STUDY OF THE RELATIONSHIP BETWEEN CHRONIC KIDNEY FAILURE WITH ANEMIA AND BACTERIAL PROFILE IN BABYLON PROVINCE, IRAQ

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Abstract

The purpose of the study was to detection the pervasiveness and clinical course that was Stimulated by types of bacterial infections and anemia in patients with chronic renal failure. Therefore, this study includes assessing the serum condition of infected patients who suffer from chronic kidney failure, as well as verification of infection, bacterial infection and anemia. This evaluation is by Cobas Integra 400 (for urea and creatinine), complete blood count (CBC) and Bacteria / ALERT 3D (FOR blood culturing) in the sera of 70 specimens that were collected during the period extending from October 2018 to March 2019. Data about individuals have collected aseptically in sterile containers, after getting all data in the special formula including, name, gender, age and history. The results of the Current study have been showed that Kidney function test (B. urea and S. creatinine) has been increased in level compared with control group, as while as, the study results showed decrease in hematology parameters (PCV, Hb) level compared with control group. While the results showed the patient’s groups were infected with bacteria (S. aureus) that was isolated from blood specimen of CKD patients, Also the study showed only 7 (14%) patients have bacterial growth While 43 (86%) patients have bacterial growth.

Key word: Renal failure, anemia and bacterial profile.

Introduction

Chronic kidney disease is one of the most complex diseases, affecting kidney functions in general, affecting human health and is considered the most prevalent in the world. Chronic kidney disease is the most common diseases that do not have primary symptoms and start from the development of symptoms of chronic renal disease until the end of the stages of renal failure, as well as secondary complications due to weak kidney functions which consider as the main reasons that lead to rise risk of Heart and Vascular diseases (Foley et al., 1999). One of the common complications of CKD is the anemia which is related with raise risk factors of Heart and Vascular diseases, increased the rate of spread of disease and death, especially on a large scale (McCullough et al., 2005). Anemia is one of the untraditional danger factor of the CVD (Vlagopoulos, 2005). Anemia complication in CKD is basically due to complete or relative deficiency of erythropoietin. Besides erythropoietin insufficiency there are many different factors that causes CKD such as blood loss, decreased half-life of RBC, iron insufficiency and in general infection and other sources, may involve in advance anemia in CKD (Nurko, 2006). Some studies referred that Hb-level begin reduced even in early Kidney failure around 70 ml/min in male and 50 ml/min in female (Jurkovitz et al., 2003).

Furthermore, anemia in CKD has been Complications gives negative results in CKD and increases the pathological conditions, which makes them worst that accompanying with diabetes and hypertension (Vlagopoulos, 2005). Early diagnosis of an anemia in CKD patients delays the evolution of renal disease at the end of the stage and thus improves morbidity and mortality of cardiovascular diseases (K/DOQI, 2006). CVD is the main causes of mortality in patients having CKD and accounts so that as the total 58% patients dies due to the CVD in CKD patients (Shulman et al., 1989). Even Mild kidney failure which has been Notified even with rise incidence of CVD. The rate of death due to infection by CVD in CKD patients has been reached between 10 to

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20 times higher when compared with general population (Foley et al., 1998). To our knowledge, the famous cases of anemia in various stages of CKD which has been unreported was happened in Nepal which is lead to the death of Patient. Thus we did this study for the purpose to discover cases of anemia in some of the patients within different stages of CKD in addition to the association of anemia and CKD in the hospital environment. (Arch Intern Med, 2002).

*Staphylococcus aureus* is the main causes of pathogens and death rate in cases of renal unit inflammations. It is the main reason why bacteria in dialysis-related peritoneal dialysis and exit infection from the exit site (Kairaitis et al., 1999; Scalamogna et al., 1991; Lentino et al., 2000). It is one of the causative agent that cause peritoneum inflammation that related to peritoneal dialysis. (Zelenitsky et al., 2000). Since many of these infections can be prevented, developing strategies that reduce infection rates is an important component of caring for this group of patients. This review examines the basis of prevention, along with its potential benefits and potential disadvantages. Also, the other more common source infection in CKD include *Staphylococcus* and *Streptococcus*, according to a study by researchers at Loyola Medicine and Loyola University Chicago. The purpose of the study is to obtain adequate information on the most important pathogens, including the most important bacterial infections, anemia and other causes that lead to CKD that causes kidney failure.

