Descriptive analytical study
use of albumin as
nutritional marker among
patients
of chronic renal failure

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بسم الله الرحمن الرحيم

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صدق الله العظيم

يوسف، الآية: 76

(وما أوتيتم من العلم إلا قليلاً)
صدق الله العظيم

الإسراء، الآية: 85
Dedication

To my father, mother, brothers… and to all those

who supported me during my life…
Acknowledgment

I would like to express my deep gratitude to Dr. Mohammed El Mozamel Hassan for his supervision and advices. And to Ustaz Tawfiq Khojali for his co-supervision. Then I would like to extend my warmth thanks to all colleagues in Dr, Salma Mohammed Suliman Kidney Dialysis and Transplant Centre, specially Osman Mohammed Osman for his useful helping, and to my colleagues in National Health Research Laboratory (STACK), Department of Clinical Chemistry, specially to Abd Motalib Mohammed Mustafa, senior staff of the department, and to my colleague Mahmoud Hassan (Al Gazar), for their support and helping and to my friend El Khair Abd Almohamoud Idris for his help and encouragement. Lastly, great thanks to all patients and statisticians who help me specially Ustaz El Shibly Mohammed Ahmed.
## ABBREVIATIONS

<table>
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<tr>
<td>ARF</td>
<td>Acute Renal Failure</td>
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<tr>
<td>CRF</td>
<td>Chronic Renal Failure</td>
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<td>ESRD</td>
<td>End Stage Renal Disease</td>
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<td>HTV</td>
<td>Hypertension</td>
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<td>GRF</td>
<td>Glomerular Filtration Rate</td>
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<td>CRP</td>
<td>C. Reactive Protein</td>
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<tr>
<td>SAA</td>
<td>Serum Amyloid A protein</td>
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<tr>
<td>TNF</td>
<td>Tissue Necrosis Factor</td>
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<tr>
<td>BUN</td>
<td>Blood Urea Nitrogen</td>
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<td>BCG</td>
<td>Bromo Cresol Green</td>
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Abstract

This study was conducted in Dr. Salma Mohammed Suliman Kidney Dialysis and Transplant Center during the period from February 2005 to December 2005.

This study aimed to detect the percentage of hypalbuminemia among Sudanese patients with chronic renal failure who were under maintenance hemodialysis treatment by using albumin as nutritional marker, and the effect of uremic syndrome on the level of serum albumin (correlation between albumin and urea and creatinine).

Hundred blood samples were collected from patients who were admitted to the Center for hemodialysis and their serum were separated and analyzed for urea, creatinine, albumin and C. Reactive protein.

This data was analyzed and the results showed that the majority of patients were males (74%) and most of the patients (54%) from central area. This study showed that (61%) of the patients have albumin level less than (3.5g/dL) the minimum reference value, also the study showed most of patients were young adults in the age group 26-37 years (42%) and only (10%) of the patients have C. reactive protein positive.

The study showed that the relation between C. reactive proteins and gender was negligible (statistically insignificant), but significant in creatinine protein and normality (P. value < 0.01). The study was distributed the patients to groups according to the severity of malnutrition, the mild-malnutrition about (45%). The study showed that the correlation between urea and creatinine was significant and (P. value is < 0.01), also it showed that the correlation was significant between creatinine and albumin the (P. value is < 0.05).
الخلاصة

إجراء الدراسة هذه في وزارة الزراعة لغسيل سليمان محمد سلامة في فبراير من السنة 2005.

نقص نسبة دراسة هذا التهديف البيومين في الكلى الفشل مريضه وبذلك يلتقط الدموي غسيل المنظم البيومين نسبة قياس في وذالك عندهم وثيقة Comparing البيولوجيا الحميمي بالنسبة إلى درجة البيومين مع صان.

تم جمع 100 عينة مريض من الكلى الفشل وبذلك واجرت الاختبارات الرائعة البيومين والبيومين تينين الرياكة سين بروتين ونurse.

الدراسة أظهرت أن الرجال عدد 74% (%) النساء عدد كبر 26% (%).

كذالك الدراسة أظهرت بأن نسبة Mرن الكمبودي نسبة كن ونurse 45% (%) نسبة Kبد لم تست_dicts الديثودة والكريتون البيومين (P. value > 0.01).

كذالك نسبة Kبد لم تست_dicts الديثودة والكَريتون البيومين (P. value < 0.05).

(P. value < 0.05)
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Chapter One
1- Introduction

It's believed that malnutrition is common in patients with chronic renal failure (CRF). They have reduced body weight depleted energy (fat tissue) stores, loss of somatic protein (low muscle mass) and low levels of serum albumin, transferrine, pre-albumin and other visceral proteins. Various studies show signs of malnutrition in 23-76% of haemodialysis and 18-50% of peritoneal dialysis patients\(^{(1,2)}\). Such variation in the prevalence of malnutrition may be related to factors such as age, case-mix, co-morbid conditions and quality of dialysis therapy. The etiology of malnutrition in CRF is decreased food intake because of anorexia, nausea, and vomiting due to uremic toxicity, hormonal derangements, acidosis and increased resting energy expenditure\(^{(1,3)}\).

While malnutrition by definition is caused by poor nutritional intake, laboratory measurements are generally used to define it clinically. Other factors can cause the same changes in body like infectious complications\(^{(3,4)}\), and chronic heart failure. In addition, factors directly associated with the dialytic procedure such as bio-incompatibility, nutrition losses in the dialysate and, during peritoneal dialysis, poor appetite due to abdominal discomfort and uptake of glucose may also contribute to what we define as malnutrition in CRF. There may exert their action either by direct nutrient loss or by triggering the inflammatory response. However since malnutrition also occurs in predialysis patients\(^{(4)}\). It is evident that dialysis – unrelated factors, e.g., infections and inflammation complications as well as co-morbidity, may also be important contributed to malnutrition CRF.
The incidence of CRF increased within the last years by observation in Khartoum, Khartoum North and Omdurman Teaching Hospital Department of Nephrology 60% of the admitted cases because of chronic renal failure\textsuperscript{(6)}. Also the increasing rate of morbidity and mortality has been reported. Most of those patients later soon they suffering from several complications, hypoalbuminemia, chronic inflammations and atherosclerosis appear to be the main complications. Hypoalbuminemia reported to be a major factor that increase the morbidity and mortality in those patients. In this study serum albumin is used as an indicator of the nutritional status of those patients and to observe the affects of uremic syndrome on the nutritional status.
Chapter Two

2- Review of Literature

In the U.K the prevalence of chronic renal failure is approximately 600 individual per population per million per year\(^{(5)}\). In Sudan there is inaccurate statistical figure, but the incidence seems to be between 140-170 individual per million population\(^{(6)}\).

In a national survey, CRF in Swedish was studied during the period from 1986-1994. the medium annual incidence of CRF was 7.7 and that of end stage renal failure was 6.4 per million.

The prevalence of CRF increased from 17.8 in 1986 to 38 per million in 1994\(^{(7)}\).

Renal failure in Sudan is a common disease, in Sudan the incidence for new cases about 140-170 million inhabitants/year. The majority were young adult patients (below 50 years old) and most die before reaching medical attention. A study carried out in 1982 by Dr. Omer to determine the causes of CRF in adult Sudanese patients show that: glomerulonephritis come top of the list 38%, renal calculi 12%, diabetes 9%, chronic interstitial nephritis 7% as sequel of ARF 5%, HTV 4% the polycytic kidney 3%, obstructive uropathy 2%, 20% of cases were unknown etiology\(^{(6)}\).

