BIO TECHNOLOGY boditech

Rheumatoid Arthritis

ichroma™ ASO

INTENDED USE

ichroma™ ASO is a fluorescence immunoassay (FIA) for the quantitative determination of Anti Streptolysin O (ASO) in human serum/plasma. It is useful as an aid in management and monitoring of scarlet fever, rheumatic fever and post infectious glomerulonephritis along with several other conditions.

For *in vitro* diagnostic use only.

INTRODUCTION

ASO is an antibody produced in human blood against streptolysin O made from an infection of Streptococcus bacteria. An elevated or rising ASO titer may demonstrate recent streptococcal infections. Some autoimmune responses, glomerulonephritis, acute tonsillitis, scarlet fever and rheumatic fever may be associated with streptococcal infections. Even if ASO titers may vary due to a number of factors including population and age.

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-antibodies 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 ASO concentration in the sample.

COMPONENTS

ichroma™ ASO consists of 'cartridges', 'detector tube' and 'detector diluent'.

- The cartridge contains the membrane called a test strip which has SLO protein at the test 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 SLO protein, anti-chicken IgY, mouse IgG as a blocker, bovine serum albumin (BSA) as a stabilizer, and sodium azide in phosphate buffered saline (PBS) as a preservative. All detection tubes are packed in a pouch.
- The detector diluent contains sodium azide as a preservative, in phosphate buffer saline (PBS) and it is pre-dispensed in a vial. The detector diluent 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 one sample only.
- The cartridge should remain sealed in its original pouch until just before use. Do not use the cartridge, if pouch is damaged or has already been opened.
- Frozen sample should be thawed only once. For shipping, samples must be packed in accordance with local regulations. Sample with severe hemolysis and/or hyperlipidemia must not be used.
- 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.
- The instrument for **ichroma™ ASO** tests may generate slight vibration during use.
- Used cartridges, detector tube, detector diluent 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 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™ ASO** will provide accurate and reliable results subject to the below conditions.
  - **ichroma™ ASO** should be used only in conjunction with the Instrument for ichroma™ tests.
  - Have to use recommended anticoagulant sample.

Recommended anticoagulant
Na-EDTA, K_2 EDTA, Sodium Citrate

STORAGE AND STABILITY

Storage condition			
Component	Storage Temperature	Shelf life	Note
Cartridge	2-30 ℃	20 months	Disposable
Detector tube	2-30 ℃	20 months	Disposable
Detector diluent	2-30 ℃	20 months	Unopened
		12 months	Opened

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

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.

MATERIALS SUPPLIED

REF CFPC-46

Components of **ichroma™ ASO**

- Cartridge box:
  - Cartridge 25
  - ID Chip 1
  - Instruction for use 1
  - Detector tube 25
  - Detector diluent 1

MATERIALS REQUIRED BUT SUPPLIED ON DEMAND

Following items can be purchased separately from **ichroma™ ASO**.

Please contact our sales division for more information.

- Instrument for ichroma™ tests
  - **ichroma™ Reader** REF FR203
  - **ichroma™ II** REF FPRR021
  - **ichroma™ III** REF FPRR037
  - **ichroma™ M3** REF FPRR035
- **Printer** REF FPRR007

SAMPLE COLLECTION AND PROCESSING

The sample type for ichroma™ ASO is human serum/plasma.

- It is recommended to test the sample within 24 hours after collection.
- The samples (serum, plasma) should be separated from the clot by centrifugation within 3 hours after the collection of whole blood.
- The samples (serum, plasma) stored frozen at -20℃ for 3 months showed no performance difference.
- However, the whole blood sample should not be kept in a freezer in any case.
- As a repeated freeze-thaw cycle may affect the test result, do not refreeze previously frozen samples.

TEST SETUP

- Check the contents of ichroma™ ASO: Sealed cartridges, detector tubes, a detector diluent, an ID chip and an instruction for use.
- Ensure that the lot number of the cartridges matches that of the detector tube, 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.
  (Please refer to the 'Instrument for ichroma™ tests Operation Manual' for complete information and operating instructions.)

