COV-GRIP®
COVID-19 and Flu Rapid Test Ref.: TR-

COG-001

User manual

INDICATION FOR USE

COV-GRIP® is an in vitro diagnostic test for the qualitative and differential detection of influenza A nucleocapsid protein antigen (including the H1N1 subtype), influenza B and/or SARS-CoV-2 in nasopharyngeal (NP) swab specimens.
COV-GRIP® is intended to facilitate the rapid diagnosis of influenza A, influenza B and/or SARS-CoV-2 infection.

INTRODUCTION

Influenza is an acute, highly contagious viral infection of the respiratory tract. The agents responsible for infection are immunologically divergent single-stranded RNA viruses, called influenza viruses. There are three types of influenza viruses: A, B, and C. Type A viruses are the most prevalent and are associated with the most severe outbreaks, while type B infection is usually milder. Type C viruses have never been associated with a large outbreak of human disease. Type A and B viruses can circulate simultaneously, but usually one type is dominant during a given season and in a particular epidemic area. The disease is easily transmitted by coughing and sneezing from droplets containing the virus. Flu epidemics normally occur each year during the fall and winter seasons.

SARS-CoV-2 was identified in 2019 and belongs to the genus of β-coronaviruses. It is the pathogen that causes an emerging atypical pneumonia, coronavirus disease 2019 (Covid-19).
Currently, patients infected with SARS-CoV-2 are the main source of transmission: infected people, who are asymptomatic, can also be an infectious source. Based on the current epidemiological investigation, the incubation period can range from 1 to 14 days but is usually 3 to 7 days.
The main symptoms are fever or feeling feverish and coughing. The sudden loss of smell, without nasal obstruction and total disappearance of taste are also symptoms that have been observed in patients. In people who develop more severe forms, there are breathing difficulties, which can lead to hospitalisation in intensive care and death.

PRINCIPLE OF THE TEST

COV-GRIP® is composed of 2 tests:

Influenza A and B rapid antigen test : A membrane-based immunochromatographic test that uses highly sensitive monoclonal antibodies to detect influenza A and B nucleoprotein antigens in nasopharyngeal specimens. The reagent buffer contains colloidal gold conjugated to monoclonal antibodies to influenza A and B viruses; the reaction membrane contains the secondary antibodies for either virus A or virus B. The entire strip is attached to the inside of a plastic device. When the sample is added to the sample well, the conjugates dried in the reagent buffer are dissolved and migrate with the sample. If influenza A is present in the specimen, a complex formed between the influenza A conjugate and the virus will be captured by the specific influenza A monoclonal antibodies that cover the A(A) region. If the specimen contains influenza B, a complex formed between the influenza B conjugate and the virus will be captured by the specific influenza B monoclonal antibodies that cover the B (B) region. The results appear after 10 minutes in the form of a red line that develops on the membrane. The control line is used as a procedural control and should always appear in the control box (C) if the test procedure is performed correctly.

SARS-CoV-2 Rapid Antigen Test : A lateral flow immunochromatographic assay that uses highly sensitive monoclonal antibodies to detect SARS-CoV-2 nucleocapsid antigen in a nasopharyngeal specimen.
The test uses monoclonal antibodies directed against the SARS-CoV-2 nucleocapsid protein attached to a nitrocellulose strip at the test area (T). A monoclonal antibody directed against the nucleocapsid protein of SARS-CoV-2 labeled with colloidal gold is used as a lyophilized conjugate.
In the test, the SARS-CoV-2 antigens in the sample interact with the monoclonal SARS-CoV-2 antibodies conjugated to the color particles to form a colored antibody-antigen complex.
This complex migrates by capillary action on the membrane to the test line (T) where it will be captured by the monoclonal anti-SARS-CoV-2 antibodies attached to the membrane.
A colored test line should appear in the results window (T) if SARS-CoV-2 antigens are present in the sample. The intensity of the colored test line will vary depending on the amount of SARS-CoV-2 antigens present in the sample. If no SARS-CoV-2 antigen is present in the sample, no color will appear on the test line (T). The control line is used as a procedural control and should always appear in the control box
(C) whether the test procedure is performed correctly.

