ichroma™ MxA/CRP

INTENDED USE

ichroma™ MxA/CRP is a fluorescence immunoassay (FIA) for the quantitative determination of Myxovirus resistance protein A (MxA) and C-Reactive Protein (CRP) in human whole blood. This test aids in identification of viral and/or bacterial infection in patients who have symptoms of acute respiratory infection within 7 days.

For *in vitro* diagnostic use only.

INTRODUCTION

Viral and bacterial respiratory infections represent a major source of morbidity, mortality, and healthcare costs. Approximately 80% of all antimicrobials are prescribed in primary care, and up to 80% of these are for respiratory tract indications[8]. But sensitive and specific diagnostic tools to aid in the diagnosis of ARIs (particularly in differentiating bacterial and viral infections) in primary and urgent healthcare settings have been lacking[9].

Mx proteins are large GTPases and belong to a group of IFN-induced GTPases involved in the control of intracellular pathogens[5]. In humans, two Mx homologs (MxA and MxB, also called MX1 and MX2, respectively) mediate antiviral activity against a broad range of viruses include Covid-19[6]. Elevated levels of MxA protein could be an indicator of endogenous interferon production mediated by an unknown viral activation and so, the MxA protein levels could be used as a general marker of viral infection[7].

CRP is one of the cytokine-induced acute-phase proteins[1] whose blood levels rise during a general, unspecific response to infections and non-infectious inflammatory processes[2]. CRP tests provide information for the diagnosis, therapy, and monitoring of inflammatory diseases.[10] During infectious or inflammatory disease states, CRP levels rise rapidly within the first 6 to 8 hours and peak at levels of up to 350–400 mg/L after 48 hours. Measurement of CRP concentration has been widely used as a clinical tool for monitoring the status of inflammation, effectiveness of treatment of various infections and autoimmune diseases such as rheumatoid arthritis.

MxA/CRP test will aid in the differentiation of viral and bacterial acute febrile respiratory infections.

PRINCIPLE

The 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 the instrument for ichroma™ tests to show MxA and CRP concentration in the sample.

COMPONENTS

ichroma™ MxA/CRP consists of 'cartridges', 'detector tubes' and 'detector diluent'.

- The cartridge contains the membrane called a test strip which has anti-MxA, and anti-CRP at the test line, CRP Ag at the antigen line, and chicken IgY at the control line. All cartridges are individually sealed in an aluminum foil pouch containing a desiccant, and they are further packaged in a box.
- The detector tube has a granule containing the anti-MxA-fluorescence complex, anti- CRP-fluorescence conjugate, anti-chicken IgY-fluorescence conjugate, and sodium azide as a preservative in phosphate buffered saline (PBS). All detector tubes are packed in a pouch.
- The detector diluent contains detergent for blood cell lysis and sodium azide as a preservative in phosphate buffered saline (PBS), and it is pre-dispensed in a vial. The detector diluent is packed in a box.

WARNINGS AND PRECAUTIONS

- For *in vitro* diagnostic use only.
- Follow the instructions and procedures described in this 'Instructions for use'.
- Use only fresh samples and avoid direct sunlight.
- Lot numbers of all the test components (cartridge, detector tube, detector diluent 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).
- Do not reuse cartridges or detector tubes. A cartridge should be used for testing one sample only. A detector tube should be used for processing of one sample only.
- Do not reuse 10 µL Capillary tube. A 10 µL Capillary tube should be used for testing one sample only.
- 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.
- After using the detector diluent, keep it closed.
- The instrument for ichroma™ tests may generate slight vibration during use.
- Used cartridges, detector tubes, detector diluent, capillary tubes and pipette tips should be handled carefully and discarded by an appropriate method in accordance with relevant local regulations.
- The detector tube and the detector diluent contain sodium azide (NaN_3), and it may cause certain health issues like convulsions, low blood pressure, low 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.
- No biotin interference was observed in **ichroma™ MxA/CRP** when biotin concentration in the sample was

below 3,500 ng/mL. If a patient has been taking biotin at dosage of more than 0.03 mg a day. It is recommended to test again 24 hours after discontinuation of biotin intake.

