PREVENTION OF PROGRESSION AND REMISSION STRATEGIES FOR CHRONIC RENAL FAILURE: A SINGLE CENTRE SOUTH AFRICAN PERSPECTIVE

Nolubabalo Unati Nqebelele

A Research Report Submitted to the Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, in partial fulfilment of the requirements for the degree of Master of Medicine in the branch of Internal Medicine.

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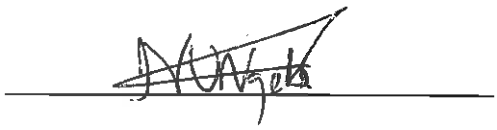
DECLARATION

I, Nolubabalo Unati Nqebelele declare that this research report is my own work. It is being submitted for the degree of Master of Medicine in the University of the Witwatersrand, Johannesburg. It has not been submitted before for any degree or examination at this or any other University.



13 day of December, 2013

DEDICATION

This work is dedicated to the following:

- my parents, Noluvuyo and David for all the years of guidance and support
- my children, Kholo and Wonga, for their never-ending love and encouragement
- my brothers and sisters, for all their support and encouragement
- Khayakazi Magadla, without whom this work would not have been possible.

PRESENTATIONS ARISING FROM THIS STUDY

1. Oral presentation at the Congress of the African Association of Nephrology 2013: February 20-23, 2013, Accra, Ghana.
2. Poster presentation at the World Congress of Nephrology 2013: May 31- June 4, 2013, Hong Kong.

ABSTRACT

INTRODUCTION:

Chronic kidney disease (CKD) is emerging as a global threat to health. In sub-Saharan Africa, most patients do not receive renal replacement therapy due to lack of funds. Measures to retard the progression of CKD are important.

METHOD:

A retrospective review of 122 patients attending a renal clinic, over a two year period was performed. Patients with CKD from hypertension, diabetes mellitus, tubulo-interstitial disease were included. Patients with CKD due to viruses, malignancies and autoimmune diseases were excluded.

RESULTS:

Diabetes mellitus and hypertension were the leading causes of CKD. BP control improved, though 76% were on ≥3 anti-hypertensives. Serum creatinine doubled in 8.2% of patients. BP, acidosis and anaemia were independent risk factors for progression of CKD. The two year renal survival rate was 82%.

CONCLUSION:

Renal function progressed in few patients, which could be related to low levels of proteinuria, good BP control and use of RAS blockers.

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I wish to extend my sincere appreciation to the following people:

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LIST OF ABBREVIATIONS

%	Percentage
AII	Angiotensin II
ACE	Angiotensin Converting Enzyme
ACE-I	Angiotensin Converting Enzyme Inhibitor
ACR	Albumin Creatinine Ratio
ADPKD	Autosomal Dominant Polycystic Kidney Disease
ARB	Angiotensin Receptor Blocker
BMI	Body Mass Index
BP	Blood Pressure
CCB	Calcium Channel Blocker
CKD	Chronic Kidney Disease
CVD	Cardiovascular diseases
CrCl	Creatinine Clearance
dL	deciliter
DBP	Diastolic Blood Pressure
DM	Diabetes Mellitus
ECM	Extracellular Matrix

eGFR	Estimated Glomerular Filtration Ratio
ESRD	End Stage Kidney Disease
FSGS	Focal Segmental Glomerulosclerosis
g	gram
GBM	Glomerular Basement Membrane
GFR	Glomerular Filtration Rate
GPF	Glomerular Capillary Plasma Flow
Hb	Haemoglobin
HCO_3	Bicarbonate
HMG-CoA	3-hydroxy-3-methyl-glutaryl-CoA
Hep B	Hepatitis B
Hep C	Hepatitis C
HIV	Human Immunodeficiency Virus
IgA-N	Immunoglobulin A Nephropathy
IFKF	International Federation of Kidney Foundations
ISN	International Society of Nephrology
KDIGO	Kidney Disease: Improving Global Outcomes (KDIGO)
K/DOQI	Kidney Dialysis Outcomes Quality Initiative
kg	kilogram

MAP	Mean Arterial Pressure
mg	milligram
min	minute
ml	millilitre
mmol	millimol
NCD	Non-Communicable disease
NDD-CKD	Non-dialysis Chronic Kidney Disease
NFκβ	Nuclear Factor κβ
NKF	National Kidney Foundation
NHANES	National Health and Nutrition Examination Survey
NHLS	National Health Laboratory Services (NHLS)
PTH	Parathyroid Hormone
PPPY	Per person per year
p value	probability value
RAS	Renin Angiotensin System
RNA	Ribonucleic Acid
RRT	Renal Replacement Therapy
SADTR	South African Dialysis and Transplantation Registry
SBP	Systolic Blood Pressure

SD	Standard Deviation
SN-GFR	Single Nephron Glomerular Filtration Rate
TGF-β	Transforming Growth Factor β
USA	United States of America
USRDS	United States Renal Data System
w	weight
WHO	World Health Organisation

CHAPTER 1.0 INTRODUCTION

1.0 INTRODUCTION

1.1 CHRONIC KIDNEY DISEASE (CKD) AS A PUBLIC HEALTH PROBLEM

Chronic diseases are diseases of long duration, generally slow progression and are the leading cause of death worldwide (World Health Organisation [WHO] report, 2011). Eighty percent of all chronic disease deaths occur in low and middle income countries, with an increasing number of affected people, families and communities. Eighty five percent of the world's population lives in low-income countries (Barsoum, 2006). These are the communities where the effects of chronic diseases are expected to be greatest.

Non communicable diseases (NCDs) have replaced communicable diseases as the most common cause of morbidity and premature mortality worldwide (WHO report, 2011). In recognition of the increasing burden and importance of chronic diseases, in 2008 the World Health Assembly endorsed a Global NCD Action Plan for NCD prevention and control (Couser *et al.*, 2011). Four NCDs (cardiovascular disease, cancer, diabetes, and chronic respiratory disease) have been prioritized in this global plan. Kidney disease is not currently identified as a separate target but there is compelling evidence that 'it is a key determinant of the poor health outcomes of diabetes and cardiovascular disease (including hypertension)' (Couser *et al.,* 2011).

Chronic kidney disease (CKD) is increasingly recognized as a global public health problem (Levey *et al.*, 2007; Schieppati and Remuzzi, 2005; Perico *et al.*, 2005). The adverse outcomes of patients with CKD include kidney failure, cardiovascular disease (CVD) and premature death (Levey *et al.*, 2005). World Kidney Day was declared in March 2006, with a mission of raising awareness of the importance of kidneys to overall health and to reduce the frequency and impact of kidney disease and its associated health problems worldwide. It is a joint initiative of the

International Society of Nephrology (ISN) and the International Federation of Kidney Foundations (IFKF). This declaration sent a clear message to the public, government health officials, physicians, allied health professionals, patients and families that 'CKD is common, harmful and treatable' (Couser *et al.,* 2011).

1.2 PREVALENCE OF CKD

The 2012 United States Renal Data System (USRDS) Annual Data Report, using data from the National Health and Nutrition Examination Survey (NHANES) revealed the following:

- The prevalence of CKD in 2005-2010, when defined by an eGFR of less than 60 ml/min/1.73m^2, was 6.3 percent. This is in comparison to 9.3 and 8.5 percent for diabetes mellitus and cardiovascular disease respectively. If CKD is defined by albumin creatinine ratio (ACR), the prevalence of CKD rises to 9.2 percent.
- Patients with CKD represent 8.4 percent of the point prevalent population and account for 17 percent of total expenditure: this figure excludes end stage renal disease (ESRD) patients on dialysis or those with a kidney transplant (these account for a further 6.4 percent of fee-for service expenditures).
- In 2010, the overall per person per year (PPPY) costs for patients with CKD reached $22,323 for Medicare patients aged 65 and older and $13 395 for patients aged 50-65 in the MarketScan database. Among Medicare patients with both CKD and diabetes mellitus, the costs were higher and accounted for 24.6 percent of the total Medicare costs of treating diabetes mellitus in 2010, totaling 22 billion.

The prevalence of ESRD in developed countries is increasing; in the United States it is currently 1500 per million population and more than 2000 per million population in Japan (Barsoum,

2006).The figures vary in developing countries. In sub-Saharan Africa; it has been reported to be less than 100 million population, whereas in Latin America, the prevalence is about 400 million per million population (Barsoum, 2006). In sub-Saharan Africa, economic factors dictate a conservative approach to therapy in most instances, with the majority of patients with ESRD dying due to lack of funds; very few can afford regular maintenance dialysis and renal transplantation is often not available (Naicker, 2003). Renal replacement therapy (RRT) places an unaffordable financial burden on poor countries and has negative social and psychological effects on communities (Barsoum, 2006).

1.3 EVALUATION, CLASSIFICATION AND STRATIFICATION OF CKD

1.3.1 DEFINITION OF CKD

In 1997, the National Kidney Foundation (NKF)/ Kidney Disease Quality Outcome Initiative (KDOQI) guidelines were published. They have since become an integral part of clinical practice in many parts of the world. In 2000, the NKF/KDOQI Advisory Board approved the development of clinical practice guidelines to define CKD and to classify stages in the progression of CKD. The Work Group defined CKD to include conditions that affect the kidney, with the potential to cause either progressive loss of kidney function or complications resulting from the decreased kidney function. CKD was thus defined as the presence of kidney damage or decreased level of kidney function for three months or more, irrespective of diagnosis.

Table 1: Definition of Chronic Kidney Disease (adapted from NKF/KDOQI)

1. Kidney damage for ≥3 months, as defined by structural or functional abnormalities of the kidney, with or without decreased GFR, manifest by either:
    - Pathological abnormalities
    - Markers of kidney damage, including abnormalities in the composition of the blood, urine, or abnormalities in imaging tests

    or

2. GFR <60mL/min/ 1.73m^2 for ≥3 months, with or without kidney damage

1.3.2 CLASSIFICATION OF CKD

The Work Group further classified CKD into 5 stages, depending on the level of kidney function, which was based on the glomerular filtration rate (GFR).

Table 2: Stages of Chronic Kidney Disease (adapted from NKF/KDOQI)

Stage	Description	GFR (mL/min/1.73m^2)
1	Kidney damage with normal or↑ GFR	≥90
2	Kidney damage with mild ↓ GFR	60-89
3	Moderate ↓ GFR	30-59
4	Severely ↓ GFR	15-29
5	Kidney failure	<15 or dialysis

Kidney Disease: Improving Global Outcomes (KDIGO) is an independently incorporated organization governed by an international board of directors, with a mission of improving the care and outcomes of kidney disease patients worldwide through promoting coordination, collaboration and integration of initiatives to develop and implement clinical practice guidelines. One of the objectives of a controversies conference by KDIGO was to provide a clear understanding to both the nephrology and non-nephrology communities of the evidence base for the definition and classification of CKD as recommended by K/DOQI. The K/DOQI definition and classification was accepted and endorsed by KDIGO in 2004, with minimal modifications/clarifications.