EQUIPMENT PROVIDED

- 20 sealed pouches each containing a cassette and a moisture absorber
- 20 sterile swabs
- 20 Extraction Tubes and 20 Dropper Tips
- 1 tube holder
- 2 x Buffer Bottles
- 1 user manual

MATERIALS REQUIRED BUT NOT PROVIDED

- Timer

STORAGE AND STABILITY

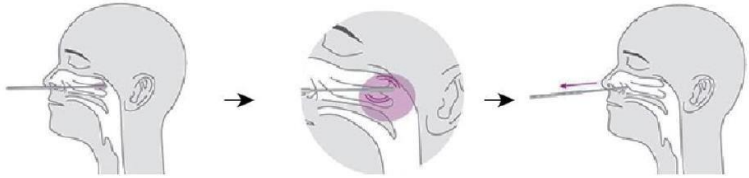
The kit can be stored at room temperature or refrigerated (2-30°C). Do not freeze the components of the kit. Do not use the test device and reagents after the expiration date. Do not use the test device when it has been out of the airtight pouch for more than one hour.

WARNINGS AND PRECAUTIONS

- For professional in vitro diagnostic use only.
- The test device should remain in the sealed pouch until it is used.
- Do not use the kit after the expiration date.
- The swabs, tubes and test cassettes are for single use.
- The extraction buffer contains a solution with a preservative (0.09% sodium azide). If the solution comes into contact with skin or eyes, rinse thoroughly with water.
- Solutions that contain sodium azide can react explosively with lead or copper pipes. Use large amounts of water to rinse the discharged solutions down a sink.
- Do not exchange or mix components from different batches.
- For nasopharyngeal swabs, use the flocked swab provided in the kit.
- Wear appropriate protective clothing, gloves, and eye/face protection when handling the contents of this kit.
- Samples should be treated as described in the "Sample Collection" and "Sample Preparation" sections of this package leaflet. Failure to follow these instructions may result in inaccurate results.
- To obtain accurate results, do not use samples that contain blood or are too viscous.
- Good laboratory practices should be followed at all times when working with samples from SARS-CoV-2 patients. Patient swabs, test cassettes, and extraction buffer vials used can be potentially infectious. Proper handling and disposal procedures should be established by the laboratory in accordance with local regulations.
- Humidity and temperature can affect the results.
- The equipment used must be disposed of in accordance with local regulations.

SAMPLE COLLECTION

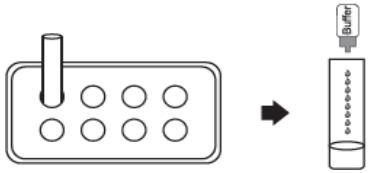
Use of the nasopharyngeal swab provided in the kit.
1. Carefully insert the swab into the patient's nostril, reaching the surface of the posterior nasopharynx.
2. Turn the swab several times.
3. Carefully remove the swab from the nasal cavity.



SAMPLE PREPARATION

1. Insert the extraction tube into the holder. Make sure the tube is securely in place and reaches the bottom of the bracket.
2. Add the extraction buffer to the extraction tube until it reaches the bottom mark (between 13-17 drops, corresponding to 0.5 ml).

13-17 drops



3. Insert the swab into the extraction tube containing the swab.
4. Turn the swab (at least 6 times) in the extraction tube for 1 minute.
5. Let the swab rest in the extraction tube for 1 minute.
6. Press the swab against the walls of the extraction tube to extract the liquid from the swab.
7. Remove and discard the swab
8. The extracted solution will be used as a sample.

