Effect of ESRD on Concentration of Serum Creatinine, Urea and Glucose in Male Patients

Abdounasser Omar ¹*, Rima Rajap Aboud², Wijdan Albakoush², Rabia Amar Anwesre ¹

¹Department of Chemistry, Gharyan’s Faculty of Science, Al-jabal Algharbi University, Gharyan, Libya.
²Undergraduate student, Department of Chemistry, Gharyan’s Faculty of Science, Al-jabal Algharbi University, Gharyan, Libya.

*Correspondence: abdounasseromar@yahoo.com

Abstract - There is a limited number of studies about end-stage renal disease (ESRD) in Libya. So, this study was carried out to inspect the effect of ESRD on the concentration of three biochemical parameters in blood serum. Concentrations of serum creatinine (SCr), serum glucose (SGlu) and serum urea (SUr) were measured in 30 males with ESRD and in 30 normal males. SCr and SUr mean values in patient group were found to be higher than the reference interval, and higher than the critical value, too. SGlu mean value was higher than the reference interval, but within the critical interval. Comparison of SCr, SUr and SGlu means, by applying the t-test, revealed significant differences (P < 0.01) between patient group and control group. Some significant relationships (P < 0.05) were found between the study parameters in patient group but not in the control group. Similarly, the significant relationships (P < 0.05) found in control group were not recorded in the patient group. These differences in relationships were accompanied by differences in similarity according to the results of cluster analysis. Briefly, the results reflected the effect of ESRD on SCr, SUr and SGlu concentrations.

Keywords: ESRD, creatinine, glucose, urea, t-test, correlation test.

1. Introduction

Chronic kidney disease (CKD) is a long-term condition usually caused by irreversible damage to both kidneys, leading to dialysis or transplantation may become necessary in some patients [1]. Progression of CKD is associated with other diseases like cardiovascular disease, anemia and metabolic bone disease [2]. CKD is defined as either a decline in glomerular filtration rate (GFR) to < 60 mL/min/1.73m² or as kidney damage for at least 3 months [3] and based on GFR, CKD is divided into five stages. These five stages are: stage 1 with GFR ≥ 90 mL/min per 1.73 m²; stage 2 with GFR between 60-89 mL/min per 1.73 m²; stage 3 with GFR between 30-59 mL/min per 1.73 m²; stage 4 with GFR between 15-29 mL/min per 1.73 m²; stage 5 with GFR < 15 mL/min per 1.73 m² [4].

The last stage of CKD is called end-stage renal disease (ESRD), an important public health issue because of its increased incidence and prevalence [5]. ESRD affects almost every system. For instance, it causes accumulation of nitrogenous wastes (i.e., azotemia), alters water excretion and regulation of body levels of some electrolytes. It also causes skeletal disorders, anemia, and alterations in cardiovascular function, neurologic disturbances and gastrointestinal dysfunction [6]. Another effect is when GFR declines the level of creatinine, urea and other chemicals in blood increases [7].

Creatinine is formed in muscles from creatine phosphate by dehydration and loss of phosphate, and it is eliminated from the body by kidneys. When creatinine level increases, this could indicate kidney impairment, so creatinine concentration in both urine and a serum specimen provides a useful clinical estimate of GFR [8]. Urea is a main nitrogenous end product of protein and amino acid catabolism. It is produced by liver and filtered out of blood in kidneys by glomeruli and partially reabsorbed with water. The most frequently determined clinical indices for
estimating renal function depends on concentration of serum urea [9]. Kidney is involved in the regulation of glucose homeostasis by three different mechanisms: (i) gluconeogenesis; (ii) uptake of glucose from the circulation; and (iii) glucose reabsorption into the circulation of from glomerular filtrate [10]. Results of some studies have revealed that serum glucose [11], serum creatinine [11-15] and serum urea [12-16] were significantly higher in CKD patient groups when compared to control groups.

In Libya, ESRD is a major public health problem, and the most common causes are diabetic kidney disease and chronic glomerulonephritis [17]. It has been reported that the prevalence of dialysis-treated ESRD in Libya is 624 per million population (pmp) [18], which is higher than those reported for other Arab countries, and is almost double the range of pmp estimated for the Mediterranean countries with similar demographic and socioeconomic characteristics [17]. Between 2007 and 2009, the number of ESRD patients on dialysis in Libya increased from 2116 to 2417 and it is expected that their number will reach 7667 in 2024 [19].