In October 2009, the leadership of KDIGO, with the endorsement of K/DOQI convened a Controversies Conference to revise the definition and classification of CKD (Levey *et al.*, 2011). The most important changes were:

- Emphasis on the cause as well as the stage
- The use of albuminuria at all stages of GFR
- The subdivision of stage 3 (at 45ml/min per 1.73 m^2)

Table 3: Revised CKD Classification based upon GFR and albumiuria

(adapted from Levey *et al.*, 2011)

Clinical Diagnosis	**GFR stages** (ml/min per $1.73m^2$)	**Albuminuria stages** (ACR, mg/g)
Diabetes Mellitus	>90	<30
Hypertension	60-89	
Glomerular disease	45-59	30-299
Many others	30-44	
Transplant	15-29	>300
Unknown	<15	

The revision of the classification of CKD has resulted in the need for an update to the KDOQI clinical practice guidelines on CKD (Levey *et al.*, 2011). These were published as a global guideline by KDIGO in 2013 as a supplementary in Kidney International, the journal of the International Society of Nephrology.

1.3.3 STAGES IN THE INITIATION AND PROGRESSION OF CKD

1.3.3.1 RISK FACTORS FOR CKD

Risk factors for CKD can be divided into initiating factors and perpetuating factors. Initiating factors are conditions that can directly cause kidney damage, which include a family history of CKD, African American ancestry, hypertension, diabetes mellitus, nephrotoxins, obesity, urological disorders *etc.*

1.3.3.2 PERPETUATING FACTORS

These are factors that influence the rate of decline in kidney function. Kidney disease can progress either as a result of the initial initiating factor or as result of pathways independent of the initial damage.

Table 4: Risk factors for CKD: perpetuating factors (adapted from Taal and Brenner, 2006)

Decreased nephron numbers
Proteinuria
High dietary protein intake
Obesity
Anaemia
Dyslipidaemia
Smoking
Nephrotoxins
Cardiovascular disease

1.4 GLOMERULAR HAEMODYNAMICS IN ANIMAL MODELS

The following landmark discoveries marked the beginning of studies into normal glomerular mechanisms:

- The discovery of a mutant strain of Wistar rats, which possess glomeruli as surface structures in the laboratory of Klaus Thurau of the University of Munich. These rats were then flown on a Lufthansa 747 jumbo jet to the laboratory of Barry Brenner in San Francisco (Brenner, 2002). Brenner and his colleagues named these rats 'Munich-Wistar rats'.
- Wiederhielm and colleagues designed and built a sensitive, servo-null microtransduction system suitable for continuous measurement of microcirculatory pressure of the frog mesentery (Wiederhielm *et al.*, 1963).

Using the above discoveries, the glomerular capillary and proximal tubular hydrostatic pressures were measured and afferent and efferent arteriolar oncotic pressures were estimated during conditions deliberately designed to increase GFR by acute expansion of plasma and extracellular volume (Brenner *et al.*, 1972). This resulted in an approximately two-fold increase in the single nephron (SN) GFR and initial glomerular capillary plasma flow (GPF) (Brenner *et al.*, 1972).

When renal mass is reduced in a rat, the remaining nephrons undergo hypertrophy, with an increase in GPF (Hayslett, 1979). The efferent arteriolar tone increases more than the afferent arteriolar tone resulting in a rise in the hydraulic pressure in the glomerular capillaries and an increase in the amount of filtrate formed by each nephron (Hayslett, 1979). This is initially beneficial as the functional consequences of a decreased renal mass are reduced, but is ultimately detrimental to the remaining glomeruli (Hostetter *et al.*, 1981).

Based on these studies, a mathematical model was developed, which has been used to investigate the influence of a variety of physiological variables on the net driving pressure for ultrafiltration in the rat glomerulus (Deen *et al.,* 1972). This enabled researchers to understand the function of the glomerulus in health and disease (Brenner, 2002).

1.5 MECHANISMS OF PROGRESSION OF CKD

When the GFR in humans falls below about half of normal, further loss of function commonly ensues, even when the original disease becomes inactive (Brenner, 2002). In some instances, the disease responsible for the initial injury may remain active throughout the progression to renal failure (Brenner, 2002). More often, total GFR continues to decline despite a well-defined initiating process having either spontaneously remitted or been controlled therapeutically (Epstein, 1982). In response to reduced renal mass, surviving nephrons undergo adaptations in structure and function that raise the single nephron GFR to meet excretory demands. Pathological studies in different forms of human kidney disease have revealed hypertrophy (presumably reflecting hyperfiltration) of the nephron units least damaged by the original disease (Gottschalk, 1971). These glomerular haemodynamic adaptations responsible for increased single nephron GFR have actually proven to initiate and perpetuate glomerular injury following experimental renal mass ablation. This suggests that similar events may occur when nephrons are lost through disease (Brenner, 1985). The increased pressures and flows acting on remnant glomerular capillaries may be responsible for progression of renal failure after an initial insult has reduced nephron mass (Brenner, 1985).

Elevated glomerular capillary pressures have been identified as the most unfavorable glomerular haemodynamic adaptation to congenital defects or focal nephron obliteration by disease. These elevated capillary pressures lead to glomerular scarring and nephron dropout (Brenner, 2002). Glomerular capillary hypertension is often maintained by angiotensin-dependent mechanisms; by increasing systemic blood pressure and causing efferent arteriolar vasoconstriction. Alleviation of glomerular capillary hypertension was found to be the most important measure in slowing the progression of experimental renal disease (Brenner, 2002)

1.5.1 THE RENIN ANGIOTENSIN SYSYEM

The renin-angiotensin system (RAS) begins with the conversion of angiotensinogen by renin into angiotensin I. Angiotensin I is then converted into angiotensin II by the angiotensin converting enzyme (ACE). Angiotensin II (AII) works via angiotensin I and angiotensin II receptors; activation of which results in haemodynamic and non-haemodynamic effects.

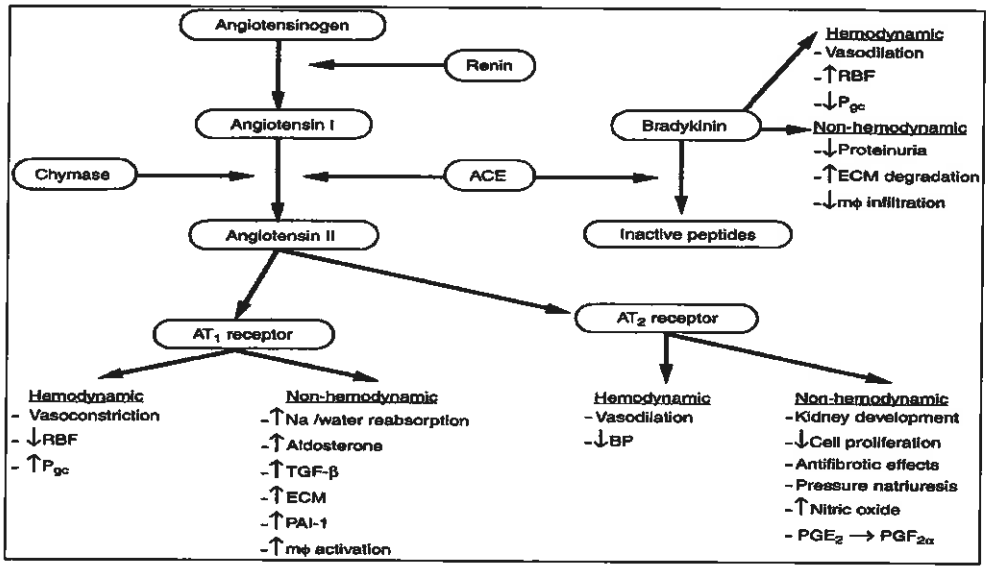

Figure 1: Schema depicting the renin-angiotensin system (adapted from Taal and Brenner, 2000)

Angiotensin also has several non-hemodynamic effects which are also important in progression of kidney disease (Brenner, 2002). These include:

- Production of reactive oxygen species
- Coordinated upregulation of cytokines, cell adhesion molecules and profibrotic growth factors, which in turn give rise to mesangial cell proliferation
- Induction of TGF-β expression
- Increased extracellular matrix proteins
- Stimulation of plasminogen activator inhibitor-1 production by endothelial and vascular smooth muscle cells
- Macrophage activation and infiltration
- Augmentation of adrenal production of aldosterone

- Augmentation of glomerular transcapillary passage of plasma proteins, the principal cause of proteinuria.

In the rat, it seems that high glomerular capillary pressures increase the radius of the pores in the glomerular basement membrane (Lapinski *et al.*, 1995). This results in increased amounts of proteins in the glomerular filtrate, which in turn increases the endocytosis of these proteins by tubular cells (Lapinski *et al.*, 1995).

1.5.1.1 BENEFITS OF RAS INHIBITION IN ANIMAL MODELS

The therapeutic advantage of ACE-I in rats was assessed using micropuncture and morphologic studies in six groups of Munich-Wistar rats who underwent right nephrectomy and segmental infarction of two-thirds of the left kidney (Anderson *et al.*, 1986). Two groups of rats received no specific therapy; two groups received the ACE-I, enalapril. The last two groups were treated with reserpine, hydralazine and hydrochlorothiazide.

In the untreated group of rats, systemic and glomerular hypertension was associated with progressive glomerular structural injury. This was detected as progressive proteinuria and a high frequency of glomerular sclerotic lesions (Anderson *et al.*, 1986). In the enalapril treated rats, control of systemic and glomerular hypertension led to a decrease in progressive proteinuria and markedly fewer glomerular structural lesions (Anderson *et al.*, 1986). In the triple-drug treated rats, there was normalization of systemic blood pressure. However, as the glomerular pressures remained high, the reduction in systemic hypertension did not provide any benefit against glomerular structural injury and glomerulosclerosis was observed with similar frequency as in the untreated group (Anderson *et al.*, 1986).

1.5.2 PROTEINURIA AND PROGRESSION OF CKD

Proteins normally cross the glomerular capillary wall in large amounts and are returned to the bloodstream by a transtubular mechanism involving proximal tubular cells (Myers *et al.,* 1975). Proteinuria has traditionally been regarded as a marker of glomerular filtration barrier integrity. A high level of proteinuria has been used as a marker indicating severe glomerular disease (Brenner, 2002). Proteinuria contributes to progressive renal injury by increasing the production of pro-inflammatory cytokines, extracellular matrix (ECM) proteins and chemoattractants (Abbate *et al.,* 1997). These ultimately lead to tubulointerstitial disease and glomerular fibrosis (Abbate *et al.*, 1997).

