



ichroma[™] β-HCG

INTENDED USE

ichroma[™] β-HCG is a fluorescence Immunoassay (FIA) for the quantitative determination of total β-hCG in human whole blood/serum/plasma. It is useful as an aid in management and monitoring of total beta human chorionic gonadotropin (total β-hCG) level in human.

For in vitro diagnostic use only.

INTRODUCTION

Human chorionic gonadotropin (hCG) is a glycoprotein hormone secreted by the developing placenta shortly after implantation. hCG can be detected in the urine and serum of pregnant women as early as 6 to 15 days after conception. The concentration of hCG increases to 50 mIU/ml one week post implantation and reaches to about 100 mIU/ml at the time of the first missed menstrual period and the peak at 100,000-200,000 mIU/ml at the first trimester.

PRINCIPLE

The test uses a sandwich immunodetection method; the detector antibody in buffer binds to antigen in sample, forming antigen-antibody complexes, and migrates onto nitrocellulose matrix to be captured by the other immobilized-antibody on test strip.

The more antigen in sample forms the more antigen-antibody complex and leads to stronger intensity of fluorescence signal on detector antibody, which is processed by instrument for ichroma[™] tests to show total β-hCG concentration in sample.

COMPONENTS

ichroma[™] β-HCG consists of 'Cartridges', 'Detection Buffer Tubes', 'Sample Diluent Tubes', and an 'ID chip'.

- The cartridge contains a test strip, the membrane which has anti human hCG at the test line, while streptavidin 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 hCG-fluorescence conjugate, biotin-BSA-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 tube. 25 detection buffer tubes are packaged in a box and further packed in a Styrofoam box with ice-pack for the shipment.
- The sample diluent pre-dispensed in a tube contains

sodium azide in phosphate buffered saline (PBS). 25 sample diluent tubes 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) 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[™] β-HCG** 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[™] β-HCG** will provide accurate and reliable results subject to the following conditions.
 - Use **ichroma[™] β-HCG** should be used only in conjunction with instrument for ichroma[™] tests.
 - Any anticoagulants other than EDTA, sodium heparin 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 dispensed in a tube is stable for 20 months if stored at 2-8 °C.
- The sample diluent buffer dispensed in a tube is stable for 24 months if stored at 4-30 °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 CFPC-36

Components of **ichroma™ β-HCG**

- Cartridge Box:
 - Cartridges 25
 - ID Chip 1
 - Instruction For Use 1
- Box containing Detection Buffer tubes
 - Detection Buffer Tubes 25
- Box containing Sample Diluent tubes
 - Sample Diluent Tubes 25

MATERIALS REQUIRED BUT SUPPLIED ON DEMAND

Following items can be purchased separately from **ichroma™ β-HCG**.

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™ β-HCG** 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. If longer storage is required, e.g. if the test could not be performed within 24 hours, serum or plasma should be immediately frozen below -20 °C. The freezing storage of sample up to 3 months does not affect the

quality of results

- However, the whole blood sample should not be kept in a freezer in any case.
- Once the sample was frozen, it should be used one time only for test, because repeated freezing and thawing can result in the change of test values.

TEST SETUP

- Check the contents of **ichroma™ β-HCG**: Sealed Cartridge, Detection Buffer Tubes, Sample Diluents and ID Chip.
- Ensure that the lot number of the cartridge matches that of the ID chip, sample diluent 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.)

TEST PROCEDURE

- 1) Transfer of sample (30 µL Human serum, plasma, control/50 µL whole blood) using a transfer pipette to a tube containing the detection buffer.
- 2) 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.)
- 3) Pipette out 75 µL of a sample mixture and load it into the sample well on the cartridge.
- 4) Leave the sample-loaded cartridge at room temperature for 15 minutes.
⚠ scan the sample-loaded cartridge immediately when the incubation time is over. If not, it will cause inexact test result.
- 5) 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.
- 6) Press 'Select' button on the instrument for ichroma™ tests to start the scanning process.
- 7) Instrument for ichroma™ tests will start scanning the sample-loaded cartridge immediately.
- 8) Read the test result on the display screen of the instrument for ichroma™ tests.

*When the concentration of a sample is higher than 50,000 mIU/mL, it can be diluted with a diluent provided.

TEST PROCEDURE-USING SAMPLE DILUENT

- 1) Transfer 30 µL of sample (Human whole blood/serum/plasma/control) to a sample diluent tube provided with pipette.
- 2) Close the lid and mix the sample thoroughly by shaking it about 10 times.
- 3) Transfer of dilution sample (30 µL Human serum, plasma, control/50 µL of whole blood) using a transfer pipette to a tube containing the detection buffer
- 4) 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.)
- 5) Pipette out 75 µL of a sample mixture and load it into the sample well on the cartridge.
- 6) Leave the sample-loaded cartridge at room temperature for 15 minutes.
△ can the sample-loaded cartridge immediately when the incubation time is over. If not, it will cause inexact test result.
- 7) 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.
- 8) Press 'Select' button on the instrument for ichroma™ tests to start the scanning process.
- 9) Instrument for ichroma™ tests will start scanning the sample-loaded cartridge immediately.
- 10) 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 hCG concentration of the test sample in terms of mIU/mL.
- The cut-off (reference value): 20 mIU/mL
- Total β-HCG level during pregnant stage

pregnant women (weeks since LMP*)	Total βhCG level [mIU/mL] range
3	5 - 50
4	5 - 426
5	18 - 7,340
6	1,080 - 56,500
7 - 8	7,650 - 229,000
9 - 12	25,700 - 288,000
13 - 16	13,300 - 254,000
17 - 24	4,060 - 165,400
25 - 40	3,640 - 117,000

* LMP is the last menstrual periods date from the first day of your last period

※The hCG levels different for each person, so the result should be consulted by a doctor about the pregnancy cycle.