2-1 The Kidneys

2-1-1 Renal anatomy:

The kidneys are paired, bean-shaped. Organs located retroperitoneally on either side of the spinal column macroscopically, each kidney is enclosed by a fibrous capsule of connective tissue, when dissected longitudinally, two regions can be clearly discerned: on outer region called the cortex and an inner region called the medulla. The pelvis
also be seen. It is a basin like cavity at the upper end of the ureter into which newly formed urine passes. The bilateral ureters are thick-walled canals connecting the kidneys to the urinary bladder. Urine is temporarily stored in the bladder until voided from the body by way of urethra. Each kidney contains approximately one million nephrons. Each nephrons is a complex apparatus comprising the following five basic parts:

1- The glomerulus, which is a capillary tuft surrounded by the expanded end of renal tubule known as Bowman's capsule. Each glomerulus is supplied by an efferent arteriole carrying the blood out. The efferent arteriole branches into peritubular capillaries that supply the tubule.

2- The proximal convoluted tubule, which is located in the cortex.

3- The long Henle's loop, which comprises the thin descending limb that spans the medulla from the corticomedullary junction to the inner medulla, and the ascending limb located in both the medulla and the cortex and is composed of a thin, then thick region.

4- The distal convoluted tubule, which is located in the cortex.

5- The collecting duct, which is formed by two or more distal convoluted tubules as they pass back down through the cortex and the medulla to collect the urine that drains from each nephron. Collecting ducts eventually merge and empty their contents into the renal pelvis(7).

2-1-2 Renal function:

The kidneys are the body's filtering system. They remove waste materials the collect in the blood as by-products of normal bodily function. Everyday the two kidneys, which are each the size of a fist, process approximately 190 liters of blood by passing it around 225 kilometers of tubes and millions of mini-filters. The kidneys also help to maintain the body's balance of chemicals, including sodium and potassium, and produce some hormones and vitamin D(8).
The kidney principle rate is the elimination of waste material and the regulation of the volume and composition of the body fluid:\(^9\):
1- Elimination of waste products.
2- Regulation of the volume and composition of body fluid.
3- Acid-base balance.
4- Water and electrolytes re-absorption.
5- Endocrinology function.
6- Gluconeogenesis.

2-1-3 Pathophysiology:

Approximately one million nephrons are present in each kidney, each contributing to the total GFR:\(^{10}\).

Regardless the etiology of renal injury with progressive destruction of nephrons, the kidney has an innate ability to maintain GFR by hyper-filtration and compensating hypertrophy of the remaining healthy nephrons. This nephron adaptability allows for continued normal clearance of plasma solutes so that substances such as urea and creatinine start to show significant increase in plasma level only after GFR has decreased 50% when the renal reserve has been exhausted. The plasma creatinine from a base line of 0.6mg/dl to 1.2mg/dl in a patient although still within the reference range, actually represents a loss of 15% of functioning nephron mass. The residual nephron hyper-filtration and hypertrophy has been hypothesized to represent a major cause of progressive renal dysfunction. This believed to occur because of increased glomerular capillary pressure which damage the capillaries and lead initially to focal and segmental glomerulosclerosis and eventually global glomerulosclerosis. This hypothesized has been on studies of five – sixthths nephrectomized react which develop this lesion that are identical to those observe in human with CRF:\(^{10}\).
2-1-4 Signs and symptoms:

Chronic renal failure (CRF) usually produces symptoms when renal function – which is measured as glomerular filtration rate (GFR) fall below 30 millimeters per minute (< 30ml/min). this is approximately 30% of the normal value. When the glomerular filtration rate slows to below 30ml/min, signs of uremia may become noticeable. When the GFR falls below 15ml/min most people become increasingly symptomatic.

Uremic symptoms can affect every organ system, most noticeably the following (11):

- Neurological system – cognitive impairment, personality change, asterixis and seizures.
- Gastrointestinal system – nausea, vomiting, food distaste.
- Blood forming system – anemia due to erythropoietin deficiency, easy bruising and bleeding due to abnormal platelets.
- Pulmonary system – fluid in the lungs, with breathing difficulties.
- Cardiovascular system – chest pain due to inflammation of the sac surrounding the heart and pericardial effusion.
- Skin – generalized itching.

2-1-5 Stages of renal failure (according to GFR):

1- Renal impairment (GFR falls at 30-70 ml/min).
2- Chronic renal failure (GFR less than 30 ml/min).
3- End stage renal disease (GFR less than 10 ml/min).

(Usually associate with sign and symptoms of urine (10)).

2-1-6 Acute renal failure:

Acute renal failure is a sudden, sharp decline in renal operation due to acute toxic or hypoxic insult to the kidneys. This has been defined as occurring when GFR is reduced to less than 10m/min (12). This syndrome
is subdivided into three types depending on the location of the precipitating defect. In pre-renal failure the defect lies in the blood supply before it reach the kidney\(^{(12)}\). Causes can include cardiovascular system failure and consequent hypovolemia.

In primary renal failure, the defect involves the kidney itself, the most common cause is acute tubular necrosis; other etiologies include vascular obstruction, inflammations and glomerulonephritis. In post-renal failure, the defect lies in the urinary tract after it exist the kidney. Generally acute renal failure occur as the sequel to a low urinary tract obstruction or to rupture of the urinary bladder.

Toxic insults to the kidney sever enough to initiate acute renal failure include hemolytic transfusion reactions, heavy metal, solvents poisonings, antifreeze ingestion, and analgesic and aminoglycoside toxicities. These conditions directly damage the renal tubules. Hypoxic insults include conditions that severely compromise renal blood flow, such as septic, hemorrhagicic shock, burns, and cardiac failure\(^{(12)}\).

The most commonly observed symptoms of acute renal failure are oliguria and anuria (<400 ml/day). The diminished ability to excrete electrolyte and water results in a marked increase in extra cellular fluid volume, leading to peripheral edema, hypertension, and congestive heart failure.

If more water is retained than sodium, hyponatremia may develop; in extreme causes, central nervous system effects are seen progressing from profound drowsiness to seizure, coma and death\(^{(12)}\). The hyperkalemia also may become sever enough to cause dangerous cardiac arrhythmias. In addition variable amounts of erythrocytic casts, hematuria, proteinuria and metabolic acidosis (with resulting respiratory acidosis at compensation) are seen.
Generally, systemic bone disease and anemia are not marked as they are in chronic renal failure. Most prominent, however, is the onset of the failure. Most or end stage renal disease (ESRD), in which increased BUN and serum creatinine values (azotimia) are observed in conjunction with the preceding symptoms. The outcome of this disease is either recovery or, in the case of irreversible renal damage, progression to chronic renal failure\(^\text{(12)}\).

**2-1-7 Chronic renal failure:**

Chronic renal failure is a clinical syndrome that occurs when there is gradual decline in renal operation overtime\(^\text{(12)}\).

Chronic renal failure is classified into four progressive stages. The first stage is marked by a period of silent deterioration in renal status. Kidney function decrease, but BUN and creatinine values stay within normal limits. The second stage is characterized by development of a slight renal insufficiency\(^\text{(12)}\). A 50% reduction in normal functioning is necessary before BUN and creatinine values are reflect the pathologic changes by increasing above references ranges. The third stage is typified by impending renal failure. Anemia begins to develop (due to the constant deficient in erythropoitin production), and systemic acidosis commences (due to the faulty clearance of indogenous metabolic acids)\(^\text{(11)}\). The fourth and the last stage commences with the onset of the classic symptoms of the uremic syndrome. The conditions that can precipitate acute renal failure also may lead to chronic renal failure\(^\text{(12)}\).

