Novel Coronavirus (2019-nCoV) IgM/IgG Antibody Combo test Kit (Colloidal Gold)

Intended Use

The Novel Coronavirus (2019-nCoV) IgM/IgG Antibody Combo test Kit (Colloidal Gold) is intended for the qualitative detection of IgM/IgG antibodies against novel coronavirus in human serum, plasma or whole blood from patients with clinical suspicion of Novel coronavirus (2019-nCoV) infection.

For *in vitro diagnostic use.*

Introduction

In the last of 2019, Novel coronavirus (2019-nCoV) was outbreak. Then the virus was characterized by genome sequencing. The latent period is about 1 to 14 days, the average is 5 days. Most of them are asymptomatic after infection. All of them are infectiousness. Therefore, early detection of 2019-nCoV infection is extremely important.

Polymerase Chain Reaction (PCR) assays have been developed and used for suspicious patients. PCR has been well accepted and is in widespread use for the detection. But there are only 30%~50% detection rate in the 2019-nCoV infected patients. This time IgM assay will be a good supplement.

IgM antibodies become detectable in a few days after the infection and reach peak levels at 10 - 14 days. These antibodies persist but rapidly diminish in concentration over the next 12 weeks until the antibody is no longer clinically detectable. Production of IgG increases rapidly for the next 7 to 30 days, then level off or even decrease in strength. IgG antibodies remain present at least 1 year. The presence of IgM antibody in a single specimen suggests that the patient has recently experienced a 2019-nCoV infection. In most cases the infection probably occurred within the preceding month. Some may continue to produce specific IgM for over 3 months.

Principle of the Assay

This product is adopted the immune colloidal gold technique to detect IgM/IgG antibodies against the novel coronavirus. The reagent binding pad is coated with colloidal gold labeled mouse anti-human IgM/IgG monoclonal antibodies and rabbit IgG antibodies, the test area of nitrocellulose membrane is coated with recombination expressed novel coronavirus antigen protein and the quality control area of nitrocellulose membrane is coated with goat anti-rabbit antibodies. When testing, IgM/IgG antibodies (specific IgM/IgG antibodies against novel coronavirus and non-specific IgM/IgG antibodies) in the specimens to be tested combine with the colloidal gold labeled mouse anti-human IgM/IgG monoclonal antibodies to form immune complexes. As a result of chromatography, immune complexes move along the membrane. If the specimens to be tested contain specific IgM/IgG antibodies against novel coronavirus, the specific IgM/IgG antibodies will be captured by the antigen

proteins of novel coronavirus coated in the test area to form a visible strip (T line) in the test area, the free colloidal gold marker or immune complexes continue to move forward and specifically bind to the goat anti-rabbit antibody coated in the quality control area to form a visible strip (C line) in the quality control area. Otherwise, no test line will appear, only quality control line(C line) will appear.

Kit Presentation

Materials Supplied

Test devices – 20 test cards containing immobilized novel coronavirus antigen proteins and goat anti-rabbit antibodies.

Dropper: 20 pcs.

Specimen diluent buffer: 1 bottles for 20 pcs/box;

Note: The **Specimen diluent buffer** cannot used mix lots.

Materials Required But Not Provided

1. Timer
2. Specimen collection container

Storage and Stability

1. Preserve in a dry place at 2-30 ℃, protected from light. The validity is tentatively 6 months.
2. In general, the kit shall be used within 30 minutes after the foil bag is opened. If the temperature is higher than 30 ℃ or the humidity of the environment is higher than 70 %, the kit shall be used as soon as possible after opening the aluminum foil bag.
3. The production date and expiry date please see in the labels on the box of kits.

Precautions

• This test is designed for in vitro diagnostic use only.

• For professional use only.

• Material should not be pipetted by mouth.

• Treat all materials as if they were infectious and dispose of all material in accordance with local regulation. Liquid waste should be disposed of in a 1%

sodium hypochlorite solution or in accordance with local requirements for disposal of infectious material.

• Liquid solutions in this kit contain Sodium Azide at a concentration less than 0.1%, these solutions should be handled with care and when disposed down the drain they should be flushed thoroughly with water.

• Do not use the kit beyond the expiration date.

• The test card is sealed in a protective foil pouch. Do not use if pouch is damaged or open. Remove test card from pouch just prior to use. Do not touch the reaction area of test card.

• Do not use damaged cards. Do not mix components from different kits.

• Use the disposable pipette, tube and card provided for each specimen tested.

• Do not re-use.

