between specimens. Handling of specimens and waste must meet relevant requirements outlined in local, state and national regulations.
5. Before use, all reagents must be fully thawed at room temperature and mixed thoroughly.
6. When testing specimens that do not contain human endogenous genes, there is no requirement for internal control (ROX channel).
7. After the addition of the sample in the tube the resulting solution is to be considered potentially biohazardous, handle the reagent with appropriate precautions and good laboratory practice.
8. The safe disposal of the reagents supplied must be carried out according to the instruction contained in the specific Safety Data Sheets and in compliance with the national regulations on disposal of potentially hazardous waste.
9. Any serious incident that has occurred in relation to the device shall be reported to the manufacturer and the competent authority of the Member State in which the user and/or the patient is established; If you have any questions about the test or the results, please contact Sansure`s customer service hotline +86-731-88883176-6116 or send an email to info@sansure.com.cn/ support@sansure.com.cn.

IVD storage, operating conditions and stability

1. The shelf life of the kit is 11 months at -25° C to -15° C and protected from light. Please see the date of manufacture and expiry date on the outer package.
2. The reagents remain stable within the expiry date if not used. When the container of the reagents was opened, the freeze/thaw cycles should not exceed three.
3. The reagents remain stable until the expiry date on the outer package when transporting for 7 days in a sealed foam box containing coolant with the temperature lower than 20°C.

Instrumentation

The kit is compatible to Fluorescence Quantitative Analysis System containing FAM, HEX/VIC, CY5 and ROX channels such as:

1. Angilent/AriaDx Real-Time PCR System
2. Applied Biosystems/7500 Real-Time PCR System
3. Bio-Rad/CFX96 Dx and CFX96 Deepwell Dx Systems
4. Hongshi/SLAN®-96P Real-Time PCR System
5. Molarray/Real-Time Quantitative Thermal Cycler (Model: MA-6000)
6. Roche/LightCycler® 480 instrument II
7. Sansure/Portable Molecular Diagnostic System (Model: S-Q37A/S-Q37B)
8. Sansure/Portable Molecular Workstation (Model: S-Q36A)
9. Sansure /Portable Molecule Workstation (Model: S-Q31A/S-Q31B)
10. Thermofisher/QuantStudio™ 5 Real-Time PCR System

Collecting and preparing specimens

1. Applicable specimen type: Nasopharyngeal swabs, oropharyngeal swabs and alveolar lavage fluid.
2. Collection of specimen:

Nasopharyngeal swabs:

a. Use sterile swabs to insert in the nostril for 3 - 6 cm in the direction of the earlobe and the tip of the nose, or on the nasal palates of both nasal passages. Stay for 15 seconds,.then slowly rotate out.
b. Put the swab into a sterile tube containing sampling solution. Plug the mouth of the tube tightly, seal and send it for detection.
c. The top part of the swab should be made from artificial material (e.g. polyester or terylene), the rod of the swab should be made from aluminum or plastics. It is not recommended to use cotton head swab with wooden handle. Calcium alginate swab sampling is not recommended.

Oropharyngeal swabs:

a. The specimen collection tube should be pasted with the barcode first;
b. The oropharyngeal swab should be collected within 3 days after the onset of the disease as far as possible;
c. A sterile flocking swab should be used for sampling, moderately wipe the posterior pharyngeal wall, avoid touching the tongue;
d. Quickly place a sterile swab into the collection tube used for collection of oropharyngeal swabs;
e. Break the sterile swab rod near the top, tighten tube cap and seal with sealing film.

Alveolar lavage fluid:

a. Severe patients or patients with pneumonia who progress rapidly;
b. Clinician extract ≥ 5 mL BALF into a 50 mL aseptic container labeled with specimen bar code and screw cap by aseptic operation;
c. Collect specimen, then tighten tube cap and seal with sealing film.

3. Storage and delivery of specimens: Specimens collected via the above-mentioned method can be stored at 4°C for detection within 48 hours, stored below -20°C for three months, or stored below -70°C for longer than three months. Multiple freeze/thaw cycles should be avoided. Specimens should be transported in a sealed frozen pitcher with ice or in a sealed foam box with ice.

