REF: GTIN : 03760097080046

Ksmart® SARS-COV2 Antigen Rapid Test

(Colloidal Gold)

IVD

This instruction for use (IFU) must be read carefully prior to use and must be carefully followed. The reliability of the result cannot be guaranteed if there are any deviations from the instructions for use.

**PACKING SPECIFICATION**

This box contains 25 Ksmart® SARS-COV2 Antigen Rapid Test individually packed in a sealed pouch. The Ksmart® tests must be used only with the LabPad® Evolution analyzer manufactured by the Avalun Company.

**INTENDED USE**

The Ksmart® SARS-COV2 Antigen Rapid Test is a rapid immunochromatographic assay for use with the LabPad® Evolution. This test is intended for the qualitative detection of 2019-nCoV antigens directly in nasopharyngeal specimens collected by a trained healthcare professional.

The antigen is generally detectable in upper respiratory specimens during the acute phase of infection. Positive results indicate the presence of viral antigens, but clinical correlation with patient history and other diagnostic information is necessary to determine the infection status. Positive results do not rule out bacterial infection or co-infection with other viruses. The agent detected may not be the definite cause of disease.

Negative results do not preclude SARS-CoV2 infection and should be considered in the context of a patient's recent exposures, history and the presence of clinical signs and symptoms consistent with COVID-19, and might need to be confirmed by a molecular assay.

**PRINCIPLE OF THE PROCEDURE**

The Ksmart® SARS-COV2 Antigen Rapid Test is a qualitative membrane-based immunoassay for the detection of 2019-nCoV antigens in human nasopharyngeal specimens. SARS-CoV2 antibody is coated in test line region. During testing, the specimen reacts with 2019-nCoV antibody-coated particles in the test. The mixture then migrates upward on the membrane by capillary action and reacts with the 2019-nCoV antibody in test line region. If the specimen contains 2019-nCoV antigens, a colored line will appear in test line region as a result of this. If the specimen does not contain 2019-nCoV antigens, no colored line will appear in the test line region, indicating a negative result. To serve as a procedural control, a colored line will always appear in the control line region, indicating that the proper volume of specimen has been added and membrane wicking has occurred.

The result of the Ksmart® SARS-COV2 Antigen Rapid Test is intended to be read out by the matching LabPad® Evolution analyzer.

**REAGENTS AND MATERIALS SUPPLIED**

**1. Main components:**

- Test card in an individual foil pouch with desiccant: x25
- Nasopharyngeal sterile swab: x25
- Antigen extraction tube: x25
- Antigen extract buffer (R1): x2
- Instruction for use: x1

**2. Ingredients of the test**

Monoclonal 2019-nCoV antibody is coated on the test line region and goat polyclonal anti-Chicken IgY antibody is coated on the control line region. Conjugate pad is coated with chicken IgY and monoclonal 2019-nCoV antibody conjugated with colloidal particles to target SARS-CoV-2 antigen.

**3. Ingredients of the sample extraction solution**

• Phosphate solution

**Note:** The components in different batches of the kit cannot be mixed.

**MATERIALS REQUIRED BUT NOT PROVIDED**

• LabPad® Evolution analyzer

• Timer, if to use the Endpoint Mode of the analyzer

• Pipette, if to control precisely the sample volume to be deposited on the Ksmart

**STORAGE AND STABILITY**

1. Store the kit at 4-30° C/ 39.2-86° F
2. Kit materials are stable until the expiration date printed on the packaging.
3. Do not freeze the kit.
4. Test should be used within 15 minutes after opening.
5. The buffer should be used immediately after dropping into the dropper.

**SPECIMEN REQUIEMENTS**

1. Nasopharyngeal swab:

To collect a nasopharyngeal swab specimen, insert a sterile swab into the nostril of the patient, reaching the surface of the posterior nasopharynx. Using gentle rotation, push the swab until resistance is met at the level of the turbinate. Rotate the swab a few times against the nasopharyngeal wall. Remove the swab from the nostril carefully.