Specimen Collection and Storage

1. Handle all blood and serum as if capable of transmitting infectious agents.

2. Optimal performance of the kit depends upon the use of fresh Plasma, serum or whole blood samples (clear, non-hemolyzed, non-lipemic, non-icteric). A minimum volume of 50 μL is recommended, in case repeat testing is required. Specimens should be collected aseptically by venipuncture.

3. Store samples between 2° and 8° C if testing will take place within two days. If specimens are to be kept for longer periods, store at -20° C or colder(except whole blood). The whole blood sample is recommended to be tested within 3 days, stored at 2 - 8 ℃ and not frozen. Do not use a frost-free freezer because it may allow the specimens to go through freeze-thaw cycles and degrade antibody. Samples that are improperly stored or are subjected to multiple freeze-thaw cycles may yield erroneous results.

4. The NCCLS provides recommendations for storing blood specimens (Approved Standard Procedures for the Handling and Processing of Blood Specimens, H18-A. 1990).15

Quality Control

A built-in procedural control on the card ensures that the test has been performed correctly; this pink/red colored line should always appear above the printed **C** line on the card. If a line does not appear in the control region discard the card as this is an invalid test and perform the test again.

It is recommended that the positive and negative controls should be run with each new kit lot number or as required by your laboratory QA standard operating procedures. If the controls do not read as expected, repeat the test. Contact your local technical support if the QC results continue to be invalid.

Test Procedure

1. Preparing

a) The specimens to be tested and the required reagents shall be removed from the storage condition and balance to room temperature;

b) The kit shall be remove from the packaging bag and placed flat on a dry desk.

2. Testing

a) Add specimen

   Serum/Plasma: Take 10 μl of specimen of serum or plasma to sample well (S), add vertically 2 drops (about 100 μl) of specimen diluent.

   Whole blood: Take 20 μl of whole blood to sample well(S), add vertically 2 drops (about 100 μl) of specimen diluent.

b) The positive specimens can be detected within 10minutes after sample addition. Relevant verification shows that the observation of the test results will be affected if the reaction time were exceed 15 minutes (calculated from after sample addition), so it is recommended to read and record the test results within 10 minutes

Interpretation of Results

POSITIVE   NEGATIVE

INVALID
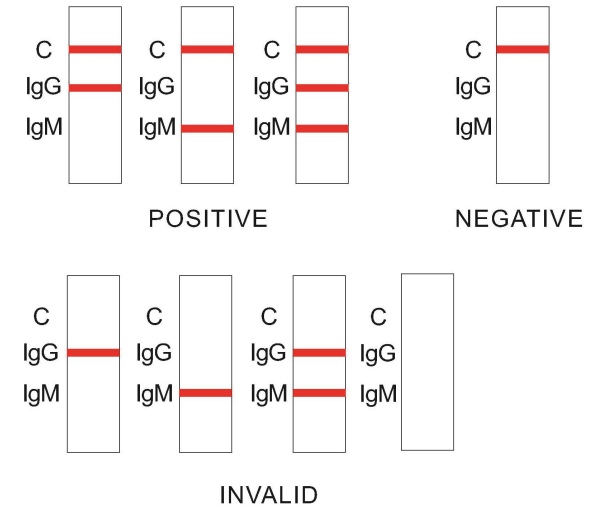
1. **IgG POSITIVE**:*Two lines appear. One colored line should be in the control line region (C),and a colored line appears in the IgG test line region. The result is positive   for IgG antibodies against the novel coronavirus.

2. **IgM POSITIVE**:*Two lines appear. One colored line should be in the control line region (C),and a colored line appears in the IgM test line region. The result is positive   for IgM antibodies against the novel coronavirus.

3. **IgG and IgM POSITIVE**:*Three lines appear. One colored line should be in the control line region (C), and two coloured lines should appear in IgG test line region and IgM test line region.

*NOTE: The intensity of the color in the test line regions will vary depending on the antibodies against the novel coronavirus present in the specimen. Therefore, any shade of color in the test line region should be considered positive.

4. **NEGATIVE**: One colored line appears in the control region (C). No apparent colored line appears in the IgG or IgM test region (T).

5. **INVALID**: Control line fails to appear. Insufficient sample volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the test with a new test cassette. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

Limitations

1. This product is only used for testing of individual serum, plasma or whole blood samples.

2. A negative result does not rule out the possibility of novel coronavirus infection.

3. A positive result does not means novel coronavirus infection. Since the IgM will last 3 months, and the IgG will last at least 1 year no matter the patients are cured or not.

4. The test results of this product are for clinical reference only and shall not be taken as the sole basis for clinical diagnosis and treatment. The clinical management of patients shall be considered in combination with their symptoms, signs, medical history, other laboratory tests (especially pathogen detection), treatment response, epidemiology and other information.