- **ichroma™ MxA/CRP** will provide accurate and reliable results subject to the below conditions.
  - **ichroma™ MxA/CRP** should be used only in conjunction with the instrument for ichroma™ tests.
  - Have to use recommended anticoagulant.

Recommended anticoagulant
K_2 EDTA, K_3 EDTA, Na_3 citrate, Sodium heparin, Lithium heparin

- **The capillary tube should be used when the following conditions are met.**
  - The capillary tube provided with the kit is recommended to obtain correct test result.
  - Whole blood should be immediately tested after collection.
  - Excess whole blood around the capillary tube should be wiped off.
  - In order to avoid cross-contamination, please do not re-use capillary tube for multiple samples.

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 the 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.
- Any clinical diagnosis based on the test result must be supported by a comprehensive judgment of the concerned physician in conjunction with clinical symptoms and other relevant test results.

※ The following cases may affect the measurement results.

- Receiving interferon therapy (e.g., MS, HIV, HBV, HCV) in the last 30 days.
- Immunocompromised state (e.g., HIV) or taking immunosuppressive or chemotherapeutic medications in the last 30 days (e.g., oral steroids, Methotrexate, Cyclosporine, Antimetabolite chemotherapy, interferon therapy).
- Taking antibiotics or antiviral therapy in the last 14 days.
- Received a live viral immunization in the last 14 days.
- Significant trauma or burns (> 5% total body surface area or full thickness (3rd°)) in the last 30 days.
- Major surgery (requiring intravenous anesthesia and/or respiratory assistance) in the last 30 days-
- History of a myocardial infarction or stroke in the last 30 days.
- Taking high biotin-containing drugs within 24 hours.

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
Detector diluent	2 – 30°C	20 months	Unopened
		20 months	Opened

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

MATERIALS SUPPLIED

REF CFPC-172

Components of **ichroma™ MxA/CRP**

- Cartridge box:
  - Cartridge 25
  - Detector tube 25
  - Detector diluent 3
  - 10 µL Capillary tube 25
  - ID chip 1
  - Instructions for use 1

MATERIALS REQUIRED BUT SUPPLIED ON DEMAND

Following items can be purchased separately with **ichroma™ MxA/CRP**

Please contact our sales division for more information.

- Instrument for ichroma™ tests
  - **ichroma™ II** REF FPRR021
  - **ichroma™ III** REF FPRR037
  - **ichroma™ M3** REF FPRR035
  - **ichroma™-50 Plus** REF FPRR036
- **Boditech MxA/CRP Control** REF CFPO-382

SAMPLE COLLECTION AND PROCESSING

The sample type for **ichroma™ MxA/CRP** is human whole blood.

- It is recommended to test the sample within 1 hour after collection.
- The samples (whole blood) may be stored for 12 hours at 2-8°C prior to being tested.
- However, the whole blood sample should not be kept in a freezer in any case.
- Whole blood sample can be collected using a capillary tube according to below:

① Wear disposable gloves and protective equipment for safety.

② Open the zipper bag which has capillary tubes.

③ Take out the capillary tube and check for damage or contamination.

④ Hold the handle of the capillary tube and touch the surface of blood with the capillary tube.

⑤ Fill it with blood completely.
(Make sure that no air bubbles are present in the capillary tube. Do not get blood on the surface of the capillary tube. If the blood gets on the surface of the capillary tube, remove it gently with gauze.)

TEST SETUP

- Check the contents of **ichroma™ MxA/CRP**: Sealed cartridges, detector tubes, a detector diluent, 10 µL capillary tubes, an ID chip and an instructions for use.
- Ensure that the lot number of the cartridge matches that of the detector tube, the detector diluent as well as an ID chip.
- If the sealed cartridge, the detector tube and detector diluent 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 instrument for ichroma™ tests operation manual for complete information and operating instructions.