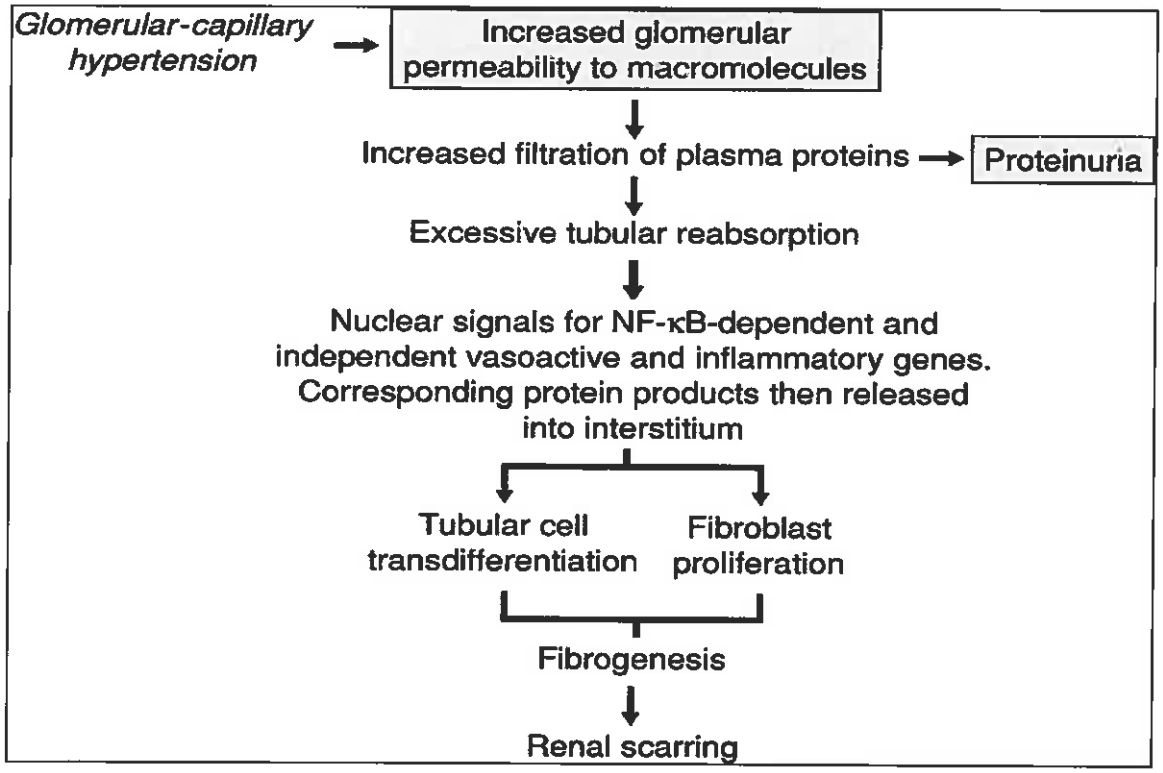

Figure 2: Pathophysiology of progressive nephropathies (adapted from Perico *et al.*, 2005)

1.5.2.1 PROTEINURIC DIABETIC NEPHROPATHY

Diabetic nephropathy, now called 'diabetes with CKD' is the leading cause of ESRD (Brenner *et al.*, 2001).

The Collaborative Study Group trial was a randomized control trial designed to assess the effects of an ACE-I (captopril) on diabetic nephropathy in patients with type 1 diabetes mellitus with proteinuria of > 500mg per 24 hours and baseline creatinine of ≤ than 221 μmol/L (Lewis *et al.*, 1993). There were 207 patients who received captopril and 202 patients who received a placebo. The target blood pressure for both groups was a systolic BP of 140mmHg and a diastolic BP of 90mmHg. The primary end point (defined as doubling of baseline serum creatinine to at least 177μmol/L) was reached in 25 patients in the captopril group, as compared to 43 patients in the placebo group (p=0.007). There was less proteinuria in the captopril group (p=0.001). Blood pressure was reduced to the same degree in both group (Lewis *et al.,* 1993). The use of captopril was associated with a reduction in the risk of doubling of serum creatinine (by almost one half), as well as the combined risk of death, dialysis or transplantation (Lewis *et al.,* 1993).

The Reduction of End Points in Type 2 DM with the Angiotensin II Receptor Antagonist Losartan (RENAAL) study was a randomized, double-blind study comparing the renoprotective effects of the angiotensin receptor blocker (ARB), losartan to placebo (Brenner *et al.*, 2001). Patients were also on additional antihypertensive agents (diuretics, calcium channel blockers, alpha blockers, beta blockers and vasodilators) but ACE-I were excluded. Throughout the study, patients received the standard of care for the treatment of diabetes and were also on lipid lowering agents. A total of 1513 patients were enrolled and followed up for an average of 3.4 years. Losartan reduced the incidence of a doubling of serum creatinine (risk reduction, 25

percent; p=0.006) and ESRD (risk reduction, 28 percent; p=0.002) but had no effect on death. Losartan was also shown to reduce proteinuria by 35 percent, compared to placebo; p=0.005 (Brenner *et al.*, 2001).

Evaluation of the risk factors for doubling of serum creatinine or the development of ESRD in this same cohort was done (Keane *et al.,* 2003). Baseline albumin:creatinine ratio (ACR) was found to be the most powerful independent predictor of doubling of serum creatinine and ESRD, independent of the blood pressure level. Anaemia and hypoalbuminaemia were also found to be independent risk factors for the doubling of serum creatinine and ESRD (Keane *et al.*, 2003).

1.5.2.2 PROTEINURIC NON-DIABETIC NEPHROPATHY

The Ramipril Efficacy in Nephropathy (REIN) study was carried out in 352 patients with chronic non-diabetic nephropathies to assess the effect of ACE-I on renal disease progression and whether the effect was dependent on blood pressure control or not (The GISEN Group, 1997). Patients with a baseline proteinuria level of less than 3g/24h had a slow rate of renal function deterioration (0.25mL/min) compared to patients with baseline proteinuria of 3g/24h or more who had a more rapid decline in GFR (0.67 mL/min). This difference was statistically significant, p=0.0001. In those patients with baseline proteinuria of ≥3g/24h, ramipril significantly slowed renal function decline and halved the combined risk of doubling of serum creatinine or end-stage renal failure (The GISEN Group, 1997). There was a substantial lowering of urinary protein excretion rate in the ramipril treated group, which appeared to exceed what would be expected for the degree of blood pressure reduction. Blood pressure reduction throughout the study was similar in the ramipril and placebo groups, which confirmed that the

renoprotective effects of ramipril were independent of the blood pressure level (The GISEN Group, 1997).

The beneficial effects of ramipril were further confirmed in the REIN follow-up study, where 97 patients from the initial REIN study formed the study cohort and were followed for a further two years (Ruggenenti *et al.*, 1998). Patients originally on ramipril were kept on the same drug, whereas patients previously on other antihypertensive agents were switched to ramipril. In the REIN follow-up study, ramipril slowed the rate of GFR decline and limited the progression to ESRD even more than what was observed in the REIN study (Ruggenenti *et al.*, 1998). Patients originally randomized to ramipril were found to have a 3-fold greater renal survival than those who were originally randomized to placebo plus conventional antihypertensive agents and then switched to ramipril (Ruggenenti *et al.*, 1998). After about 36 months of treatment with ramipril, no additional patients progressed to the point of requiring dialysis, whereas patients switched from conventional therapy to ramipril continued to develop ESRD. This suggests that the sooner treatment with an ACE-I is started, the greater is the capacity of these drugs to protect against a decline in eGFR and ESRD (Ruggenenti *et al.*, 1998).

1.5.3 RENOPROTECTION BY BLOOD PRESSURE CONTROL

Hypertension, together with proteinuria is a major factor contributing to the progression of CKD. The rate of renal progression seems to be related to the level of the mean arterial pressure (MAP); with high MAP values associated with faster progression of renal disease (Locatelli *et al.*, 2005). In animal models, systemic hypertension is associated with increased intraglomerular

pressures and lowering of the blood pressure results in slowing of progression and reduction of renal damage (Anderson *et al.*, 1986).

The situation is similar in human kidney disease. The Multiple Risk Factor Intervention Trial (MRFIT) followed up a large cohort of over 300 000 men for 16 years. Elevated BP was found to be a strong independent risk factor for ESRD, which was independent of associations between age, race, income, treatment for diabetes, history of myocardial infarction, serum cholesterol and cigarette smoking (Klag *et al.,* 1996). This increase in the risk associated with a raised blood pressure was continuous throughout the distribution of BP levels above the optimal level of <120/80 mmHg (Klag *et al.*, 1996).

Similarly, Alvestrand showed that the higher the blood pressure, the faster the progression of renal disease (Alvestrand *et al.*, 1988). Twenty eight patients with CKD due to different aetiologies were studied for a period of 7-24 months. There were retrospective and prospective phases to the study. Comparisons of the rate of progression and level of blood pressure were made between the two phases. They used β-blockers, diuretics and vasodilators to maintain the blood pressure below 160/90mmHg. The patients were divided into two groups based on whether the blood pressure increased or decreased. In the first group, where the MAP decreased by 7±1 mmHg, the progression of renal failure was retarded. The mean decline in creation clearance (CrCL) decreased from 1.1±0.2 ml/min/month in the retrospective phase compared to 0.4± 0.1ml/min/month in the prospective phase ($p<0.01$) (Alvestrand *et al.*, 1998). In the second group, the MAP increased by 3±1mmHg. In this group where the blood pressure increased, there was no difference in the rate of decline of renal function (CrCl in the prospective phase was similar to that in the retrospective phase (0.7±0.1 ml/min per month and 0.6 ml/min per month respectively).

The Modification in Diet in Renal Disease (MDRD) trial was the first large, randomized trial performed to assess the degree of BP reduction needed to achieve renoprotection (Locatelli *et al.*, 2005). There were 840 patients with CKD who were stratified into 2 groups according to baseline renal function. Patients in the first group had a GFR of 25-55 ml/min/1.73m^2, whilst patients in the second group had a GFR 13-25ml/min/1.73m^2. The MAP was similar in both groups at ≤125mmHg. The patients were then randomized to usual BP control (MAP ≤ 107 mmHg in patients aged≤ 60 years or ≤ 113mmHg in patients older than 60 years) or stricter BP control (MAP ≤ 92 mm Hg in participants aged ≤ 60 years or ≤98mmHg in participants older than 60 years) (Peterson *et al.*, 1995). The antihypertensive agents used consisted of diuretics, ACE-I and calcium channel blockers.

Glomerular filtration rate declined faster in patients with higher achieved blood pressure in both groups (over 3 years, r= -0.2, p<0.001 for group 1 and r= -0.34, p<0.001 for group 2) (Peterson *et al.*, 1995). A low blood pressure had a greater effect in persons with higher baseline proteinuria, resulting in significantly reduced proteinuria (Peterson *et al.*, 1995). For patients with baseline proteinuria of > 1g/d, the mean rates of decline in GFR were 6.6ml/min/year and 5.7ml/min/year in group 1 and group 2 respectively. The rate of decline in GFR was unrelated to follow-up BP in patients with baseline proteinuria of less than 1 g/dL. Based on the results from this study, targets for BP control were set based on the level of proteinuria; that is, for patients with proteinuria of more than 1g/d, a target BP of less than 125/75mmHg (MAP 92mmHg) was suggested. For patients with proteinuria of 0.25 to 1g/d, a target BP of less than 130/80mmHg (MAP 98mmHg) was advised (Peterson *et al.*, 1995).

