Infection

ichroma™ Syphilis

INTENDED USE

ichroma™ Syphilis is a fluorescence Immunoassay (FIA) for the qualitative determination of Syphilis total antibody (Treponema pallidum- IgG, IgM, IgA) in human whole blood/serum/plasma. It is useful as an aid in screening of Syphilis infection.

For *in vitro* diagnostic use only.

INTRODUCTION

Syphilis is an infectious and sexually transmitted chronic disease caused by Treponema pallidum. The progression of syphilis is classified into early symptomatic (primary, secondary, early latent) and late asymptomatic (late latent, tertiary) stage.[1)]

These reflect the infectious period, thus early syphilis is infectious and at late stage the infection is not transmittable. Early syphilis can be divided again into primary, secondary and early latent syphilis depending on clinical presentation.[3)]

Late syphilis consists of late latent (asymptomatic) and tertiary syphilis. Approximately 30- 40% of cases untreated syphilis will develop late symptomatic disease. Tertiary syphilis is the manifestation of long term syphilis and consists of cardiovascular, neurological or gummatous involvement. syphilis can cause benign lesions in skin, bone, liver and upper respiratory tract. Cardiovascular syphilis primarily involves the aorta leading to aortitis, aortic regurgitation or aneurysm. Late neurosyphilis manifests as meningitis, stroke, cranial nerve palsies, myeopathy (including tabes dorsalis), seizures or progressive dementia (general paresis). Late latent syphilis is diagnosed in the absence of neurosyphilis and other symptoms and signs of disease. [3)]

The infection can also be passed from mother to her fetus during pregnancy. Most infected individuals have no symptoms or have transient lesions and therefore a serological test must be used to screen for infection. [2)]

The control of syphilis requires early identification and treatment of cases. This calls for tests that are easily administered and interpreted, and treatment that is fast, efficacious and side effect free.3) If syphilis patients are not diagnosed properly, it may lead to a serious public health problem. [2)]

The "**ichroma™ Syphilis**" is an immunoassay for the detection of Syphilis total anti-body in human whole blood/serum/plasma.

PRINCIPLE

The test uses a sandwich immunodetection method; Dried detector antigen in the detection buffer, once diluted with the diluent, bind with antibody in the sample to form antigen-antibody complexes. These complexes then migrate through the nitrocellulose matrix and are captured by another sets of immobilized antigen on test line.

The more antibody in the sample, the more antigen-antibody complexes, which leads to stronger fluorescence signal. This signal then is interpreted by the reader to display 'Syphilis positive' in the sample.

COMPONENTS

ichroma™ Syphilis consists of 'Cartridges', 'detection Buffers', 'Diluent' and an 'ID chip'.

- The cartridge contains a test strip, the membrane which has recombinant syphilis antigen at the test line, with chicken IgY at the control line.
- Each cartridge is individually sealed in an aluminum foil pouch containing a desiccant. 25 sealed cartridges are packed in a box which also contains an ID chip.
- The detection buffer contains dried detection buffer and granulated ball. There are contain recombinant Treponema pallidum antigen-fluorescence conjugate, anti chicken IgY-fluorescence conjugate, bovine serum albumin (BSA) as a stabilizer and sodium azide as a preservative in phosphate buffered saline (PBS).
- The dried detection buffer is pre-dispensed in a tube. 25 detection buffer tubes are packed in an aluminum foil pouch.
- The diluent contains a detergent and bovine serum albumin (BSA) as a stabilizer and sodium azide as a preservative in PBS. The diluent is dispensed in a vial.
- The detection buffer (while sealed in an aluminum foil pouch) and the diluent vial are packed in a box.