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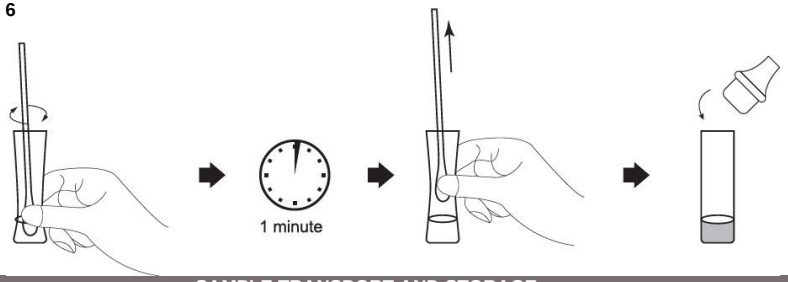


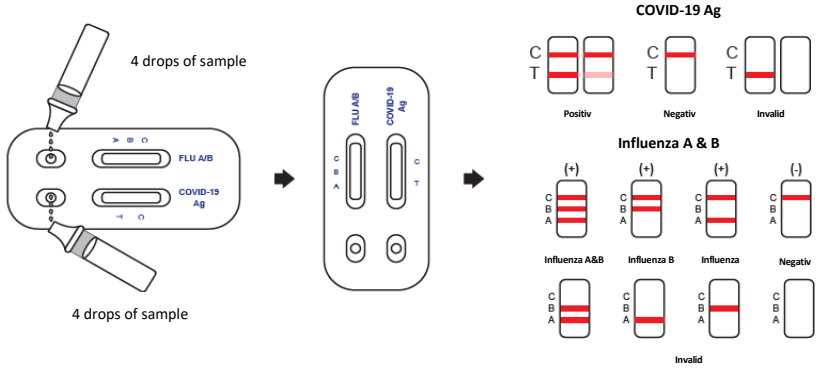
SAMPLE TRANSPORT AND STORAGE

Do not return the nasopharyngeal swab to its original packaging.
Nasopharyngeal swabs should be tested as soon as possible. If this is not possible, in order to preserve the integrity of the sample, it is strongly recommended to place the nasopharyngeal swab in a clean, unused plastic tube, labeled with the patient's information, and seal tightly and then store it at room temperature (15-30°C) for up to 1 hour before the test is performed. Make sure that the swab fits firmly into the tube and that the cap is tightly closed. If the delay exceeds 1 hour, the sample must be disposed of. A new sample must be taken to perform the test.

PROCEDURE

Ensure that the sample and test components are at room temperature (15-30°C) before proceeding with the test.

1. Remove the cassette from the sealed pouch just before performing the test and place it on a flat surface.
2. Turn the extraction tube upside down and place 4 drops (100µL) of sample, by pressing the tube, into <u>each</u> of the sample wells (S) of the cassette.
3. Wait for the coloured stripe(s) to appear. The result should be read at 15 minutes. Do not interpret the result after 20 minutes.



INTERPRETATION OF THE RESULTS

<u>For the influenza A&B rapid antigen test</u> **:**

Positive / Influenza A Positive : The presence of two bands, the control band (C) and the test band A (A), in the result window indicates a positive result for the influenza A viral antigen.

Positive / Influenza B positive: The presence of two bands, the control band (C) and test band B (B), in the result window indicates a positive result for the influenza B viral antigen.

Positive / Influenza A+B positive : The presence of three bands, the control band (C) and the test bands (A) and (B), in the result window indicates a positive result for the viral antigen of influenza A and influenza B.

Negative : Only the control strip (C) appears in the result window. This indicates a negative result.

Invalid : If the control band (C) is not visible in the result window after performing the test, the result is considered invalid. Some causes of invalid results are due to not following the instructions correctly. It is recommended that the sample be retested using a new test.

<u>For the COVID-19 rapid antigen test:</u>

Positive: The presence of two bands, the control band (C) and the test band (T), in the result window indicates a positive result.

Negative : Only the control strip (C) appears in the window indicating a negative result.

Invalid: If the control band (C) is not visible in the result window after performing the test, the result is considered invalid. Some causes of invalid results are due to not following the instructions correctly. It is recommended that the sample be retested using a new test.

Note :
The intensity of the color in the test line region may vary depending on the concentration of the sample. Therefore, any color shade in the region of the test line (T) should be considered positive. Note that this is a qualitative test and cannot determine the concentration of antigen in the sample.
Insufficient sample volume, incorrect operating procedure, or outdated testing are the most likely reasons for control tape failure.