**Material and methods**

This study was carried out in the department of Medical Laboratory Techniques in Al-Mustaqbal University College, during the period extended from October 2018 to March 2019.

**Subjects**

A total of Seventy blood sample, fifty renal failures and twenty healthy subject’s bloods samples were collected aseptically in sterile containers, after getting all data in special formula including gender, age, medical history and result of analysis.

**Ethical Approval**

The agreements of all subjects’ intended in this study was obtained before taking the study specimens. Furthermore, the study design was approved by research Ethical committee at Medical Laboratory Techniques in Al-Mustaqbal University College.

**Sample Collection**

**Sera Samples Preparation and Preservation**

Blood specimens were collected by drawing 3ml of blood from each subject included in this study using sterile 10ml syringes with sterile needle G-22. Blood samples of kidney failure and healthy control that has been collected in sterile 10ml capacity sterile gel tubes which has been labeled as with name, age, gender as well as date of their collection.

When the blood clot formation at room temperature within 30 minutes, the blood clot sample will be centrifuged at 2,500 RPM for five minutes.

Now all of the separated sera specimens were collected and become ready to use in the lab.

**Whole blood samples Preparation**

The first step to prepare whole blood samples by cleaning the skin by cotton containing alcohol, collected 2 ml of blood venous in disposable syringe then with drawn in EDTA container and mixed with the anticoagulant to avoid clot formation.

**Methods**

**Complete blood count**

Complete blood count {HB, RBCs, WBC (neutrophil and lymphocyte), MCV, MCH, MCHC, HCT, platelet indices} was tested by automated method Sysmex.

**Principles of the examination method**

Blood sample collected in EDTA anticoagulant is diluted with CELLPACK in a WBC counting container then a fixed volume of STROMATOLYSER-WH solution (1 volume of STROMATOLYSER-WH to 2 volumes of CELLPACK) is added automatically to obtain a final dilution of 1:500. The addition of STROMATOLYSER-WH lyses the RBC and so the remaining cell stroma is at a level undetectable by the instrument. The WBC membrane is preserved and WBC are stabilized at a level detectable by the instrument. They are then counted by the DC method. Hemoglobin is released during RBC lysis, and is converted to the red methemoglobin. A portion of this diluted sample is transferred automatically to the hemoglobin detector where the absorbance of the red pigment is measured to give blood hemoglobin level.

**Examination procedure**

1. Use the STROMATOLYSER-WH at a temperature of 15-30°C. Measuring at a temperature above 30°C or below 15°C may give inaccurate WBC count, WBC tri-model size distribution analysis and hemoglobin level.

2. Loosen and remove the cap of the bottle of STROMATOLYSER-WH and connect to the instrument.

3. Refer to the guide and tool’s operator’s for detailed information.
Cobas Integra 400 (for urea and creatinine)

The kit of Cobas Integra can be Analyst is the best solution for laboratories that use up to 400 specimens daily. The extensive list of tests includes more than 120 tests and applications that integrate clinical chemistry with special proteins, monitor medicinal drugs, and drug use testing. This compact table top Analyst gives greatest diversity to get better effectiveness and decrease the costs. The cobas c pack has been used widely because it is Commensurate with all the laboratories, which standardizes patient results across integrated laboratory networks. Over 140 requests for all types of sample matrices are measured using one of 4 different measurement techniques for optical absorption measurement, turbidity measurement, fluorescence polarization and selective ion voltage measurement.