In Gharyan, a Libyan city, there is an also an increase of ESRD patient numbers, with limited attention being paid to this issue. So, considering the lack of information about any parameter in serum of ESRD patients in Gharyan, the aim of this study was to provide some information for the physicians and researchers about concentrations of serum creatinine (SCr), serum urea (SUr) and serum glucose (SGlu) in males from Gharyan suffering from ESRD. This will help in diagnosing ESRD and more understanding of the disease itself.

To the best of our knowledge, this is the first study to inspect the effect of ESRD on SCr, SUr and SGlu concentrations in males with ESRD in Gharyan. This was done by comparing SCr, SUr and SGlu concentrations in serum of ESRD male patients (patient group) with regard to their reference and critical intervals. Also the obtained data of the patient group and was statistically compared with similar data of healthy males free from ESRD (control group). To achieve this, correlation test (r), t-test and cluster analysis were applied to the data obtained from patient and control group.

II. EXPERIMENTAL

Steps of sampling and analysis were carried out at Gharyan’s Educational Hospital, Libya, and the analysis of each sample was done directly after obtaining its serum.

A. Subjects

Sixty male volunteers from Gharyan city were subjected to blood analysis to determine SCr, SGlu and SUr concentrations. Thirty of the volunteers were normal males representing the control group, and the others (30 males) were suffering from ESRD and on dialysis, representing the patient group. Age range for the control group was 21-54 years with a mean of 31 years, and for the patient group was 22-85 years with a mean of 51 years. All subjects were informed of the nature of the study and they agreed to volunteer and take part in it.

B. Sampling

Two ml of the venous blood was collected from each volunteer, transferred to a plain tube and then left at room temperature for 30 minutes to allow the clot to form. Then, each sample was subjected to centrifugation at 4000 rpm for 10 minutes. After that, the serum was obtained and analyzed twice for its content of creatinine (Cr), glucose (Glu) and urea (Ur).
C. Reagents

Reagents used to determine the concentration of SCr, SGlu and SUr were obtained from Spinreact (Spinreact, Girona, Spain) and were provided by Gharyan’s Educational Hospital, Libya.

D. Instruments and Methods

Determination of SCr, SGlu and SUr concentrations was done photometrically, at 505, 505, and 340 nm, respectively, by using the Selectra Prom chemistry analyzer equipped with a Quartz-iodine lamp 12V-20W (ELITech Group, Puteaux, France). All analysis steps for the three biochemical parameters were carried out according to the instructions supplied with the kit from Spinreact.

Determination of SCr concentration was based on the reaction of creatinine with alkaline sodium picrate. The reaction forms a red complex (creatinine picrate) with an intensity proportional to the concentration of SCr in the sample.

In case of SGlu, the quantification was based on the oxidation of glucose to gluconic acid. This reaction also produces hydrogen peroxide, which reacts with phenol and 4-aminophenazone in the presence of peroxidase to form quinone, which its color intensity is proportional to the SGlu concentration in the sample.

Concentration of SUr in each sample was determined by hydrolyzing it with urease into carbon dioxide and ammonium ions. In the presence of nicotinamide adenine dinucleotide (NADH), ammonium ions react with α-ketoglutarate to form L-glutamate. In this reaction NADH is converted to its oxidized form (NAD⁺), and the decrease in NADH concentration is related to the SUr concentration in the sample.

E. Statistical Analysis

Minitab software (version 17) and Microsoft Excel 2013 were used to carry out all the statistical analyses. Besides descriptive analysis and interval plots to compare means, other tests were applied to the data. Those tests included: correlation test (r) at a significance level (α) of 0.05 to test if there were positive or negative relationships between age and the data of biochemical parameters in patient group, also in control group for comparison; cluster analysis was applied to the data of biochemical parameters as a measurement of similarity between them; t-test was run to test the difference between the biochemical means (patient group versus control group) at α = 0.01.