TEST PROCEDURE

▶ ichroma™ II, ichroma™ M3

Multi test mode / Read now mode

1) Take 400 µL of detector diluent using a pipette and dispense it to the detector tube containing granules. When the granule form is completely dissolved in the tube, it becomes detection buffer.
   (The detection buffer must be used immediately. Do not exceed 30 seconds.)
2) Take 10 µL of sample (Human whole blood/control) using a pipette and dispense it to the detector tube.
   *If you use a capillary tube (10 µL), put it into the detector tube after collecting whole blood sample.
3) Close the lid of the detector tube and mix the sample thoroughly by shaking it about 20 times.
   (The sample mixture must be used immediately. Do not exceed 30 seconds.)
4) Take 75 µL of the sample mixture and dispense it into the sample well of the cartridge.
5) Leave the cartridge at room temperature for 12 minutes.
   ⚠ *Scan the sample-loaded cartridge immediately when the incubation time is over. If not, it will cause inaccurate test result.*
6) 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.
7) Tap the 'Start' button on the instrument for ichroma™ tests to start the scanning process.
   (ichroma™ M3 will start the test automatically after inserting.)
8) The instrument for ichroma™ tests will start scanning the sample-loaded cartridge immediately.
9) Read the test result on the display screen of the instrument for ichroma™ tests.

Single test mode/ Walk away mode

1) The test procedure is same with the 'Multi test mode 1) – 4)'.
2) Insert the sample-loaded cartridge into the 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.
3) Press the 'Start' button on the instrument for ichroma™ tests.
   (ichroma™ M3 will start the test automatically after inserting.)
4) The cartridge goes inside the instrument for ichroma™ tests and will automatically start scanning the sample-loaded cartridge after 12 minutes.
5) Read the test result on the display screen of the instrument for ichroma™ tests.

▶ ichroma™ III

1) The test procedure is same with the 'Single test mode'.

▶ ichroma™-50 Plus

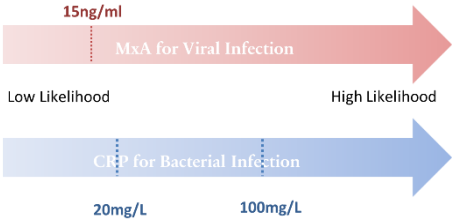
1) Insert the tip array in the tip station.
2) Insert the detector tube in the reagent station and cover the reagent station to hold the detector tubes in place.
3) Open the lid of the detector diluent and insert the detector diluent in the diluent station.
4) Insert the cartridge magazine with the cartridges into the magazine station.
5) Insert the sample tube into the blood collection tube rack and load the blood collection tube rack into the sampling station (loading part).
6) Tap the button located in the upper side of the No. of test cartridge region to select the ID chip that you want to use.
7) When the selected cartridge slot is activated, set the number of the detector tube 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.

INTERPRETATION OF TEST RESULT

- The instrument for ichroma™ tests calculates the test result automatically and displays MxA/CRP concentration of the test sample in terms of of ng/mL and mg/L respectively.

Unit	MxA [ng/mL]	CRP [mg/L]
Clinical Cut-off	15.0	20.0
Measuring range	10.0 – 300.0	1.0 -200.0

MxA [ng/mL]	CRP [mg/L]	Interpretation
MxA ≥ 15.00	CRP < 20.00	viral infection
	20.00 ≤ CRP < 100.00	viral infection and bacterial infection likely
	CRP ≥ 100.00	viral infection and bacterial infection very likely
MxA < 15.00	20.00 ≤ CRP < 100.00	bacterial infection likely
	CRP ≥ 100.00	bacterial infection very likely
	CRP < 20.00	non-infection