**2-1-8 Laboratory diagnosis:**

Chronic renal failure is diagnosed by the observation of a combination of symptoms and elevated blood urea and creatinine levels. The following abnormalities found in the blood may signal chronic renal failure\(^\text{(11)}\).
• Anemia.
• High level of parathyroid hormone.
• Hypocalcemia.
• Hyperphosphatemia.
• Hyperkalemia.
• Hyponateremia.
• Low blood level of bicarbonate.
• Low plasma pH.

2-2 Creatinine (Biochemistry and disease correlations):

Creatine is synthesized mainly in the liver from arginin, glycine and methionine. It’s then transported to other tissues, such as muscle, where it is converted to phosphocreatine, which serves as a high-energy source. Creatine phosphate loss phosphoric acid and creatine loss water to form creatinine, which is released into the plasma\(^{(13)}\).

Creatinine is exerted into the circulation at a relatively constant rate that has been shown to be proportional to the individuals muscle filtration. It’s removed from the circulation by glomerular filtration and excreted by the proximal tubule. Small amounts may also be reabsorbed by the renal tubules, especially at low flow rate\(^{(13)}\).

Plasma levels of creatinine are related to the relative muscle mass, the rate of creatine turnover, and renal function. It has been accepted for many years that the plasma level of creatinine is relatively unaffected by diet\(^{(14)}\).

When plasma creatinine is elevated, GFR is decreased, indicating renal damage. Unfortunately, plasma creatinine is a relatively insensitive monitor and may not be measurable increased until renal function has deteriorated more than 50\%\(^{(15)}\). The absorbed relationship between plasma creatinine and the GFR and the observation that plasma creatinine
levels are relatively constant and unaffected by diet should make it an excellent analyze for the assessment of renal function\(^{(15)}\).

2-3 Urea (biochemistry and disease correlations):

Urea constitutes nearly half the non protein nitrogenous substances in the blood. It is synthesized in the liver from CO\(_2\) and the ammonia arising from the deamination of amino acids by means of the ornithine or krebs-henseliet cycle. Urea constitutes the major excretory product of protein metabolism following synthesis in the liver, urea is carried in the blood to the kidney, where it is readily filtered from the plasma by the glomerulus. Most of the urea in the glomerulus is excreted in the urine, although up to 40% is reabsorbed by passive diffusion during passage of the filtrate through the renal tubular the amount reabsorbed depends on urine flow rate and level of hydration. Small amounts of urea (< 10% of total) are excreted through the gastrointestinal tract and skin. The level of urea in the plasma is governed by renal function and perfusion. The protein content of the diet, and the amount of protein catabolism\(^{(16)}\).

An elevated level of urea in the blood is called azotemia very high levels of plasma urea accompanied by renal failure is called uremia or the uremic syndrome which is eventually fatal if not treated by dialysis\(^{(16)}\).

Conditions causing elevations of plasma urea are classified according to cause into three main categories, pre-renal, renal and post-renal\(^{(16)}\).

Pre-renal is caused by reduced renal blood flow, reduction in blood flow delivers less urea to the kidney and therefore less urea is filtered. Causative factors include: congestive heart failure, shock, hemorrhage dehydration, and any other factors resulting in a marked decrease in blood volume. The level of protein metabolism also causes pre-renal changes in blood urea concentration. A high protein diet or increase protein
Catabolism such as occurs in fever, major illness, stress, corticosteroid therapy and gastrointestinal hemorrhage, may increase urea levels. Levels will be decreased during periods of low protein intake or increased protein synthesis, such as late pregnancy and infancy\(^{(16)}\).

Decreased renal function causes an increase of plasma urea concentration due to compromised urea excretion renal causes of an elevated urea include acute and chronic renal failure, glomerular nephritis. Tubular necrosis, and other intrinsic renal disease\(^{(16)}\).

Post-renal elevations of urea are due to obstruction to the urine flow anywhere in the urinary tract by renal stones, tumor of the bladder or prostate or severe infection\(^{(16)}\).

2-4 Albumin:

Albumin is a protein manufactured by the liver, and it perform many functions including maintaining the osmotic pressure that cause fluid to remain within the blood stream instead of leaking out into the tissue\(^{(17)}\). The normal value of albumin is depends on the laboratory running the test. Most laboratories consider roughly 3.5 to 5.5 grams per deciliter to be normal. Liver disease, kidney disease and malnutrition are the major causes of low albumin. A diseased liver produces insufficient albumin. Diseased kidney sometimes lose large amounts of albumin into the urine faster than the liver can produce it (this is found in nephritic syndrome). In malnutrition there is not enough protein in the patients diet for the liver to make new albumin\(^{(17)}\).

2-5 Hypoalbuminemia in dialysis, is it a marker for malnutrition or inflammation?

Hypoalbuminemia in dialysis is a highly prevalent condition associated with morbidity and mortality. Hypoalbuminemia although not synonymous of malnutrition, is highly related to it. poor nutrient intake,
hypoalbuminemia. In addition, it has been recently reported that a systemic inflammatory response may participate in developing hypoalbuminemia in chronic renal failure\textsuperscript{(18)}. Uremia \textit{perse}, or through mechanisms stimulated by the use of current dialysis membranes and/or solution, seems to trigger the inflammatory process, remarkably associated with hypoalbuminemia\textsuperscript{(18)}, infections, to which patients on dialysis are particularly pre-disposed, stimulate productions of the inflammatory response as well\textsuperscript{(18)}, such as the protein losses through dialysis, may cause and increase malnutrition\textsuperscript{(18)}. Patients with chronic renal failure develop hypoalbuminemia due to a complex setting of conditions, with systemic inflammatory response as a major cause, not with standing. Other factors such as malnutrition and over-hydration can also play a relevant role\textsuperscript{(18)}.

Serum albumin is a valid and clinically useful measure to protein energy nutritional status in maintenance pre-dialysis patients. The pre-dialysis stabilized serum albumin is a measure of visceral protein pool size and individuals with pre-dialysis or stabilized serum albumin that is zoom should be evaluated for protein energy malnutrition\textsuperscript{(19)}. Also the presence of acute and chronic inflammation limits the specificity of serum albumin as nutritional marker\textsuperscript{(19)}.

2-6 \textbf{Albumin as nutritional marker:}

Serum albumin levels have been used extensively to assess the nutritional status of individual with and without chronic renal failure (CRF). Malnutrition is common in the end stage renal disease population, and hypoalbuminemia is highly predictive of failure mortality risk when present at the line of maintenance dialysis. It follows nutritional interventions that maintain or increase serum albumin levels may fall modestly with sustained decrease in dietary protein or energy intake.
Conversely, serum albumin levels may fall actually with inflammation or acute or chronic stress and increase following resolution or recovery\(^{(20)}\).

Although no single ideal measure of nutritional status exists, the serum albumin concentration is considered to be a useful indicator of protein – energy nutritional status in maintenance, dialysis and the powerful association between hypoalbuminemia and mortality risk in the maintenance dialysis population, strongly support this condition. In addition, the measurement of serum albumin levels is inexpensive, easy to perform, and widely available\(^{(20)}\).

**2-7 Is serum albumin a marker of nutritional status?**

Hypoalbuminemia is a strong predictor of mortality and morbidity in hemodialysis patients, can be consequence of a combination of malnutrition and inflammatory reactions. The purpose of this study which carried in Sao Paulo, Brazil, to analyze serum albumin as a marker of nutritional status in maintenance hemodialysis\(^{(21)}\). Patient with no signs of inflammation.