Test procedure

Please process according to the following steps for AriaDx Real-Time PCR System, SLAN®-96P Real-Time PCR System, 7500 Real-Time PCR System, QuantStudio™ 5 Real-Time PCR System, LightCycler® 480 instrument II, Real-Time Quantitative Thermal Cycler (Model: MA-6000), CFX96 Dx and CFX96 Deepwell Dx Systems instrument:

1.1 Preparation of reagent (performed at "reagent preparation room")

1.1.1 Take out all the components out off the kit and equilibrate them at room temperature. Allow the reagents to equilibrate at room temperature, then vortex each of them for later use.

1.1.2 Mix Six RP-Internal Control and Six RP-Negative Control in proportion : Add 10μL Six RP-Internal Control into every 200 μL Six RP-Negative Control, then fully mix them to make a Six RP-Negative Control containing internal control, centrifuge instantaneously for later use.

1.1.3 According to the quantity of test specimens, Six RP-Positive Control and Six RP-Negative Control, pipette appropriate quantity of Six RP-PCR Mix A and Six RP-Enzyme Mix (Six RP-PCR Mix A 43.5 μL/test + Six RP-Enzyme Mix 1.5 μL/test), mix them thoroughly to make a PCR-Mastermix A, and centrifuge instantaneously for later use. Pipette appropriate quantity of Six RP-PCR Mix B and Six RP-Enzyme Mix (Six RP-PCR Mix B 43.5 μL/test + Six RP-Enzyme Mix 1.5 μL/test), mix them thoroughly to make a PCR-Mastermix B, and centrifuge instantaneously for later use.

	1 specimen	10 specimens	24 specimens
Six RP-PCR Mix A or B (μL)	43.5	435	1044
Six RP-Enzyme Mix (μL)	1.5	15	36

Note: The above configuration is just for your reference and to ensure enough volume of the PCR-Mastermix A or B. More volume may be required.

1.2. Processing and loading of specimens (performed at "specimen processing room")

1.2.1 Processing of specimens

Add 200 μL of test specimen, Six RP-Negative Control and Six RP-Positive Control respectively into 1.5 mL centrifuge tubes. Use Multi-type Sample DNA/RNA Extraction-Purification Kit (Magnetic beads method) (Reference Number :S1006E/S10016E Series) manufactured by Sansure Biotech Inc. to extract the nucleic acid as the product instructions for use.

1.2.2 Loading of specimens

1.2.2.1 Add 5 μL of above prepared test specimen, Six RP-Positive Control and Six RP-Negative Control respectively into corresponding 0.2mL PCR reaction tube, then add 45 μL PCR-Mastermix A to every tube, cover the tube lid.

1.2.2.2 Add 5 μL of above prepared test specimen, Six RP-Positive Control and Six RP-Negative Control respectively into corresponding 0.2mL PCR reaction tube, then add 45 μL PCR-Mastermix B to every tube, cover the tube lid.

1.2.2.3 It is suggested to use the spotting method as shown in Table 1 to detect the specimen.

Table 1 Proposed Specimen Plate Layout (96-well-plate)

PCR-Mastermix A		PCR-Mastermix B	
Positive Control	Specimen 5	Positive Control	Specimen 5
Negative Control	Specimen 6	Negative Control	Specimen 6
Specimen 1	Specimen 7	Specimen 1	Specimen 7
Specimen 2	Specimen 8	Specimen 2	Specimen 8
Specimen 3	Specimen 9	Specimen 3	Specimen 9
Specimen 4	...	Specimen 4	...

1.3. PCR Amplification (Refer to user manual of each instrument to adjust the settings.)

1.3.1 Place PCR reaction tubes into the specimen wells of the amplification instrument. Set up the Six RP-Positive Control, Six RP-Negative Control and unknown specimens in the corresponding sequence and input specimen information.

1.3.2 Select PCR test channel:(take ABI7500 as example)

1.3.2.1 PCR-Mastermix A

a) Select FAM channel (Reporter: FAM, Quencher: None) to test influenza A virus.
b) Select HEX or VIC channel (Reporter: HEX/VIC, Quencher: None) to test influenza B virus.
c) Select CY5 channel (Reporter: CY5, Quencher: None) to test respiratory syncytial virus.
d) Select ROX channel (Reporter: ROX, Quencher: None) to test internal control.
e) Set Sample Volume: 50.

1.3.2.2 PCR-Mastermix B

a) Select FAM channel (Reporter: FAM, Quencher: None) to test adenovirus.
b) Select HEX or VIC channel (Reporter: HEX/VIC, Quencher: None) to test human rhinovirus.
c) Select CY5 channel (Reporter: CY5, Quencher: None) to test *Mycoplasma pneumoniae*.
d) Select ROX channel (Reporter: ROX, Quencher: None) to test internal control.
e) Set Sample Volume: 50.