Bact / ALERT 3D (FOR blood culturing)

Bact / ALERT® 3D is a modern microbial detection tool. Bact / ALERT® System provides benefits in all dimensions of the microbial detection test. From space-saving modular design to easy-to-touch touch screen operation and flexible data management options, every size laboratory can perform microbial detection with Bact / ALERT® 3D that allows you to achieve consistent, accurate and objective results with minimal effort:

1- Choose suitable BACT/ALERT® culture medium bottle
2- Inoculate bottle with sample
3- Load bottle inside BACT/ALERT® system

The BACT / ALERT® bottle contains sterile cultures and has a color sensor that changes from gray to yellow in the presence of carbon dioxide from growing microorganisms. Once The bottles are loaded, the color sensors are checked every ten minutes. Once growth is detected, the system alerts the sample data is recorded. BACT / ALERT 3D is used to detect the presence or absence of microorganisms in blood-sterilized body fluids.

Bacterial profile

Cultivation of bacterial profile

A group of culture media has been used in this project, such as Brain-heart infusion broth, macConkey agar, blood agar, and mannitol salt agar according to specific instruction of the company.

Colonial morphology and microscopic examination

As is the practices in all medical laboratories such as diagnostic methods, and the most common in the laboratories in general, types of bacteria have been diagnosed according to the method Bruckner (2016). The bacterial profile in specimens which has been isolated and identification are performed in all the positive sample in culture media has been identify according morphology for the colonies and microscopic examination test which is used Gram staining.

Biochemical test of bacterial profile

The detection of biochemical test must be including of the following: catalase test, coagulase test, mannitol fermentation test and haemolysin production according to manual procedure with bacterial profile in CKD patients (Forbes et al., 2007).

Results and Discussion

Relationship between B. urea and S. creatinine with chronic renal failure according to gender and age

Fifty samples has been taken from patients suffer chronic kidney failure (30 males and 20 females) and twenty samples were taken from healthy people as a unit of measurement (10 males and 10 females). There are many differences has been observed between Gender and age in many patients and subjects being monitored. The intermediate comparison of the different Properties of the study societies is shown in table 1.

It was observed that the levels of the (B. urea and S. creatinine) increased in some patients with chronic nephritis in tests of the normal kidney function in comparison with the ratios of levels of the (B. urea and S. creatinine) in healthy people as a standard unit, thus these results were similar to the expected results as previous studies such as (Shivananda, 2007). It was reported that a very significant increase in urea and creatinine was found in the serum of patients with CRF compared to standard groups. An increase in urea in kidney failure is caused by a weak ability to secrete the protein copulate due to a marked decrease in the glomerular filtration rate (GRF). Increases in creatinine in the blood are also a result of decreased kidney secretion.

The results regarding age and gender has been distributed in certain patients who suffer from CKD in the current study was confirmed that reported from other authors because the renal diseases were found at all age groups and both gender (Miftari et al., 2017).

Relationship anemia with chronic renal failure according to gender and age

The results of the study showed a decrease in the level of hematology parameters (PCV, Hb) compared to the standard groups due to the complete or relative deficiency of erythropoietin that affects by anemia in some the patients who suffer from chronic kidney disease.
Table 1: Illustrate the distribution of study according to alternation \( B. \text{urea} \) and \( S. \text{creatinine} \) with chronic renal failure.

<table>
<thead>
<tr>
<th>Factor</th>
<th>( B. \text{urea} ) (Mean)</th>
<th>( S. \text{creatinine} ) (Mean)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td>Male</td>
<td>Female</td>
</tr>
<tr>
<td>Age</td>
<td>Patient</td>
<td>Control</td>
</tr>
<tr>
<td>30-39</td>
<td>29.60</td>
<td>5.22</td>
</tr>
<tr>
<td>40-49</td>
<td>16.55</td>
<td>4.78</td>
</tr>
<tr>
<td>50-59</td>
<td>25.40</td>
<td>5.33</td>
</tr>
<tr>
<td>Total</td>
<td>23.85</td>
<td>5.11</td>
</tr>
</tbody>
</table>

Table 2: Illustrate the distribution of study according to alternation anemia with chronic renal failure.

