Serum Creatinine Determination: Comparison between Colorimetric (Jaffé) and Enzymatic Methods in Renal Transplant Recipients

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ABSTRACT

Background: Serum creatinine is the most requested parameter in current practice in the context of functional exploration of the kidney. Its contribution to medical care is particularly decisive for kidney transplant patients. The aim of the study is to compare the colorimetric and enzymatic techniques available in our laboratory for the determination of creatinine in samples from kidney transplant patients.

Methods: Prospective study carried out at the Biochemistry laboratory of the University Hospital Center (CHU) Ibn Rochd in Casablanca Morocco, over a period of 8 months, carried out on a
series of 45 samples from kidney transplant patients who simultaneously benefited from a creatinine assay by colorimetric and enzymatic techniques. The samples were taken on dry tubes and analyzed on the Alinity® Abbott analyzer. The statistical study of correlation was made by the SPSS software with calculation of the p-value and the coefficient of Pearson (r).

**Results:** During the study period 45 samples were collected. Seventy percent of the prescriptions came from the Nephrology Department. The mean serum creatinine value by enzymatic technique was $39.04 \pm 11.7$ mg/l, and by colorimetric technique $38.24 \pm 12.4$ mg/l. The creatinine level by the two techniques was pathological in 69% of cases and normal in 31%. The correlation between the serum creatinine assay by enzymatic Versus colorimetric technique is positive with $r=0.994$ and $P$-value $<0.001$.

**Conclusion:** According to literature, colorimetric techniques are sensitive to interference with pseudochromogenic substances, while enzymatic techniques are standardized due to their specificity and performance but remain more expensive. In our study the correlation between the two techniques was positive with a correlation equation: $(y = 0.82 + 1\times x)$. According to our results, no significant difference between the two techniques was observed in our laboratory.

**Keywords:** Enzymatic creatinin; colorimetric creatinine; renal transplant recipient.

### 1. INTRODUCTION

Serum creatinine is the most requested parameter in current practice in the context of the exploration of renal function. Its contribution to medical care and clinical follow-up is decisive, particularly for kidney transplant patients [1].

The techniques for assaying this parameter are various. The current variability in serum creatinine measurements affects all equations for estimating Glomerular filtration rate (GFR), including the MDRD equation [1]. Accurate and precise measurements are necessary for more reliable estimation of GFR as a support for reliable clinical decision making.

Two major types of techniques widely described in the literature are the colorimetric method and the enzymatic method [2]. Several studies claim that enzymatic techniques measure serum creatinine with much less variability than Jaffe techniques when compared to reference values [3,4]. However, their high cost limits their use in the majority of laboratories.

In this context, we aimed to compare the two techniques available in our laboratory on samples from known kidney transplant patients.

### 2. MATERIALS AND METHODS

This is a prospective study carried out in the Biochemistry laboratory of the University Hospital Center (CHU) Ibn Rochd in Casablanca spread over a period of 8 months from May 2022 to January 2023, on a series of 45 blood samples from kidney transplant patients who benefited from serum creatinine assay by both colorimetric (Jaffé) and enzymatic techniques. The samples were taken on dry tubes and analyzed on the Alinity® Abbott analyzer.

The analysis of the data was carried out by the Excel software version 2016 and the correlation analysis by the SPSS software with calculation of the p-value and the Pearson correlation coefficient (r).

### 3. RESULTS

During the study period, 45 samples were collected, 33% were women and 67% men (Fig. 1).

The requests came from the Nephrology Department in 70% of cases and from the various medical departments, in 30% (Fig. 2).

The mean serum creatinine value by enzymatic technique was $39.04 \pm 11.7$ mg/l, and by colorimetric technique (Jaffé) $38.24 \pm 12.4$ mg/l (Table 1).

The creatinine level by the two techniques was pathological in 69% of cases and normal in 31% of cases.