The study, which is cross-sectional, showed that the serum albumin did not discriminate well, between nourished and malnourished hemodialysis patients without evidence of inflammation\(^{(21)}\).

**2-8 Nutritional status in chronic renal failure:**

Malnutrition is a decreased intake of calories or micronutrients (vitamins and trace elements) resulting in a risk of impaired physiologic function, associated with increased morbidity and mortality\(^{(22)}\).

Malnutrition is frequently present in early stages of chronic renal failure but is often diagnosed in later stages of disease\(^{(23)}\). The systemic follow up of patients combined with the use of biochemical and
biophysical markers well help to identify the onset of nutritional disorders and rapid assessment of ongoing treatments.

During the evaluation of chronic renal failure, malnutrition can appear when glomerular filtration assessed by creatinine clearance becomes lower than 40ml/min/1.73m$^2$(23). Different mechanism can explain this state of malnutrition: reduction in protein and caloric intakes, increasingly marked of renal function is altered; disorders in metabolism of the main nutrients; increased protein catabolism due to acidosis and related infections or inflammations (more frequent by age)(23). 40% of patients show malnutrition symptoms catabolism factors. Malnutrition represents the main cause of morbidity and mortality in dialysis patients(23).

It may seem puzzling that where as hypoalbuminemia$^{(24,25)}$ and inflammation$^{(26,27)}$, have been shown to be important predictors of mortality complications from malnutrition and inflammation as well are not common causes of mortality on dialysis patients$^{(28)}$.

Serum albumin level is the most widely used indicators of malnutrition. However since its half-life is about 20 days, serious protein and caloric depletion is already present by the time it is shown. It is therefore useless to measure it every 15 days$^{(29)}$. Some states are accompanied by hypoalbuminemia during which albumin leaks in the interstitial fluid: nephrotic syndrome, old age and inflammation$^{(30)}$.

During inflammation hepatic synthesis of inflammatory proteins such as CRP, SAA, $\alpha_1$-antitrypsine and $\alpha_1$-globulin is increased is stimulated by pro-inflammatory cytokines such as tumour necrosis factor-$\alpha$, interkin-1 and interkin-6, while the synthesis of malnutritional serum albumin remains a good prognostic indicators in dialysis chronic renal failure patients, regardless of the cause of hypoalbuminemia$^{(31)}$. 
2-9 Pathophysiology of protein–energy wasting in chronic renal failure:

There is a high prevalence of protein-energy malnutrition in both non-dialyzed patients with advanced chronic renal failure and those end stage disease who are receiving maintenance hemodialysis or chronic peritoneal dialysis therapy. Approximately one-third of maintenance dialysis patients have mild to moderate protein energy malnutrition and about 6 to 8 percent of those individuals have severe malnutrition. The statistics of major concern because of morbidity and mortality\(^{(32)}\).

It was recently, in a large number of patients, demonstrated that as glomerular filtration rate declines, \(< 20\text{-}28\text{ml/min}\) signs of nutritional deterioration develop with declining levels of serum albumin\(^{(33)}\) one component of this decline in the nutritional status may be due to spontaneous reduction indicative caloric intake\(^{(34)}\). Albumin has long been used in the assessment of hospitalized patients. Low levels of serum albumin may reflect low hepatic production or protein loss from the vascular component. Low levels have been identified as a common abnormality in patients in long-term care facilities\(^{(35)}\). Hospitalized patients with low serum albumin levels experienced a four-fold increase in mortality\(^{(36)}\) and six-fold increase in mortality. Serum albumin so far has been the most common to assess malnutrition and hypoalbuminemia has sometimes, perhaps erroneously, been used to diagnose malnutrition\(^{(37)}\). Despite those findings and the fact that today several alternative methods are available for assessing nutritional status, serum albumin still seems to be, by far the most commonly used nutritional marker in CRF patients\(^{(38)}\). Serum albumin concentration is best seen as being regulated by several factors especially protein malnutrition, inflammation and external losses\(^{(38)}\). But serum albumin is not good indicator of short-term protein
and energy deprivation, however, albumin levels are good indicator of chronic deficiency\(^{39}\). Traditionally, albumin has been used to help in determination two important nutritional status, first, it help to identify chronic protein deficiency under conditions of adequate non-protein calorie intake, which lead to marked hypoalbuminamia this may result from the net loss of albumin from both in the intravascular and extravascular pools, causing Kwashiorker\(^{39}\).

Second, albumin concentration may help to define marasmus, this is caused by caloric insufficiency without protein insufficiency, so that the serum albumin levels remains normal but there is considerable loss of body weight.

Studies have classified various levels of malnutrition by using albumin levels. Serum albumin levels of 3.5g/dL or greater are considered normal\(^{39}\). Albumin levels of 2.7g/dL indicate moderate malnutrition, and levels less than 2.1g/dl indicate severely depleted levels of albumin\(^{40}\). Serum albumin is an accurate marker of the catabolic stress of infection\(^{41,42}\).

**2-10 C. Reactive Protein (CRP):**

C. Reactive Protein (CRP) appears in the blood of patients with diverse inflammatory disease, but it is undetectable in healthy individuals. It's synthesized in the liver CRP was so named because it precipitates with C. Substance, a polysaccharide of pneumococci. However, it was found that CRP vises sharply whenever their is tissue necrosis, whether the damage originates from a pneumococcal infection or some other source.

This lead to discover that CRP recognizes and binds to molecular groups found on a wide variety of bacteria and fungi. CRP bound to bacteria promotes the binding of complement. CRP is elevated in acute
rheumatic fever, bacterial infections, myocardial infection, rheumatoid arthritis, cersonomatosis, gout and viral infections\(^{43}\). In recent years, several reports have suggested that inflammation, alone or in combination with a low protein intake, plays a significant role in causing hypoalbuminemia in chronic renal failure patients\(^{44,45}\). The prevalence of an increased CRP (>8 – 10mg/L) has been reported to be high in dialysis\(^{17,46,47}\) and predialysis patients\(^{20}\). Serum levels of CRP appear to reflect generation of pro-inflammatory cytokines (interleukin-1 (14-1)), IL-6 and tumor necrosis factor-\(\alpha\) (TNF-\(\alpha\)) which have also been reported to be increased in CRF patients\(^{17,28,48}\).

It's well documented that high levels of proinflammatory cytokines may cause muscle wasting by stimulating protein catabolism via the ubiquitin – profeosome pathway\(^{49}\) by reducing albumin synthesis and by inhibiting appetite\(^{50}\).

Hypoalbuminemia is a major risk factor for morbidity and mortality in the ESRD population. The core indicators project that the serum albumin value is a measure of the patients nutritional status. While the serum albumin level is a measure of the visceral protein size, a decrease in albumin synthesis is due to more than poor nutritional intake (in part related to inadequate dialysis). A cute phase reactants and the plasma volume status are other major factors that impact on serum albumin deformation. It's recommended that the serum albumin level be eliminated as an indicator of nutritional status in the ESRD patients\(^{51}\). During chronic renal failure, malnutrition is responsible for increased morbidity and mortality. Both protein and energy intakes decrease during the cause of renal insufficiency\(^{52}\). During dialysis, severe malnutrition is found in 25% of patients and compromises the prognosis. Indicators of protein nutrition such as protein catabolic rate, serum albumin and pre-albumin, which are the best markers of the prognosis, must be integrated
in the follow up of these patients\textsuperscript{(52)}. Malnourished patients had significantly higher serum creatinine levels\textsuperscript{(53)}.