- This product is intended for screening tool only. A negative result does not preclude a viral or bacterial infection. Any clinical diagnosis based on the test result must be supported by a comprehensive judgment of the concerned physician in conjunction with clinical symptoms and other relevant test results.
- NICE(National Institute for Health and Care Excellence) recommend CRP test for antibiotics prescription as follows.
  Pneumonia in adults: diagnosis and management (CG191)
  [Presentation with lower respiratory tract infection10]
  For people presenting with symptoms of lower respiratory tract infection in primary care, consider a point care C-reactive protein test if after clinical assessment a diagnosis of pneumonia has not been made and it is not clear whether antibiotics should be prescribed. Use the results of the C-reactive protein test to guide antibiotic prescribing in people without a clinical diagnosis of pneumonia as follows:
  - Do not routinely offer antibiotic therapy if the C-reactive protein concentration is less than 20 mg/liter.
  - Consider a delayed antibiotic prescription (a prescription for use at a later date if symptoms worsen) if the C-reactive protein concentration is between 20 mg/liter and 100 mg/liter
  - Offer antibiotic therapy if the C-reactive protein concentration is greater than 100 mg/liter.

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.
- 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 with **ichroma™ MxA/CRP.** For more information regarding obtaining the control materials, contact Boditech Med Inc.'s Sales Division for assistance.
  (Please refer to the instructions for use of control material.)

PERFORMANCE CHARACTERISTICS

- **Analytical sensitivity**

Unit	MxA [ng/mL]	CRP [mg/L]
LOB	4.84	0.45
LOD	6.34	0.66
LOQ	9.7	0.96

- **Analytical specificity**
  - Cross reactivity
  Biomolecules listed in the following table were added to the test sample(s) at concentrations much higher than their normal physiological levels in the blood. **ichroma™ MxA/CRP** test results did not show any significant cross-reactivity with these biomolecules.

Cross reactants	Concentration
MxB	100 ng/ml
Human IRGM (Immunity-related GTPase family M protein)	100 ng/ml
Human GBP1 (guanylate-binding protein 1)	100 ng/ml
Human Gvin1 (Interferon-induced very large GTPase 1)	100 ng/ml
Human Interferon-γ-Inducible Protein-10	100 ng/ml
Interferon α	100 pg/ml
Interferon β 1a	100 pg/ml
Interferon λ	100 pg/ml
TNF-α	100 pg/ml
IL-10	100 pg/ml
IL-6	100 pg/ml
IL-2	100 pg/ml
PCT	50 ng/ml

  - Interference
  Interferents listed in the following table were added to the test sample at the concentration mentioned below. **ichroma™ MxA/CRP** test results did not show any significant interference with these materials.

Interference material	Concentration
Ascorbic acid	350 µmol/L
Bilirubin (conjugated)	475 µmol/L
Albumin	60 g/L
Glucose	1,000 mg/dL
Triglycerides mixture	1,500 mg/dL
Hemoglobin	10 g/L
K_2 EDTA	5.45 mg/mL
K_3 EDTA	5.45 mg/mL
Li-Heparin	10.41 mg/mL
Na-Heparin	10.44 mg/mL
Sodium Citrate	32 mg/mL

- **Precision**
  - Single-site study
  Repeatability (within-run precision)
  within-laboratory precision (Total precision)
  Lot to lot precision
  3 Lots of **ichroma™ MxA/CRP** were tested for 20days. Each standard material was tested 2 times per day. For each test, each material was duplicated.