2. Specimen extraction:

Place the swab in the Antigen extraction tube, rotate the swab for about 10 seconds, and press the swab head against the tube wall to release the antigen in the swab.

3. Timing for testing:

Specimen should be tested as soon as possible after collection (within half an hour).

4. Samples should not be inactivated.

**APPLICABLE EQUIPMENT**

This product is only applicable to the LabPad® Evolution analyzer.

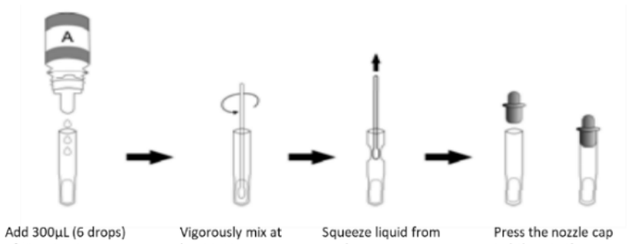
**TEST PROCEDURE**

**Caution:**

1. Do not open the pouch until you are ready to perform a test.
2. Remove the rapid test from the foil pouch and place on a clean dry surface.
3. Identify the test card for each specimen.
4. Discard used test, extraction tube and swab in a suitable biohazards waste container.
5. Wear personal protective equipment, such as gloves and lab coats when handling kit reagents. Wash hands thoroughly after the tests are done.
6. Clean up spills thoroughly using an appropriate disinfectant.
7. Handle all specimens as if they contain infectious agents.
8. Observe established precautions against microbiological hazards throughout testing procedures.

**Sample processing:**

1. Place the extraction tube on the workbench. The bottle of antigen extraction buffer (R1) is pressed vertically downward to allow the solution to drip freely into the extraction tube without touching the edge of the tube. Add 6 drops of R1 to the extraction tube.
2. Put the swab specimen into the extraction tube, rotate the swab for about 10 times, and press the swab head against the tube wall to release the antigen in the swab. Squeeze the swab over the head to remove the swab so as to remove as much liquid as possible from the swab. Dispose of swabs according to biohazard waste disposal method.
3. Press the nozzle cap tightly onto the extraction tube




**Test operation:**

Before performing the test, you must read the instruction manual of the product and the operating manual of the LabPad® Evolution analyzer completely.

1. Allow all kit components and specimens to reach room temperature between 18°C~26°C prior to testing. Ksmart® Test and extraction buffer that are not balanced to room temperature (18℃~26℃) can affect the accuracy of the results.
2. Instrument preparation: Turn on the power of the LabPad® Evolution analyzer, and check the detection mode set for SARS-COV2 Antigen (Standard or Endpoint Mode). If necessary, go to the set up menu to switch to the desired mode (see LabPad® Evolution user manual for details).
3. Unpack the Ksmart® from the pouch and place it horizontally on a dry table.
4. Collect the specimen and process the sample as describe above (see respectively "Specimen requirements" and "Sample Processing"
5. Add 80μL (2 drops) of the processed sample to the sample well of the Ksmart®
   **Note :** You may want to use a pipette if you want a precise control of the sample volume.
6. Proceed to Ksmart® reading according to the selected detection mode:

➔ Standard mode: Insert the Ksmart® into the analyzer as soon as the absorption of the sample is complete in the sample well of the Ksmart® and within 2 minutes after sample deposition. The reader will automatically take care of the timing of the test and the result will be displayed as soon as available if positive, or when 20 minutes have elapsed if negative.

➔ Endpoint mode: count 20 minutes with a timer before inserting the Ksmart® into the reader. Reading takes about 10 seconds until result is displayed.
**Note**: It is essential to insert the Ksmart® in a time window of 20 to 30 minutes after depositing the sample in the Ksmart® sample well, as invalid result may occur otherwise. After 30 minutes, do not insert the Ksmart®: use another Ksmart® to restart test operations from step 1.

