Infection ichroma™ COVID-19 Ag

INTENDED USE

ichroma™ COVID-19 Ag is a fluorescence Immunoassay (FIA) for the qualitative detection of novel corona virus (eg, SARS-CoV-2, 2019-nCoV) in human nasopharyngeal swab. It is helpful as an aid in the screening of early mild, asymptomatic, or acute patients for identification of 'Novel Coronavirus' infection.

For *in vitro* diagnostic use only.

INTRODUCTION

The third zoonotic human coronavirus (CoV) of the century emerged in December 2019, with a cluster of patients connected to Wuhan, Hubei Province, China. This virus, the newly identified coronavirus 2019 nCOV, could cause risky pneumonia so that prevention and control of the infection has become highly required. The 2019-nCoV is a member of the Betacoronavirus Genus, that also includes Severe Acute Respiratory Syndrome coronavirus (SARS-CoV) and Middle East Respiratory Syndrome coronavirus (MERS-CoV). Since it is identified that symptoms become rapidly severe without a proper treatment after onset of illness, early diagnosis of the virus infection is quite crucial. Currently, the spread of the viral transmission become fast so that the prevention of local transmission requires a point-of care test (POCT), which shows quick outcome within 20 minutes.

ichroma™ COVID-19 Ag is an *in vitro* diagnostic medical device that helps you to diagnose novel coronavirus infections by detecting the specific antigen of SARS-CoV-2.

PRINCIPLE

This test uses a sandwich immunodetection method. The detector antibodies in buffer bind to antigens in the sample, forming antigen-antibody complexes, and migrate onto nitrocellulose matrix to be captured by the other immobilized streptavidin on a test strip.

More antigens in the sample will form more antigen-antibody complexes which lead to stronger fluorescence signal by detector antibodies, which is processed by instrument for ichroma™ tests to display the SARS-CoV-2 antigens 'Positive'/ 'Negative' in the sample.

COMPONENTS

ichroma™ COVID-19 Ag consists of 'cartridges', 'detector tubes' and 'extraction tube sets'.

- The cartridge contains the membrane called a test strip which has streptavidin at the test line, and chicken IgY at the control line. All cartridges are individually sealed in an aluminum foil pouch containing a desiccant in a box.
- The detector tube has 2 granules containing anti-COVID19 fluorescence conjugate, anti-COVID19-Biotin conjugate, anti-chicken IgY-fluorescence conjugate, bovine serum albumin (BSA) and sucrose as a stabilizer and sodium azide as a preservative in Tris-HCl buffer.
- The extraction buffer tube contains sodium chloride, sodium azide as a preservative in Tris-HCl buffer.

WARNINGS AND PRECAUTIONS

- For *in vitro* diagnostic use only.
- Follow the instructions and procedures described in this 'Instructions for use'.
- Avoid direct sunlight.
- Do not reuse cartridge, detector tube and extraction tube set. They should be used for testing one sample only.
- Lot numbers of all the test components (cartridge, detector tube, extraction tube set and ID chip) must match each other.
- Do not interchange the test components between different lots or use the test components after the expiration date, either of which might yield incorrect test result(s).
- The cartridge should remain sealed in its original pouch until just before use. Do not use cartridge, if pouch is damaged or has already been opened.
- If test components and/or sample are stored in refrigerator, then allow cartridge, detector tube, detector diluent and sample to be at room temperature for approximately 30 minutes before use.
- An exposure to larger quantities of sodium azide may cause specific health issues like convulsions, low blood pressure and heart rate, loss of consciousness, lung injury and respiratory failure.
- The instrument for ichroma™ tests may generate slight vibration during use.
- Used cartridges, detectors, extraction tubes, nozzles and swabs should be handled carefully and discarded by an appropriate measure in accordance with the relevant local regulations.
- The detector tube and the extraction buffer contain sodium azide (NaN_3), and they may cause certain health issues like convulsions, low blood pressure and heart rate, loss of consciousness, lung injury and respiratory failure. Avoid contact with skin, eyes, and clothing. In case of contact, rinse immediately with running water.
- **ichroma™ COVID-19 Ag** will provide accurate and reliable results when it is used only in conjunction with the instrument for ichroma™ tests.

WARNINGS AND PRECAUTIONS FOR SAMPLE

- Use the fresh samples.
- It is recommended to test the sample immediately after sample collection.
- Refrain from smoking or eating, while sample is collected.
- Do not collect samples outside of the nasopharynx. In any cases, pre-education for user is required for the proper sample collection.
- Please use fresh swab to avoid the cross-reactivity between samples. Never reuse the sterile swab.
- The improper samples such as those from an individual who has recently taken any interfering medicine or samples mistakenly mixed up with different patients shall cause inaccurate test results.