III. RESULTS AND DISCUSSION

As shown in Table 1 the age range for the control group was 21-54 years (mean = 31 years, standard deviation = 10 years), and for the patient group it was 22-85 years (mean = 51 years, standard deviation = 17 years). The standard deviation (SD) and the coefficient of variance (CV%) of the three biochemical parameters, shown in Table 1, were found to be higher in patient group, reflecting that the concentration values of SCr, SUr and SGlu are more dispersed in patient group than in control group. It is clear that the highest CV% value was for SGlu in patient group, and the highest difference in CV% between each pair (one parameter in both groups) was between SGlu pair, suggesting that ESRD has more effect on data dispersion in case of SGlu concentration compared to SCr and SUr concentrations.
As shown in Table 1, the range of SCr concentrations in patient group was 6.11-14.86 mg/dL with a mean of 9.63 mg/dL, which was higher than that of control group as shown in Fig. 1; the range of SGlu concentrations in patient group was 71.5-349 mg/dL with a mean of 136 mg/dL which, as SCr, was higher than that of control group (Fig. 2); the range of SUr concentrations in patient group was 73.5-226.5 mg/dL with a mean of 144.4 mg/dL which, as SCr and SGlu, was higher than that of control group (Fig. 3).

| Table 1: Some Descriptive Statistics for Age & the Biochemical Parameters of Patient and Control Groups |
|-----------------------------------------------|----------------|----------------|----------------|------|------|
| Age (years)                                  | Mean | Minimum | Maximum | SD   | CV%  |
| Patient group                                | 51   | 22      | 85       | 17   | 34.2 |
| Control group                                | 31   | 21      | 54       | 10   | 32.1 |

| SCr (mg/dL)                                  | Mean | Minimum | Maximum | SD   | CV%  |
| Patient group                                | 9.63 | 6.11    | 14.86   | 2.25 | 23.3 |
| Control group                                | 0.76 | 0.55    | 1.12    | 0.12 | 16   |

| SGlu (mg/dL)                                 | Mean | Minimum | Maximum | SD   | CV%  |
| Patient group                                | 136  | 71.5    | 349     | 72.7 | 53.5 |
| Control group                                | 89.9 | 62      | 116.5   | 13.2 | 14.7 |

| SUr (mg/dL)                                  | Mean | Minimum | Maximum | SD   | CV%  |
| Patient group                                | 144.4| 73.5    | 226.5   | 41.31| 28.6 |
| Control group                                | 27.05| 16.5    | 43      | 7.17 | 26.5 |

As shown in Table 1, the range of SCr concentrations in patient group was 6.11-14.86 mg/dL with a mean of 9.63 mg/dL, which was higher than that of control group as shown in Fig. 1; the range of SGlu concentrations in patient group was 71.5-349 mg/dL with a mean of 136 mg/dL which, as SCr, was higher than that of control group (Fig. 2); the range of SUr concentrations in patient group was 73.5-226.5 mg/dL with a mean of 144.4 mg/dL which, as SCr and SGlu, was higher than that of control group (Fig. 3).

Figure 1. Interval plot of SCr concentration for patient and control groups with a diagonal line connecting the means.
Comparing the data of each biochemical parameter in patient group with its reference interval [20], and its critical interval [20], revealed that all values of SCr concentrations and their mean were higher than the reference interval (0.9-1.3 mg/dL). These values also exceeded the critical value (5 mg/dL). Similar to SCr, all values of SUr
concentrations were higher than the reference interval (6-20 mg/dL), and nearly 93% of them were higher than the critical value (80 mg/dL). This surpass of the critical value, in creatinine and urea concentrations, shows that most patients are in a life-threatening condition if not treated.

In case of glucose, more than half values (60%) of SGlu concentrations, and their mean, were higher than the reference interval (74-100 mg/dL), while 37% were within the reference interval, and in only one sample SGlu concentration was lower than the reference interval. Although that SGlu readings in nearly 63% of the patients were beyond the reference interval, this shouldn’t be considered as a life threat because all SGlu readings and their mean were within the critical interval (40-450 mg/dL).