1.5.3.1 CALCIUM CHANNEL BLOCKERS (CCB) AND RENOPROTECTION

Calcium channel blockers are used frequently in patients with renal disease (Ruggenenti *et al.*, 2001). Their effect on renoprotection is controversial.

The effects of an ACE-I/ CCB combination on proteinuria in diabetic nephropathy were studied in 37 patients in a randomized, open label study (Bakris *et al.*, 1998). Patients with type 2 diabetes mellitus and CKD were divided into three groups: ACE-I trandolapril group, non-dihydropyridine CCB verapamil group and a group on combination of both agents. The authors were able to demonstrate that proteinuria was significantly reduced from baseline in the group on both agents (ACE-I plus verapamil) compared to the groups on either drug alone. This antiproteinuric effect was independent of any reductions in BP, as there were no significant differences in arterial pressure, or even GFR, between the groups. They were also able to use a significantly lower dose of each individual component in the group on two agents (Bakris *et al.*, 1998).

In rats CCBs (nifedipine), though providing significant BP reductions, failed to provide renoprotection as they adversely affected renal autoregulation (Griffin *et al.*, 1995).This failure to provide adequate renoprotection was replicated in humans in the REIN study, where patients on dihydropyridine CCBs (nifedipine or amlodipine) had more proteinuria and faster decline in GFR than those on the ACE-I, ramipril (The GISEN Group 1997).

1.5.4 METABOLIC ACIDOSIS

1.5.4.1 EFFECTS OF METABOLIC ACIDOSIS

Metabolic acidosis is often detected as a low serum bicarbonate (HCO_3^-). This is a common complication in patients with advanced CKD, particularly when GFR falls below 30ml/min (Jeffery and Kurtz, 2005). There is usually reduced synthesis of bicarbonate and decreased ability of the kidneys to excrete nonvolatile acids (Kopple *et al.*, 2005). The main complications of chronic metabolic acidosis include decreased protein synthesis and increased protein catabolism resulting in a negative protein balance. Other deleterious effects of chronic metabolic acidosis in patients with CKD include bone demineralization, cell mediated bone resorption, hypertriglyceridaemia and accumulation of β-2 microglobin (Kopple *et al.*, 2005). Correction of the metabolic acidosis with bicarbonate supplementation can result in improvement in bone metabolism and nutritional status.

1.5.4.2 METABOLIC ACIDOSIS AND MORTALITY

In patients with moderate and advanced non dialysis CKD (NDD-CKD), both lower and higher serum bicarbonate levels have been associated with an increased all-cause mortality, which seems to be highest in those with baseline serum bicarbonate levels < 22mmo/L (Kovesdy *et al.*, 2009). The lowest mortality was observed in those with baseline bicarbonate of 26-29mmo/L.

1.5.4.3 METABOLIC ACIDOSIS AND PROGRESSION OF CKD IN ANIMAL MODELS

Wistar rats underwent a 5/6 nephrectomy and were randomly assigned to four groups: non-treated, treated with calcium citrate, treated with captopril or treated with both captopril and calcium citrate groups (Gadola *et al.*, 2004). In the first and 20th week, calcium citrate was shown to slow the progression of chronic kidney injury with less glomerular and tubulointerstitial cellular proliferation ($p<0.005$) and with higher bicarbonate levels at ten weeks ($p<0.005$). Inulin clearances were higher ($p<0.005$) and urine protein excretion rates were lower ($p<0.005$) in the group treated with calcium citrate compared to the untreated group (Gadola *et al.*, 2004).

1.5.4.4 EFFECT OF TREATMENT OF METABOLIC ACIDOSIS WITH SODIUM BICARBONATE SUPPLEMENTATION IN HUMANS

The effects of oral bicarbonate supplementation in humans were investigated in a randomized, controlled trial of 134 adult patients with stages 4 to 5 CKD and serum bicarbonate levels between 16 and 20 mmol/L (Bristol-Ashurt *et al.*, 2009). The rate of decline in the CrCl was slower with bicarbonate supplementation, compared with the control group (1.88 versus 5.93 ml/min 1.73m^2, $p<0.0001$). Patients supplemented with oral bicarbonate were also less likely to experience rapid decline of CrCl, which was defined as a drop in CrCl >3ml/min per 1.73m^2 (9% versus 45%; $p<0.0001$). Fewer patients progressed to ESRD in the bicarbonate group compared to controls (6.5% versus 33%; $p<0.001$). Current guidelines suggest treating patients with alkali when the serum bicarbonate level decreases to <22 mEq/l (Turner *et al.*, 2011).

1.5.5 LIPID NEPHROTOXICITY THEORY

The possible involvement of lipids in the progression of kidney disease was first proposed and investigated by Moorhead and his colleagues (Moorhead *et al.*, 1982).Their hypothesis was that abnormalities of plasma lipid metabolism could be key in the understanding and prevention of renal failure. Initial glomerular basement membrane (GBM) injury resulted in increased permeability and loss of lipoprotein lipase activator, with resultant hyperlipidemia. The filtered lipoproteins accumulated in mesangial cells with stimulation of mesangial cell proliferation and production of excess GBM (Moorhead *et al.*, 1982). The apolipoproteins in the lumen would precipitate and result in worsening of tubulo-interstitial disease (Moorhead *et al.,* 1982).

This 'lipid nephrotoxicity hypothesis' was recently revisited (Ruan *et al.,* 2009). A direct effect of the dyslipidaemia on tubular cells was proposed as the possible mechanism for the acceleration of CKD. Oxidative stress, which is increased in the presence of hyperlipidaemia, was thought to be a possible mediator of the damage to renal tubular cells (Ruan *et al.*, 2009). Activation of the nuclear factor κβ (NFκβ) pathway, with increased inflammatory events in the glomerulonephritis would then result in progression of CKD (Guijarro and Egido, 2001). Other effects of lipids on renal function include increased expression of transforming growth factor β (TGF-β) messenger RNA, with increased matrix expansion (Ruan *et al.*, 2009) and induction of podocyte damage (Fried, 2008).

1.5.5.1 STATINS

Statins are a group of drugs that work by inhibiting the HMG-CoA (3-hydroxy-3-methyl-glutaryl-CoA) reductase. This is the rate-limiting enzyme in the production of mevalonic acid, which is essential for cholesterol synthesis.

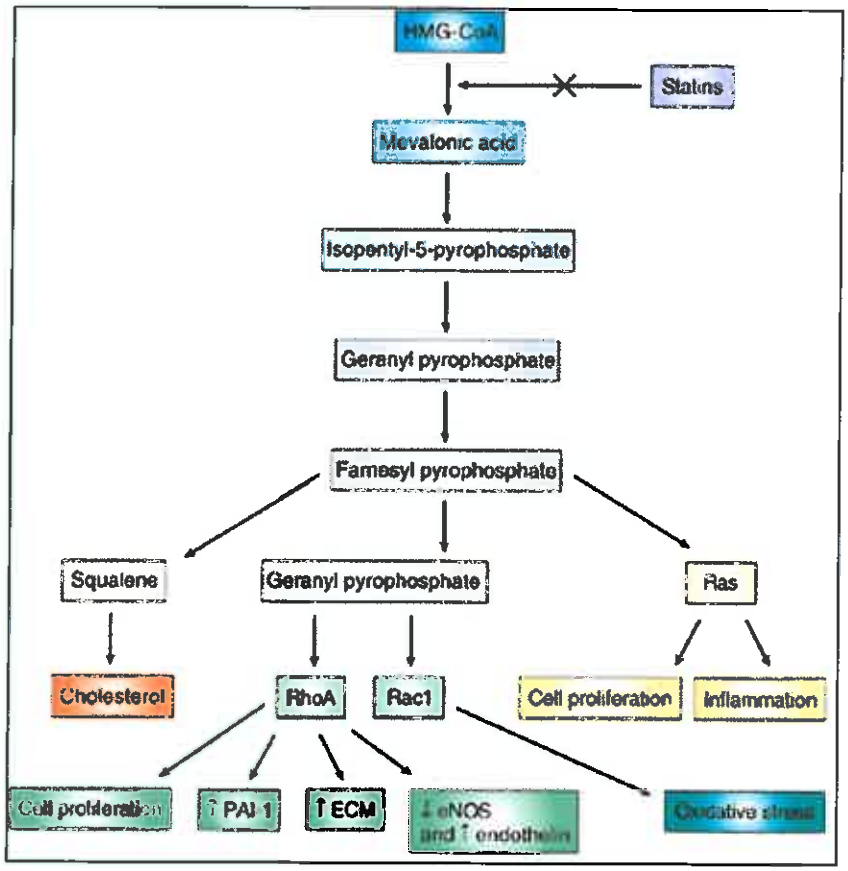

Figure 3: Mevalonic acid pathway (adapted from Fried, 2008)

As they inhibit cholesterol synthesis, statins have been shown to decrease the lipoprotein burden within tissues. They also have additional pleitrophic effects which might explain their renoprotective effects (Fried, 2008).

These pleitrophic effects, which might be of benefit in kidney disease and renoprotection include:

- Inhibition of mesangial proliferation
- Inhibition of induction of TGF-β
- Decreased inflammation and oxidative stress
- Ameriolation of podocyte damage
- Ameriolation of renal vascular disease

1.5.5.2 CLINICAL BENEFITS OF STATINS IN SLOWING PROGRESSION OF CKD

Dyslipoproteinaemia of CKD presents from the early stages of renal functional impairment. This could lead to accelerated atherosclerosis and promote progression of renal disease (Ruggenenti *et al.*, 2001).

There have been conflicting results from human trials looking at the benefits of statins in progression of CKD. In a meta-analysis of 27 randomized controlled and crossover studies, there was a small but statistically significant favourable effect on annual decline in eGFR (1.22ml/min/year slower in patients on statins compared to placebo) (Sandhu *et al.*, 2006). Statins were associated with a reduction in proteinuria, with a resultant reduction in the rate of progression of CKD, especially in populations with cardiovascular disease (Sandhu *et al.*, 2006).

The Study of Heart and Renal Protection (SHARP) trial is the largest randomized double blind trial to assess the efficacy and safety of the combination of simvastatin plus ezetimibe in patients with moderate-to-severe kidney disease. There were 4650 patients on simvastatin plus ezetimibe and 4620 patients on placebo. In the non-dialysis CKD group, the use of statins plus ezetimibe

was not associated with any reduction in the progression of CKD. (Baigent *et al.*, 2011). However, the use of statins was shown to be useful in the reduction of cardiovascular disease complications in patients with CKD (Baigent, Landray *et al.*, 2011).

1.5.6 SMOKING

Cigarette smoking is a modifiable risk factor for cardiovascular disease. Smoking, working via nicotine receptors, causes tachycardia, increases blood pressure, increases catecholamine levels and results in a decrease in GFR (De Fransisco *et al.*, 2005).