TEST PROCEDURE

▶ **ichroma™ Reader, ichroma™ II, ichroma™ M3**

Multi test mode

1) Take 500 µL of detector diluent using a pipette and dispense it to the detector tube containing a granule. 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 5 µL of sample (serum/plasma) using a pipette and dispense it to the detector tube.
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) Press the 'Select' or tap the 'Start' button on the instrument for ichroma™ tests to start the scanning process.
   (ichroma™ M3 is tested 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

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 'Select' or tap the 'Start' button on the instrument for ichroma™ tests.
(ichroma™ M3 is tested 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'.

INTERPRETATION OF TEST RESULT

- The instrument for ichroma™ tests calculates the test result automatically and displays ASO concentration of the test sample in terms of IU/mL.
- The cut-off (reference range): 160 IU/mL

Upper Limit of Normal ASO	
Age	Concentration of ASO
Adult	<166 IU/mL
Preschool age	<100 IU/mL
School age	<250 IU/mL

- Working range: 25-800 IU/mL.

PERFORMANCE CHARACTERISTICS

- **Analytical sensitivity**

Limit of Blank (LoB)	5.86 IU/mL
Limit of Detection (LoD)	9.18 IU/mL
Limit of Quantification (LoQ)	25.0 IU/mL

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

Cross Reactivity	Concentration
CRP	0.4 mg/dL
RF IgM	2.5 IU/mL

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

Interference	Concentration
Hemoglobin	2 mg/ml
Unconjugated bilirubin	342 µmol/L
Phospholipid	1.11 mmol/L
Triglyceride	3.7 mmol/L

- **Precision**
  - **Single-site study**
    Repeatability (within-run precision)
    Within-laboratory precision (Total precision)
    Lot to lot precision
    3 Lots of **ichroma™ ASO** were tested for 21 days. Each standard material was tested 2 times per day. For each test, each material was duplicated.
  - **Between person**
    Three different persons tested three lots of **ichroma™ ASO**, ten times at each concentration of the control standard.
  - **Between site**
    One lot of **ichroma™ ASO** was tested at three different sites; ten times at each concentration of the control standard.
  - **Between reader**
    One lot of **ichroma™ ASO** was tested with three different instruments; five times at each concentration of the control standard.

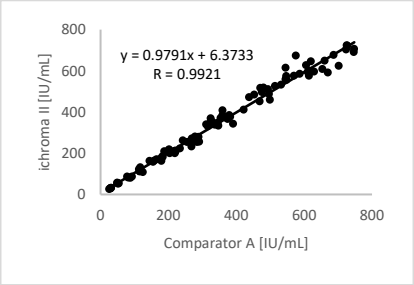
Samples (IU/mL)		Repeatability (within-run)		Total precision (within-laboratory precision)		Lot to lot precision	
		AVG	CV(%)	AVG	CV(%)	AVG	CV(%)
Cal. 5	110	111.71	7.58	111.96	7.64	110.87	8.43
Cal. 6	187.5	183.14	7.09	184.81	7.54	185.87	7.39
Cal. 8	500	495.38	8.23	490.13	8.08	493.76	8.05

Samples		Between-person		Between-site		Between-Reader	
		AVG	CV(%)	AVG	CV(%)	AVG	CV(%)
Cal. 5	110	111.43	7.57	110.88	8.52	110.48	8.93
Cal. 6	187.5	187.19	7.96	191.34	6.35	183.07	8.29
Cal. 8	500	501.88	7.98	505.80	7.99	483.57	8.52

- **Accuracy**
  The accuracy was confirmed by testing with 3 differents lots of **ichroma™ ASO.** The tests are repeated 10 times in each different concentration.

No.	Expected value [IU/ml]	Lot 1	Lot 2	Lot 3	AVG	Recovery
1	24.47	24.04	24.99	25.18	24.74	101%
2	49.64	47.59	48.48	48.75	48.27	97%
3	102.11	107.27	106.06	110.59	107.97	106%
4	194.07	176.89	188.26	193.11	186.09	96%
5	275.89	253.65	239.59	251.66	248.30	90%
6	515.33	463.63	501.38	476.85	480.62	93%
7	581.36	594.14	584.45	577.08	585.22	101%
8	671.29	649.98	680.58	712.87	681.14	101%
9	805.34	814.92	792.33	788.57	798.61	99%
10	908.77	873.46	900.23	892.92	883.19	98%
11	983.63	1014.16	969.04	999.98	994.39	99%

- **Comparability**
  ASO concentrations of 100 clinical samples were quantified independently with **ichroma™ ASO (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

Σ	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

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