# LiliF™ GBN COVID-19 Ag Rapid Kit

REF IRH41071 Σ 25 1℃ 30℃

## Intended Use

LiliF™ GBN COVID-19 Ag rapid kit is a chromatographic immunoassay for the qualitative test for the detection of COVID-19 antigens in nasopharyngeal swab specimen. This kit should only be used as a research use only (RUO) purposes. In the case of positive results, confirmation tests through alternative diagnostic methods are required. After analyzing these test results and other clinical data, the Principal Investigator should perform a final diagnosis.

## Development Background

There are four genes in the Coronavirus family. Those are known to alpha, beta, gamma, and delta. Alpha and beta corona viruses can cause illness in both humans and animals, whereas others, such as gamma and delta coronaviruses, only infect animals. Reported illnesses have ranged from mild cold symptoms by Coronavirus 229E, NL63, OC43, or HKU1 to severe illness (e.g., pneumonia) by MERS-CoV and SARS-CoV. COVID-19 is a new coronavirus that has not previously identified. The new coronavirus (COVID-19) belongs to beta and is one of the new infectious corona viruses that infects the human body as a pathogen of mass pneumonia that occurred in Wuhan, Hubei, China in December 2019. Common characteristics of people infected with coronavirus are fever, cough, shortness of breath and dyspnea. In more severe cases, it can cause pneumonia, severe acute respiratory syndrome, kidney failure, and even death.

## Principle

LiliF™ GBN COVID-19 Ag Rapid Kit is a lateral flow chromatographic immunoassay for the detection of SARS-CoV-2 specific antigens in human nasopharyngeal swab specimens. It is intended to be used by professionals as an aid in the diagnosis of infection with SARS-CoV-2 coronavirus, which causes COVID-19 disease. After applying the extracted swab specimen onto the hole of testing device, human's sample first react with gold conjugated mouse monoclonal antibody to SARS-CoV-2 Nucleocapsid antibody and rabbit anti-chicken IgY antibody. After that, each reactant diffuses on the membrane to the another monoclonal antibody to SARS-CoV-2 Nucleocapsid antibody marked as test lines and the control line is coated with chicken IgY antibody. If positive, the test line turns to red color because antigen-antibody-gold conjugate complex is formed on the test lines. The control line is intended for procedural control and should always appear if the test procedure is performed correctly and the control line reagents are operated.

## Material Composition

- Chicken IgY antibody
- Rabbit anti-chicken IgY antibody
- Monoclonal antibody to SARS-CoV-2 Nucleocapsid

## Storage & Shelf Life

All components should be stored at 1 ~ 30 ℃ with protection from direct light. The test device(unopened) and sample diluent buffer are stable for 18 months when stored in the recommended condition.

## Kit Contents

| No | Contents | 25 tests/kit |
|---|---|---|
| 1 | Test device | 25 ea/kit |
| 2 | Extraction Buffer tube | 25 ea/kit |
| 3 | Nozzle cap | 25 ea/kit |
| 4 | Flocked swab(Manufactured by a 3rd company) | 25 ea/kit |
| 5 | Instrcution for use | 1 ea/kit |

[Description]

1. **Test device :** There is a circular sample spot on the outside of the plastic cassette. The rectangular display window shows the location of the control line (C) and the test line (T). To block moisture, each device is packaged in one pouch.
2. **Extraction Buffer tube** : Colorless opaque plastic tube sealed with foil material at the top of the tube.
3. **Nozzel cap** : Lid combined with filter for drip purpose
4. **Flocked swab** : The Flocked swap for collecting surface samples near the back of the nasopharyngeal. (Manufactured by a 3rd company)

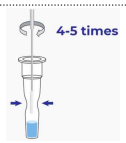
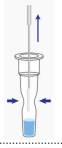
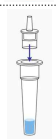
EC REP Intermundien-Lemon Europe GmbH
Brueckstr.47, D-44787 Bochum / Germany
Tel : 49.234.8901.773
Fax : 49.234.8901.774
Mail : info@intermund.com

PMW corp.
19, World cup-daero 243beon-gil, Yeonje-gu, Busan, Rep. of Korea
Tel : +82-51-507-6364,
Fax : +82-51-507-6365
Mail : info@pmwglobal.co.kr