WARNINGS AND PRECAUTIONS

- For *in vitro* diagnostic use only.
- Carefully follow the instructions and procedures described in this 'Instruction for use'.
- Use only fresh samples and avoid direct sunlight.
- Lot numbers of all the test components (cartridge, ID chip and detection buffer, diluent) should agree.
- Do not interchange test components between different lots or use test components after the expiration date, either of which might yield misleading of test result(s).
- Do not reuse. A detection buffer tube should be used for processing one sample only. So should a cartridge.
- The cartridge should remain sealed in its original pouch before use. Do not use the cartridge, if is damaged or already opened.
- Frozen sample should be thawed only once. For shipping, samples must be packed in accordance with applicable local requirement. A Sample with severe hemolytic and hyperlipidemia cannot be used and should be recollected.
- Just before use, allow the cartridge, the detection buffer and the sample reach the room temperature by leaving them in the room for approximately 30 minutes at the least.
- **ichroma™ Syphilis** as well as the instrument for ichroma™ tests should be used away from vibration and/or magnetic field. During normal usage, it can be noted that instrument for ichroma™ tests may produce minor vibration.
- Used detection buffer tubes, pipette tips and cartridges should be handled carefully and discarded by an appropriate method in accordance with relevant local regulations.
- An exposure to larger quantities of sodium azide may cause certain health issues like convulsions, low blood pressure and heart rate, loss of consciousness, lung injury and respiratory failure.
- **ichroma™ Syphilis** will provide accurate and reliable results subject to the following conditions.
  - **ichroma™ Syphilis** should be used only in conjunction with instrument for ichroma™ tests.
  - Any anticoagulants other than **K_2EDTA, heparin, sodium citrate** should be avoided.

STORAGE AND STABILITY

- The cartridge is stable for 20 months (while sealed in the original aluminum foil pouch) if stored at 4-30 °C.
- The detection buffer and the diluent are stable for 20 months if stored at 2-8 °C.
- 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 antigens.

- The test may yield false negative result. The non-responsiveness of the antibodies to the antigens is most common where the epitope is masked by some unknown components, so as not to be detected or captured by the antibodies. The instability or degradation of the antigen with time and/or temperature may cause the false negative as it makes antibody unrecognizable by the antigens.
- 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 including clinical symptoms and other relevant test results.

MATERIALS SUPPLIED

REF CFPC-85

Components of **ichroma™ Syphilis**

- Cartridge Box:
  - Cartridges 25
  - ID Chip 1
  - Instruction For Use 1
- Buffer box
  - Detection Buffers 25
  - Diluent Vial 1

MATERIALS REQUIRED BUT SUPPLIED ON DEMAND

Following items can be purchased separately from **ichroma™ Syphilis**. Please contact our sales division for more information.

- **ichroma™ II** REF FPRR021
- **Boditech Syphilis Control** REF CFPO-210

SAMPLE COLLECTION AND PROCESSING

The sample type for **ichroma™ Syphilis** is human whole blood/ serum/plasma.

- It is recommended to test the sample within 24 hours after collection.
- The serum or plasma should be separated from the clot by centrifugation within 3 hours after the collection of whole blood.
- Samples may be stored for up to a week at 2-8 °C prior to being tested. If testing will be delayed more than a week, samples should be frozen at -20 °C.
- Samples stored frozen at -20 °C for 2 months showed no performance difference.
- However, the whole blood sample should not be kept in a freezer in any case.
- Once the sample was frozen, it should be thawed one time and only for test, because repeated freezing and thawing can result in the changed test values.
- Samples containing precipitates must be clarified by centrifugation.

TEST SETUP

- Check the contents of **ichroma™ Syphilis**: Sealed Cartridge, Detection Buffer Tubes, Diluent Vial and ID Chip.
- Ensure that the lot number of the cartridge matches that of the ID chip as well as the buffer box.
- Keep the sealed cartridge (if stored in refrigerator) and the detection buffer tube at room temperature for at least 30 minutes just prior to the test. Place the cartridge on a clean, dust-free and flat surface.
- Turn on the Instrument for ichroma™ tests.
- Insert the ID Chip into the ID chip port of the Instrument for ichroma™ tests**.**
- Press the 'Select' button on the Instrument for ichroma™ tests. (Please refer to the 'Instrument for ichroma™ tests Operation Manual' for complete information and operating instructions.)

TEST PROCEDURE

1) Open the diluent vial and transfer 150 µL of diluent buffer using a transfer pipette to the detection buffer tube.
2) Transfer 30 µL of sample (whole blood/serum/plasma/control) using a transfer pipette to the detection buffer tube.
3) Mix well by pipetting 10-20 times. (The sample mixture must be used immediately.)
4) Pipette out 75 µL of a sample mixture and dispense it into the sample well on the cartridge.
5) Leave the sample-loaded cartridge at room temperature for 12 minutes.
   ⚠ *Scan the sample-loaded cartridge immediately when the incubation time is over. If not, it will cause inexact 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 has been marked on the cartridge especially for this purpose.
7) Tab the 'Start' icon on the screen.
8) 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.