The elevation of SCr and SUr levels (relative to the reference interval) should be considered together, as indicators of ESRD in Libyan males, particularly when these results are linked to the results of t-test, which revealed that there are significant differences (P = 0.000) between SCr and SUr means of the patient and the control group. In agreement with these findings, some researchers [21-23] in Iraq have reported that SCr and SUr concentrations were found to be significantly higher in female and male patients with CKD when compared to control groups.

Considering that SGlu concentration in 60% of the patients was higher than reference interval, SGlu concentration can help in ESRD diagnosis. This is because there was significant difference (P = 0.002) between patient and control group in SGlu mean. In a study [24] carried out in Iraq there has been significant increase in SGlu of patient group compared to control group.

For correlation test, Table 2 shows that there were two significant positive relationships in patient group, one of them was between the age and SGlu concentration (r = 0.499, P < 0.05), and the other was between SCr and SUr concentrations (r = 0.404, P < 0.05). In addition to these two relationships, a significant negative relationship (r = -0.392, P < 0.05) was observed between SGlu and SCr concentrations.

In contrast, these three relationships weren’t observed in control group, correlation factor values for those pairs was so small and P-values were > 0.05. Moreover, two different significant positive relationships were observed in the control group, one of them was between SUr and SGlu concentrations and the other one was between SUr and age.

<p>| TABLE 1: CORRELATION COEFFICIENTS (r) AND P-VALUES BETWEEN AGE AND THE THREE PARAMETERS |
|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|</p>
<table>
<thead>
<tr>
<th></th>
<th>Age</th>
<th>SCr</th>
<th>SUr</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age</strong></td>
<td><strong>r</strong></td>
<td><strong>P</strong></td>
<td><strong>r</strong></td>
<td><strong>P</strong></td>
<td><strong>r</strong></td>
<td><strong>P</strong></td>
</tr>
<tr>
<td>Patient group</td>
<td>-0.284</td>
<td>0.128</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control group</td>
<td>0.181</td>
<td>0.319</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>SCr</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Patient group</td>
<td>-0.215</td>
<td>0.254</td>
<td>0.404</td>
<td>0.027</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control group</td>
<td>0.380</td>
<td>0.038</td>
<td>0.018</td>
<td>0.924</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>SUr</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Patient group</td>
<td>0.499</td>
<td>0.013</td>
<td>-0.392</td>
<td>0.032</td>
<td>-0.258</td>
<td>0.127</td>
</tr>
<tr>
<td>Control group</td>
<td>0.152</td>
<td>0.422</td>
<td>-0.023</td>
<td>0.884</td>
<td>0.495</td>
<td>0.005</td>
</tr>
</tbody>
</table>

In contrast, these three relationships weren’t observed in control group, correlation factor values for those pairs was so small and P-values were > 0.05. Moreover, two different significant positive relationships were observed in the control group, one of them was between SUr and SGlu concentrations and the other one was between SUr and age.
These observed differences in relationships were also accompanied by differences in similarity ratio of the three biochemical concentrations estimated by cluster analysis as shown in Fig. 4 and Fig. 5. The highest similarity (70%) in patient group (Fig. 4) was between SUr and SCr concentrations, while the highest similarity (75%) in the control group (Fig. 5) was between SUr and SGlul concentrations.

Figure 3. Dendrogram of biochemical parameters for patient group.

Figure 3. Dendrogram of biochemical parameters for control group.
To sum up, these findings reflect the role of ESRD on altering serum concentration of the studied biochemical parameters of Libyan males with ESRD, and on the relationships between them.

IV. CONCLUSION

It is clear that ESRD has a significant effect on the studied biochemical parameters. Thus, these parameters should be taken into account when a patient is expected to be in ESRD stage. Patients with ESRD receiving dialysis in Gharyan’s educational hospital should be treated for elevated concentrations of serum creatinine and serum urea. Further investigation of other chemical parameters in ESRD patients will provide more information to help in diagnosing ESRD, and lessen its impact on those patients. Also, other studies could be conducted to investigate the efficiency of dialysis on concentrations of SCR, SUr and SGlu ESRD patients.

ACKNOWLEDGMENT

We thank the staff at Gharyan’s Educational Hospital, Libya, where sampling and analysis were carried out.

REFERENCES