1.3.3 Set cycle parameters (the time parameter varies according to instruments):

	Step	Temperature	Time	Cycle No.
1	Reverse transcription	50°C	30 min.	1
2	Pre-denaturation	95°C	1 min.	1
3	Denaturation	95°C	15 sec.	45
	Annealing, extension and fluorescence detection	60°C	30 sec.*	
4	Device cooling (Optional)	25°C	10 sec.	1

***Note:** Due to ABI 7500's technical specification, it can not be set at 30 sec., but at 31 sec.

When the settings are completed, save the settings and carry out the reaction procedure.

2.Please process according to the following steps for Portable Molecule Workstation (Model: S-Q31A/S-Q31B):

2.1 Preparation of consumables and reagents

(1) Take out the reaction tube carrier, PCR reaction tube and Tip.
(2) Put the **Tip** into **Well H**,and **PCR reaction tube** into **Well PCR** (The well location information has been marked on the reaction tube carrier).
(3) Put Sample Release Reagent (Reference Number: S1014E) into the **Well B**; Put Six RP-PCR Mix A/B into the **Well C**; Put Six RP-Enzyme Mix into the **Well D**; Put internal control into the **Well A** (if necessary).
(4) Add 20μL sample to be tested or Six RP-Positive Control or Six RP-Negative Control into the Well B (To avoid bubbles during operation, it is recommended to pipet deeply and release slowly).

2.2 Test Procedure (Refer to user manual of each instrument to adjust the settings.)

2.2.1 Gently press the front door to open it.
2.2.2 Place the **Well A** of reagent strip into the instrument towards the outside of the instrument,and close the front door of the instrument.
2.2.3 Click the "**Lab task**" on the instrument display screen to enter the interface of setting new lab task.
2.2.4 Select the required Lab project in the drop-down menu of **Lab project**, enter the corresponding task name in the **Task Name** bar, and input and select other items that should be input or selected.
2.2.5 Click **"Submit"** to submit the lab task and **"OK"** to run the instrument and start the lab task successively.
2.2.6 When the Portable Molecule Workstation (Model: S-Q31B) shows "**Please transfer the PCR tube to the 1/2/3/4"**(The S-Q31A shows "Please transfer the PCR tube") on the interface, take out the PCR tube and cover it well, then centrifuge it instantaneously.
2.2.7 Insert the PCR tube into the PCR amplification module (the "PCR 1/2/3/4" cover has been automatically opened at this time), close the PCR lid of the amplification module, then click "**OK**" for amplification detection.

3.Please process according to the following steps for Portable Molecular Workstation (Model: S-Q36A):

3.1 Preparation of consumables and reagents

(1)Take out the Consumables kits and reagents.
(2) Put Sample Release Reagent (Reference Number: S1014E) into the **Well B**; Put Six RP-PCR Mix A/B into the **Well C**; Put Six RP-Enzyme Mix into the **Well D**; Put internal control into the **Well A** (if necessary), (The well location information has been marked on the carrier set).
(3) Add 20μL sample to be tested or Six RP-Positive Control or Six RP-Negative Control into the **Well B** (To avoid bubbles during operation, it is recommended to pipet deeply and release slowly).

3.2 Test Procedure

3.2.1 Click the "" and "" button on the instrument display screen to open the door of the instrument and put the prepared consumables into the designated position of the instrument.
3.2.2 Click the "**New**" on the instrument display screen to enter the new experiment task setting interface.
3.2.3 Select the required Lab project in the drop-down menu of **Lab project**, enter the corresponding task name in the **Task Name** bar, and input and select other items that should be input or selected.
3.2.4 Click "**Submit**" to submit the lab task and "**OK**" to run the instrument and start the lab task successively.

4. Please process according to the following steps for Portable Molecular Diagnostic System (Model: S-Q37A/S-Q37B):

4.1 Pre-run preparation

4.1.1 Load the amplification reagent component assembly into the extraction reagent component (Nucleic Acid Extraction-Purification Kit, Reference Number: S50016E-12A) to compose the test reagent cartridge;
4.1.2 Open the seal plug of the sample loading hole, add 350μL sample or Six RP-Positive Control or Six RP-Negative Control into the sample loading hole (To ensure Diagnostic System have 300μL samples for nucleic acid extraction); or use transfer pipet from the extraction reagent kit to pipette sample into the sample loading hole (When sample enter the lower bubble of transfer pipet indicates enough sample has been taken). Then close the seal plug.