With possibly are exception, all of the approximately 30 to 40 surveys to the nutritional status of patients undergoing maintenance hemodialysis or peritoneal dialysis indicate that there was high incidence of protein-energy malnutrition; the prevalence rate is about 16 to 54 in various reports\textsuperscript{(54,55,56,57)}. Several studies in maintenance dialysis patients indicate that part of the predictive value of serum albumin can be accounted for by co morbid conditions\textsuperscript{(58)}, and that serum albumin levels correlate with dietary protein intake\textsuperscript{(59)}, and that serum albumin levels correlate inversely with certain acute phase proteins including C. Reactive protein\textsuperscript{(60)}. The observations have engendered the hypothesis that the direct correlation between serum albumin and mortality reflects the presence of co morbid conditions in patients with low serum albumin concentrations, the low serum albumin is caused by increased tissue levels or actions of inflammatory and catabolic cytokines due to the co morbid conditions or possibly to uremia \textit{perse}. Indeed, plasma concentrations or leukocyte production of interleukin-6 and tumor necrosis factor-\textit{\alpha} are increased in advanced chronic renal failure\textsuperscript{(60,61,62)}. 
Chapter Three

3- Objectives

3-1 Objectives:
To detect the percentage of hypoalbuminemia among Sudanese patients with chronic renal failure who under maintenance hemodialysis treatment by using albumin as a marker.

3-2 Specific Objectives:
1- To measure serum albumin level in patients with chronic renal failure who have maintenance dialysis.
2- The effect of uremic syndrome on the level of serum albumin (correlation urea and/or creatinine level and serum albumin).
Chapter Four
4- Materials & Methods

4-1 Study design:

It is prospective descriptive study conducted in Khartoum State during the period from Feb. 2005 to Dec. 2005, the patients suffered from chronic renal failure and admitted to Dr. Salma Mohammed Suliman dialysis and kidney transplant center.

4-2 Collection and Preparation of Blood Samples:

Hundred blood samples were collected from a chronic renal failure patients, 5ml of venous blood samples were withdrawn into clean plane container and allowed to clot, serum was stored in the refrigerator while analyzed for, urea, creatinine, albumin and C. reactive protein.

4-3 Methods:

4-3-1 Determination of urea by Berthelot reaction (Enzymetic, colormetric, end point, Berthelot method)

4-3-1-1 Principle:

Urase catalyses the conversion of urea to ammonia. In a modified Berthelot reaction. The ammonium ions react with a mixture of salicylate, Hypochlorite and nitroprusside to yield a blue-green dye (indophenol). The intensity of this dye is proportional to the concentration of urea in the sample\(^{63,64,65}\).

\[
Urea + H_2O \xrightarrow{\text{Urase}} 2NH_3 + CO_3
\]

\[
NH_2 + \text{Salicylate} + \text{Hypochlorite} \xrightarrow{\text{Nitroprusside}} 2.2 - \text{dicarboxy indophenol}
\]
4-3-1-2 Assay:

wavelength : Hg 578nm
optical path : 1cm
temperature : 20-25°C or 37°C
measurement: against reagent blank

4-3-1-3 Procedure:

<table>
<thead>
<tr>
<th></th>
<th>Test</th>
<th>STD</th>
<th>Control</th>
<th>Blank</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample</td>
<td>0.01mL</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Standard</td>
<td></td>
<td>0.01mL</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td></td>
<td>0.01mL</td>
<td></td>
</tr>
<tr>
<td>Reagent 1</td>
<td>1.0mL</td>
<td>1.0mL</td>
<td>1.0mL</td>
<td>1.0mL</td>
</tr>
</tbody>
</table>

Mix incubate for 10 min at 20-25°C or 5 min at 37°C

Reagent 2 | 1.0mL | 1.0mL| 1.0mL  | 1.0mL |

Absorbance of the sample (Aₕ) against the reagent blank.

4-3-1-4 Calculation:

\[
\text{Urea (mg/dl)} = \frac{Aₕ}{A_{STD}} \times \text{concentration of standard}
\]

4-3-1-5 Normal values  10-45 mg/dl

4-3-2 Creatinine:

Creatinine will be determined by the chemical method which is described by jaffe (modified jaffe reaction with deproteinization)

4-3-2-1 Principle:

Creatinine reacts with picric acid in alkaline pH to give red-orange chromogen. The intensity of the produced colour is directly proportional to the concentration of the creatinine in the sample.

4-3-2-2 Procedure:

Deproteinization step.
- Pipette into test tubes labeled, test, standard, blank 2.5 ml of distilled water in each tube.
- 1.0 ml of serum in the test tube.
- 1.0 ml of standard reagent in tube labeled STD.
- 0.5 ml of 10% sodium tungstate in each tube.
- 1.0 ml of 2/3N sulphuric acid in each tube.
- Centrifuge all tubes for 3-5 min.

From the supernatant
1) 1.0ml of supernatant into test tube.
2) 1.5ml of standard into standard tube.
3) 1.5ml of blank into blank tube.
4) 0.5ml of 0.75N sodium hydroxide in each tube.
5) 0.5ml of picric acid in each tube.
6) Mix, incubate at room temperature for 10 min.

Read the test absorbance of the test \( (A_S) \) and of standard \( (A_{STD}) \) calculation.

\[
\frac{(A_S)}{A_{STD}} \times \text{con. of standard} = \text{concentration of sampe (mg/dl)}
\]

4-3-2-3 Normal values:
0.5-1.3mg/dL

4-3-3 Measurement of serum albumin by Bromocersol green method (BCG) (end point, colorimetric method)

4-3-3-1 Principle:
The measurement of serum albumin is based on its quantitative binding to the indicator 3,3\( \mathcal{S}, 5\mathcal{C} \) - tetrabromo - M cresol sulphone phthalein (Bromo Cresol Green BCG) the albumin – BCG – complex absorbs – maximally at 578nm\(^{(69, 70, 71)}\).
4-3-3-2 Assay:
wave length Hg578nm or Hg 623nm
optical path 1cm
temperature 20°C, 25°C, 37°C
measurement against reagent blank

4-3-3-3 Procedure:

<table>
<thead>
<tr>
<th></th>
<th>Test</th>
<th>STD</th>
<th>Control</th>
<th>Blank</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample</td>
<td>0.02mL</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Standard</td>
<td>-</td>
<td>0.02mL</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td>0.02mL</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reagent</td>
<td>1.0ml</td>
<td>1.0ml</td>
<td>1.0ml</td>
<td>1.0ml</td>
</tr>
</tbody>
</table>

Mix incubate for 10 min at 37°C, read absorbance of sample (A_S) against the reagent blank at 623nm.

4-3-3-4 Calculations:

\[
\frac{(A_S)}{A_{STD}} \times \text{con. of standard} = \text{concentration of sample (mg/dl)}
\]

4-3-3-5 Normal value:

Adults:

3.5 – 5.5g/dl

4-3-4 C. Reactive protein latex test:

(Latex agglutination slide test for the qualitative and semi quantitative in vitro determination of C. reactive protein (CRP) in serum)\(^{(72)}\).

4-3-4-1 Principle:

The CRP reagent contain Latex particles coated with anti-human CRP antibodies. When the reagent is mixed with serum containing CRP at level greater than 6mg/l the particles with agglutinate this is interpretated as being a positive sample\(^{(73, 74)}\).
4-3-4-2 Sample collection and storage:
Serum is used undiluted, CRP remains stable in serum for at least 3 days at +15 to +25°C, 6 days at +2 to +8°C or 6 months at -20°C. Frozen samples should be thawed at room temperature and mixed thoroughly before use. Refreezing thawed samples is not recommended. Reject and contaminated. Lipaemic or haemolyzed sera and avoid plasma whose fibrinogen cause non-specific agglutination.