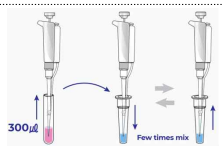
## Procedure

**1. Sample collection and Preparation**

**[Nasopharyngeal swab]**

| | | |
|---|---|---|
| ① | Insert a Flocked swab into the patient's nostril, rotate the swab 4 to 5 times against the nasopharyngeal surface and remove it from the nasal cavity. |   |
| ② | Open the extraction buffer tube and insert the Flocked swab with the cotton part first. Stir the swab at least 5 times while squeezing the buffer tube. |   |
| ③ | Squeeze the side of the tube to remove the swab while extracting the liquid from the swab. |  |
| ④ | Combine the corresponding nozzle cap to the extraction buffer tube accurately. |  |

**[Specimens in transport media]**

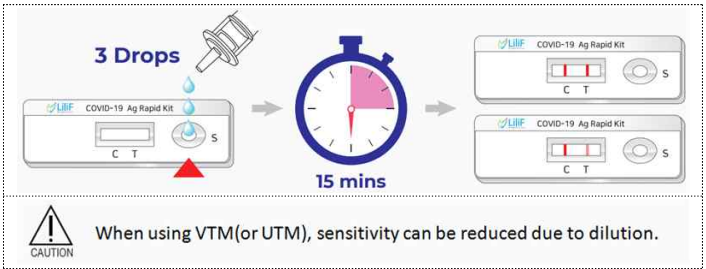
| | | |
|---|---|---|
| ① | Collect the 300µl of specimen from the VTM or UTM. |   |
| ② | Combine the corresponding nozzle cap to the extraction buffer tube accurately. |  |

# LiliF™ GBN COVID-19 Ag Rapid Kit

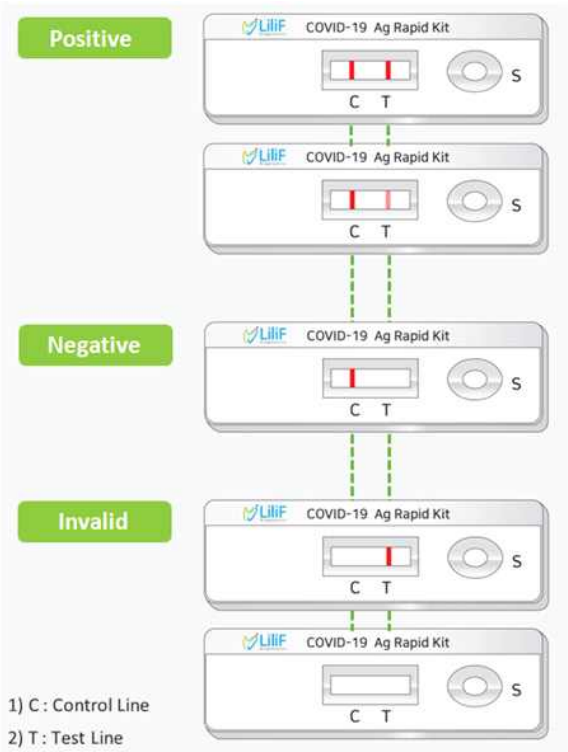
REF IRH41071 Σ 25 1℃ 30℃

## 2. Methods

1) Ensure that specimen and test components are equilibrated to room temperature.
2) After pulling out the testing cassette from the closed pouch, place it on the flat position. And label the device with specimen's ID or number.
3) Apply 3 drops (90 ~ 100ul) of specimen and extraction buffer mixture prepared for sample preparation method directly to the sample pad in the hole and ensure that the specimen and extraction buffer mixture are fully absorbed.
4) Wait for 15 min until the red line appears on the control line. Read and interpret the result at 15 min after applying this diluted sample.
5) Do not read test results after 15 minutes.




## 3. Result Interpretation




1) Positive : A colored band will appear on both the control line(C) and is test line (T) in the detection window.
2) Negative : A colored band only will appear on the control line(C) in the detection window. The meaning of this line is that it is working correctly.
3) Invalid : If a colored band does not appear on the control line(C), this result is interpreted as invalid and test it again. Or if the only T line is shown alone, re-testing is required.

## Performance Specifications

### 1. Limit of Detection (LoD)

The SARS-CoV-2 positive specimen was prepared by Heat-Inactivated SARS-Related Coronavirus 2, Isolate USA-WA1/2020. LoD is determined as 1.0 x $10^2$ $TCID_{50}$/ml by testing serially diluted the Heat-Inactivated positive specimen.

