Creatinine Plus
Jaffé

Kinetic-Colorimetric Assay
Store at 2 - 8 °C.
« in vitro » use only

INTRODUCCIÓN

Creatinine is a final product of muscular metabolism resulting from the loss a water molecule by creatine. On the other hand, creatine is formed by the hydrolysis of phosphocreatine by phosphocreatinase (CPK), with activated phosphorous and creatine as metabolites of said reaction. The phosphate radical can contribute energy directly to the reaction or by coupling with a molecule of ADP to form ATP which is later hydrolyzed by ATPase.

In humans, creatinine is eliminated exclusively through glomerular filtration and is therefore an important indicator of kidney function. As opposed to urea, the elimination of creatinine through the urine is not accompanied by diuresis. Although a person’s daily secretion of urine is constant, it does not depend on the food ingested but rather on the muscle mass, which is the greatest conditioning factor of a person’s total daily urine excretion. In all, it is possible to affirm that the elimination creatinine during an interval of 24 is a very constant value which depends principally on the individual's muscle mass. Calculation of creatine clearance is a direct parameter of renal function.

USE PROPOSED

The method is valid for determining creatinine clearance in human sera.

MATERIALS

Micropipettes
Photometric equipment capable of reading up to 492 nm (490-510) or of greater quality (programmable or automatic)

FUNDAMENT OF THE METHOD

The chemical reaction used in photometry was described by Jaffé, based on the color (orange) of the solution following the reaction of creatinine with alkaline picrate. There are a number of substances both in the serum and in the urine that behave as non-specific chromogens. This can be a problem especially when calculating clearance, hence the importance of adjusting all of the reaction variables, especially the pH, in order to obtain maximum sensitivity to creatinine and minimum interference by the chromogens. By adapting the reaction to a kinetic value, it is possible to achieve high specificity since creatinine reacts with alkaline picrate more quickly than with the chromogens (methyl guanidine, picramate, etc.). Consequently, by measuring the increase in color during a short initial reaction time it is possible to obtain creatinine values with little influence of the chromogens. That is why we recommend, if possible, the determination of kinetic parameters.

CONTENT

<p>| | |</p>
<table>
<thead>
<tr>
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</thead>
<tbody>
<tr>
<td>R1 Picric Acid</td>
<td>Picnic acid</td>
</tr>
<tr>
<td>Creatinine</td>
<td>Sodium</td>
</tr>
<tr>
<td>R2 Sodium hydroxide</td>
<td>Sodium hydroxide</td>
</tr>
<tr>
<td>Creatinine</td>
<td>Creatinine</td>
</tr>
</tbody>
</table>

CALIBRATION:

The Standard has been assessed against a Reference Standard BCR-574 de IVMM/BCR

PREPARATION AND STABILITY

The reagents are ready to use. They will remain stable until the date of expiry indicated on the label. Mix both reagents, R1 and R2, in equal portions and according needs. This mixture will remain stable at room temperature during a period of 10 days.

SAMPLE

Human serum.

The creatinine in the serum will remain stable for at least 24 hours at 2 - 8 °C.
**PROCEDURE**

<table>
<thead>
<tr>
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<th>Standard</th>
<th>Sample</th>
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<tbody>
<tr>
<td>Sample</td>
<td>--</td>
<td>100 µL</td>
</tr>
<tr>
<td>Standard</td>
<td>100 µL</td>
<td>--</td>
</tr>
<tr>
<td>Reagents</td>
<td>1.0 mL</td>
<td>1.0 mL</td>
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Mix the reagents and start the timer. Note Abs. after 30 seconds (A₁) and after 90 seconds (A₂). Read at 492 nm (490-510).

**CALCULATION**

\[
\text{µmol/L Creatinine} = \frac{(A₂-A₁) \text{ Sample}}{(A₂-A₁) \text{ Standard}} \times \text{ Standard Conc.}
\]

\[
\text{µmol/L} \times 0.0113 = \text{mg/dL}
\]

**FUNCTIONAL CHARACTERISTICS**

The most important functional characteristics of the product are the following:

1- **Linearity:** 2200 µmol/L

2- **Inaccuracy:** Reproducibility during assay:
   Variation coefficient < 5%

3- **Recovery (Accuracy):** 80 – 120 %

4- **Parallelism:**

   The trial was satisfactory indicates a lack of systematic error in the assay (0.988x – 11.76).

   The reagents do not exhibit significant differences when compared to other commercial reagents.

   The following results were obtained in 42 samples:

   Regression Straight Line Equation: \( y = 0.94x + 4.16 \)

The characteristics of the method can vary according to the analyzer used.

**REFERENCE VALUES**

<table>
<thead>
<tr>
<th>µmol/l</th>
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<tr>
<td>61.8 – 132.6</td>
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</table>

**NOTES**

Uric acid, ascorbic acid, glucose, ketone and cephalosporin antibiotics, when present in high concentrations can interfere with the assay giving rise to false high values.

Do not use lipemic sera.

**PRECAUTIONS**

Contains small amounts of sodium azide.

Avoid contact with irritated skin and mucosa.

**REFERENCE**


**QUALITY CONTROL**

SPINTROL. Normal and pathological. Assessed controlled sera.

**PRESENTATION**

<table>
<thead>
<tr>
<th>Code: 1001111</th>
<th>2 x 150 mL</th>
<th>300 Minimum Determinations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Code: 1001112</td>
<td>2 x 1000 mL</td>
<td>2000 Minimum Determinations</td>
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