BIO TECHNOLOGY
boditech

Hormone

ichroma™ Testosterone

INTENDED USE

ichroma™ Testosterone is a fluorescence Immunoassay (FIA) for the quantitative determination of Testosterone in human serum/plasma. It is useful as an aid in management and monitoring of androgen level.

For *in vitro* diagnostic use only.

INTRODUCTION

Testosterone (17ß-hydroxyandrost-4-en-3-one) is an anabolic steroid synthesized primarily by Leydig cells in the testes of male, the ovary of female, and adrenal glands of both sexes[1]. It is synthesized from cholesterol, androstenediol, Dehydroepiandrosterone (DHEA), progesterone, and pregnenolone acting as some of the intermediate substrates. Testosterone level in male increase 10 to 20-fold during puberty, driving the physiological changes associated with male puberty. It also exerts a powerful, wide-ranging influence over emotional well-being, sexual function, muscle mass and strength, energy, cardiovascular health, bone integrity, and cognitive ability throughout a man's entire life. In the blood only 1 to 15 % of testosterone is in its unbound or biologically active form. The remaining testosterone is bound to serum proteins.

PRINCIPLE

The test uses a competitive immunodetection method. In this method, the target material in the sample binds to the fluorescence (FL)-labeled detection antibody in detection buffer, to form the complex as sample mixture. This complex is loaded to migrate onto the nitrocellulose matrix, where the covalent couple of testosterone and bovine serum albumin (BSA) is immobilized on a test strip, and interferes with the binding of target material and FL-labeled antibody. If the more target material exists in blood, the less detection antibody is accumulated, resulting in the less fluorescence signal.

COMPONENTS

ichroma™ Testosterone consists of 'Cartridges', 'Detection Buffer Tubes', 'Sample Mixing Tubes', an 'ID chip' and 'Displacing Reagents'.

- The cartridge contains a test strip, the membrane which has BSA conjugated human testosterone at the test line, while KLH 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 anti human testosterone-fluorescence conjugate, bovine serum albumin (BSA) as a stabilizer and sodium azide in phosphate buffered saline (PBS) as a preservative.
- The detection buffer is pre-dispensed in a separate tube.
- The displacing reagent contains anti KLH-fluorescence conjugate as a stabilizer and sodium azide in phosphate buffered saline (PBS) as a preservative.
- The displacing reagent is dispensed in a vial.
- 25 detection buffer tubes and displacing reagent vial are packaged in a box and further packed in a Styrofoam box with ice-pack for the shipment.

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) 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 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 the regulations. Sample with severe hemolytic and hyperlipidemia cannot be used and should be recollected.
- Just before use, allow the cartridge, detection buffer and sample to be at room temperature for approximately 30 minutes.
- **ichroma™ Testosterone** 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™ Testosterone** will provide accurate and reliable results subject to the following conditions.
  - Use **ichroma™ Testosterone** should be used only in conjunction with instrument for ichroma™ tests.
  - Any anticoagulants other than **EDTA** should be avoided.

STORAGE AND STABILITY

- The cartridge is stable for 20 months (while sealed in an aluminum foil pouch) if stored at 4-30 °C.
- The detection buffer pre-dispensed in a tube is stable for 20 months if stored at 2-8 °C.
- The displacing reagent dispensed in a vial is 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 antibodies.
- The test may yield false negative result. The non-responsiveness of the antigen to the antibodies 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 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 including clinical symptoms and other relevant test results.

MATERIALS SUPPLIED

REF 13012

Components of **ichroma™ Testosterone**

- Cartridge Box:
  - Cartridges 25
  - ID Chip 1
  - Instruction For Use 1
  - Sample Mixing Tubes 25
- Detection Buffer Box:
  - Detection Buffer Tubes 25
  - Displacing Reagent Vial (1 mL) 1

MATERIALS REQUIRED BUT SUPPLIED ON DEMAND

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

Please contact our sales division for more information.

- Instrument for ichroma™ tests
  - **ichroma™ Reader** REF FR203
  - **ichroma™ II** REF FPRR021
  - **ichroma™ D** REF 13303
- **ichroma™ Printer** REF FPRR007
- **Boditech Hormone Control** REF CFPO-95

SAMPLE COLLECTION AND PROCESSING

The sample type for **ichroma™ Testosterone** is human 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 3 months showed no performance difference.
- 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.

TEST SETUP

- Check the contents of **ichroma™ Testosterone**: Sealed Cartridge, Detection Buffer Tubes, Displacing Reagent Vial, Sample Mixing Tubes and ID Chip.
- Ensure that the lot number of the cartridge matches that of the ID chip, displacing buffer as well as the detection buffer.
- 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.)