4-3-4-3 Procedure:
Qualitative determinations:
Place successful on the slide:

<table>
<thead>
<tr>
<th>Latex reagent</th>
<th>Sample</th>
<th>Negative control</th>
<th>Positive control</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.05ml</td>
<td>0.05ml</td>
<td>0.05ml</td>
</tr>
</tbody>
</table>

1- Using a suitable pipette drop 0.05ml of undiluted test sample or control beside each drop of latex.
2- Mix and spread to fill the test circle with the stirring bar. Rotate the slide for 2 minutes and observe for any agglutination. Do not read beyond 3 minutes, as drying out of reagent may give the appearance of false agglutination

4-3-4-4 Interpretation of results:
1- Marked agglutination indicates a CRP concentration above 6mg/l (positive).
2- A smooth haemogenous milky suspension indicator a CRP concentration of less than 6mg/l (negative).

4-3-4-5 Normal values in serum\(^{(5, 6)}\):
Normal levels can be up to 6mg/l in healthy adults. There are no significant differences in mean CRP values between adult males and non-pregnant females.
Chapter Five

5- Result

In this prospective descriptive study, hundred blood samples were collected from a chronic renal failure patients.

Table (1), figure (1) represented the distribution of the patients by gender, 74% (74 cases) were males and the rest were females.

The results showed that the major age group 26-32 years constituting 42% (42 cases), 15-26 years, 37-48 years, 59-70 years, 48-59 years, 4-15 years and also 81-92 years constituting, 22% (22 cases), 18% (18 cases), 7% (7 cases), 6% (6 cases), 1% (1 case) and 1% (1 case) respectively, as shown in table (2), figure (2). The distribution of patients by region were represented by table (3), figure (3), the results showed that 45 cases (45%) from central region, 23 cases (23%) from east, 20 cases (20%) from west, 9 cases (9%) from north and 3 cases (3%) from south.

The results showed that 61 cases (61%) have albumin level less than 3.5g/dL and 39 cases (39%) above 3.5g/dL as shown in table (4), figure (4).

Table (5), figure (5) represented the relationship between normality and gender, 30 cases (40.5% within gender, 76.9% within normal albumin level) were males, and 9 cases (34.6% within gender and 23.1% within normal albumin level) were females. 44 cases (59.5% within gender, 72.1% within hypoalbuminemia) were males and 17 cases (65.4% within gender, 27.9% within hypoalbuminemia) were females.

The results showed that 90 cases (90%) of patients with C. reactive protein level less than 6mg/dL (negative) and 10 cases (10%) with C. reactive protein level more than 6mg/dL (positive), as shown in table (6), figure (6).
Table (7), figure (7) represented the relationship between C. reactive protein with gender.

Table (8), figure (8) represented the relationship between C. reactive proteins and normality.

The results showed that 45 cases (45%) have mild-malnutrition, 39 cases (39%), 14 cases (14%), 2 cases (2%), have normal albumin level, moderate-malnutrition and sever-malnutrition respectively as shown in table (9), figure (9).

Table (10) represented the relationship between urea, albumin and creatinine.

Table (11) represented the relationship between urea and creatinine.

Table (12) represented the relationship between urea and age.
Table (5.1): Distribution of patients by gender

<table>
<thead>
<tr>
<th>Gender</th>
<th>Frequency</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>male</td>
<td>74*</td>
<td>74.0</td>
</tr>
<tr>
<td>female</td>
<td>26</td>
<td>26.0</td>
</tr>
<tr>
<td>Total</td>
<td>100</td>
<td>100.0</td>
</tr>
</tbody>
</table>

* Majority of cases were males.
Table (5.2): Distribution of patients by age

<table>
<thead>
<tr>
<th>Age</th>
<th>Frequency</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>4 to 15</td>
<td>1</td>
<td>1.0</td>
</tr>
<tr>
<td>15 to 26</td>
<td>22</td>
<td>22.0</td>
</tr>
<tr>
<td>26 to 37</td>
<td>42</td>
<td>*42.0</td>
</tr>
<tr>
<td>37 to 48</td>
<td>18</td>
<td>18.0</td>
</tr>
<tr>
<td>48 to 59</td>
<td>6</td>
<td>6.0</td>
</tr>
<tr>
<td>59 to 70</td>
<td>7</td>
<td>7.0</td>
</tr>
<tr>
<td>70 to 81</td>
<td>3</td>
<td>3.0</td>
</tr>
<tr>
<td>81 to 92</td>
<td>1</td>
<td>1.0</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>100</strong></td>
<td><strong>100.0</strong></td>
</tr>
</tbody>
</table>

* Most of cases were between the group age of 26-37 years.
Table (5.3): Distribution of patients by region

<table>
<thead>
<tr>
<th>Region</th>
<th>Frequency</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Meddle Sudan</td>
<td>45*</td>
<td>45.0</td>
</tr>
<tr>
<td>Northern Sudan</td>
<td>9</td>
<td>9.0</td>
</tr>
<tr>
<td>Southern Sudan</td>
<td>3</td>
<td>3.0</td>
</tr>
<tr>
<td>Western Sudan</td>
<td>20</td>
<td>20.0</td>
</tr>
<tr>
<td>Eastern Sudan</td>
<td>23</td>
<td>23.0</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>100</strong></td>
<td><strong>100.0</strong></td>
</tr>
</tbody>
</table>

* Great majority of patients from central region.
Table (5.4): Distribution of patients by normality

<table>
<thead>
<tr>
<th>Albumin level</th>
<th>Frequency</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>39</td>
<td>39.0</td>
</tr>
<tr>
<td>Hypoalbuminemia</td>
<td>*61</td>
<td>*61.0</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td>100</td>
<td>100.0</td>
</tr>
</tbody>
</table>

* Most cases were have hypoalbuminemia.
Table (5.5): Normality * Gender Cross tabulation

<table>
<thead>
<tr>
<th>Albumin Level</th>
<th>Gender</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1 male</td>
<td>2 female</td>
</tr>
<tr>
<td>Normal</td>
<td>30</td>
<td>9</td>
</tr>
<tr>
<td>% within Normality</td>
<td>%76.9</td>
<td>23.1%</td>
</tr>
<tr>
<td>% within Gender</td>
<td>%40.5</td>
<td>%34.6%</td>
</tr>
<tr>
<td>% of TOTAL</td>
<td>%30.0%</td>
<td>%40.0%</td>
</tr>
<tr>
<td>Hypoalbuminemia</td>
<td>44</td>
<td>17</td>
</tr>
<tr>
<td>% within Normality</td>
<td>72.1%</td>
<td>27.9%</td>
</tr>
<tr>
<td>% within Gender</td>
<td>59.5%</td>
<td>65.4%</td>
</tr>
<tr>
<td>% of TOTAL</td>
<td>44.0%</td>
<td>17.0%</td>
</tr>
<tr>
<td>Total</td>
<td>74</td>
<td>26</td>
</tr>
<tr>
<td>% within Normality</td>
<td>74.0%</td>
<td>26.0%</td>
</tr>
<tr>
<td>% within Gender</td>
<td>100.0%</td>
<td>100.0%</td>
</tr>
<tr>
<td>% of TOTAL</td>
<td>74.0%</td>
<td>26.0%</td>
</tr>
</tbody>
</table>
Table (5.6): Distribution of patients by C. Reactive Protein level