The correlation between serum creatinine assay by colorimetric technique (Jaffé) Versus enzymatic; is positive with a Pearson correlation coefficient at $r = 0.994$ and a $P$-value $< 0.001$ with the correlation equation: $(y = 0.82 + 1\times x)$ (Fig. 3).
Fig. 1. Breakdown of requests by gender

Fig. 2. Breakdown of requests by department

Fig. 3. Linear regression curve = Correlation between serum creatinine assay by colorimetric (Jaffé) and enzymatic technique
Table 1. Mean values of serum creatinine determination by both colorimetric and enzymatic techniques

<table>
<thead>
<tr>
<th>Serum creatinine</th>
<th>Mean Value (mg/l)</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enzymatic technique</td>
<td>39,04</td>
<td>11,7</td>
</tr>
<tr>
<td>Colorimetric technique</td>
<td>38,24</td>
<td>12,4</td>
</tr>
</tbody>
</table>

4. DISCUSSION

Serum creatinine is one of the most prescribed laboratory tests in clinical practice. It is the main parameter used for the functional evaluation of the kidney to estimate the glomerular filtration rate [2,3].

Its contribution is decisive in the medical management allowing to ensure the follow-up and to adapt the treatment, particularly for the kidney transplant patients, requiring a reliable dosage.

There are different techniques for measuring creatinine [4-6]. Chromatography coupled with mass spectrometry with isotopic dilution (SM-DI) is the reference method; it is a cumbersome technique and not used routinely [7-9].

The two techniques routinely used in medical biology laboratories are: colorimetric and enzymatic.

Colorimetric methods based on the Jaffe reaction were described by Jaffé since 1886, its principle is based on the interaction between picrate and creatinine in an alkaline medium, which gives the red-orange “Janovsky” complex whose optical density measurement at 505 nm is proportional to the creatinine concentration [4,10]. This technique is the most used in current practice, characterized by its low cost and ease of use [4,6]. However, its main drawback is its lack of specificity, well-illustrated in the literature, linked to interference with so-called "pseudochromogenic" substances, in particular bilirubin, proteins and acetoacetate [11,12].

The general principle of enzymatic methods includes a succession of enzymatic reactions which give a colored final product and whose intensity of coloring is directly proportional to the concentration of creatinine. The most widespread enzymatic method uses the enzyme "creatininase" which degrades creatinine into creatine which is then converted into sarcosine by creatinase [13]. Then the enzyme sarcosine peroxidase transforms sarcosine into formaldehyde, glycine and hydrogen peroxide which will be quantified by a final enzymatic reaction which varies according to the suppliers [14].

Several studies have shown that enzymatic methods have less interference and excellent analytical performance, and are therefore the most recommended [5-16].

However, they are still little used, and are much more expensive than colorimetric methods [17,18], which has probably limited their use in analytical laboratories [9,8].

In our study the correlation between the two techniques was positive with a correlation equation: \( y = 0.82 + 1^*x \). It is very clear from these results that no significant differences between the two techniques were observed in our laboratory.

In 2009, Cobart et al. [19] clarified that according to Abbott Diagnostics, the new uncompensated Jaffe method is a modified version that exhibits negligible interference with bilirubin.

They add that hemolysis does not interfere in enzymatic methods, and only relatively slightly modifies the value of creatinine in the Jaffé method [9].

Delatour et al. [17] state that according to the results of the 2009 national quality control by principle of method, the bias does not exceed 2% for routine methods (colorimetric Jaffé type and enzymatic), and that their accuracy is very satisfactory.

5. CONCLUSION

Renal function is assessed by estimating the Glomerular filtration rate (GFR) obtained from the serum creatinine assay. This parameter has a valuable contribution in the diagnosis and monitoring of patients with renal insufficiency and particularly renal transplant recipients. Its accuracy depends on the analytical performance of the assay methods used.

According to literature, colorimetric techniques are sensitive to interference with
pseudochromogenic substances, while enzymatic techniques are standardized due to their specificity and performance but remain more expensive.

In our study, which concerns kidney transplant patients, no significant difference was observed. Thus, in these patients requiring close and regular long-term biological monitoring, the Jaffé colorimetric method provides an excellent contribution at a lower cost.

CONSENT
As per international standard or university standard, patient(s) written consent has been collected and preserved by the author(s).

ETHICAL APPROVAL
As per international standard or university standard written ethical approval has been collected and preserved by the author(s).

COMPETING INTERESTS
Authors have declared that no competing interests exist.

REFERENCES


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