※To confirm the exact pregnancy, retest 2-3 days after the first measurement.

- Working range: 5-50,000 mIU/mL
- In case of the test is performed with sample dilution procedure, please follow the below equation to obtain

correct result.

[Final Sample Concentration = Reported Concentration x 10]

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™ β-HCG. 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 L-Ascorbic acid, hemoglobin, triglycerides, cholesterol, glucose, bilirubin, heparin were shown as below for interference study. But this doesn't interfere with the ichroma™ β-HCG test measurements, nor occurs any significant interference.

Interfering Substance	Concentration added
L-Ascorbic acid	3 mg/dL
hemoglobin	500 mg/dL
triglycerides	1000 mg/dL
cholesterol	70 mg/dL
glucose	120 mg/dL
bilirubin	20 mg/dL
heparin	143 U/mL
EDTA	15 mg/L

- **Precision:** The intra-assay precision was calculated by one evaluator, who tested different concentration of control standard ten times each with three different lots of ichroma™ β-HCG. The inter-assay precision was confirmed by 4 different evaluators with 3 different lots, testing ten times each different concentration.

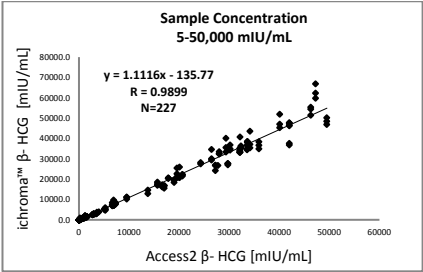
Whole blood (mIU/mL)	Intra assay			Inter assay	
	Mean (mIU/mL)	CV (%)		Mean (mIU/mL)	CV (%)
7.5	6.9	22.1		6.9	21.0
15.1	15.9	16.8		15.7	14.9
178.4	189.4	3.6		191.8	4.4
2,039	2038.4	5.7		1972.4	4.8
22,322	22680.4	7.1		22775.6	5.3
35,416	34019.5	4.4		33519.2	4.0
98,563*	100845.8	3.8		102486.5	4.6
225,491*	226324.4	4.6		230127.8	6.0

Serum (mIU/mL)	Intra assay			Inter assay	
	Mean (mIU/mL)	CV (%)		Mean (mIU/mL)	CV (%)
* 195,560	192,890.8	6.8		193,959.2	6.6
* 155,400	151,555.1	5.7		159,474.5	4.9
* 77,700	77,064.8	6.0		76,175.9	6.8
38,850	39,470.4	5.9		41,666.8	7.2

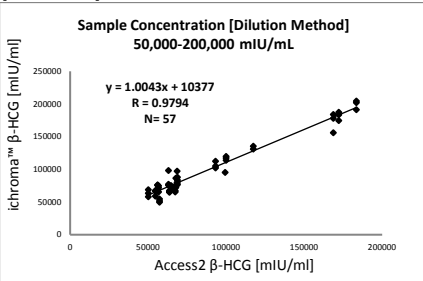
12,007	10,431.0	3.8	10,490.6	6.3
1,214	1,238.2	2.4	1,222.3	4.2
152	157.2	3.4	157.6	6.3
19	17.8	8.2	17.4	9.0
5	4.8	10.7	4.7	13.5

* For samples with the concentration of higher than 50,000 mIU/mL, they were diluted with diluent as described above.

■ **Comparability:** hCG concentrations of 284 serum samples were quantified independently with **ichroma™ β-HCG** and Beckman Coulter Access2 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 $Y = 1.1116X - 135.779$ and $R=0.9899$ respectively at the test without dilution. Linear regression and coefficient of correlation between the two tests were $y = 1.0043x + 10377$ and $R = 0.9794$ respectively at the test with dilution.



[With dilution]



* The values were obtained with dilution as described above.

REFERENCES

1. Braunstein GD, JL Vaitukaitis, PP Carbone, GT Ross
“Ectopic production of human chorionic gonadotropin by neoplasms”, Ann. Intern Med. 1973; 78(1): 39-45.
2. Steier JA, P Bergsjo, OL Myking “Human chorionic gonadotropin in maternal plasma after induced abortion, spontaneous abortion and removed ectopic pregnancy”, Obstet. Gynecol. 1984; 64(3): 391-394.
3. Lenton EA, LM Neal, R Sulaiman “Plasma concentration of human chorionic gonadotropin from the time of implantation until the second week of pregnancy”, Fertil.

Steril. 1982; 37(6): 773-778.
4. Batzer FR. “Hormonal evaluation of early pregnancy”, Fertil. Steril. 1980; 34(1): 1-13

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
	This product fulfills the requirements of the Directive 98/79/EC on in vitro diagnostic medical devices