4.2 Test Procedure

4.2.1 Click the "Specimen" button on the instrument display screen to open the door of the instrument and enter the new experiment task setting interface.
4.2.2 Put the prepared consumables into the designate position of the instrument.
4.2.3 Enter specimen information, select the required experimental project in the drop-down menu of Experimental project, enter the corresponding task name in the Task Name bar, and input and select other items that should be input or selected.
4.2.4 Click "Submit" to submit the experimental task and "OK" to run the instrument.

Reading test results

1. Result Analysis (Refer to user manual of each instrument to adjust the settings, take ABI7500 as example.)
Results will be saved automatically when reactions are completed. Adjust Start, End and Threshold values of Baseline of the graph according to analysis result (Users can adjust the values according to the actual situation. Start value can be set between 3-15, and End value between 5-20. Adjust the amplification curve of negative control to be flat or below threshold). Click "**Analyze**" to implement the analysis and make sure each parameter satisfy the requirements given in "**2. Quality Control**". Go to "Plate" window to record Ct value.

2. Quality Control

The test results below should be met to validate the experiment. Otherwise the test is invalid and needs to be re-tested.

	Six RP-Negative Control	Six RP-Positive Control
Ct value	Respectively use PCR-Master mix A and PCR-Master mix B to validate the experiment by using Six RP-Negative Control and Six RP-Internal Control. The result of RP-Internal Control should be detected positive at the ROX channel with a typical S-shape amplification curve and Ct ≤ 40. The result of Six RP-Negative Control at FAM, HEX(or VIC) , CY5 channels should display no amplification curve (no Ct) and Ct >40.	Respectively use PCR-Mastermix A and PCR-Mastermix B validate the experiment by using Six RP-Positive Control. The result should be a typical S-shape amplification curve at FAM, HEX(or VIC) , CY5 channels and 27≤ Ct ≤33. And it should be no amplification curve (no Ct) or Ct >40 at ROX channel .

Reference Range

ROC curves based on clinical data show that the Ct value of target gene is determined to be 40, and the Ct value of internal control is determined to be 40.

Interpretation of test results

1. Determination for positive and negative result: it should be determined as positive if there is a typical S-shape amplification curve and the Ct ≤ 40; and be determined as negative if there is no amplification curve (No Ct) or the Ct > 40. The details are as below:

Amplification	FAM channel		HEX/VIC channel		CY5 channel		ROX channel	
results Mix type	Ct ≤ 40	No Ct or Ct > 40	Ct ≤ 40	No Ct or Ct > 40	Ct ≤ 40	No Ct or Ct > 40	Ct ≤ 40	No Ct or Ct > 40
PCR-Mastermix A	Inf. A **Positive**	Inf. A **Negative**	Inf. B **Positive**	Inf. B **Negative**	RSV **Positive**	RSV **Negative**	Internal Control	Internal Control
PCR-Mastermix B	ADV **Positive**	ADV **Negative**	HRV **Positive**	HRV **Negative**	MP **Positive**	MP **Negative**	**Positive**	**Negative**

2. For the positive specimen, there is no requirement on the internal control test results. For the negative specimen (none of the six types of respiratory pathogens is detected), the internal control test should be positive (Ct ≤ 40). If the internal control Ct >40 or no Ct is detected, the test result is treated as invalid. An investigation should be performed to find out the reasons. Please collect specimen and retest the specimen. (If repeated tests still produce invalid results, please contact Sansure Biotech.)
3. Determination of gray zone: If the fluorescence signal of the specimen increased significantly at FAM or HEX/ VIC or CY5 channel, but the Ct > 40, then the specimen is in the gray zone, and needs to be retested. If the retest result is still in gray zone, then it is positive.

Limitations of the procedure

The detection result is related to the quality of specimen collection, delivery, processing and storage. Mistakes may lead to an incorrect result. Cross-contamination occurring in specimen processing may result in a false positive result. This kit is designed for detection of conservative areas of pathogens. Mutations are rare, but it is not excluded that gene mutations occur during the epidemic, which may lead to false negative results.