The HUNT II study was a large population-based study on Norwegian individuals which looked at the role of smoking in renal function (Hallan and Orth, 2011). There were 65 000 participants in the study, which were divided into four groups; never-smokers (45%), former smokers (26.1%), currents smokers (27.7%) and the last group was made up of individuals for whom the smoking status was missing (1.2%). The patients were followed up for 10.3 years. Using Cox proportional hazard regression analysis, for subjects under the age of 70, a former smoker had a 3.3 times higher risk of future kidney failure compared with never-smokers ($p=0.02$). Current smokers had a 4 times higher risk compared with never smokers ($p=0.01$). The risk increased with an increase in the number of cigarettes smoked and was similar for both sexes.

In patients over the age of 70 years, neither former nor current smokers had a significant increased risk of kidney failure.

The effects of cigarette smoking on renal function in patients with diabetes mellitus was investigated in 180 patients with type 1 diabetes mellitus (Christiansen, 1978). There was a

significant difference in the prevalence of proteinuria between smokers (including ex and current smokers) and non-smokers, with a p value of 0.005.

A retrospective, case control study was done to assess whether smoking increased the risk of progression to ESRD in non- diabetic patients with IgA Nephropathy (IgA-GN) and autosomal dominant polycystic kidney disease (ADPKD) (Orth *et al.*, 1998). They found a significant dose-dependent increase in the risk of progression to ESRD in those patients who smoked. Patients who had a 5 to 15 pack-year history, the odds ratio was 3.5 (95% CI 1.3 to 9.6), whilst for patients who had a pack year history of more than 15 years, the odds ratio was 5.8 (95% CI 2 to 17).

1.5.7 OBESITY

Obesity is a pandemic. The modern lifestyle of overconsumption and decreased energy expenditure is responsible for this phenomenon. Exercise reduces blood pressure and modest weight loss reduces the incidence of type 2 diabetes. Obese patients can develop proteinuria, which is due to an obesity-related glomerulopathy. This is a distinct form of focal and segmental glomerulosclerosis (FSGS). Weight loss can reduce the amount of proteinuria in these patients. Similarly weight loss can result in an improved lipid profile and increased insulin sensitivity. A group of 35 morbidly obese patients underwent biliopancreatic diversion. One year after surgery, there were fewer patients with hypertension (from 61% pre-surgery to 12 % post-surgery, $p<0.05$), diabetes (from 20 % pre-surgery to 0 patients post-surgery, $p<0.005$), hyperlipidemia and there was a reduction in microalbuminuria (De Francisco *et al.*, 2005).

Popular weight-loss diets are commonly high in potassium and protein and can result in hyperkalemia in patients with advanced CKD and are not recommended.

Robust data on the contribution of obesity on progression of CKD, particularly non-dialysis CKD is sparse. Current guidelines recommend that CKD ND patients should maintain a healthy BMI of 20-25 kg/m^2.

CHAPTER 2.0 MATERIALS AND METHODS

2.1 AIM

i. To analyse the progression of CKD in a subset of patients attending the Renal Clinic at Charlotte Maxeke Johannesburg Academic Hospital (CMJAH).

ii. To identify the factors that may be associated with progression of CKD.

2.2 STUDY DESIGN

Retrospective case review study, using available clinical records of patients attending the renal clinic. These records are kept at the Division of Nephrology, Department of Internal Medicine at CMJAH hospital.

2.3 RESEARCH METHODS

Ethics approval was obtained from the Human Research Ethics Committee, University of the Witwatersrand; clearance certificate number M111168 (Appendix A). Permission for the study was granted by the CMJAH medical superintendent, Dr Mofokeng.

All available clinical records were assessed. Records of patients who had attended the renal clinic for a minimum of 2 years with regular follow-up (arbitrarily defined as at least two visits per year) between 1 January 2005 and the 30 December 2008 formed the study cohort. In those who had regular follow-up for a minimum of two years, the following were considered inclusion criteria:

- Age > 18 years
- Diagnosis of chronic kidney disease- the aetiology of which was hypertension, diabetes mellitus, tubulointerstitial diseases or CKD of unknown aetiology.

The following exclusion criteria were applied:

- Acute kidney injury
- Malignancy (history of/or current active malignancy)
- Glomerulonephritides
- Viral infections including human immunodeficiency virus (HIV), hepatitis B and hepatitis C infections
- Intravenous drug use (history of/or current use)

The following variables were collected from the study population in the first year (beginning of the study period) and again at the end of the second year (end of the study period):

- Demographic data: Age, gender, race
- Aetiology of chronic kidney disease
- Weight and height (with calculation of the body mass index using the ratio of the weight to the height squared)
- Blood pressure (this was averaged annually)

All urine and blood results were retrieved from the computers that are managed by the CMJAH laboratory, National Health Laboratory Services (NHLS).

The following blood/urine results were retrieved:

1. Urine protein quantification of a spot urine sample:

- protein: creatinine ratio (g/mmol)

2. Haemoglobin level (from the full blood count)

3. Serum creatinine

4. Serum corrected calcium

5. Serum phosphate

The following were calculated based on some of the above results:

- Estimated Glomerular Filtration Rate (eGFR)

  The Cockroft and Gault formula was used (Cockroft and Gault 1976) as follows:

  $$CCr\ (ml/min) = \frac{(140-age) \times \text{lean body weight (kg)} \times \text{Constant}}{\text{Serum creatinine } (\mu mol/L)}$$

  Where Constant is 1.23 for men and 1.04 for women

- Calcium Phosphate product

  This was calculated by multiplying the calcium level by the phosphate level (both in mmol/L)

For these calculations, a calculator FC-100V (CASIO Corporation) also capable of performing natural logarithms was used.

- Depending on the eGFR, the National Kidney Foundation/Kidney Disease Outcomes Quality Initiative (NKF/KDOQI) classification system for the staging of patients with CKD was used.

The medications that the patients were taking were retrieved from the prescription charts. The different agents were grouped according to mechanism of action/ class of the drug into the following:

- Renin-angiotensin system (RAS) blockers :
    - Angiotensin converting enzyme inhibitors (ACE-I): examples include enalapril maleate (Renitec®, Adcock Ingram Pharmaceuticals, South Africa), perindopril tert-butylamine (Prexum®, Biogaran, South Africa)
    - Angiotensin receptor blockers (ARB): telmisartan (Micardis®, Boehringer Ingelheim, South Africa)
- Calcium channel blockers (CCB): amlodipine besilate (Austell Laboratories (Pty) Ltd, South Africa), nifedipine (Adalat® XL, Bayer Schering Pharma, Germany)
- Beta blockers: Adco-atenolol (Adcock Ingram Limited, South Africa), carvedilol (Vediblok®, Arrow Pharma (Pty) Ltd, South Africa
- Alpha blockers- doxazocin mesylate (Cardugen®, Mylan (Pty)Ltd, South Africa)
- Diuretics-furosemide (Lasix®, Sanofi Aventis, South Africa), hydrochlorothiazide (Ridaq®,Pharmacare, South Africa)
- HMG-CoA reductase inhibitors (Statins)- atorvastatin (Lipitor®, Pfizer, South Africa), simvastatin (Simvotin® MERCK, South Africa)

2.4 DATA COLLECTION

Information was recorded on a data collection form and placed onto an Excel program worksheet. Data was analysed using SPSS version 16.

Statistical analyses were performed as follows:

- Descriptive analyses were carried out with continuous variables being summarised using means, standard deviations and ranges.
- Student T-test was used to compare means.
- Bivariate correlation analysis of creatinine and clinical parameters (such as blood pressure) as well as laboratory markers (such as haemoglobin, bicarbonate and proteinuria) was done using Spearman's correlation.
- Multiple regression analysis was done to assess which factors were independent risk factors for progression of CKD.

Hypothesis testing was considered statistically significant when the 2-sided probability value, p, was less than 0.05.

CHAPTER 3.0 RESULTS

3.1 DEMOGRAPHIC DATA

There were a total of 122 patients enrolled onto the study.

3.1.1 GENDER

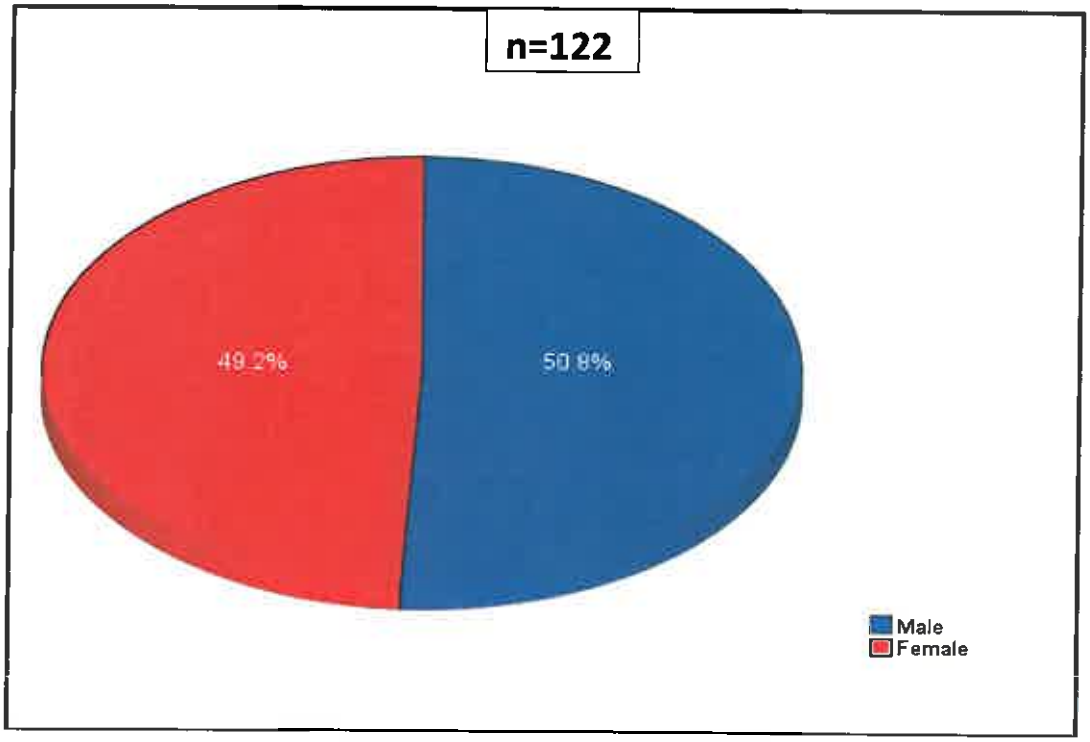

Figure 4: Gender of adult patients with CKD

50.8% of adult patients that formed the study population were males and 49.2% were female.