QUALITY CONTROL

Internal quality control is included in the test. A red line appearing in the control region (C) is the internal procedure control. It confirms that a sufficient sample volume was used and that the test procedure was correctly followed.
External quality checks are not provided in this kit.

BOUNDS

- **COV-GRIP®** is intended for professional in vitro diagnostic use and should only be used for the qualitative detection of influenza A, B and/or SARS-CoV-2 in nasopharyngeal (NP) swab specimens.
- The test is capable of detecting viable and non-viable particles of influenza and SARS-CoV-2. The performance of the test depends on the antigenic load and may not correlate with the results of the viral culture performed on the same sample.
  The etiology of respiratory infection caused by microorganisms other than influenza A, B or SARS-CoV-2 will not be established with this test.
- If the test result is negative and clinical symptoms persist, additional testing is recommended to use other clinical methods. A negative result does not exclude the presence of influenza A, influenza B, or SARS-CoV-2 viral antigens in the specimen as they may be present below the detection limit of the test or if the specimen was collected or transported incorrectly.
- As with all diagnostic tests, a confirmed diagnosis should only be made by a physician after all clinical and laboratory results have been evaluated.
- Inadequate or inappropriate specimen collection, storage, and transportation may result in a false negative result.
- Failure to follow the test procedure may have a negative effect on the performance of the test and/or invalidate the test result.
- Although this test has been shown to detect cultured avian influenza viruses, including H5N1 avian influenza subtype A, the performance characteristics of this test with samples from humans infected with H5N1 or other avian influenza viruses are unknown.
- Performance characteristics for influenza A were established when influenza A/H3 and A/H1 viruses were the major circulating influenza A viruses. When other influenza A viruses emerge, performance characteristics may vary.
- The positive and negative predictive values are highly dependent on prevalence. False-positive results are more likely during periods of low influenza activity when prevalence is moderate to low. Positive results do not exclude co-infections with other pathogens.
- For the COVID-19 rapid antigen test, positive test results do not exclude co-infections with other pathogens and do not differentiate between SARS-CoV and SARS-CoV-2.
- The test must be used according to the regulations in force.

PERFORMANCE

For the influenza A&B rapid antigen test

Limit of Detection (LOD)
The lowest detectable concentration of influenza A virus is **1.5 x 104 TCID50/test**. The lowest detectable concentration of influenza B virus is **1.5 x 105 TCID50/test**.

Analytical responsiveness
The influenza A strain listed tested positive by the influenza A&B rapid antigen test.
Specific strains of influenza, causing infection in humans, can all contain the conserved nucleoproteins targeted by the influenza A&B rapid antigen test.