**INTERPRETATION OF TEST RESULTS**

LabPad® Evolution automatically determines the positive or negative result according to the comparison between the intensity value of the control and detection lines and a reference intensity value, and displays the qualitative result on the screen.

1. Positive (+): indicates that the control line is positive, and that the test line value is above the cut-off value, which is interpreted as positive.

2. Negative (-): indicates that the control line is positive, and that the test line value is below the cut-off value, which is interpreted as negative.

3. Invalid: indicates that the control line is negative, even if the test line value is above the cut-off value.

**LIMITATIONS**

1. The test procedure, precautions and interpretation of results for this test must be followed strictly when testing.
2. The result of the test should be used for screening. Diagnosis should be established along with clinical symptoms, epidemic condition, and further clinical data. For more accuracy of immune status, additional follow-up testing using other laboratory methods may be recommended.
3. This reagent can only qualitatively detect 2019-nCoV antigens in human nasopharyngeal swab. It cannot determine the antigen concentration in the samples.
4. A negative result may occur if the concentration of antigen in a specimen is below the detection limit of the test or if the specimen was collected or transported improperly. Therefore, a negative test result does not eliminate the possibility of SARS-CoV-2 infection, and might need to be confirmed by viral culture or a molecular assay or an ELISA test.
5. Positive test results do not rule out co-infections with other pathogens.
6. The accuracy of the test depends on the sample collection process. Improper sample collection, improper sample transportation and storage or freezing and thawing of the sample will affect the test results.
7. Optimum sensitivity is obtained when eluting swabs with the R1 extraction buffer. Using other diluents may result in wrong results and lower sensitivity.
8. The solution and test card must be equilibrated to room temperature (18℃~26℃) before used, otherwise the results may be incorrect.
9. Sensitivity maybe decrease if the sample did not test directly. Please test the sample as soon as possible.
10. Cross reactions maybe exist due to the N protein in SARS has a high homology with the new coronavirus (2019-nCoV). However, the interpretation of the results is not affected during seasons without SARS infection.
11. Mutations in viral genes may lead to changes in antigen epitope, leading to false negative results.

**PERFORMANCE CHARACTERISTIC**

**1. Limit of Detection (LOD)**

The LOD for the reagent of the Ksmart® SARS-COV2 Antigen Rapid Test was established using dilutions of an inactivated virus culture. The LOD was established to be 30 TCID50/mL.

**2. Clinical study**

The sensitivity of the reagent of the Ksmart® SARS-COV2 Antigen Rapid Test was established in prospective studies conducted at a trial site in China during the spring 2020 SARS-CoV-2 pandemic. A total of 176 positive specimens were tested. These specimens consisted of nasopharyngeal swabs.

The specificity of reagent of the Ksmart® SARS-COV2 Antigen Rapid Test was tested using 441 negative specimens. The sensitivity and specificity of the reagent of the Ksmart® SARS-COV2 Antigen Rapid Test was compared to a commercialized molecular assay.

| 2019-nCoV Antigen | PCR Test | | Total |
|---|---|---|---|
| Rapid Test | Positive | Negative | |
| Positive | 164 | 3 | 167 |
| Negative | 12 | 438 | 450 |
| Total | 176 | 441 | 617 |

Analysis of coincidence rate of the reagent of Ksmart® SARS-COV2 Antigen Rapid Test and PCR Test in nasopharyngeal samples:

Sensitivity: 164/ (164+12) × 100 % = 93.18 %

CI95% = [89.32 % - 97.04 %]

Specificity: 438 / (3+438) × 100 % = 99.32 %

CI95% = [95.55 % - 100 %]

Accuracy: (164+438) / (164+12+3+438) × 100% = 97.57 %

### 3. Cross reaction testing

Cross-reaction is mainly used to verify the influence of common respiratory pathogens on the detection performance of the test. The following respiratory pathogens were selected for cross-reactivity tests: influenza A virus, H1N1, influenza B virus, Mycoplasma pneumoniae, Rhinovirus A, Rotavirus, Large intestine Escherichia, respiratory syncytial virus, adenovirus, etc.