INTERPRETATION OF TEST RESULT

- Instrument for ichroma™ tests calculates the test result automatically and displays "Positive"/"Negative"/ "Indeterminate".
- Ancillary value is served in the form of a cut-off index (COI).

Cut-off index (COI)	Result	Note
< 0.90	Negative for Syphilis.	No need to additional test.
≥ 0.90, < 1.0	Indeterminate.	Dilute the clinical sample with suitable diluent (2 times).
≥ 1.0	Positive for Syphilis.	Need to confirmation test.

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 not provided with **ichroma™ Syphilis**. 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**
  - Cut-off

ichroma™ Syphilis decides between positive and negative through COI calculated by ichroma™ II algorism.

Cut-off Index (COI)	Result
COI ≥ 1.0	Positive
0.90 ≤ COI < 1.0	Indeterminate
COI < 0.90	Negative

- **Analytical Specificity**

- Cross-reactivity

There was no false positive result from 264 samples containing potentially interfering substances with the **ichroma™ Syphilis** test. The overall specificity was 100 %.

Clinical category	ichroma™ Syphilis results		
	Number of samples	Negative	Positive
CMV	20	20	0
EBV	20	20	0
HAV	29	29	0
Anti-HBs	11	11	0
HBsAg	15	15	0
HSV	20	20	0
Rubella	20	20	0
VZV	20	20	0
Anti-HCV	20	20	0
ANA	23	23	0
RF	24	24	0
Early stage of pregnancy	19	19	0
Middle stage of pregnancy	23	23	0
Total	264	264	0

- Interference

There was no significant interference from these material with the **ichroma™ Syphilis** test.

Materials	Concentration
Heparin	100,000 U/L
EDTA	5 µM
Sodium citrate	25 mg/mL
Bilirubin	0.5 mM/L
Hemoglobin	2 g/L
Triglycerides	1.5 mg/mL
Cholesterol	20 mM
Albumin	60 mg/mL

- **Precision**

- Between LOT

One person tested three different lots of **ichroma™ Syphilis**, ten times at each concentration of the control standard.

- Between person

Three different persons tested same LOT of **ichroma™ Syphilis**, five times at each concentration of the control standard.

- Between Site

Three different site tested same LOT of **ichroma™ Syphilis**, five times at each concentration of the control standard.

Cal.	Between LOTs		Between person	
	Positive/ Sample number	Positive	Positive/ Sample number	Positive
Negative	0/10	0 %	0/5	0 %
Mid	10/10	100 %	5/5	100 %
Low	10/10	100 %	5/5	100 %
Cal.	**Between day**		**Between site**	
	Positive/ Sample number	Positive	Positive/ Sample number	Positive
Negative	0/5	0 %	0/5	0 %
Mid	5/5	100 %	5/5	100 %
Low	5/5	100 %	5/5	100 %

- **Comparability with reference product**

		Reference product		
		Positive	Negative	Total
ichroma™ Syphilis	Positive	54	1	55
	Negative	1	102	103
	Total	55	103	158

- Positive Comparability: 98.2 %
- Negative Comparability: 99 %
- Total Comparability: 98.7 %

REFERENCES

1. Recent Trends in Clinical Observation of Syphilis and Consideration for Laboratory Tests. Choi *et al.*, *J Korean Med Assocl*. 2009; 52(11): 1100 - 1106
2. Syphilis - Recognition, Description and Diagnosis. NS Sato *et al.*, InTech. 87-88 pp. ISBN 978-953-307-554-9, 2011
3. Syphilis: A Review of the Diagnosis and Treatment. CR. Emerson *et al.*, The Open Infectious Diseases Journal. 2009, *3,* 143-147

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

For technical assistance; please contact:
Boditech Med Inc.'s Technical Services
Tel: +82 33 243-1400
E-mail: sales@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

EC REP **Obelis s.a**
Bd. Général Wahis 53,
1030 Brussels, BELGIUM
Tel: +(32) -2-732-59-54
Fax: +(32) -2-732-60-03
E-Mail: mail@obelis.net

CE