3.1.2 PATIENT POPULATION RACE GROUP

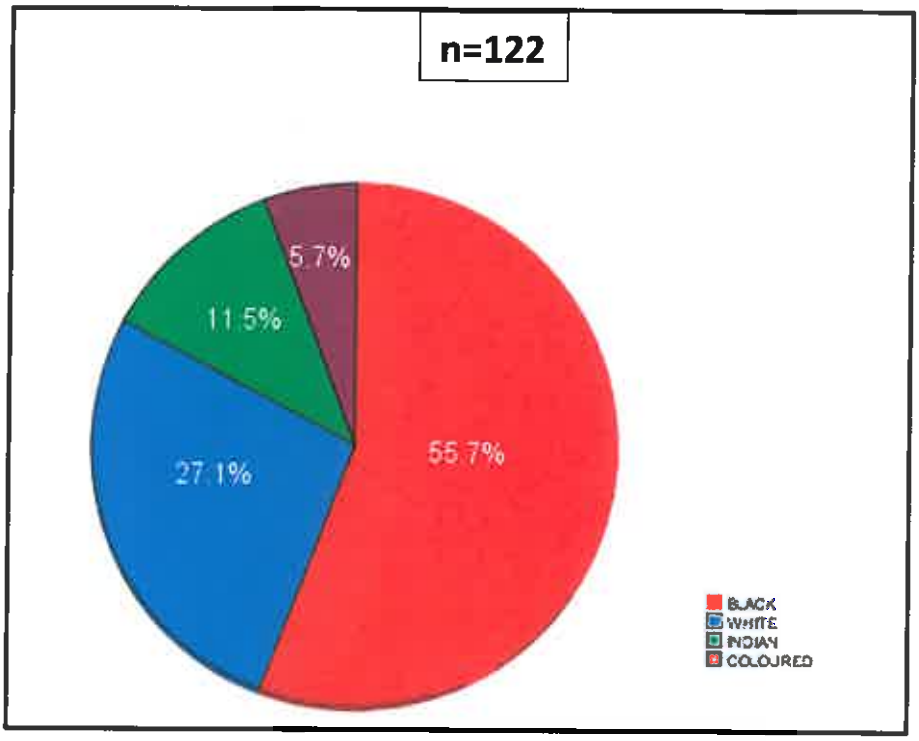

Figure 5: Racial profile of the CKD population

The majority of patients that formed the study population were black Africans (55.7%), the rest being 27.1% Caucasian, 11.5% Indian and 5.7% Coloured.

3.1.3 PATIENT POPULATION AGE RANGES

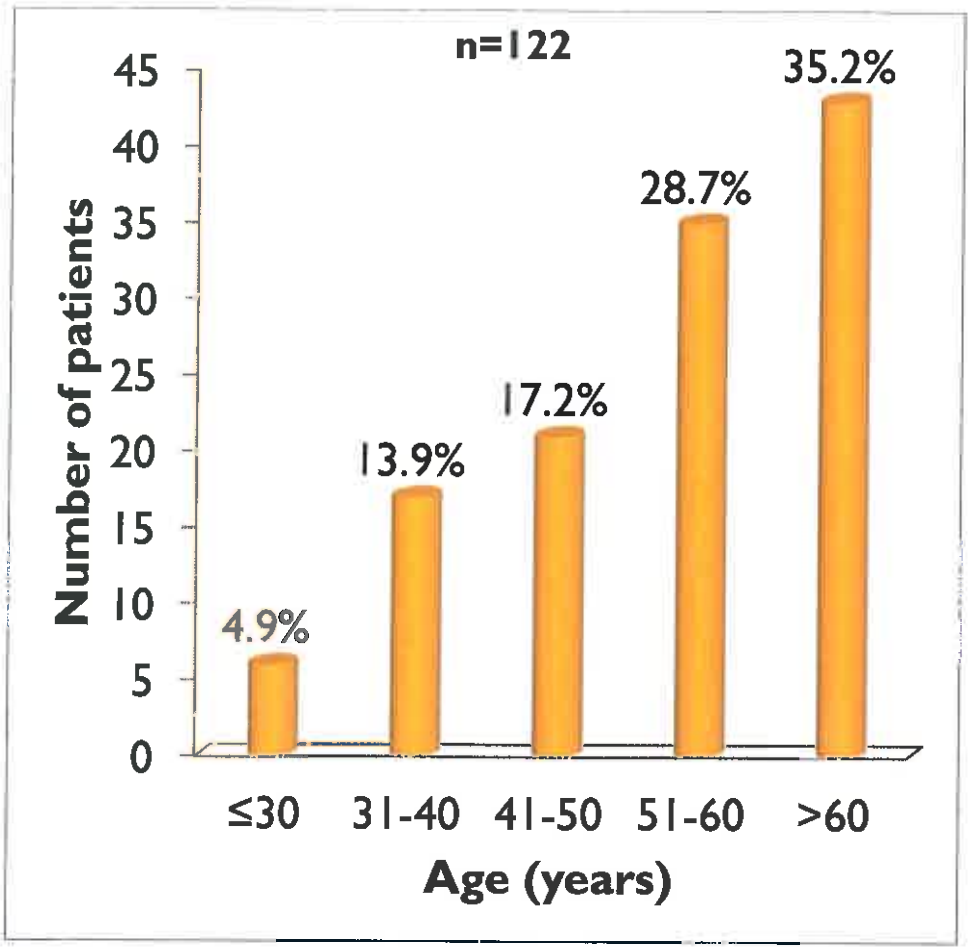

Figure 6: Age of the study population

The mean age at the beginning of the study was 54.1±1.3 years. Most of the adult patients enrolled onto the study were older than 50 years, with the biggest group belonging to the age group of patients older than 60 years (35.2%, n=43). Only 6 patients were younger than 30 years.

3.2 CAUSE OF CKD

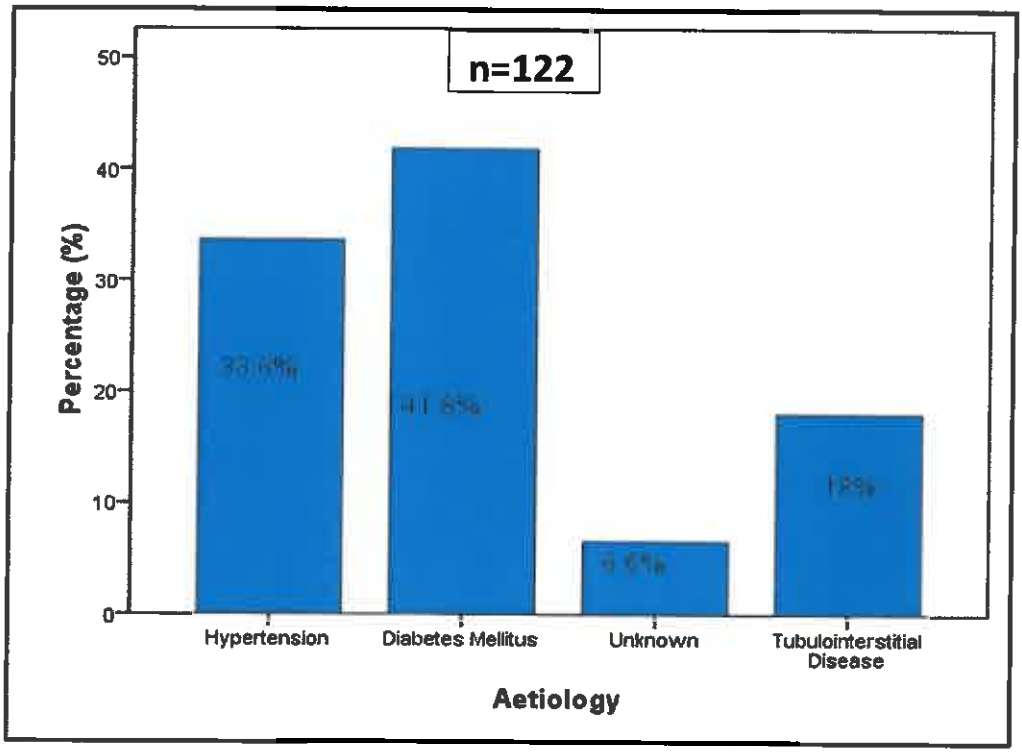

Figure 7: Aetiology of CKD in the study population

Diabetes mellitus was the commonest cause of CKD (41.8%), followed by hypertension (33.6%). Tubulointerstitial diseases were a cause of CKD in 18% of patients. The cause of CKD could not be ascertained in 6.6% of patients.

3.3 BODY MASS INDEX

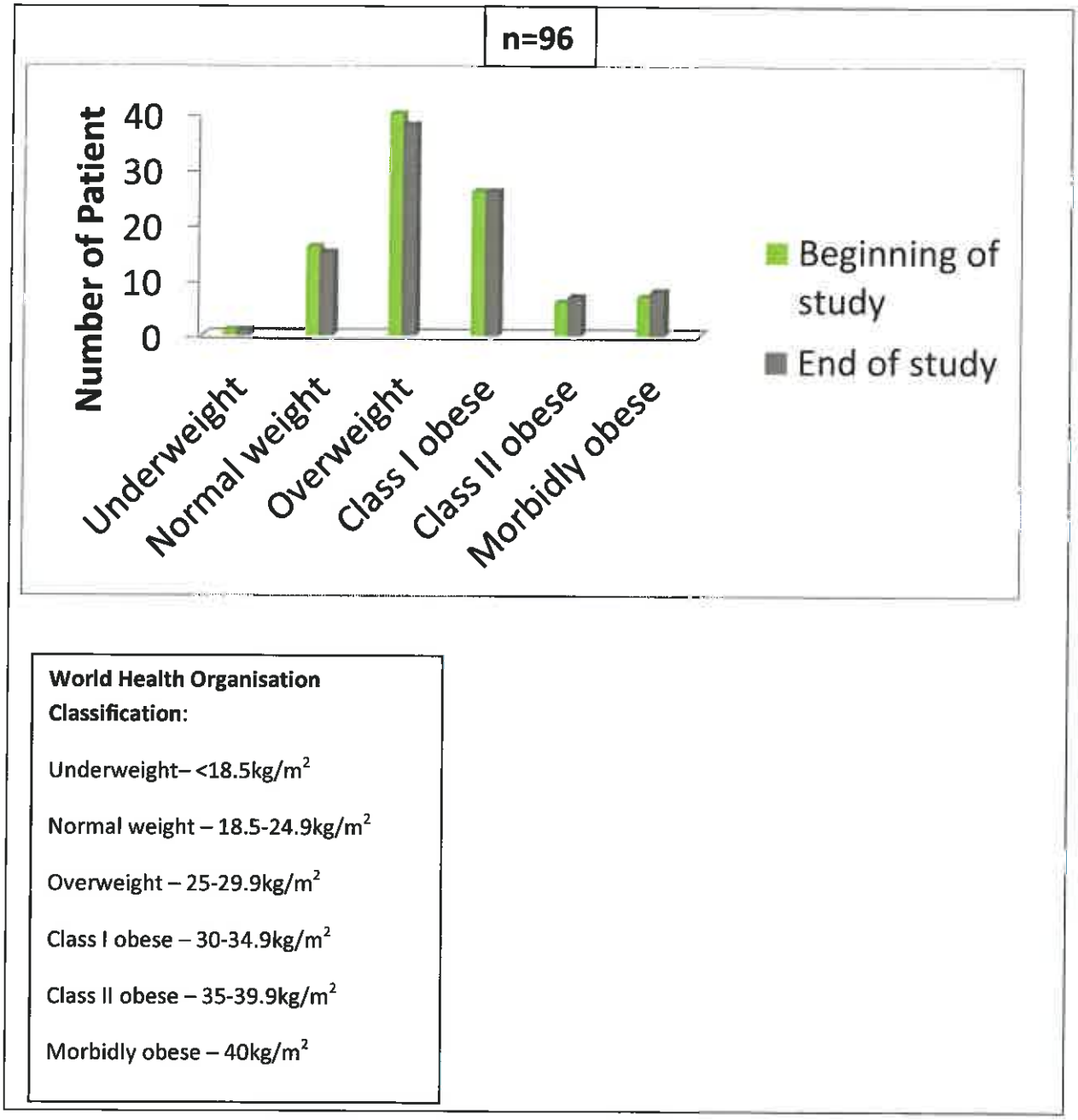

Figure 8: BMI (kg/m^2) of the patients enrolled onto the study

BMI could be calculated in 96 patients. This was due to the unavailability of reliable height or weight measurements in 26 patients. The mean BMI at the beginning of the study was 29.6 $\pm 6 kg/m^2$, whereas at the end of the study the BMI was 30.3 ± 6.31 kg/m^2. This difference was statistically significant (p=0.007).

