BIO TECHNOLOGY boditech

Diabetes

ichroma™ Microalbumin

INTENDED USE

ichroma™ Microalbumin is a fluorescence Immunoassay (FIA) for the quantitative determination of Microalbumin in human urine. It is useful as an aid in management and monitoring of determination of kidney damage from diabetes.

For *in vitro* diagnostic use only.

INTRODUCTION

A Microalbumin test evaluates urine for the presence of a protein called albumin[1]. Albumin is normally found in the blood and filtered by the kidneys[2]. When the kidneys are working properly, albumin is not present in the urine. However, when the kidneys are damaged, small amounts of albumin leak into the urine. This condition is called Microalbumin[1, 2,3,4].

Microalbumin is most frequently caused by kidney damage from diabetes. However, many other conditions can lead to kidney damage, such as high blood pressure, heart failure, cirrhosis, or systemic lupus erythmatosus(SLE). If kidney damage is not treated at an early stage, larger amounts of albumin and protein may leak into the urine[5,6]. This condition is called macroalbuminuria or proteinuria. When the kidneys spill protein, it can mean serious kidney damage is present. This can lead to chronic kidney disease. A microalbumin urine test can be done on a sample of urine collected randomly (usually after the first time you urinate in the morning), a sample collected over a 24-hour period, or a sample collected over a specific period of time, such as 4 hours or overnight[7].

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 microalbumin concentration in the sample.

COMPONENTS

ichroma™ Microalbumin consists of 'cartridges', 'detector tubes', and 'detector diluents.'

- The cartridge contains the membrane called a test strip which has anti human microalbumin at the test line, human serum albumin 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 contains a granule containing anti human microalbumin-fluorescence conjugate, anti-chicken IgY-fluorescence conjugate, anti-human microalbumin, bovine serum albumin (BSA) as a stabilizer and sodium azide as a preservative in potassium phosphate buffer. All detector tubes are packed in a pouch.
- The detector diluent contains sodium azide as preservative in potassium phosphate buffer, and it is pre-dispensed in tubes. The detector diluents are packed in a box.

WARNINGS AND PRECAUTIONS

- For *in vitro* diagnostic use only.
- Follow 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, detector tubes, or detector diluents. A cartridge should be used for testing one sample only. A detector tube should be used for testing one sample only. A detector diluent 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.
- 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™ tests may generate slight vibration during use.
- Used cartridges, detector tubes, detector diluents 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 (NaN3), 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™ Microalbumin** will provide accurate and reliable results subject to the below conditions.
- **ichroma™ Microalbumin** should be used only in conjunction with instrument for ichroma™ tests.

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 the 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.

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	Disposable

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

MATERIALS SUPPLIED

REF i-CHROMA MAU-25

Components of **ichroma™ Microalbumin**

- Cartridge Box:
  - Cartridge 25
  - Detector tube 25
  - ID chip 1
  - Instruction for use 1
- Box containing detector diluent
  - Detector diluent 25

MATERIALS REQUIRED BUT SUPPLIED ON DEMAND

Following items can be purchased separately from **ichroma™ Microalbumin**.
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
- **Boditech MAU Control** REF CFPO-4

SAMPLE COLLECTION AND PROCESSING

The sample type for **ichroma™ Microalbumin** is human urine.

- It is recommended to test the sample within 24 hours after collection.
- The samples(urine) may be stored for up to two days at 2-8 °C prior to being tested. If testing will be delayed more than two days, samples should be frozen at -20 °C.
- Samples stored frozen at -20 °C for 8 weeks showed no performance difference
- As a repeated freeze-thaw cycle may affect the test result, do not refreeze previously frozen samples.

TEST SETUP

- Check the contents of **ichroma™ Microalbumin**: Sealed cartridges, detector tubes, detector diluents, ID chip and instruction for use.
- Ensure that the lot number of the cartridge matches that of the detector tube, detector diluent, as well as an ID chip.
- If the sealed cartridge, the detector tube and the 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.
- 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™ Reader, ichroma™ II, ichroma™ M3**

Multi test mode

1) Take 10 µL (Human urine/control) of sample using a pipette and dispense it to the detector diluent tube.
2) Close the lid of the detector diluent tube and mix the sample thoroughly by shaking it about 10 times.
   (The sample mixture must be used immediately. Do not exceed 1 minute.)
3) Take 150 µL of the sample mixture using a pipette and dispense it to the detector tube. When the granule form is completely dissolved in the tube, it becomes detection buffer.
   (The detection buffer must be used immediately. Do not exceed 1 minute.)
4) Take 75 µL of the 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 microalbumin concentration of the test sample in terms of mg/L.
- Reference value: 18 mg/L
- Working range : 2-300 mg/L

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™ Microalbumin**. 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**
  - Limit of Blank (LoB) 0.455 mg/L
  - Limit of Detection (LoD) 0.931 mg/L
  - Limit of Quantification (LoQ) 2 mg/L
- **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. **ichroma™ Microalbumin** test results did not show any significant cross-reactivity with these biomolecules.