5. Serological antibody testing is of limited reference value in patients with impaired immune function or receiving immunosuppressive therapy.

6. Positive IgM antibodies occurs not only in primary infection but also in secondary infection.

7. The target detection object of this product is IgM/IgG antibodies of novel coronavirus, which does not directly reflect the presence of novel coronavirus in the specimen.

Performance Characteristics

Clinical Sensitivity and Specificity

The sensitivity performance of the Novel coronavirus (2019-nCoV) IgM/IgG Antibody test was evaluated at three different sites using both under treatment (97 samples) and convalescent samples (114 samples). All of the samples were from the patients between 7 to 90 days after the infection.

The specificity performance of the LYHER® Novel coronavirus (2019-nCoV) IgM/IgG Antibody test was evaluated at four different sites using 200 retrospective (frozen) and 192 prospective (fresh) negative samples. 282 where from patients with symptoms of pneumonia from a non-2019-nCoV, 110 from patients suffering other respiratory tract infections.

The results are stated below:

Serum, plasma and whole blood samples (n=604)		Clinical Diagnosis	
		Positive	Negative
2019-nCoV IgM	Positive	202	3
	Negative	9	389

2019-nCoV IgM total Sensitivity= 95.73% (95%CI 92.06−98.03)

Total Specificity = 99.23% (95%CI 97.78 – 99.84)

Total conformity =98.01% (95%CI 95.03 – 99.76)

Serum, plasma and whole blood samples (n=571*)		Clinical Diagnosis	
		Positive	Negative
LYHER® 2019-nCoV IgG	Positive	175	3
	Negative	3	389

2019-nCoV IgG total Sensitivity= 98.31% (95%CI 95.15 −99.65)

Total Specificity = 99.23% (95%CI 97.78 – 99.84)

Total conformity =98.95% (95%CI 96.21 – 99.73)

Note- Because the IgG level become detectable were 10 days later after the infection, we removed the 33 samples that collected within 10 days after the infection.

CROSS-REACTIVITY

A series of 63 samples confirmed positive for the different antinuclear antibodies (ANA),

rheumatoid factor (RF), and other common viruses, such as Influenza (H1N1, H3N2, H5N1, H7N9, Yamagata, Victoria), RSV, RUB, CMV, HSV, VZV, HIV, EBV, adenovirus, rotavirus, Mumps, enterovirus, and measles) , and Legionella were obtained from outside clinical laboratories. All samples were negative to 2019-nCoV antibodies. These sera were then run in the 2019-nCoV IgM/IgG test. The results of this study indicate that the 2019-nCoV IgM/IgG test contains no cross-reacting proteins to other common viruses, Legionella, ANA or rheumatoid factor.

References

1. World Health Organization (WHO). WHO Statement Regarding Cluster of Pneumonia Cases in Wuhan, China. Beijing: WHO; 9 Jan 2020. [Accessed 26 Jan 2020].

2. Weiss SR, Leibowitz JL. Coronavirus pathogenesis. Adv Virus Res 2011;81:85-164.

3. Su S, Wong G, Shi W, et al. Epidemiology, genetic recombination, and pathogenesis of coronaviruses. Trends Microbiol 2016;24:490-502.

4. Cui J, Li F, Shi ZL. Origin and evolution of pathogenic coronaviruses. Nat Rev Microbiol 2019;17:181-192.

5. https://www.who.int/health-topics/coronaviru

6. Diagnosis and Treatment plan of Corona Virus Disease 2019 (Tentative seventh edition)

http://www.nhc.gov.cn/yzygj/s7653p/202003/46c9294a7dfe4cef80dc7f5912e b1989.shtml

ORDERING INFORMATION

Item ： Novel Coronavirus (2019-nCoV) IgM/IgG Antibody Combo test Kit (Colloidal Gold)

Specimen：Whole blood/Serum/Plasma

Format: Device

Guide to Symbols

Consult instructions for use

Temperature limit

REF — Catalogue number

IVD — *In vitro diagnostic medical device*

Manufacturer

LOT — Batch code

Use–by date
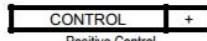
0.00% NaN₃ — Sodium Azide

Caution

GHS06 Acute Toxicity

19nCoV-GⅡ — Novel Coronavirus (2019-nCoV) IgM/IgG

eIFU indicator

CONTROL + — Positive Control

EXT BUF — Extraction Buffer
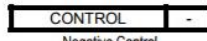
CONTROL - — Negative Control
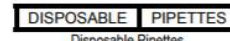
DISPOSABLE PIPETTES — Disposable Pipettes
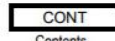
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