<table>
<thead>
<tr>
<th>Factor</th>
<th>PCV (Mean)</th>
<th>Hb (Mean)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender</td>
<td>Male</td>
<td>Female</td>
</tr>
<tr>
<td>Age</td>
<td>Patient</td>
<td>Control</td>
</tr>
<tr>
<td>30-39</td>
<td>28.12</td>
<td>43.89</td>
</tr>
<tr>
<td>40-49</td>
<td>22.86</td>
<td>40.78</td>
</tr>
<tr>
<td>50-59</td>
<td>31.41</td>
<td>43.46</td>
</tr>
<tr>
<td>Total</td>
<td>27.46</td>
<td>42.71</td>
</tr>
</tbody>
</table>

These results were approved by previous studies who expressed that the mean concentrations of Hb and PCV. Also the study showed there is no notable difference (\( p > 0.05 \)) in the concentration of Hb in males and females of CKD patients under hemodialysis. In contrast the Studies have shown great importance for measurement of decrease (\( p < 0.05 \)) in the concentration of Hb between male patients with control group males as well as female patients With female control. The study show no Noticeable difference (\( p > 0.05 \)) in PCV for both of males and females the are Suffering from chronic renal insufficiency patients who were subject to dialysis. The study also show a significant decrease (\( P < 0.05 \)) in the PCV in both male patients and female patients with male and female control group sequentially (Abd Alzahraa and Wafaa, 2013).

The results of present study indicate a decrease for both concentration of Hb, the percentage of PCV which used as indicators of patients with anemia who Suffering from chronic renal disability. For there many studies have indicated that anemia is one of the most significant complications associated with chronic renal disability. Where some cases clarified that the severity of anemia patients with disability Renal failure was directly proportional to the state of functional deterioration of the kidney. The study results were consistent with most previous researchs (Costa \textit{et al.}, 2008).

In some cases of the patients who is suffer from anemia with chronic renal impairment is due to a lack of secretion of the hormone erythropoietin, which is responsible for stimulating and forming red blood cells within the bone marrow. The lack of RBCs cell count leads to a decrease in the concentration of Hb and the percentage of PCV. Surrounding the renal tubules in the formation of the hormone erythropoietin and any damage to these cells will cause a decrease in the secretion of this hormone (Brunelli and Berns, 2009).

Relationship chronic renal failure with bacterial infection

It was possible to study the inter-relationship of infections at sites in 50 patients. In 7(14%) of these, the \( S. \text{aureus} \) was isolated from the blood specimen of chronic renal failure cases Fig. 1.

The results showed the patient’s groups were infected with bacteria (\( S. \text{aureus} \)) that was isolated from the blood specimen of chronic renal failure cases, just 7 (14%) patients have bacterial growth and 43 (86%) patients have no bacterial growth. This finding is an agreement with Borzio \textit{et al.}, (2001) stated that fundamentals of urine and body fluid analysis Bunnzel, nancy A.

The conclusions for the documented is a high widespread presence of anemia in all stages of CKD, so that \( B. \text{urea} \) and \( S \text{creatinine} \), a strong association between anemia and CKD was found. Anemia in CKD is the established risk factor for the development of the cardiovascular disease and also bacterial infections frequently precipitate renal failure.

Therefore, we recommended that Necessary to identify anemia in CKD as soon as possible and manage them properly before development of cardiovascular disease. Conduct extensive studies on the prevalence of bacteria was only blood or urine specimen. Spread

![Fig. 1: Percentage of bacterial growth with chronic renal failure patients.](image-url)
community awareness and provide quality services in the field of Public Health which is the most appropriate way to eradicate this disease. Monitoring blood dialysis units to reduce the spread of the disease through the sterilization of sanitary ware, especially the tools used for the purpose of dialysis. Conduct periodic examinations for the purpose of early detection and prevention of access to acute and chronic cases. The need to expand studies in this field to find out the reality of the disease in Iraq, especially in the province of Babylon, because of the deficiency of studies on this subject. Encourage researchers to find advanced prevention and treatment methods that differ from what exists to eliminate or reduce the spread of this disease.

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References


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