CAUTION

- To minimize erroneous test results, we suggest that the ambient temperature of the cartridge should be 25 °C during the reaction time after loading sample mixture to the cartridge.
- To maintain the ambient temperature to 25 °C, you can use various devices such as an i-Chamber or an incubator and so on.

TEST PROCEDURE

1) Transfer 30 µL of displacing reagent to the sample mixing tube.
2) Transfer 75 µL of sample (Human serum/plasma/control) using a transfer pipette to a sample mixing tube containing the displacing reagent.
3) Close the lid of the sample mixing tube and mix the sample thoroughly by shaking it about 10 times.
4) Incubation the tube at room temperature for 3 minutes.
5) Pipette out 75 µL of a sample mixture and load it into the detection buffer tube.
6) Close the lid of the detection buffer tube and mix the sample thoroughly by shaking it about 10 times. (The sample mixture must be used immediately.)
7) Pipette out 75 µL of a sample mixture and load it into the sample well on the cartridge.
8) Insert the sample-loaded cartridge into the slot of the i-Chamber or an incubator (25 °C).
9) Leave the sample-loaded cartridge in the i-Chamber or an incubator for 12 minutes.
   ⚠ *Scan the sample-loaded cartridge immediately when the incubation time is over. If not, it will cause inexact test result.*
10) 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.
11) Press 'Select' button on the instrument for ichroma™ tests to start the scanning process.
12) Instrument for ichroma™ tests will start scanning the sample-loaded cartridge immediately.
13) 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 testosterone concentration of the test sample in terms of ng/mL.
- The cut-off (reference range): 2-8 ng/mL
- Working range: 1-10 ng/mL

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™ Testosterone**. 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

- **Specificity**: There, in test samples, are biomolecules such as the below table in higher concentration than their normal physiological levels. But this doesn't interfere with the **ichroma™ Testosterone** test measurements, nor occurs any significant cross-reactivity.

Compound	Spiked concentration (ng/mL)	Cross-reactivity (%)
Androstenedione	10	1
Cortisol	1,000	ND*
DHEA	1,000	ND
Estradiol	50	ND
Oxymetholone	100	2.14
Progesterone(17- α)	1,000	0.52
Hydroxyprogesterone	1,000	0.21
Cortisone	1,000	ND

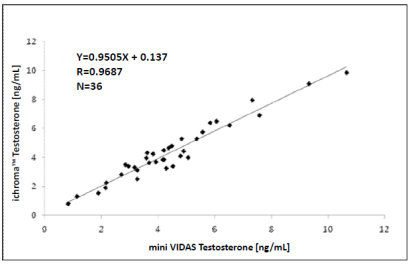
Dihydrotestosterone	50	16

*ND : Not Detected
*None of the substance tested interfered in the assay

- **Precision**: The intra-assay precision was calculated by one evaluator, who tested different concentration of control standard twenty times each with three different lots of **ichroma™ Testosterone.** The inter-assay precision was confirmed by 3 different evaluators with 3 different lots, testing ten times each different concentrations.

Sample	Intra-assay		Inter-assay	
(ng/mL)	Mean (ng/mL)	CV (%)	Mean (ng/mL)	CV (%)
1.17	1.17	16.14	1.22	16.4
2.5	2.47	7.58	2.4	14.67
5.5	5.52	4.59	5.72	6.83
9.43	9.43	1.88	9.43	3.39

- **Comparability:** Testosterone concentrations of 36 clinical samples were quantified independently with **ichroma™ Testosterone** and mini VIDAS (BioMerieux Inc. France) as per prescribed test procedures. Test results were compared and their comparability was investigated with linear regression and coefficient of correlation (R). Linear regression and coefficient of correlation between the two tests were Y = 0.9505X + 0.137 and R = 0.9687 respectively.



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1. Wilson, J.D., George, F.W., and Griffin, J.E. The hormonal control of sexual development. Science, 1981, 211: 1278 – 1284.
2. Vining, R.F., and McGinley, R.A. The measurement of hormones in saliva: Possibilities and pitfalls. Journal of Steroid Biochemistry, 1987, 27: 81-94.
3. Tulsidas G. Shrivastav. Matrix interference in direct total Testosterone enzyme immunoassay and It's elimination with the use of non-cross reactivity steroids in serum based standards. Health and Population Perspectives and Issues, 2002,25(2):55-64.

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