Performance characteristics

Use this kit to detect the enterprise references:
Detect 20 enterprise positive references (P1 - P20): Positive coincidence rate 100%.
Detect 12 enterprise negative references (N1 - N12): Negative coincidence rate 100%.
Detect 11 enterprise precision references (R0 - R10): Repeatedly for 10 times, the coefficient of variation (CV, %) of the Ct value of the corresponding pathogen detection channel for R1 - R6 is not greater than 5.0%.
Detect 20 enterprise detection limit references (SQ1 - SQ20): All meet the minimum detection limit claim.
The minimum detection limit of this kit:

Pathogen	Limit of Detection	Pathogen	Limit of Detection
Influenza A virus	2.0 $TCID_{50}$/mL	Adenovirus	500.0 copies/mL
Influenza B virus	2.0 $TCID_{50}$/mL	*Mycoplasma pneumoniae*	500.0 copies/mL
Respiratory syncytial virus	500.0 copies/mL	Human rhinovirus	500.0 copies/mL

Use this kit to detect National reference for influenza A/B virus nucleic acid test reagent (batch no. : 370006-201501) : **Positive conformity rate :** The test results of PC01 and PC02 are both positive for influenza B virus and negative for influenza A virus. The test results of PC03, PC04, PC05 and PC06 are positive for influenza A virus and negative for influenza B virus. The positive conformity rate (+/+) is 6/6.

Negative conformity rate: The test results of NC01-NC06 are both negative for influenza virus. The negative conformity rate (-/-) is 6/6.
Precision (CV, %): Detect precision references CV1 and CV2 respectively and repeatedly for 10 times, the test results of CV1 are positive for influenza B virus and negative for influenza A virus, the test results of CV2 are positive for influenza A virus and negative for influenza B virus, and the CV of Ct value is not greater than 5.0%.
Limit of Detection: The titer of sensitivity reference (S1) is no less than $2.1x10^{1}TCID_{50}$/L, the test result is positive for influenza B virus and negative for influenza A virus. The titer of S2 is no less than $2.0TCID_{50}$/L,the test result is positive for influenza B virus and negative for influenza A virus. The titer of S3 is no less than $2.5X10^{-1}TCID_{50}$/L, the test result is positive for influenza A virus and negative for influenza B virus. The titer of S4 is no less than $1.0TCID_{50}$/L, the test result is positive for influenza A virus and negative for influenza B virus. The titer of S5 is no less than $9.8X10^{-1}TCID_{50}$/L, the test result is positive for influenza A virus and negative for influenza B virus.
Interfering substance references tests show that the common drugs for respiratory infections (such as oxymetazoline hydrochloride, dexamethasone, cefmenoxime hydrochloride, menthol, zanamivir, ribavirin, azithromycin) under normal dosage will not interfere with this kit. There is no cross reaction with common respiratory pathogens, like measles virus, mumps virus, rubella virus, *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, human parainfluenza virus Type I, cytomegalovirus, enterovirus, human metapneumovirus, *Bordetella pertussis*, *Chlamydia pneumonia*, *Haemophilus influenzae*, *Streptococcus salivarius*, *Streptococcus pneumoniae*, *Neisseria meningitidis*, *Mycobacterium tuberculosis*.

List of references

1. Computational analysis identifies human adenovirus type 55 as a re-emergent acute respiratory disease pathogen. J. Clin.Microbiol, 2010.
2. Development of a Respiratory Virus Panel Test for Detection of Twenty Human Respiratory Viruses by Use of Multiplex PCR and a Fluid Microbead-Based Assay. Journal Of Clinical Microbiology, 2007.
3. Analytical Sensitivity Comparison between Singleplex Real-Time PCR and a Multiplex PCR Platform for Detecting Respiratory Viruses, Plos One, 2015.

Symbol key

Symbols	Meanings	Symbols	Meanings
IVD	*In Vitro* Diagnostic Medical Device	LOT	Batch Code
	Use-by date	REF	Reference Number
	Manufacturer		Date of Manufacture
Σ	Contains sufficient for <n> tests		Temperature Limit
	Caution		Consult Instructions for Use
21 PAP	PAP21: Not corrugated cardboard		Do not re-use
EC REP	Authorized representative in the European Community	CE	This product fulfills the requirements of the European Directive 98/79/EC for *in vitro* diagnostic medical devices.
PCR MIX A	PCR Mix	ENZYME MIX	Enzyme Mix
PCR MIX B	PCR Mix B	INTER CONTROL	Internal Control
CONTROL -	Negative Control	CONTROL +	Positive Control
PRE	Prepackaging		Keep away from light
VERSION	Version	UDI	Unique device identifier

Contact information

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CE IVD