**RENISCHEM® L-FABP POC Kit**

A kit for the detection and semi-quantitative determination of human L-FABP in urine.

**INTENDED USE**

RENISCHEM® L-FABP POC Kit is capable of the detection and semi-quantitative determination of human L-FABP in urine. It is intended to surveise diagnosis of renal disease accompanied by renal tubule dysfunction.

**INTENDED USER**

Lab Professionals

**KIT COMPONENTS**

<table>
<thead>
<tr>
<th>SYMBOL</th>
<th>Name</th>
<th>Quantity</th>
<th>Chief ingredients</th>
</tr>
</thead>
<tbody>
<tr>
<td>TC</td>
<td>L-FABP Test Cassette</td>
<td>x 1</td>
<td>L-FABP mouse monoclonal antibody (Clone2), Gold colloidal anti-human L-FABP mouse monoclonal antibody (Clone1), forms an antigen-antibody complex, move through the membrane by a capillarity action, and reacts to the anti-human L-FABP mouse monoclonal antibody on the Test Line (Clone2), visualizing a burgundy color. <strong>RENISCHEM®</strong></td>
</tr>
<tr>
<td>PT</td>
<td>Pre-treatment Micro Tube</td>
<td>x 1</td>
<td>Pre-treatment reagent (blue-colored reagent)</td>
</tr>
<tr>
<td>DP</td>
<td>Dropper</td>
<td>x 1</td>
<td>Disposable plastic pipette</td>
</tr>
<tr>
<td>DS</td>
<td>Dry sheet</td>
<td>x 1</td>
<td>Drying reagent</td>
</tr>
</tbody>
</table>

All kit components are pre-packed into aluminum pouch (10 units/box) which each contains the following components in sufficient quantities for 10 individual determinations.
- 10 individual test kits
- 10 reference cards
- 1 Instructions For Use

**SUMMARY AND EXPLANATION OF THE TEST**

L-FABP is expressed as a low-molecular-weight soluble protein of approximately 14 kDa in a proximal tubule, which is responsible for reabsorption function in a kidney, playing an important role in metabolism of energy and lipid. 1. L-FABP is excreted to the urine in response to proteinuria observed by the progression of kidney disease or ischemic and oxidative stresses. 2, 3. L-FABP can be detected in the early stage of the progression of kidney disease, although most of biomarkers reflect the result of failure of kidney function 3, 4. Many of clinical trials using the CE-marked product “RENISCHEM™” report that excretion levels of L-FABP into urine can reflect the degree of progression of diabetic nephropathy and L-FABP, the diagnostic index is a highly sensitive biomarker for kidney disease 5-9.

**PRINCIPLE**

**RENISCHEM®** L-FABP POC Kit is an immunochromatographic test for detection and semi-quantitative determination of L-FABP. 

L-FABP levels in the urine samples is affected by the presence and severity of renal disease, and L-FABP, the diagnostic index is a highly sensitive biomarker for kidney disease.

**L-FABP POC Kit** allows semi-quantitative measurement by comparing the color intensity of Test Line with that of reference card. Additionally, the gold colloidal anti-human L-FABP mouse monoclonal antibody (Clone1) moves further on the membrane and binds with the anti-mouse IgG polyclonal antibody on the Control Line, visualizing a burgundy color. This line is an index for checking that the test is performed properly.

**WARNINGS AND PRECAUTIONS**

**General Precautions**

1. The user should read Instructions For Use and keep it if there is a need.

2. This kit is intended to be used for in vitro diagnostic use only. Do not use for any other purpose.

3. Comprehensive diagnosis should be made results of lab tests, consideration of clinical symptoms, etc., in addition to the result of the measurement by this kit.

4. Components stored under conditions over 30°C or below 1°C may not perform properly and may adversely affect the assay result.

5. Do not interchange reagents or L-FABP Test Cassette from any different production lots.

**Handling Cautions**

1. Wear disposable protective gloves while handling reagents and specimen. Do not pipette by mouth.

2. Some reagents contain a substance derived from animals. Wash hands thoroughly after measurement procedures.

3. If reagent gets into eyes or mouth, take necessary emergency procedures, such as washing hands with water, and seek doctor’s treatment as needed.

4. Take care to avoid injury with aluminum pouch when opening.

5. This product may jump out of the aluminum pouch if it is pulled too hard when opening.

**Operating Precautions**

1. Follow the operation method described Instructions For Use when using the product.

2. Use this product at 10-25°C.

3. Be sure to use the reference card enclosed in the kit for result assessment.

4. Do not mix/replace any components, including reference card, with/from that of other kits or any kits from different production lots.

5. Use L-FABP Test Cassette as quickly as possible after opening aluminum pouch.

6. Handle this product and specimen under the conditions described in Instructions For Use. The measurement result may be affected by reaction time and temperature.

7. No visible line on the Control line indicate specimen failed to be reacted correctly. In the case, confirm operation method and perform a retest.

**Disposal Cautions**

1. Dispose of reagents and accessories in compliance with local authorities requirements.

2. Specimen should be considered potentially infectious. Dispose of used instruments (e.g. pipettes, tubes), waste solution and sampling tips after decontamination according to the following methods:

   - Soak in 0.05% of formalin solution at 37°C for 72 hours or more.
   - Soak in 2% of glacial acetic acid solution for 1 hour or more.
   - Autoclave at a temperature of 121°C for 20 minutes.

**STORAGE AND STABILITY**

1. Store this kit at 1 - 30°C. Do not freeze.

2. Performance of the kit is quality assured until the expiry date shown on the label, provided that it is unopened and stored under correct conditions.