LIMITATION OF THE TEST SYSTEM

- The test may yield false positive result(s) due to the cross-reactions and/or non-specific adhesion of certain sample components to the capture/detector antibodies.
- The test may yield false negative result(s) due to the non-responsiveness of the antigen to the antibodies which is most common if the epitope is masked by some unknown components, so therefore not being able to be detected or captured by the antibodies. The instability or degradation of the antigen with time and/or temperature may also cause false negative result as it makes antigen unrecognizable by the antibodies.
- Other factors may interfere with the test and cause erroneous results, such as technical/procedural errors, degradation of the test components/reagents or presence of interfering substances in the test samples.
- If the test result is Negative even though the patient has significant infectious symptoms, it should be recommended to conduct additional test including PCR or culture test.
- The accurate determination of test result as Positive should be confirmed by additional clinical evaluation.
- Negative result should be considered with possibilities of other infections. Positive result should be considered with additional infections by another pathogenic bacterium.
- If the product has positive results, any clinical diagnosis based on the test result must be supported by a comprehensive judgment of the concerned physician including clinical symptoms and other relevant test results.
- In case of antigen concentration is low, the test may yield false negative results. Therefore, the negative results cannot exclude the possibility of infection completely.
- This product is only to detect the presence of a SARS-CoV-2 antigen.

STORAGE AND STABILITY

Storage condition			
Component	Storage Temperature	Shelf life	Note
Cartridge	2 - 30 °C	20 months	Disposable
Detector tube	2 - 30 °C	20 months	Disposable
Extraction buffer tube	2 - 30 °C	20 months	Disposable

- After the cartridge pouch is opened, the test should be performed immediately.

MATERIALS SUPPLIED

REF CFPC-115

Components of **ichroma™ COVID-19 Ag**

- Cartridge Box:
  - Cartridge 25
  - Detector tube 25
  - Extraction tube set
    - Extraction buffer tube 25
    - Nozzle 25
  - ID chip 1
  - Instructions for use 1

MATERIALS REQUIRED BUT SUPPLIED ON DEMAND

Following items can be purchased separately from **ichroma™ COVID-19 Ag.**

Please contact our sales division for more information.

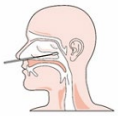
- Instrument for ichroma™ tests
  - **ichroma™ II** REF FPRR021
  - **ichroma™ 50** REF FPRR022
  - **ichroma™ M2** REF FPRR031
  - **ichroma™ III** REF FPRR037
  - **ichroma™ 50 PLUS** REF FPRR036
- **Boditech COVID-19 Ag control** REF CFPO-293
- **Boditech COVID-19 Ag Control Swab** REF CFPO-354

SAMPLE COLLECTION AND PROCESSING

The sample type for **ichroma™ COVID-19 Ag** is human nasopharyngeal swab.

- **Collection method for sample**

  To collect samples, insert a sterile swab in the nasal cavity and spin it smoothly in the nasopharynx.



< Nasopharyngeal swab>

- It is recommended to test the sample immediately after collection. If do not use sample immediately, it should be stored at 2-8 °C.
- Samples stored at 2-8 °C for 3 days showed no performance difference.
- It is highly recommended that **ichroma™ COVID-19 Ag** test would be performed on the nasopharyngeal swab specimens directly collected from patients with provided extraction buffer.

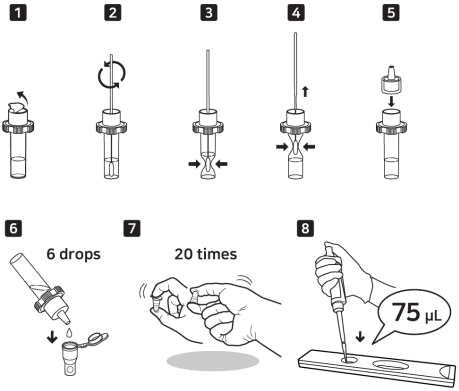
TEST SETUP

- Check the contents of **ichroma™ COVID-19 Ag**: cartridges, detector tubes, Extraction tube sets, ID chip and Instructions for use.
- Ensure that the lot number of the cartridge matches that of the detector tube and the extraction buffer as well as an ID Chip.
- If the sealed cartridge, the detector tube and the extraction buffer have been stored in a refrigerator, place them on a clean and flat surface at room temperature for at least 30 minutes before testing.
- Turn on the instrument for ichroma™ tests.
- Insert the ID chip into the 'ID chip port'.

