Intended use
The Wako Creatinine M L-Type reagent is an in vitro assay for the quantitative determination of creatinine in serum, plasma or urine.

Summary and explanation of the test
Creatinine is produced directly from creatine phosphate or by the dehydration of creatine in the muscles and nerves. The amount of metabolically produced creatinine in the urine is conveniently used to test glomerular function. Therefore, creatinine measurement is one of the essential clinical tests in the diagnosis of uremia and renal diseases, such as renal insufficiency and nephritis, and in monitoring renal diseases. The enzymatic methods with high specificity are widely used to measure creatinine. Creatinine M L-Type is a reagent kit for creatinine assay, based on the enzymatic method employing creatininase, creatinase, sarcosine oxidase and N-(3-sulfopropyl)-3-methoxy-5-methylxylamine (HMMPS) as a new color agent.

Principle of the method
When a sample is mixed with R1 and R2, creatinine in the sample is converted to creatine by the action of creatininase. The creatine formed is hydrolyzed by creatinase to produce sarcosine and urea. The sarcosine produced is then decomposed by sarcosine oxidase to form glycine, formaldehyde and hydrogen peroxide. In the presence of peroxidase (POD), the hydrogen peroxide formed yields a blue pigment by quantitative oxidation condensation with N-(3-sulfopropyl)-3-methoxy-5-methylxylamine (HMMPS) and 4-aminoantipyrine (4-AA). The creatinine concentration is obtained by measuring the absorbance of the blue color.

Reactions
Creatinine + H2O \rightarrow Creatininase \rightarrow Creatine
Creatine + H2O \rightarrow Creatininase \rightarrow Sarcosine + Urea
Sarcosine + H2O + O2 \rightarrow Sarcosine oxidase \rightarrow Glycine + Formaldehyde + H2O2
POD + H2O2 + 4-aminoantipyrine + HMMPS \rightarrow Blue pigment

Specimen collection and preparation
Serum, plasma or urine can be used as a specimen for testing. Urine samples should be collected without a preservative.

Physical or chemical indications of instability
The presence of precipitates in the reagents or values of control sera outside the manufacturer’s acceptable range may be an indication of reagent’s instability.

Standard procedure
Temperature: 37°C (Hach/Pf17)
Sample blank
Measurement

Results
The final results are automatically calculated and printed in concentration. The results are given in mg/dL. Always use the same unit for the calibrator.

Expected values
Serum: Men: 0.55 - 1.10 mg/dL (49 - 97 µmol/L), Women: 0.47 - 0.90 mg/dL (42 - 80 µmol/L).

Performance characteristics
(1) Accuracy
When a sample of known concentration is assayed, the assay value falls within the range of ± 10% of the known concentration.

(2) Sensitivity
a) When saline is assayed, the absorbance is not more than 0.02.
b) When a calibrator of given concentration (creatinine 5 mg/dL) is assayed, the absorbance is 0.01 - 0.11.

(3) Precision
When a sample is assayed 5 times in a run, CV is not more than 5%.

(4) Measurable range
0.05 - 100 mg/dL (0.10 - 100 µmol/L) Creatinine (In the case of using the standard procedure).

Correlation
Specimen: Serum, Plasma, Urine
Correlation coefficient
Regression equation
y = Wako Creatinine M L-Type (Creatinase - HMMPS method, mg/dL)
Wako Creatinine F L-Type (Creatinase - F DAOS method, mg/dL)

Manufactured by:
Wako Chemicals GmbH
Fuggerstraße 12, D-41468 Neuss
Telephon(e): +49-2131-311-0
Facsimile: +49-2131-311-100
URL: www.wako-chemicals.de
Interfering substances
a) Do not use sodium azide as an additive. It gives positive interference.
b) Anticoagulants such as heparin, citrate, oxalate and EDTA, and sodium fluoride as a glycolytic inhibitor, have no influence on the assay when they are used in their usual quantities.
c) Hemolysis, ascorbic acid and bilirubin do not have significant effects on the assay.

Warnings and precautions
- For in vitro diagnostic use.
- The usage and application of this test is reserved for professional use only. Please refer to respective national and local regulations and legislation.
- Not to be used internally in humans or animals.
- Do not use the reagents described above for any purpose other than described herein. Performance cannot be guaranteed if the reagents are used in other procedures or for other purposes.
- Operate the instruments according to operator’s manuals under appropriate conditions. Consult the instrument manufacturer for details.
- When using enzymatic methods for the determination of cholesterol esters, contamination and interference to other clinical chemistry assays on the same instrument in principle cannot be excluded. In the rare event of such a problem occurring, please refer to the instrument’s manual for channel setting and washing procedure options.
- Clinical diagnosis must be determined with clinical symptoms and other test results by a physician.
- URINE: High concentrations of homogentisic acid in urine (alkaptonuria) can lead to falsely decreased results.

Overdose of Paracetamol (Acetaminophen): Intravenous treatment with N-Acetylcysteine can lead to falsely decreased results.
- Store the reagents under the specified conditions. Do not use reagents past the expiration date stated on each reagent container label.
- Do not use reagents which were frozen in error. Such reagents may give false results.
- After opening the reagents, it is recommended to use them immediately. When the opened reagents are stored, cap the bottles and keep them under the specified conditions.
- Do not use the containers and other materials in the package for any purposes other than those described herein.
- If the reagents come in contact with the mouth, eyes or skin, wash off immediately with a large amount of water. Consult a physician if necessary.
- When discarding the reagents, dispose of them according to local or national regulations.
- All the devices including reagents and reagent bottles contacted with specimen should be considered potentially infectious.
- This assay should not be used as the sole determinant for clinical diagnosis.
- R2 contains 0.095% sodium azide as a stabilizer. Sodium azide may react with lead or copper plumbing to form explosive compounds. Even though the reagent contains minute quantity of sodium azide, drains should be flushed well with a large amount of water, when discarding the reagents.
- Separate instrument application is available for the measurement of urinary creatinine.

Quality control
A quality control program is recommended for all clinical laboratories.

References

Ordering information

<table>
<thead>
<tr>
<th>Code No.</th>
<th>Products</th>
<th>Package</th>
</tr>
</thead>
<tbody>
<tr>
<td>419-08199</td>
<td>Creatinine M L-Type R1</td>
<td>R1: 4 x 70 mL</td>
</tr>
<tr>
<td>419-08299</td>
<td>Creatinine M L-Type R2</td>
<td>R2: 4 x 70 mL</td>
</tr>
</tbody>
</table>