Strains	Sources	Subtype	Concentration
Flu A/Hubei/PR8/2001	Human	H1N1	1.8x104 TCID50/test
Flu A/New Kaledonia/20/99	Human	H1N1	1.8x104 TCID50/test
Flu A/Yamagata/32/89	Human	H1N1	1.8x104 TCID50/test
Flu A/Beijing/262/95	Human	H1N1	1.8x104 TCID50/test
Flu A/Singapore/1/57	Human	H2N2	3.0x104 TCID50/Test
FLu A/Hubei/3/2005	Human	H3N2	3.0x104 TCID50/Test
Flu A/Akita/1/94	Human	H3N2	3.0x104 TCID50/Test
Flu A/Kita Kyusyu/159/93	Human	H3N2	3.0x104 TCID50/Test
Flu A/Iowa/15/30	Pig	H1N1	3.0x104 TCID50/Test
Flu A Hongkong/168/93	Pig	H1N1	3.0x104 TCID50/Test
Flu A/Anhui/24/2004	Pig	H5N1	6.0x104 TCID50/Test
Flu A/Hubei/134/2000	Pig	H9N2	6.0x105 TCID50/Test
Flu A/Hubei/251/2001	Pig	H9N2	6.0x105 TCID50/Test
Flu A/Yuyao/1/2006	Chicken	H5N1	6.0x104 TCID50/Test
Flu A/Yuyao/2/2006	Chicken	H5N1	6.0x104 TCID50/Test
Flu A/Jiangs u/2/2004	Chicken	H5N1	6.0x104 TCID50/Test
Flu A/Hubei/216/83	Duck	H7N8	3.0x105 TCID50/Test
Flu A/Hubei/118/2003	Duck	H9N2	1.5x105 TCID50/test
Flu A/Hubei/155/2003	Duck	H9N2	6.0x105 TCID50/Test
Flu A/Hubei/137/1982	Duck	H10N4	3.0x105 TCID50/Test
Flu A/Singapore/3/97	Duck	H5N3	6.0x104 TCID50/Test
Flu A/Henan/1/2004	Tree Sparrow	H5N1	6.0x105 TCID50/Test
Flu A/Henan/2/2004	Tree Sparrow	H5N1	3.0x105 TCID50/Test
Flu A/Henan/4/2004	Tree Sparrow	H5N1	6.0x104 TCID50/Test
Flu A/Wisconsin/66	Turkey	H9N2	6.0x104 TCID50/Test
Flu A/England/1/63	Turkey	H7N3	6.0x104 TCID50/Test
Flu A/Singapore/1/57	Bird	H5N1	6.0x104 TCID50/Test
Flu A/Hunan/71/2004	Bird	H5N1	6.0x104 TCID50/Test
Flu A/Shanxi/50/2006	Bird	H5N1	6.0x104 TCID50/Test
Flu A/Shanxi/42/2006	Bird	H5N1	6.0x104 TCID50/Test
Flu A/Fujian/320/2004	Bird	H5N1	3.0x105 TCID50/Test

The influenza A&B rapid antigen test can detect all nine strains of influenza B.

Clinical performance
The performance of the influenza A&B rapid antigen test compared to cell culture.

Types of samples	Type	Sensitivity (%)	Specificity (%)	Accuracy (%)
Nasopharyngeal swab	Has	92.6 (25/27)	96.4 (81/84)	95.5 (106/111)
	B	90.0 (27/30)	95.8 (91/95)	94.4 (118/125)
Oropharyngeal swab	Has	83.3 (20/24)	95.2 (59/62)	91.9 (79/86)
	B	82.6 (19/23)	91.8 (67/73)	89.6 (86/96)
Nasal suction	Has	88.9 (48/54)	93.3 (125/134)	92.0 (173/188)
	B	91.2 (52/57)	95.4 (98/103)	93.8 (150/160)
Runny nose /Nasal mucus	Has	80.7 (46/57)	94.9 (93/98)	89.7 (139/155)
	B	89.6 (62/69)	94.6 (87/92)	92.5 (149/161)

Cross-reactions
The influenza A&B rapid antigen test was evaluated on a total of 30 bacterial and viral isolates. Bacterial isolates were evaluated at a concentration between 107 and 109 org/ml.
Viral isolates were evaluated at a concentration of at least 104-108 TCID50/mL. Adenovirus 18 and Parainfluenza virus 3 were tested at a concentration of 102 TCID50/ml.
None of the organisms or viruses listed below have tested positive for influenza A&B rapid antigen testing.

Bacterial panel:

Acinetobacter calcoaceticus	Bacteroides fragilis
Neisseria gonorrhoeae	Neisseria meningitidis
Pseudomonas aeruginosa	Staphylococcus aureus
Streptococcus pneumoniae	Streptococcus sanguis
Proteus vulgaris	Streptococcus sp. Gp. B
Streptococcus sp. Gp. C	Streptococcus sp. Gp. G
Mycobacterium tubercolosis	Oral Mycoplasma

Viral Panel:

Human Adenovirus B	Human Rhinovirus 2
Human Adenovirus C	Human Rhinovirus 14
Adenovirus type 10	Human Rhinovirus 16
Adenovirus type 18	Measles
Human Coronavirus OC43	Mumps
Human Coxsackievirus A9	Sendai virus
Coxsackievirus B5	Parainfluenza virus 2
Human herpesvirus 2	Parainfluenza virus 3

Interference
The substances listed below were tested: whole blood (2%); three over-the-counter mouthwashes (25%); three over-the-counter throat drops (25%); three over-the-counter nasal sprays (25%); 4-Acetamidophenol (10mg/mL); Acetylsalicylic acid (20mg/mL); Chlorpheniramine (5mg/mL); Dextromethorphan (10mg/mL); Diphenhydramine (5mg/mL); Ephedrine (20mg/mL); Gaiacol glyceryl ether (20 mg/mL); Oxymetazoline (20mg/mL); Phenylephrine (100mg/mL) and Phenylpropanolamine (20mg/mL).
The results showed no interference.