The concentration of bacterial samples is set to $10^6$ cfu/ml or higher, and the concentration of virus samples is set to $10^6$ pfu/ml or higher. The results of the reagent testing are shown in the table below.

| Pathogen | Concentration | Testing results |
|---|---|---|
| HKU1 | $10^6$ pfu/mL | - |
| OC43 | $10^6$ pfu/mL | - |
| NL63 | $10^6$ pfu/mL | - |
| 229E | $10^6$ pfu/mL | - |
| MERS-coronavirus | $10^6$ pfu/mL | - |
| Human Metapneumovirus | $10^6$ pfu/mL | - |
| Influenza A virus H1N1 | $10^6$ pfu/mL | - |
| Influenza A virus H3N2 | $10^6$ pfu/mL | - |
| Influenza A virus H5N1 | $10^6$ pfu/mL | - |
| Influenza A virus H7N9 | $10^6$ pfu/mL | - |
| Influenza B virus | $10^6$ pfu/mL | - |
| Mycoplasma pneumoniae | $10^6$ cfu/mL | - |
| Rhinovirus A | $10^6$ pfu/mL | - |
| Rhinovirus B | $10^6$ pfu/mL | - |
| Rhinovirus C | $10^6$ pfu/mL | - |
| Adenovirus 1 | $10^6$ pfu/mL | - |
| Adenovirus 2 | $10^6$ pfu/mL | - |
| Adenovirus 3 | $10^6$ pfu/mL | - |
| Adenovirus 4 | $10^6$ pfu/mL | - |
| Adenovirus 5 | $10^6$ pfu/mL | - |
| Adenovirus 7 | $10^6$ pfu/mL | - |
| Adenovirus 55 | $10^6$ pfu/mL | - |
| Enterovirus A | $10^6$ pfu/mL | - |
| Enterovirus B | $10^6$ pfu/mL | - |
| Enterovirus C | $10^6$ pfu/mL | - |
| Enterovirus D | $10^6$ pfu/mL | - |
| EB Virus | $10^6$ pfu/mL | - |
| Measles virus | $10^6$ pfu/mL | - |
| Human cytomegalovirus | $10^6$ pfu/mL | - |
| Rotavirus | $10^6$ pfu/mL | - |
| Norovirus | $10^6$ pfu/mL | - |
| Mumps virus | $10^6$ pfu/mL | - |
| Varicella-zoster virus | $10^6$ pfu/mL | - |
| Respiratory syncytial virus | $10^6$ pfu/mL | - |
| Mycoplasma pneumoniae | $10^6$ cfu/mL | - |
| Escherichia Coli | $10^6$ cfu/mL | - |
| Staphylococcus aureas | $10^6$ cfu/mL | |
| 20 samples of normal people | | All negative |

These experiments have confirmed that the concentration of microorganisms or viruses set above has no effect on the detection performance of the reagent of the Ksmart® SARS-COV2 Antigen Rapid Test.

### 4. Interference testing

Some clinically commonly used substances may affect product performance. The following substances were spiked into negative and weakly positive samples to detect the impact on the test results (see the table below).

| Materials | Concentration | Negative sample | Weak positive samples |
|---|---|---|---|
| Mucin | 200 mg/ml | - | + |
| Hemoglobin | 10 mg/ml | - | + |
| Histamine Hydrochloride | 4.0mg/L | - | + |
| Human albumin | 60 mg/ml | - | + |
| α- interferon | 2 ng/ml | - | + |
| Lopinavir | 2 μg/ml | - | + |
| Tobramycin | 10 mg/L | - | + |
| Ribavirin | 40 mg/L | - | + |
| Tramadol | 12 μg/ml | - | + |
| Azithromycin | 5 μg/ml | - | + |
| Meropenem | 10 mg/ml | - | + |
| Oseltamivir | 1000 ng/ml | - | + |
| Benzocaine | 1.5 mg/ml | - | + |
| Peramivir | 20 μg/ml | - | + |

These experiments confirm that the above substances have no effect on the detection performance of the reagent of the Ksmart® SARS-COV2 Antigen Rapid Test. However, high concentrations of hemoglobin have an effect on the elimination of the background, suggesting that strong hemolysis can affect the observation of weakly positive samples.