<table>
<thead>
<tr>
<th>C.R.P. Level</th>
<th>Frequency</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>10</td>
<td>10.0</td>
</tr>
<tr>
<td>Negative</td>
<td>*90</td>
<td>90.0</td>
</tr>
<tr>
<td>Total</td>
<td>100</td>
<td>100.0</td>
</tr>
</tbody>
</table>

* Most cases were negative (less than 6mg/dL).
### Table (5.7): C. reactive proteins * Gender Cross tabulation

<table>
<thead>
<tr>
<th>C. Reactive proteins</th>
<th>Gender</th>
<th>1 male</th>
<th>2 female</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>Count</td>
<td>7</td>
<td>3</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>% within C. Reactive</td>
<td>70.0%</td>
<td>30.0%</td>
<td>100.0%</td>
</tr>
<tr>
<td></td>
<td>C. reactive proteins</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>% within Gender</td>
<td>9.5%</td>
<td>11.5%</td>
<td>10.0%</td>
</tr>
<tr>
<td></td>
<td>% of Total</td>
<td>7.0%</td>
<td>3.0%</td>
<td>10.0%</td>
</tr>
<tr>
<td>Negative</td>
<td>Count</td>
<td>67</td>
<td>23</td>
<td>90</td>
</tr>
<tr>
<td></td>
<td>% within C. Reactive</td>
<td>74.4%</td>
<td>25.6%</td>
<td>100.0%</td>
</tr>
<tr>
<td></td>
<td>C. reactive proteins</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>% within Gender</td>
<td>90.5%</td>
<td>88.5%</td>
<td>90.0%</td>
</tr>
<tr>
<td></td>
<td>% of Total</td>
<td>67.0%</td>
<td>23.0%</td>
<td>90.0%</td>
</tr>
<tr>
<td>Total</td>
<td>Count</td>
<td>74</td>
<td>26</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>% within C. Reactive</td>
<td>74.0%</td>
<td>26.0%</td>
<td>100.0%</td>
</tr>
<tr>
<td></td>
<td>C. reactive proteins</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>% within Gender</td>
<td>100.0%</td>
<td>100.0%</td>
<td>100.0%</td>
</tr>
<tr>
<td></td>
<td>% of Total</td>
<td>74.0%</td>
<td>26.0%</td>
<td>100.0%</td>
</tr>
</tbody>
</table>
Table (5.8): C. reactive proteins * Gender and Normality Cross tabulation

<table>
<thead>
<tr>
<th>C. Reactive proteins</th>
<th>Gender</th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>1 male</td>
<td>2 female</td>
<td>Total</td>
</tr>
<tr>
<td>Positive</td>
<td>Count</td>
<td>3</td>
<td>7</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>% within C. Reactive C. reactive proteins</td>
<td>30.0%</td>
<td>70.0%</td>
<td>100.0%</td>
</tr>
<tr>
<td></td>
<td>% within Normality</td>
<td>7.7%</td>
<td>115%</td>
<td>10.0%</td>
</tr>
<tr>
<td></td>
<td>% of Total</td>
<td>3.0%</td>
<td>%7.0</td>
<td>%10.0</td>
</tr>
<tr>
<td>Negative</td>
<td>Count</td>
<td>36</td>
<td>54</td>
<td>90</td>
</tr>
<tr>
<td></td>
<td>% within C. Reactive C. reactive proteins</td>
<td>40.0%</td>
<td>60.0%</td>
<td>100.0%</td>
</tr>
<tr>
<td></td>
<td>% within Normality</td>
<td>92.3%</td>
<td>88.5%</td>
<td>90.0%</td>
</tr>
<tr>
<td></td>
<td>% of Total</td>
<td>%36.0</td>
<td>54.0%</td>
<td>%90</td>
</tr>
<tr>
<td>Total</td>
<td>Count</td>
<td>39</td>
<td>61</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>% within C. Reactive C. reactive proteins</td>
<td>39.0%</td>
<td>61.0%</td>
<td>100.0%</td>
</tr>
<tr>
<td></td>
<td>% within Normality</td>
<td>100.0%</td>
<td>100.0%</td>
<td>100.0%</td>
</tr>
<tr>
<td></td>
<td>% of Total</td>
<td>39.0%</td>
<td>%61.0</td>
<td>100.0%</td>
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</tbody>
</table>
Table (5.9): Distribution of variable by albumin concentrations

<table>
<thead>
<tr>
<th>Type of malnutrition</th>
<th>Frequency</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sever malnutrition</td>
<td>2</td>
<td>2.0</td>
</tr>
<tr>
<td>Moderate malnutrition</td>
<td>14</td>
<td>14.0</td>
</tr>
<tr>
<td>Mild malnutrition</td>
<td>45</td>
<td>*45.0</td>
</tr>
<tr>
<td>Normal</td>
<td>39</td>
<td>39.0</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>100</strong></td>
<td><strong>100.0</strong></td>
</tr>
</tbody>
</table>

* Most of patients have mild-malnutrition
Table (5.10): Correlation between urea, creatinine and albumin

<table>
<thead>
<tr>
<th></th>
<th>Conc. Urea</th>
<th>Conc. Creatinine</th>
<th>Conc. Albumin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Person correlation</td>
<td>1.000</td>
<td>74.0</td>
<td>0.212</td>
</tr>
<tr>
<td>Sig. (2-tailed)</td>
<td>0.212</td>
<td>0.054</td>
<td></td>
</tr>
<tr>
<td>N</td>
<td>100</td>
<td>100.0</td>
<td>83</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>Conc. Creatinine</th>
<th>Conc. Albumin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Person correlation</td>
<td>0.488**</td>
<td>0.276*</td>
</tr>
<tr>
<td>Sig. (2-tailed)</td>
<td>0.000</td>
<td>0.012</td>
</tr>
<tr>
<td>N</td>
<td>100</td>
<td>83</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>Conc. Albumin</th>
</tr>
</thead>
<tbody>
<tr>
<td>Person correlation</td>
<td>0.212</td>
</tr>
<tr>
<td>Sig. (2-tailed)</td>
<td>0.54</td>
</tr>
<tr>
<td>N</td>
<td>83</td>
</tr>
</tbody>
</table>

** Correlation is significant at the 0.01 level
* Correlation is significant at the 0.05 level
Table (5.11): Correlation between urea and creatinine

<table>
<thead>
<tr>
<th></th>
<th>Conc. Urea</th>
<th>Conc. Creatinine</th>
</tr>
</thead>
<tbody>
<tr>
<td>CO_U Com. Urea</td>
<td>Person correlation</td>
<td>1.000</td>
</tr>
<tr>
<td></td>
<td>Sig. (2-tailed)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>100</td>
</tr>
<tr>
<td>Conc. _Creatinine</td>
<td>Person correlation</td>
<td>0.488*</td>
</tr>
<tr>
<td></td>
<td>Sig. (2-tailed)</td>
<td>.000</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>100</td>
</tr>
</tbody>
</table>

* Correlation is significant at the 0.01 level
Table (5.12): Correlation between urea and age