3. Do not use reagents which are already expired or stored in frozen condition.

**SPECIMEN COLLECTION**

1. Urine should be collected in a clean and dry sample container.

2. Measure the specimen immediately after urine collection. When measuring the specimen that has been stored, measure it within two days after being put in cold storage or when the specimen is frozen (-30 to -20°C), do not repeat freezing and thawing the specimen. In addition, the specimen frozen is to be thawed, leave it under room temperature or in a water bath (2 to 28°C), and mix it sufficiently prior to measurement. Specimen frozen at -80°C is verified to be stable for 1 year.

3. Apply the specimen mixed with the Pre-treatment reagent immediately to the L-FABP Test Cassette.

4. If the reagent has a foreign body, high viscosity, or deep coloration, the result of assessment may not be accurate.

5. Use the specimen with the Pre-treatment reagent slowly to avoid bubbling as much as possible. In the case that the mixture with the Pre-treatment reagent shows excessive bubbling, the amount of specimen may be insufficient.

6. Urine collection with added hydrochloric acid may affect measurement results. Urine collection with added sodium does not affect measurement results.

**INTERFERING SUBSTANCES**

1. Lactate dehydrogenase, hemoglobin, glucose or ascorbic acid in urine specimen in the following ranges can interfere with assay performance. However the following levels can be present in the urine with no effect on assay value:

   - Conjugated bilirubin up to 19.7 mg/dL
   - Conjugated bilirubin up to 19.7 mg/dL
   - Conjugated bilirubin up to 21.8 mg/dL
   - Hemoglobin up to 24.8 mg/dL
   - Glucose up to 1000 mg/dL
   - Ascorbic acid in urine up to 1.6 mg/mL
   - Acid urine below pH 5 may affect measurement result.

**TEST PROCEDURE**

**Instruments and Equipment Required**

Multichannel micropipette: adjustable to 100μL.

**Preparation**

1. Open aluminium pouch and place L-FABP Test Cassette on a flat table.

2. Check that the cap of the Pre-treatment Micro Tube containing Pre-treatment reagent (blue solid) is closed tightly.

3. Check that the Pre-treatment reagent is at the bottom of the tube. When the Pre-treatment reagent is not at the bottom of the tube, hold the middle part of the tube and shake lightly down to move the Pre-treatment reagent to the bottom of the tube.

4. Prepare urine sample stored in collection cup.

**Operation**

1. Take 100μL of urine sample (fill to the line of the Dropper) and add to Pre-treatment Micro Tube (See Illustration 1).

2. Mix the sample with the Pre-treatment reagent slowly 10 times by using Dropper to avoid bubbling as much as possible (See Illustration 2).

3. Take 100μL of the mixed sample (fill to the line of the Dropper). Place the mixed sample (100μL) into L-FABP Test Cassette (See Illustration 3).

4. Keep L-FABP Test Cassette flat and stable. After 15 minutes, read Test Line of L-FABP Test Cassette. The L-FABP concentration range of the sample is determined.

5. Do not read Test Line shown on L-FABP Test Cassette which has been left for more than 15 minutes following addition of sample.

**ASSESSMENT PROCEDURE**

The L-FABP concentration range is determined by comparing the color intensity of the Test Line with the color blocks of the reference card (See Illustration 4). It is necessary to use the reference card supplied with the kit. Comparing to color density printed on the reference card, tick appropriate box of the L-FABP concentration level which is close to the color density displayed on the Test Line of the L-FABP Test Cassette.
CLINICAL IMPLICATIONS

1. Background of development and usefulness in clinical diagnosis

CMIC HOLDINGS Co., Ltd. developed the CE-marked product “RENISCHEM® L-FABP TMB Kit” as the world-first in vitro diagnostics that are based on enzyme-linked immunosorbent assay (ELISA) method. Moreover, the usefulness of urinary L-FABP has also been recently indicated in the international guideline and review papers in acute kidney injury (contrast medium-induced nephropathy, cardiogenic shock, renal transplantation, hemostatic stem cell transplantation, sepsis, the area of intensive care including newborns) and the establishment of POC (point of care) assay that swiftly evaluates prognosis of cardio-renal syndrome and the area of intensive care is expected. On these backgrounds, we developed RENISCHEM® L-FABP POC Kit, which is based on immunochromatographic method. This product can measure urinary L-FABP easily at bedside and gives a negative result under the reference values determined by the clinical trial data for “RENISCHEM®,” which has already been approved result score makes it possible to diagnose acute kidney injury swiftly or to estimate the degree of severity.

2. Range of reference value

The range of reference value, which are determined by the distribution of urinary L-FABP corrected by urinary creatinine levels in 431 healthy adults in the clinical performance trial of the CE-marked product, “RENISCHEM®,” is 8.4 μg/gCr or below. The range of reference value of urinary L-FABP concentration, which is not corrected by urinary creatinine levels, is 10.1ng/mL or below.

3. Accuracy of diagnosis

As shown in Table 1 and Figure 1 the true negative ratio to the range of reference value (below 8.4 μg/gCr) of the CE-marked product was 100% (38/38), and the true positive ratio beyond the upper limit of the range of reference value was 66% (28/43).

4. Result of correlation test

When the correlation between this product and the CE-marked product “RENISCHEM® L-FABP TMB Kit” was evaluated, the following excellent correlation was obtained.

<table>
<thead>
<tr>
<th>Concentration range</th>
<th>RENISCHEM® L-FABP PoC Kit</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;12.5</td>
<td>&lt;12.5</td>
<td>68</td>
</tr>
<tr>
<td>≥12.5, &lt;100</td>
<td>≥12.5, &lt;100</td>
<td>36</td>
</tr>
<tr>
<td>≥100</td>
<td>≥100</td>
<td>31</td>
</tr>
</tbody>
</table>

Concordance rate 91.9% (124/135)