Most of the patients throughout the study period were overweight; 41.7% (n=40) and 40% (n=38) at the beginning and at the end of the study respectively. Only 16.7% (n=16) at the beginning of the study and 15.6% (n=15) at the end of the study were of normal weight.

3.4 CHARACTERISTICS OF THE STUDY POPULATION

Table 5: Clinical and laboratory characteristics of the study population

Characteristic	Baseline: mean (SD)	End of follow-up : mean (SD)	p value
Serum creatinine (µmol/L)	209.5 (161-266)	228 (176-344)	<0.001
GFR (ml/min per 1.73m^2)	38 (12.1)	33.7 (16)	<0.001
Urinary protein excretion (g/24h)	1.6 (2)	1.37 (1.94)	0.269
BMI (kg/m^2)	29.64 (6)	30.27 (6.31)	0.007
Systolic BP (mmHg)	143.9 (25.6)	136.3 (17.1)	<0.001
Diastolic BP (mmHg)	83.3 (13.9)	75.9 (10)	<0.001
Mean arterial pressure (mmHg)	131.1 (21.1)	121.4 (14.5)	<0.001
Serum potassium (mmol/L)	4.42 (0.6)	4.59 (0.6)	0.306
Serum haemoglobin (g/dl)	13 (11-14.6)	12.5 (11-13.9)	0.032
Serum bicarbonate (mmol/L)	23.6 (3.3)	23.6 (4.5)	1.00

3.5 PROTEINURIA

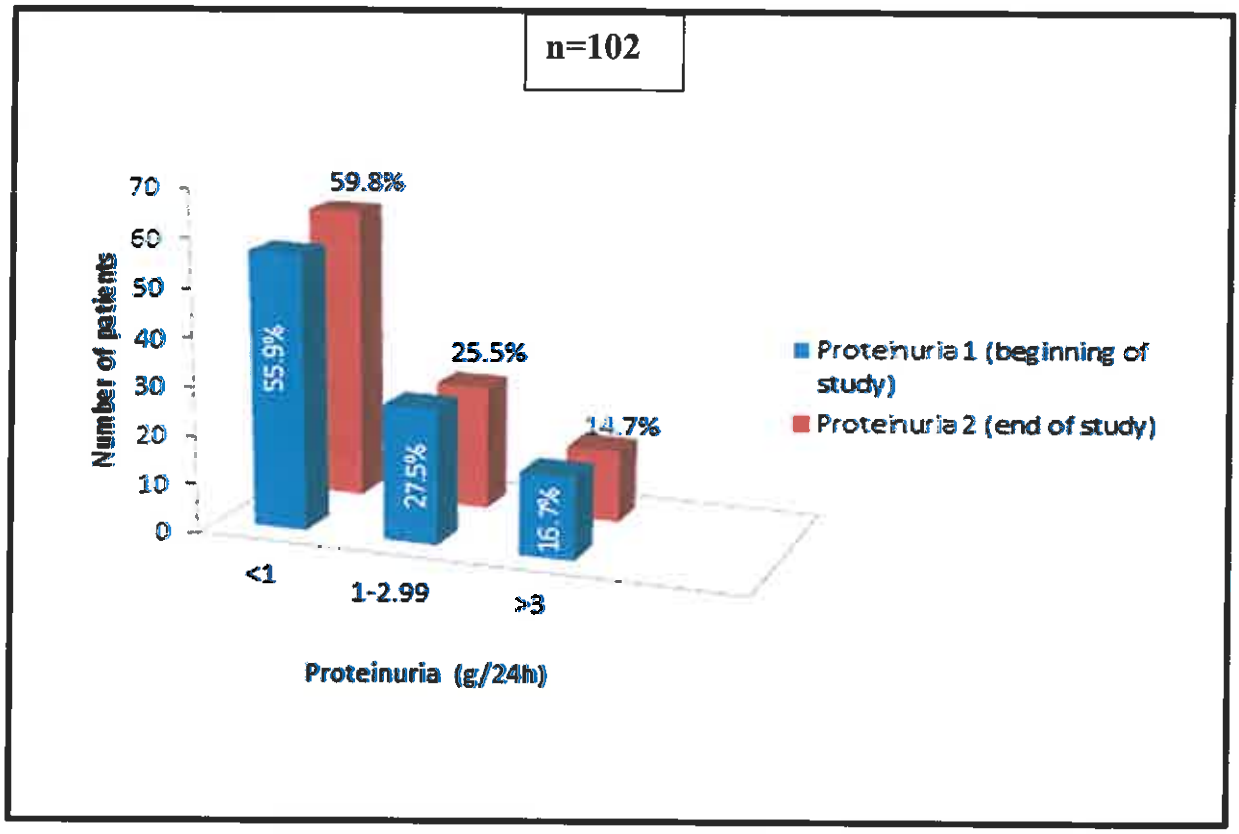

Figure 9: Level of proteinuria in patients with CKD

Laboratory data on proteinuria was available in 102 patients. The mean proteinuria at the beginning of the study was 1.5±1.98 g/24h. At the end of the study, the mean proteinuria level was 1.37±1.94 g/24h. This difference was not statistically significant (p=0.269).

At the beginning of the study period, 55.9% (n=57) of patients had low levels of proteinuria, with proteinuria being less than 1 g/24hours.This figure improved at the end of the study, with 59.8% (n=61) of patients passing less than 1g protein/24h.

3.5.1 NEPHROTIC RANGE PROTEINURIA

Nephrotic range proteinuria (proteinuria > 3g/24h) was present in 16.7% (n=17) at the beginning of the study.

Further analysis of these patients revealed the following:

Table 6: Causes of CKD in patients with nephrotic range proteinuria

Aetiology	Number of patients	Percentage (%)
Hypertension	3	17.6
Diabetes mellitus	11	64.7
Unknown	3	17.6

Diabetes mellitus was the cause of CKD in most of the patients with nephrotic range proteinuria, accounting for 65.7% (n=11) of patients. None of the patients with CKD due to tubulointerstitial disease had proteinuria in the nephrotic ranges.

Table 7: Clinical and laboratory characteristics of CKD patients with nephrotic range proteinuria

Characteristic	Age (years)	BMI (kg/m^2)	Serum creatinine (µmol/L)	Systolic BP (mmHg)	Diastolic BP (mmHg)	Mean Arterial pressure (mmHg)
Median	53	29	260	148	80	133
Interquartile range	40-62	26-30	154-300	125-175	80-99	121-159

The mean BMI for the patients with nephrotic range proteinuria was 29 (26-30) kg/m^2, which makes them overweight according to the WHO classification. The blood pressure in this group was inadequately controlled with a median systolic BP of 148 (125-175) mmHg and a median diastolic BP of 80 (80-99) mmHg. The mean arterial pressure was 133 (121-159) mmHg.

Table 8: Use of RAS blockers in CKD patients with nephrotic range proteinuria

Agent used	Number of patients	Percentage (%)
ACEI	13	76.5
ACEI + ARB	2	11.8
None	2	11.8

Fifteen patients (88.3%) with nephrotic range proteinuria were on agents that block the RAS system, with only 2 patients (11.7%) not on RAS blockers.

3.5.2 CORRELATION BETWEEN PROTEINURIA AND SERUM CREATININE

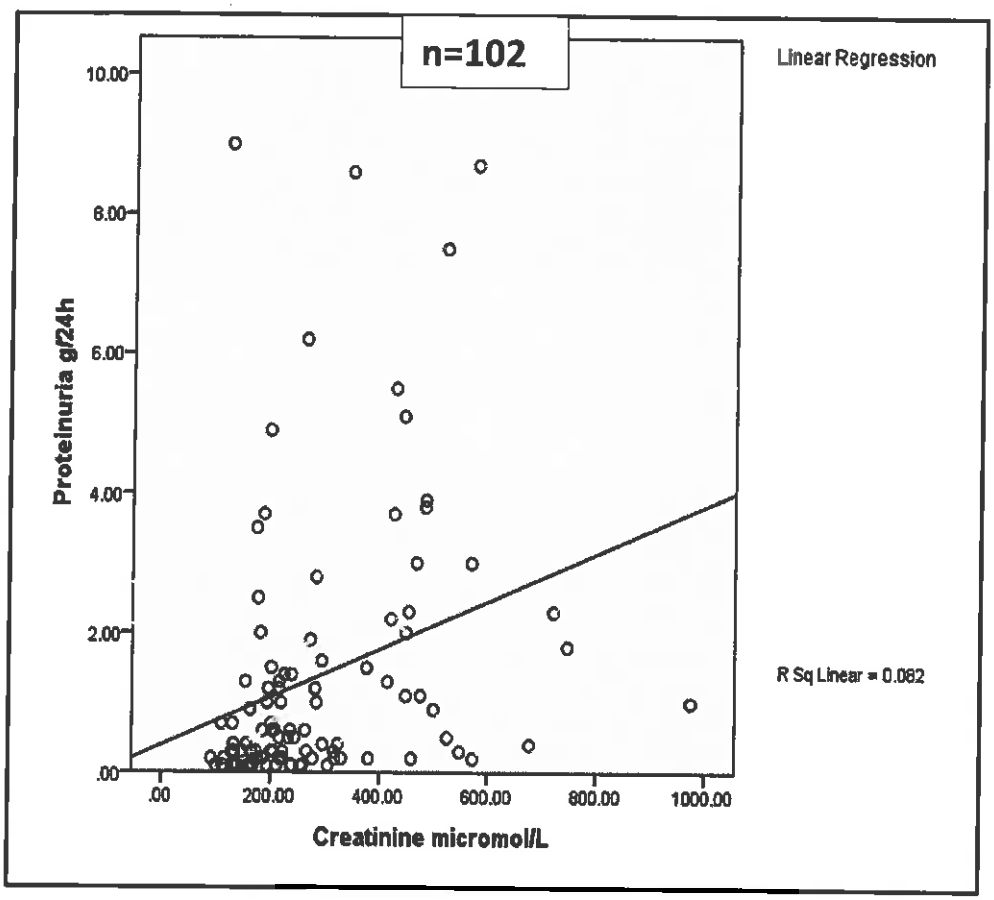

Figure 10: Correlation of proteinuria to serum creatinine

(r=0.286; p=0.004; n=102)