### 2. Cross-Reactivity

For the cross-reactivity of the LIilF™ GBN COVID-19 Ag rapid kit, virus samples of the pathogens listed below were used. This test had no observable cross reactivity with the following virus.

| Potential cross reacting substance | Concentration | Potential cross reacting substance | Concentration |
|---|---|---|---|
| *Coronavirus* OC43 | 9.8 x $10^7$ pfu/ml | Respiratory syncytial virus A | 8.0 x $10^6$ pfu/ml |
| *Coronavirus* 229E | 1.0 x $10^6$ pfu/ml | Respiratory syncytial virus B | 2.4 x $10^7$ pfu/ml |
| Influenza A virus H3N2 | 8.0 x $10^5$ pfu/ml | *Parainfluenza virus* 1 | 2.8 x $10^5$ pfu/ml |
| Influenza A virus H1N1 | 1.2 x $10^6$ pfu/ml | *Parainfluenza virus* 2 | 2.0 x $10^8$ pfu/ml |
| Influenza B virus | 9.8 x $10^8$ pfu/ml | *Parainfluenza virus* 3 | 1.6 x $10^6$ pfu/ml |
| Adenovirus 7 | 6.0 x $10^7$ pfu/ml | *Metapneumovirus* | 1.4 x $10^5$ pfu/ml |

### 3. Evaluation

Performance characteristics for the LiliF™ GBN COVID-19 Ag rapid kit for rapid detection of SARS-CoV-2 antigen was established in retrospective, randomized, single-blinded study conducted at a trial site in KOREA during the 2021 SARS-CoV-2 pandemic situation. A total of 146 retrospective specimens were tested using the LiliF™ GBN COVID-19 Ag rapid kit. These specimens consisted of nasopharyngeal swabs from symptomatic patients. Prior to clinical trials, specimens identified as RT-PCR were used.

| LiliF™ GBN COVID-19 Ag rapid kit | | COVID-19 RT-PCR | | |
|---|---|---|---|---|
| | | Positive | Negative | Total |
| Kit Result | Positive | 44 | 3 | 47 |
| | Negative | 3 | 96 | 99 |
| | Total | 47 | 99 | 146 |
| Sensitivity | | 93.6% (44/47) | | |
| Specificity | | 97.0% (96/99) | | |

## Precautions for handling the product

1. Do not use components beyond the expiry date of this product.
2. Solid and liquid waste from experiments should be discarded after high-pressure autoclaving for 15 min at 121℃.
3. Avoid moisture, direct sunlight, heat and keep it in room temperature.
4. Due to deterioration in quality when testing device is exposed to moisture in storage, pull out the device from the closed pouch right before use and use it within 30 min.
5. If the kit's protective packaging is damaged upon receipt, please contact manufacturer for instructions. Attention should be paid to the "use by" date specified on the pack label and individual tube labels.
6. Dispose of all equipment, including reagents and human samples used or not used in testing, in accordance with local, state, and federal regulations.

# LiliF™ GBN COVID-19 Ag Rapid Kit

REF IRH41071 Σ 25 1℃ 30℃

## Precautions for testing

1. Be careful with handling specimens which may impregnate dangerous sources of unknown viral or bacterial infection. Use a disposable glove when handle the specimen and wash your hands after handling. In case of human exposure, the part shall be immediately cleansed with running tap water and medical attention shall be sought immediately for symptoms including high fever and rashes.
2. Do not touch the membrane in the detection window of this kit by hands directly, or it affects the test result.
3. Do not drink or eat while handling specimen.
4. Take precautions with tall specimen because hey may contain infectious agents.
5. Established precautions against microbiological hazards should be prepared and followed throughout the test.
6. Do not use contaminated, heated or improperly stored(or prepared) specimen for testing.
7. Serious hemolysis or microbial contaminated specimen may cause inaccurate result.
8. This kit cannot exclude the possibility of false positive and false negative results due to various factors, completely. The final diagnosis should not be determined solely by the kit, and should be combined with clinical observation, patient history, and epidemiological information.

## Quality control

Red lines should appear on the control line for each testing.