  (Please refer to the operation manual of instrument for ichroma™ tests for complete information and operating instructions.)

TEST PROCEDURE

- **ichroma™ II**



<Single mode>

① Open the lid of the aluminum foil of the extraction buffer tube.
② Collect samples with a sterile swab and then put it into the extraction buffer tube. Spin the sterile swab 5 times and squeeze the sterile swab to extract the sample into the buffer.
③ Squeeze the bottom to extract the sample into the buffer and start pushing the swab to the top.
④ Continue squeezing and pushing the swab to the top of extraction buffer tube to pull it out of tube.
⑤ Assemble a nozzle to the top of the extraction buffer tube.
⑥ Load six drops of sample mixture onto the detector tube.
⑦ Close the lid of the detector tube and mix the sample thoroughly by shaking it about 20 times. The sample mixture must be used within 30 seconds.
⑧ Pipette out 75 μL of a sample mixture and load it into the sample well on the cartridge.
⑨ Insert the sample-loaded cartridge into the instrument for ichroma™ test
⑩ Tap the "Start" button on the instrument for ichroma™ tests.
⑪ Cartridge goes inside the Instrument for ichroma™ tests and will automatically start scanning the sample-loaded cartridge after 12 minutes.
⑫ Read the test result on the display screen of the instrument for ichroma™ tests.

<Multi mode>

① The test procedure is same with "ichroma™ II Single test mode ① – ⑧"
② Leave the Cartridge at room temperature for 12 minutes before inserting the cartridge into the holder.
⚠ *Scan the sample-loaded cartridge immediately when the incubation time is over. If not, it will cause inaccurate test result.*
③ To scan the sample-loaded cartridge, insert it into the cartridge holder of the instrument for ichroma™ tests. Ensure proper orientation of the cartridge before pushing it all the way inside the cartridge holder. An arrow is marked on the cartridge especially for this purpose.
④ Tap the "Start" button on the instrument for ichroma™ tests.
⑤ The Instrument for ichroma™ tests will start scanning the sample-loaded cartridge immediately.
⑥ Read the test result on the display screen of the instrument for ichroma™ tests.

※ It is highly recommended that **ichroma™ COVID-19 Ag** test would be performed on the nasopharyngeal swab specimens directly collected from patients with provided extraction buffer.

▪ **ichroma™ 50, ichroma™ 50 PLUS**

① Insert the tip array in the tip station.
② Insert the cartridges in the cartridge magazine individually.
(The cartridges of ichroma 50 plus are packaged in a magazine. Insert the magazine in the magazine station.)
③ Insert the detector tube in the reagent station.
④ The test procedure is same with "ichroma™ II Single test mode ① – ⑤"
⑤ Put all sample mixture onto the test tube.
⑥ Insert the test tube including the extraction buffer into the tube rack.
⑦ Set the number of the test cartridge by tapping.
⑧ Set the number of pipette tips by tapping.
⑨ Tap the 'START' button on the left upper of the main screen to start test.

▪ **ichroma™ M2**

< Read Now mode >

① Check the display "Read Now" on the ichroma™ M2 screen and set the sample type.
② The test procedure is same with "ichroma™ II Single test mode ① – ⑧".
③ Leave the cartridge at room temperature for 12 minutes before inserting the cartridge into the cartridge holder of ichroma™ M2.
⚠ *Scan the sample loaded cartridge immediately when the incubation time is over. If not, it will cause inexact test result.*
④ To scan the sample-loaded cartridge, insert it into the cartridge holder of the instrument for ichroma™ tests. Ensure proper orientation of the cartridge before pushing it all the way inside the cartridge holder. An arrow is marked on the cartridge especially for this purpose.
⑤ The instrument will automatically start scanning the cartridge. Do not remove the cartridge or touch the reader during scanning.
⑥ Read the test result on the display screen of the instrument.
⑦ When the cartridge is removed from cartridge holder, the display will show "Read Now" as a standby state.

< Walk Away mode >

① Check the display "Walk Away" on the ichroma™ M2 screen and set the sample type.
② The test procedure is same with "ichroma™ II Single test mode ① – ⑧".
③ After loading the sample mixture, insert the mixture

loaded cartridge into the holder. Ensure proper orientation of the cartridge before pushing it all the way inside the cartridge holder. An arrow is marked on the cartridge especially for this purpose.

④ The instrument will automatically start scanning the cartridge after reaction time. When the cartridge is inserted, reaction time is displayed.

⑤ Read the test result on the display screen of the instrument.

⑥ When the cartridge is removed from cartridge holder, the display will show "Walk Away" as a standby state.

▪ **ichroma™ III**

① The test procedure is same with "ichroma™ II Single test mode ①–⑧".