Cross reactivity materials	Conc.
CEA	500 μg/mL
PSA	50 μg/mL
AFP	500 μg/mL
ALT	500 μg/mL
Troponin I	500 μg/mL
CRP	500 μg/mL
Myoglobin	500 μg/mL

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

Interference materials	Conc.
Creatinine	442 μmol/L
L-ascorbic acid	298.31 μmol/L
Bilirubin, unconjugated	684 μmol/L
D-glucose	55 mmol/L
Urea	42.9 mmol/L
Hemoglobin	2 g/mL

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

Conc. [mg/L]	Repeatability			Total precision		
	Mean (mg/L)	SD	CV (%)	Mean (mg/L)	SD	CV (%)
4.5	4.33	0.41	9.5	4.36	0.42	9.7
100	98.28	8.91	9.1	99.55	9.50	9.5
258	256.95	23.29	9.1	256.27	22.00	8.6
Conc. [mg/L]	Lot to lot precision			Reproducibility		
	Mean (mg/L)	SD	CV (%)	Mean (mg/L)	SD	CV (%)
4.5	4.43	0.43	9.8	4.55	0.41	8.91
100	99.78	8.78	8.8	100.45	6.66	6.63
258	258.66	22.50	8.7	259.75	17.76	6.84

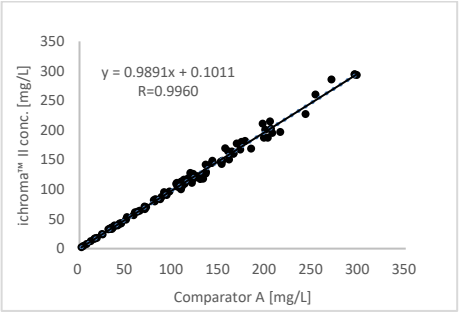
- **Accuracy**

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

Expected value (mg/L)	Lot 1	Lot 2	Lot 3	AVG	Recovery (%)
4.5	4.33	4.54	4.48	4.45	98.9
23.0	23.31	23.09	22.38	22.92	99.7
50.0	48.65	49.56	50.58	49.60	99.2
100.0	100.36	101.12	97.05	99.51	99.5
180.0	185.25	185.58	179.04	183.29	101.8
258.0	261.14	254.57	260.74	258.82	100.3

■ **Comparability**

Microalbumin concentrations of 100 urine samples were quantified independently with **ichroma™ Microalbumin (ichroma™ II)** and Comparator A as per prescribed test procedures. Test results were compared, and their comparability was investigated with linear regression and coefficient of correlation (R). The regression equation and correlation coefficient are as



REFERENCES

1. Rowe DJF, Dawnay A, Watts GF. Microalbumin in diabetes mellitus: review and recommendations for the measurement of albumin in urine. Ann Clin Biochem 1990; 27: 297-312.
2. Doumas BT, Peters T. Serum and urine albumin: a progress report on their measurement and clinical significance. Clin Chim Acta 1997; 258:3-20.
3. Mogensen CE. Microalbumin, a marker for organ damage. 1993. London: Science Press.
4. Waugh J, Kilby M, Lambert P, Bell SC, Blackwell CN, Shennan A, et al. Validation of the DCA 2000 microalbumin:creatinine ratio urinanalyzer for its use in pregnancy and preeclampsia. Hypertens Pregnancy 2003; 22(1): 77-92.
5. Mogensen CE, Christnesen CK. Predicting diabetic nephropathy in insulin dependent diabetes. New Eng J Med 1984; 311:89-93.
6. Viberti GC, Hill RD, Jarrett RJ. Microalbumin as a predictor of clinical nephropathy in insulin dependent diabetes mellitus. Lancet 1982;I: 1430-2.
7. Mathiesesen ER, Ronn B, Jensen T, and Deckert, T. Relationship between blood pressure and urinary excretion in the development of microalbuminurea. Diabetes 1990; 39:245-9.
8. Brooks DE, Devine DV, Harris PC, et al. RAMP(TM): A rapid, quantitative whole blood immunochromatographic platform for point-of-care testing.Clin Chem 1999;45:1676-1678.
9. Oh SW, Moon JD, Park SY, et al. Evaluation of fluorescence hs-CRP immunoassay for point-of –care testing. Clin Chim Acta 2005; 356:172-177.

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