## Traceability of the calibration

**1. Appearance test**

| No. | Test Standard |
|---|---|
| 1 | Check that there is a circular specimen spot on the outside of the plastic device, and the position of the control line (C) and the inspection line (T) are marked on the oval display window. |
| 2 | Check that the unfolded state is evenly raised and that the thickness and thickness of the band are appropriate. |
| 3 | Check the kit's Lot No. and the expiration date. |

| Test Method |
|---|
| Visually check whether it meets the inspection criteria. |

**2. Efficacy test (Positive)**

| Test Standard |
|---|
| All the results should be positive after repeating the test three times using a standard product. |

| No. | Test Method |
|---|---|
| 1 | Put the standard product at room temperature (1~30℃) before starting the test, and then mix it homogeneously (voltexing). |
| 2 | Immediately before the inspection, open the silver foil, take out the inspection device, and place it on a flat surface. |
| 3 | Put 10 µl of the standard product on the drop site using a micropipette, and drop 100 µl of the sample extract using a macro pipette. |
| 4 | Check whether it meets the test standards 15 minutes from the start of the inspection.t. |
| 5 | Results that appear 15 minutes after the start of the inspection are not included in the judgment. |

**3. Efficacy test (Negative)**

| Test Standard |
|---|
| All results of repeated tests 3 times using a negative sample (sample extract) should be negative. |

| No. | Test Method |
|---|---|
| 1 | Place the negative sample (sample extract) at room temperature (1~30℃) before starting the test, and then mix it homogeneously (voltexing). |
| 2 | Immediately before the inspection, open the silver foil, take out the inspection device, and place it on a flat surface. |
| 3 | Use a micropipette to drop 100 µl of a negative sample (sample extract) onto the drop site. |
| 4 | Conforms to the test standard 15 minutes from the start of the inspection |
| 5 | Results that appear 15 minutes after the start of the inspection are not included in the judgment. |

## Limitation of procedure

1. The test procedures, precautions, and interpretation of the results of this test shall be strictly followed during the test.
2. This test should be used to detect SARS-CoV-2 antigens in human nasopharyngeal swab specimens.
3. Failure to correctly follow the test procedures and interpretation of the test results may have a negative effect on the test performance or produce invalid results.
4. Negative results may occur if the concentration of antigen in the specimen is below the detection limit of the test or if the specimen is improperly collected or transferred, so the negative results do not eliminate the possibility of SARS-CoV-2 infection and should be identified by further analysis.
5. A positive test does not preclude co-infection with other pathogens.
6. When using VTM(UTM), the sensitivity may be reduced due to dilution.
7. If the result of the test line is positive, it means antigen positive. This kit should only be used as reaserch use only (RUO) purposes. In the case of positive results, confirmation tests through alternative diagnostic methods are required. After analyzing these test results and other clinical data, the Principal Investigator should perform a final diagnosis.

## Storage and Expiration Date

| Contents | Condition | Storage | Shelf life | note |
|---|---|---|---|---|
| Test device | unopened | 1 ~ 30℃ | 18 months | - |
| | Opened | 1 ~ 30℃ | 1 hour | - |
| Extraction Buffer tube | unopened | 1 ~ 30℃ | 18 months | - |

## PACKAGING UNIT

| Cat. No. | Name | Package |
|---|---|---|
| IRH41071 | LiliF™ GBN COVID-19 Ag Rapid Kit | 25 tests/kit |

# LiliF™ GBN COVID-19 Ag Rapid Kit

REF IRH41071 Σ 25 1℃ 30℃

## Symbols

| Symbol | Description | Symbol | Description |
|---|---|---|---|
| CE | CE marking | IVD | In vitro diagnostic Medical device |
| | Use by<br>Synonym for this:<br>Expiry Date | LOT | Batch Number |
| REF | Catalogue Number | | Attention.<br>See Instruction for use |
| | Storage Temperature Limitation | | Keep Away From Sunlight |
| | Manufactured by | | Manufacturing Date |
| EC REP | Authorized Representative in the European Community | Σ | Contains sufficient for Tests |
| | Consult Instructions For Use | | Do not reuse |

EC REP **Obelis s.a**
Bd. Général Wahis 53 B-1030 Brussels, Belgium
Tel : 32.2.732.59.54
Fax : 32.2.732.60.03
Mail : mail@obelis.net

**iNtRON Biotechnology, Inc.**
#604, Sangdaewon-dong, Jungang Induspia, 137, Sagimakgol-ro, Jungwon-gu, Seongnam-si, Gyeonggi-do, 13202, Korea
Tel : +82-31-739-5062,
Fax : +82-31-739-5264
Mail : intronbio@intronbio.com

Revised date : 11, May, 2021