② Insert the sample-loaded cartridge into the ichroma™ III.

③ Tap the "Start" button on the ichroma™ III.

④ Cartridge goes inside and the ichroma™ III will automatically start scanning the sample-loaded cartridge after 12 minutes.

⑤ Read the test result on the display screen of the ichroma™ III.

INTERPRETATION OF TEST RESULT

- The instrument for ichroma™ tests calculates the test result automatically and displays 'Positive' or 'Negative'.

Display	Interpretation
Positive	Positive test for SARS-CoV-2 (SARS-CoV-2 antigen present)
Negative	Negative test for SARS-CoV-2 (No SARS-CoV-2 antigen detected)
Invalid	Result invalid. Need to retest.

- This product cannot be used as a tool for confirmation. The false positive and false negative results can be caused by various causes.
- Any clinical diagnosis based on the test result must be supported by a comprehensive judgment of the concerned physician including clinical symptoms and other relevant test results.

QUALITY CONTROL

- Quality control tests are a part of the good testing practice to confirm the expected results and validity of the assay and should be performed at regular intervals.
- The control tests should be performed immediately after opening a new test lot to ensure the test performance is not altered.
- Quality control tests should also be performed whenever there is any question concerning the validity of the test results.
- Control materials are provided on demand. For more information regarding obtaining the control materials, contact Boditech Med Inc.'s Sales Division for assistance. (Please refer to the instruction for use of control material.)

PERFORMANCE CHARACTERISTICS

- **Analytical sensitivity**

The Limit of Detection (LOD) of ichroma™ COVID-19 Ag was determined using of Heat-inactivated SARS-CoV-2 (USA-WA1/2020). LoD is determined as 35.15 $TCID_{50}$/ml through 20 repetitive tests for 3 days.

- **Analytical specificity**

- **Cross-reactivity**

ichroma™ COVID-19 Ag test results did not show any significant cross-reactivity with these 33 various respiratory viruses and bacteria except Recombinant SARS-CoV NP.

No.	Cross-reactivity materials	Conc.
1	*Corona virus – NL63*	1.7 X 10^5 $TCID_{50}$/ml
2	*Corona virus – 229E*	4.17 X 10^5 $TCID_{50}$/ml
3	*Corona virus – OC43*	5.01 X 10^5 $TCID_{50}$/ml
4	*MERS-CoV*	1.7 X 10^5 $TCID_{50}$/ml
5	*Influenza A virus H3N2 Hongkong*	1.4 x 10^6 PFU/ml
6	*Influenza B virus B/Lee/40*	7.6 x 10^6 PFU/ml
7	*Respiratory Syncytial virus A*	8.5 x 10^5 PFU/ml
8	*Respiratory Syncytial virus B*	1.65 x 10^3 PFU/ml
9	*Adenovirus type1*	1 X $10^{5.8}$ $TCID_{50}$/ml
10	*Adenovirus type2*	1 X $10^{6.8}$ $TCID_{50}$/ml
11	*Adenovirus type3*	1 X $10^{4.3}$ $TCID_{50}$/ml
12	*Adenovirus type4*	1 X $10^{3.8}$ $TCID_{50}$/ml
13	*Adenovirus type6*	1 X $10^{6.8}$ $TCID_{50}$/ml
14	*Adenovirus type7*	1 X $10^{4.6}$ $TCID_{50}$/ml
15	*Coxsackievirus A2*	1 X 10^8 $TCID_{50}$/ml
16	*Coxsackievirus A4*	1 X $10^{5.7}$ $TCID_{50}$/ml
17	*Coxsackievirus B1 - conn5*	8.7 X 10^{-7} $CCID_{50}$/ml
18	*Coxsackievirus B3 – nancy (5A1)*	2.3 X 10^{-8} $CCID_{50}$/ml
19	*Rhinovirus - RV21*	5.6 X 10^{-4} $CCID_{50}$/ml
20	*Rhinovirus -RV14*	3.7 X 10^{-6} $CCID_{50}$/ml
21	*Rhinovirus -RV71*	1.1 X 10^{-4} $CCID_{50}$/ml
22	*Human Metapneumovirus type A1*	9 x 10^5 PFU/ml
23	*Parainfluenza virus Type 1*	5.01 X 10^5 $TCID_{50}$/mL
24	*Parainfluenza virus Type 2*	1.51 X 10^6 $TCID_{50}$/mL
25	*Parainfluenza virus Type 3*	4.57 X 10^6 $TCID_{50}$/mL
26	*Parainfluenza virus Type 4B*	1.70 X 10^5 $TCID_{50}$/mL
27	*Legionella spp*	6 x 10^8 CFU/mL
28	*Mycobacterium tuberculosis*	6 x 10^8 CFU/mL
29	*Mycoplasma pneumoniae*	5.62 x 10^7 CFU/mL
30	*Streptococcus pneumoniae*	6 x 10^8 CFU/mL
31	*Streptococcus pyogenes*	6 x 10^8 CFU/mL
32	*Recombinant SARS-CoV NP*	200 ng/mL
33	*Recombinant HCoV-HKU1 NP*	0.44 mg/mL
34	*Recombinant human bocavirus protein*	0.6 mg/mL