MxA Conc. [ng/ml]	Repeatability (within-run)		within-laboratory		Lot to lot precision	
	Mean [mg/L]	CV (%)	Mean [mg/L]	CV (%)	Mean [mg/L]	CV (%)
18.2	17.88	9.3	18.07	8.1	17.91	7.8
71.67	69.12	8.5	70.42	8.7	70.95	8.0
144.28	143.06	7.6	143.42	7.2	141.85	7.6
CRP Conc. [mg/L]	Repeatability (within-run)		within-laboratory		Lot to lot precision	
	Mean [mg/L]	CV (%)	Mean [mg/L]	CV (%)	Mean [mg/L]	CV (%)
9.1	9.18	6.4	9.16	6.6	9.11	6.8
42.2	42.06	6.1	42.21	6.3	41.82	6.5

93	92.54	6.5	92.40	6.5	93.29	6.5

- Multi-site study

Reproducibility

1 Lot of **ichroma™ MxA/CRP** was tested for 5 days in 3 different sites (1 person per 1 site, 1 instrument per 1 site). Each standard material was tested 1 time per and 5 replicates per day.

Multi-site study			
MxA [ng/ml]	Mean [ng/ml]	SD	CV (%)
18.2	18.04	1.21	6.7
71.67	70.61	4.75	6.7
144.28	140.52	10.91	7.8
CRP [mg/L]	Mean [mg/L]	SD	CV (%)
9.1	9.15	0.63	6.9
42.2	41.64	2.56	6.1
93	92.70	5.89	6.4

- **Accuracy**

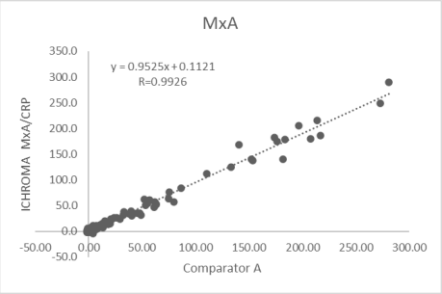
The accuracy was confirmed by testing with 3 different lots of **ichroma™ MxA/CRP.** The tests were repeated 10 times at each concentration of the control standard.

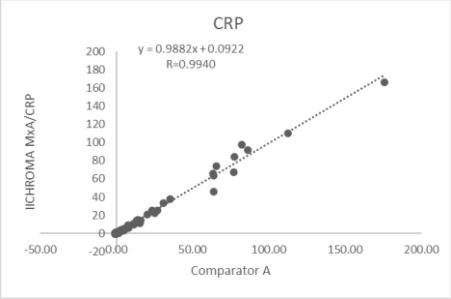
MxA [ng/ml]	Lot 1	Lot 2	Lot 3	3 LOTs of AVG [ng/ml]	recovery (%)
18.21	18.4	18.7	18.8	18.6	102.3
43.42	43.5	44.1	44.3	44.0	101.3
68.64	66.8	68.5	70.5	68.6	100.0
93.85	95.7	93.1	96.7	95.2	101.4
119.1	118.9	120.0	120.2	119.7	100.5
144.3	145.2	140.7	143.3	143.1	99.2

CRP [mg/L]	Lot 1	Lot 2	Lot 3	3 LOTs of AVG [mg/L]"	Bias (%)
41.20	40.27	42.48	41.58	41.44	**0.6%**

- **Comparability**

MxA/CRP concentration of 165 clinical samples were quantified independently with **ichroma™ MxA/CRP (ichroma™ II)** and **comparator A** as per prescribed test procedures. Test results were compared, and their comparability was investigated with linear regression and correlation coefficient (R). The regression equation and correlation coefficient are as follows.





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

	Sufficient for <n> tests
	Read instruction for use
	Use by Date
	Batch code
	Catalog number
	Caution
	Manufacturer
	Authorized representative of the European Community
	In vitro diagnostic medical device
	Temperature limit
	Do not reuse

For technical assistance, please contact:
Boditech Med Inc.'s Technical Services
Tel: +(82) -33-243-1400
E-mail: TS@boditech.co.kr

 **Boditech Med Incorporated**
43, Geodudanji 1-gil, Dongnae-myeon, Chuncheon-si,
Gang-won-do, 24398, Republic of Korea
Tel: +(82) -33-243-1400
Fax: +(82) -33-243-9373
www.boditech.co.kr

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