### 5. HOOK effect

**Determination of Hook effect**

The Hook effect can appear in immunochromatographic tests at high antigen concentration, when the intensity of the test line can become weaker as the antigen concentration increases. The Hook effect was tested here using a strongly positive sample which was diluted up to 1:64. The experiment started from the lowest concentration up to highest concentration, each concentration being repeated 5 times.

| Specimen | 1:2 | 1:4 | 1:8 | 1:16 | 1:32 | 1:64 |
|---|---|---|---|---|---|---|
| Detection intensity | +++ | +++ | +++ | ++ | >+ | + |
| Detection times | 5 times | 5 times | 5 times | 5 times | 5 times | 5 times |

No obvious hook or prozone effect was detected for the reagent of the Ksmart® SARS-COV2 Antigen Rapid Test.

**Prozone effect verification**

5 strongly positive samples were used for verification.

| Specimen | 1:2 | 1:4 | 1:8 | 1:16 | 1:32 | 1:64 |
|---|---|---|---|---|---|---|
| Specimen 1 | +++ | +++ | +++ | ++ | >+ | + |
| Specimen 2 | +++ | +++ | +++ | ++ | >+ | + |
| Specimen 3 | +++ | +++ | +++ | ++ | ++ | + |
| Specimen 4 | +++ | +++ | +++ | ++ | >+ | + |
| Specimen 5 | +++ | +++ | +++ | ++ | ++ | + |

No obvious prozone effect was observed for the reagent of the Ksmart® SARS-COV2 Antigen Rapid Test.

### PRECAUTIONS

1. The reagent of the Ksmart® SARS-COV2 Antigen Rapid Test is a disposable in vitro diagnostic reagent, which is only used for the detection of human nasopharyngeal swab. The operations should be carried out strictly according to the instructions above. Do not use expired or damaged products.
2. The kit should be sealed and kept away from moisture. Reagents or samples stored at low temperature should be balanced to room temperature before they can be used.
3. In order to avoid dampness to affect results, Ksmart® should be used as soon as possible after removal from their humidity free individual pouches.
4. Do not use a Ksmart® that laid for too long or was contaminated after being removed from its individual pouch.
5. Please operate in accordance with laboratory testing procedures for infectious diseases. In particular, used materials must be disposed of properly in accordance with applicable procedures to infectious substances.
6. Results accuracy may be affected by incorrect operations such as insufficient sample mixing, insufficient amount of solution, inaccurate detection time, etc.
7. Components from different batches should not be mixed.
8. There should be appropriate biosafety assurance procedures for those substances containing or suspected to contain sources of infection, especially:
    1) Handle samples and reagents with gloves;
    2) Do not suck samples with your mouth;
    3) Do not smoke, eat, drink, make up or handle contact lenses while handling these items;
9. In case of spilled sample or reagent, immediately use a disinfectant for cleaning. Disinfect and treat all samples, reagents and potential pollutants in accordance with relevant local regulations;

**BIBLIOGRAPHY**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4. https://www.medcalc.org/calc/relative_risk.php



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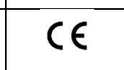
**INSTRUCTIONS OF SYMBOL**

| | | | |
|---|---|---|---|
|  | Consult instructions for use |  | Number of tests per kit |
|  | Store between |  | Lot number |
|  | For single use |  | In vitro diagnostic medical device |
|  | Manufacturer |  | CE Mark |
|  | Expire date | | |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