<table>
<thead>
<tr>
<th></th>
<th>CO_U Conc. Urea</th>
<th>Age</th>
</tr>
</thead>
<tbody>
<tr>
<td>CO_U Con. Urea</td>
<td>Person correlation</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1.000</td>
<td>.200*</td>
</tr>
<tr>
<td>Sig. (2-tailed)</td>
<td></td>
<td>.046</td>
</tr>
<tr>
<td>N</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>AGE</td>
<td>Person correlation</td>
<td></td>
</tr>
<tr>
<td></td>
<td>.200*</td>
<td>1.000</td>
</tr>
<tr>
<td>Sig. (2-tailed)</td>
<td>.046</td>
<td></td>
</tr>
<tr>
<td>N</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>

* Correlation is significant at the 0.05 level
Figure (5.1): Distribution of patients by gender
Figure (5.2): Frequency distribution of variable group of albumin concentration
Figure (5.3): Distribution of patients by region
Figure (5.4): Distribution of patients by serum albumin level
(normality)
Figure (5.5): Distribution of patients by normality and gender

<table>
<thead>
<tr>
<th>Normality</th>
<th>Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>no</td>
<td>30</td>
</tr>
<tr>
<td>up</td>
<td>44</td>
</tr>
</tbody>
</table>

GENDER
- male
- female

normality

- 43 -
Figure (5.6): Distribution of patients by C. reactive protein level

- 44 -
Figure (5.7): Distribution of patients by C. reactive protein and gender
Figure (5.8): Distribution of patients by C, reactive and Normality

C. reactive proteins

Normality

no

up

Count

+ ve

- ve

54

36

7

3

- 46 -
Figure (5.9): Frequency distribution of variable group of albumin concentration

Percentage

Severe malnutrition: 2
Moderate malnutrition: 14
Mild malnutrition: 45
Normal: 39

Group of conc_albumine
Chapter Six  
6- Discussion

In this prospective descriptive study conducted in Dr. Salma Mohammed Suleiman Kidney Dialysis and Transplant Center from February 2005 to December 2005, hundred chronic renal failure patients were represent study population.

The results showed that most of patients were males 74 cases (74%) and the rest were females, this according to the random selection of patients, our results agrees with the results of Aboud Omar\(^6\), who showed that 72 cases (72%) of study population were females. In this study male : female ratio is 2.8 : 1 and also agrees with that ration obtained by Aboud Omar\(^6\) 2.6 : 1.

Most of patients were in the age group 26-37 years and this result agrees with the result that obtained by Aboud Omar (1982)\(^6\) which showed that age group 20-35 years constituting 40 cases (40%) of study populations.

The result showed that 45 cases (45%) from central Sudan, 23 cases (23%) from east Sudan, 20 cases (20%) from west Sudan, 9 cases (9%) from north Sudan and 3 cases (3%) from south Sudan.

This results agrees with the result of Aboud Omar\(^6\) which showed that the majority of patients of CRF were from central Sudan constituting 40 cases (40%).

Markmann (1998)\(^{56}\), which showed that 23-67% of his study population with hypoalbuminemia (level between 3.5g/dL) and this agrees with results which showed that 61 cases (61%) with albumin level between 3.5g/dL and the rest (39 cases, 39%) with normal albumin level. This result is disagree with that obtained by (Aparico M., 1997)\(^{23}\). The
result showed that only 40% of study population have hypoalbuminemia and malnutrition. The difference depend mainly on the number of study population used by Aparico M. which is 1000 patients with chronic renal failure who have maintenance hemodialysis and they take nutritional interventions that maintain or increase serum albumin levels.

The study showed that 10 cases (10%) have C. reactive protein level more than 6mg/dL and the rest were have serum levels below 6mg/dL. 7 cases (70%) with C. reactive protein positive (above 6mg/dL) show hypoalbuminemia and the albumin level is less than 3.5g/dL this is agrees with the results that obtained by Cianciaruso B. and et al., (1995)(54) which showed that more than 54% of study population show hypoalbuminemia with signs of chronic inflammation.

There is statistical association between chronic inflammation with C. reactive protein positive (above 6mg/dL) and hypoalbuminemia, this result agrees wit the results obtained by Cuto Manzano (2001)(18) and also agrees with the results obtained by Kopple (2000)(33).

The above two previous study showed that P. value less than 0.01.

The results showed most of cases had mild malnutrition constituting 45 cases (45%). Their results agrees with the results obtained by Joel D. (1999)(32), which showed that one-third (40%) of study population have mild malnutrition, the results showed 2 cases (2%) had sever malnutrition.

This result is seems to be near to that obtained by Joel D. (1999)(32), which show that 6-8% of study population have sever malnutrition this because he had a large study population.

Michael (1982)(12), showed that there is a statistical association between urea and creatinine P. value (< 0.01) this agrees with our study results.
The results showed that there is statistical association between creatinine and albumin (the P. value < 0.01) and this results agrees with the result of Michael (1982)\(^{(12)}\).

The results showed that there are a statistical association between creatinine and albumin level (P. vale <0.01) and this agrees with results obtained by Djunkavic L. (1998)\(^{(53)}\) which showed that there is a high serum creatinine levels in malnourished patients.
Chapter Seven
7- Conclusion and Recommendations

This study showed that there were 61% of patients with hypoalbuminemia, the clinical investigation diagnosed them as malnourished patients.

The study also showed there were two types of malnutrition exist in chronic renal failure: one without (type 1) and one with (type 2) a concomitant inflammatory response and significant co-morbidity. Type 1 malnutrition is due to uremic toxicity mainly, type 2 usually due to the chronic inflammations.

We recommend that:

- Raising the awareness of the community concerning renal failure.
- Great effort and funding should be directed toward improvement of investigation and detection of the cases and early treatment.
- Further more researches needed to confirm the effect of inflammatory complications on serum albumin level.

Further more researches also needed to establish an national plane using albumin as a marker to detect an early sign of malnutrition during renal failure.
Chapter Eight

References


10. Mauro Verreli, MD., FRCPS, assistant professor, Department of Medicine, Section of Nephrology, University of Manitoba; Canada.


40. Storker P. M., Gumb F. E., Askanazi J. et al., Serum albumin levels as index of nutritional support, surgery; 1982; 91:194.


Appendix

**Urea reagents:**

1- Phosphate buffer 120 mmol/L  
   Sodium salicylate 60 mmol/L  
   Sodium nitroprusside 5 mmol/L  
   EDTA 1 mmol/L  
   Urease 5Kul/L

2- Phosphate buffer 120 mmol/L  
   Sodium hydroxide 400 mmol/L  
   Sodium hypochlorite 10 mmol/L

3- Urea standard 80 mg/dL (13.3 mmol/L)

(Reagents from creasent diagnostic company)

**Creatinine reagents (Jaffe reaction with deproteinization):**

Reagent (1) creatinine standard 2.0mg/dL  
Reagent (2) picric acid 38mmol/L  
Reagent (3) sodium hydroxide 1.2mmol/L  
Reagent (4) sodium tungstate

All reagents are ready for use and stable (manufactured by Vitro scient.)

**Albumin reagent:**

R₁ Albumin standard 4.0g/dl  
R₂ Succinate buffer, pH 4. 75.0 mmol/l  
   Bromcresol green 0.26 mmol/l

All reagents are ready for use (manufactured by Vitro scient.)
C. reactive protein latex test reagent:

1- Latex reagent:
   An aqueous suspension of latex particles coated with anti-human C. reactive protein antibody (1 drop 0.05 ml).

2- Diluents:
   Glycine buffered saline pH 8.2.

3- Positive control:
   Established liquid containing C. reactive protein at a concentration of 30± 6mg/L (1 drop 0.05ml).

4- Negative control:
   Established liquid containing C. reactive protein at a concentration of 6mg/L (1 drop 0.05ml).

All reagent are ready for use (manufactured by Randox Company).