For SARS-CoV-2 Rapid Antigen Test Limit of Detection (LOD)
The minimum detectable concentration of SARS-CoV-2 is **1.15 x 102 TCID50/ml.**

Clinical performance
The performance of the SARS-CoV-2 rapid antigen test was evaluated at the Orléans Regional Hospital (Infectious Diseases Department – publication of results in progress) as part of a prospective comparative clinical study involving 226 individuals of unknown status with regard to SARS-CoV-2 infection, recruited consecutively or randomly.

Performance of the SARS-CoV-2 rapid antigen test compared to PCR

Reference method	PCR SARS-CoV-2 Positive
Number of positive samples	113
Total number of samples	117
Sensitivity Result	**96.6%**
95% CI	*93.3-99.8%*

Reference method	PCR SARS-CoV-2 Negative
Number of negative samples	109
Total number of samples	109
Specificity Result	**100%**

Precision
Within: 3 samples (one negative, one weak positive (LOD) and one strong positive (LODx4)) were tested 10 times each. The results were all correct.
Inter-batch: 3 samples (one negative, one weak positive (LOD) and one strong positive (LODx4)) were tested 10 times on 3 different batches. The results were all correct.

Cross-reactions

Samples containing the pathogens listed below were tested. The results showed no cross-reaction.

Pathogen	Concentration
Respiratory syncytial virus Type A	5.5×107PFU/ml
Respiratory syncytial virus Type B	2.8×105TCID50/ml
Novel influenza A H1N1 virus (2019)	1×106PFU/mL
Seasonal influenza A H1N1 virus	1×105PFU/ml
Influenza A H3N2 virus	1×106PFU/mL
Influenza A H5N1 virus	1×106PFU/mL
Influenza B Yamagata	1×105PFU/ml
Influenza B Victoria	1×106PFU/mL
Rhinovirus	1×106PFU/mL
Adenovirus 3	5×107.5TCID50/ml
Adenovirus 7	2.8×106TCID50/ml
EV-A71	1×105PFU/ml
Mycobacterium tuberculosis	1×103bacteria/ml
Mumps virus	1×105PFU/ml
Human coronavirus 229E	1×105PFU/ml

Pathogen	Concentration
Human coronavirus OC43	1×105PFU/ml
Human coronavirus NL63	1×106PFU/mL
Human coronavirus HKU1	1×106PFU/mL
Parainfluenza virus 1	7.3×106PFU/mL
Parainfluenza virus 2	1×106PFU/mL
Parainfluenza virus 3	5.8×106PFU/ml
Parainfluenza virus 4	2.6×106PFU/ml
Haemophilus influenzae	5.2×106CFU/ml
Streptococcus pyogenes	3.6×106CFU/ml
Streptococcus pneumoniae	4.2×106CFU/ml
Candida albicans	1×107CFU/ml
Bordetella pertussis	1×104bacteria/ml
Mycoplasma pneumoniae	1.2×106CFU/ml
Chlamydia pneumoniae	2.3×106IFU/mL
Legionella pneumophila	1×104bacteria/ml

Interference

The substances listed below were tested: blood (EDTA), antiviral drugs, antibiotics/antibacterial drugs, nasal sprays or nasal drops, nasal corticosteroids.

The results showed no interference.

SYMBOL LEGEND

	Read the user manual		Number of tests per kit		Maker
IVD	For use in *in vitro diagnostics* only		Expiry date		Do not reuse
	Store between 2 and 30°C	LOT	Lot number	REF	Reference
CE	CE marking				

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