- **Interference**

ichroma™ COVID-19 Ag test results did not show any significant interference with these interference materials except biotin.

No.	Interference materials	Conc.
1	Nasal sprays drop (Mometason Furoate)	0.00096 mol/L
2	Nasal corticosteroids (Fluticasone)	0.00011 mol/L
3	Homeopathic allergy relief medicine (Cetrizine)	0.012 mol/L
4	Mouth wash (Listerine)	5 mg/ml
5	Throat lozenges, oral anesthetic & analgesic (Dextromethorphan)	0.01845 mol/L
6	Antiviral drugs (Tamiflu; Oseltamivir)	0.01603 mol/L
7	Antibiotic nasal ointment (Bactroban; mupirocin)	5 mg/ml
8	Whole blood	1% (v/v)
9	Analgesic (Acetaminophen)	0.06623 mol/L
10	Analgesic (Ibuprofen)	0.04854 mol/L

11	Povidone-iodine	1% (v/v)
12	Acetylsalicylic acid (Aspirin)	0.1111 mol/L
13	Antibacterial (cefadroxil)	0.01377 mol/L
14	Mucin (Porcine stomach)	0.5% (v/v)
15	Throat lozenge (VICKS; cetylpyridinium chloride)	20 mg/ml
16	Throat lozenge (dipotassium glycyrrhizinate)	20 mg/ml
17	Throat lozenge (Nandina extraction)	20 mg/ml
18	Biotin	200 ng/mL

■ **Precision**

- Between days
  One person tested one lot of **ichroma™ COVID-19 Ag** during five days, six times at each concentration of the control standard.
- Between lots
  One person tested three different lots of **ichroma™ COVID-19 Ag**, during five days, four times at each concentration of the control standard.
- Between persons
  Three different persons tested one lot of **ichroma™ COVID-19 Ag**, during five days, four times at each concentration of the control standard.
- Between sites
  One person tested one lot of **ichroma™ COVID-19 Ag** at three different site, during five days, four times at each concentration of the control standard.

Standard material	Between day		Between lot	
	Positive / No.	Positive rate	Positive / No.	Positive rate
Negative	0/30	0%	0/60	0%
Low positive	30/30	100%	60/60	100%
Milddle positive	30/30	100%	60/60	100%
Standard material	Between person		Between site	
	Positive / No.	Positive rate	Positive / No.	Positive rate
Negative	0/60	0%	0/60	0%
Low positive	60/60	100%	60/60	100%
Milddle positive	60/60	100%	60/60	100%

■ **Clinical performance evaluation**

ichroma™ COVID-19 Ag has demonstrated the following clinical performance results.

		RT-PCR		
		Positive	Negative	Total
ichroma™ COVID-19 Ag	Positive	123	2	125
	Negative	12	158	170
	Total	135	160	295

- Clinical sensitivity: 91.1%
- Clinical specificity: 98.8 %

REFERENCES

1. Laboratory testing for 2019 novel coronavirus (2019-nCoV) in suspected human cases Interim guidance (2020) 17 Jan, WHO.
2. Wölfel et al. Virological assessment of hospitalized cases of coronavirus disease 2019 (2020) Nature. [Epub ahead of print]
3. Trivedi SU et al. Development and Evaluation of a Multiplexed Immunoassay for Simultaneous Detection of Serum IgG Antibodies to Six Human Coronaviruses (2019) Sci Rep. 9: 1390
4. Yongchen et al. Different longitudinal patterns of nucleic acid and serology testing results based on disease severity of COVID-19 patients (2020) Emerg Microbes Infect 20: 1

Note: Please refer to the table below to identify various symbols.

	Sufficient for <n> tests
	Read instruction for use
	Use by Date
LOT	Batch code
REF	Catalog number
	Caution
	Manufacturer
EC REP	Authorized representative of the European Community
IVD	In vitro diagnostic medical device
	Temperature limit
	Do not reuse
CE	This product fulfills the requirements of the Directive 98/79/EC on in vitro diagnostic medical devices

CE IVD