The correlation between the level of proteinuria and creatinine is statistically significant (p=0.004, r=0.286) (Pearson's correlation coefficient)

3.6 RENAL FUNCTION

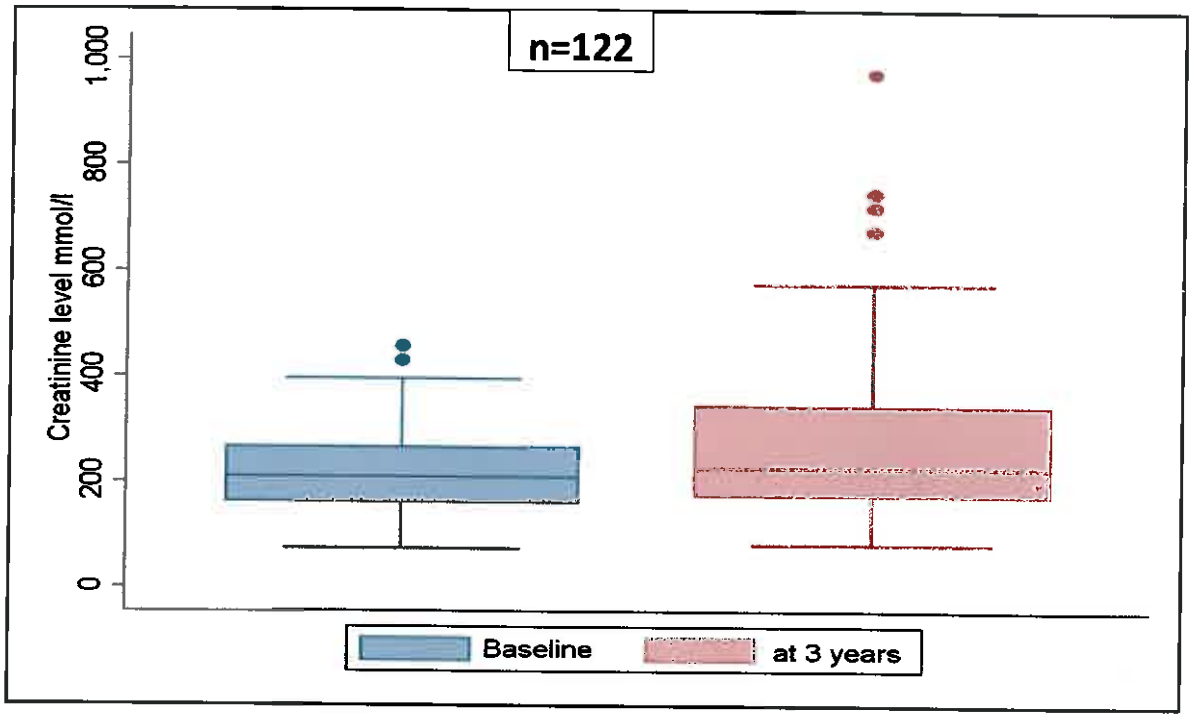

Figure 11: Changes in serum creatinine levels

The baseline median serum creatinine was 209.5 µmol/L with an interquartile range (IQR) of 161-266 µmol/L. At 3 years, the median creatinine level was 228.5 µmol/L, with an IQR of 176-344 µmol/l. This was significant with a p value of <0.001.

The baseline median glomerular filtration rate (GFR) was 36.7 ml/min per1.73m^2, (IQR 28.2-47.4 ml/min per1.73m^2). At the end of the study period, the median GFR was 30.8 ml/min per1.73m^2 (IQR 20.4-45.1 ml/min per1.73m^2). This was significant with a p value <0.001.

3.6.1 STAGES OF CKD

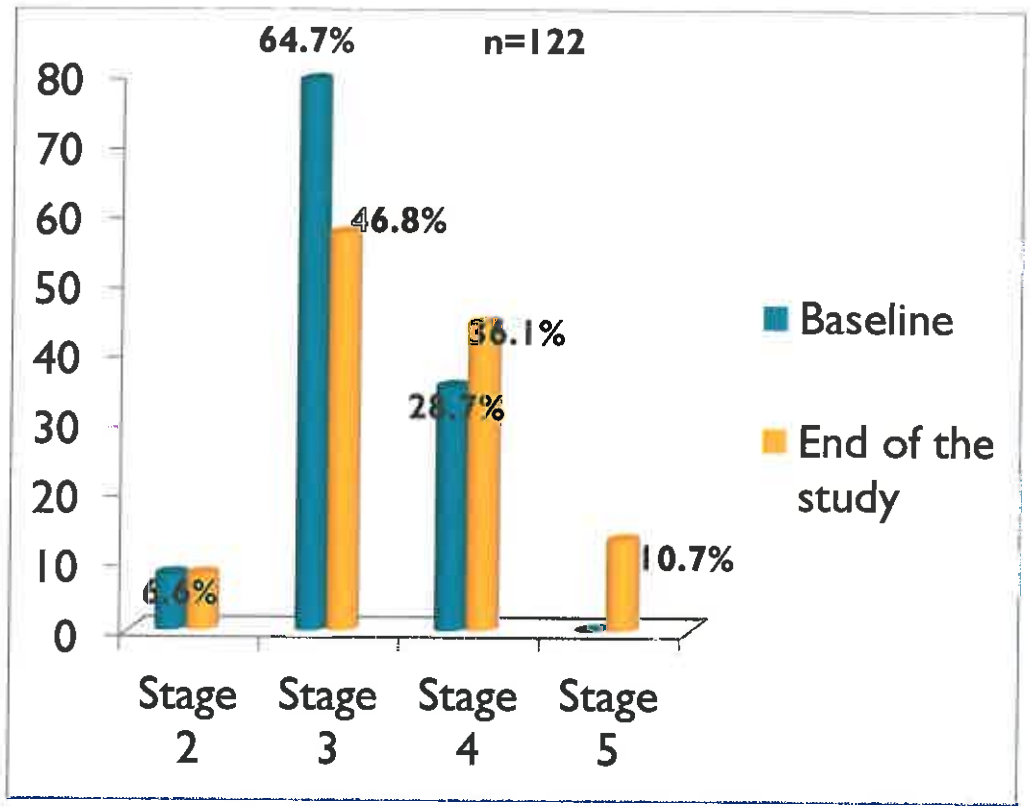

Figure 12: CKD stages at baseline and at the end of the study period

At the beginning of the study period, most of the patients (64.7%) had stage 3 CKD. Only 6.6% of the patient had stage 2 CKD. There were no patients with stage 5 CKD (end stage kidney disease), as this was an exclusion criterion.

At the end of the study period, the majority of patients had stage 3 CKD. There was evidence of worsening of CKD with 10.7% (n=13) of patients having progressed to stage 5 CKD.

3.6.2 CORRELATION OF SERUM CREATININE TO HAEMOGLOBIN

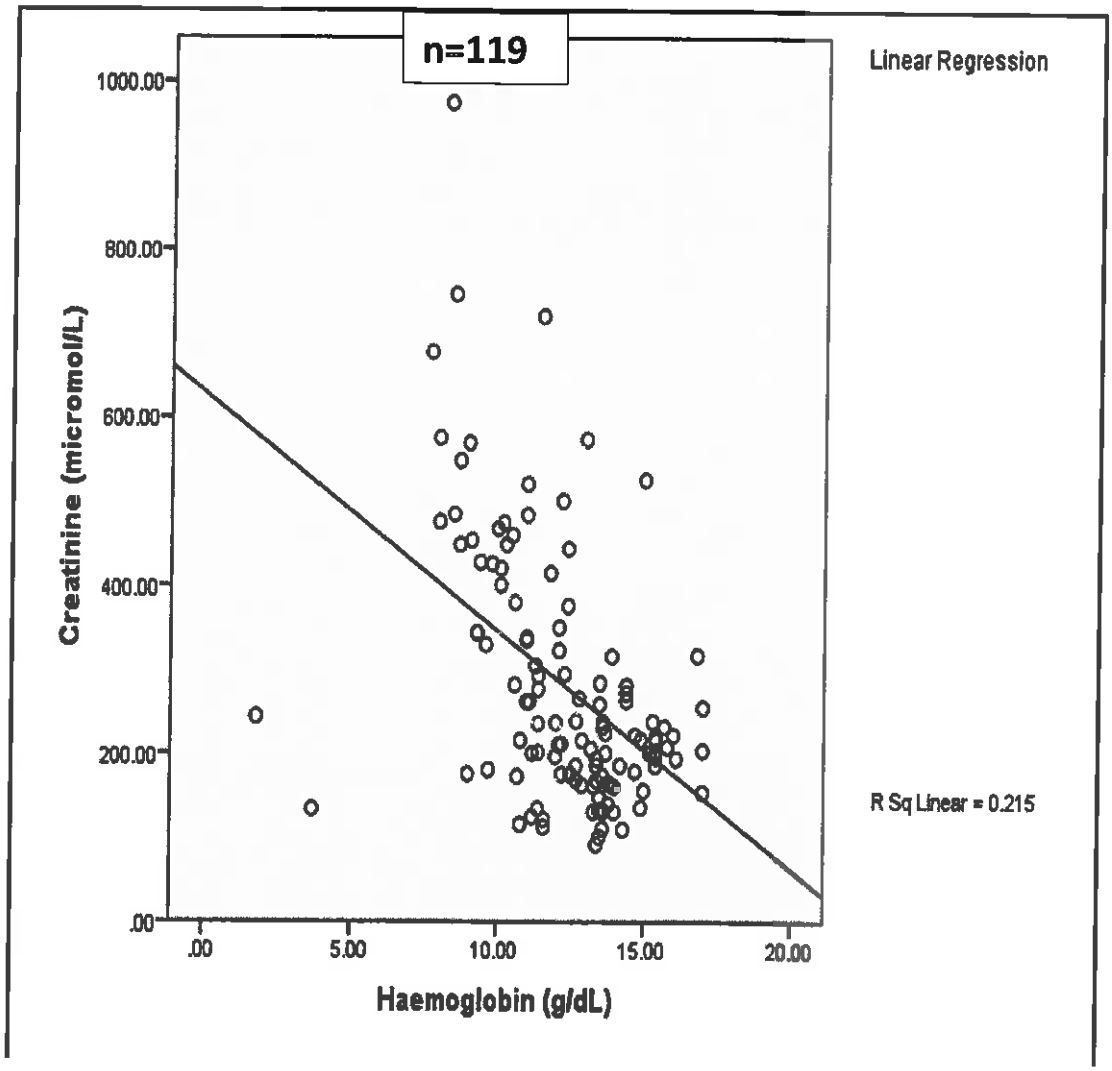

Figure 13: Correlation of serum creatinine to haemoglobin

(r=-0.464; p<0.001; n=119)

There was a statistically significant correlation (p<0.001) between creatinine and haemoglobin (Pearson's correlation coefficient).

3.6.3 CORRELATION OF SERUM CREATININE TO SERUM BICARBONATE LEVEL

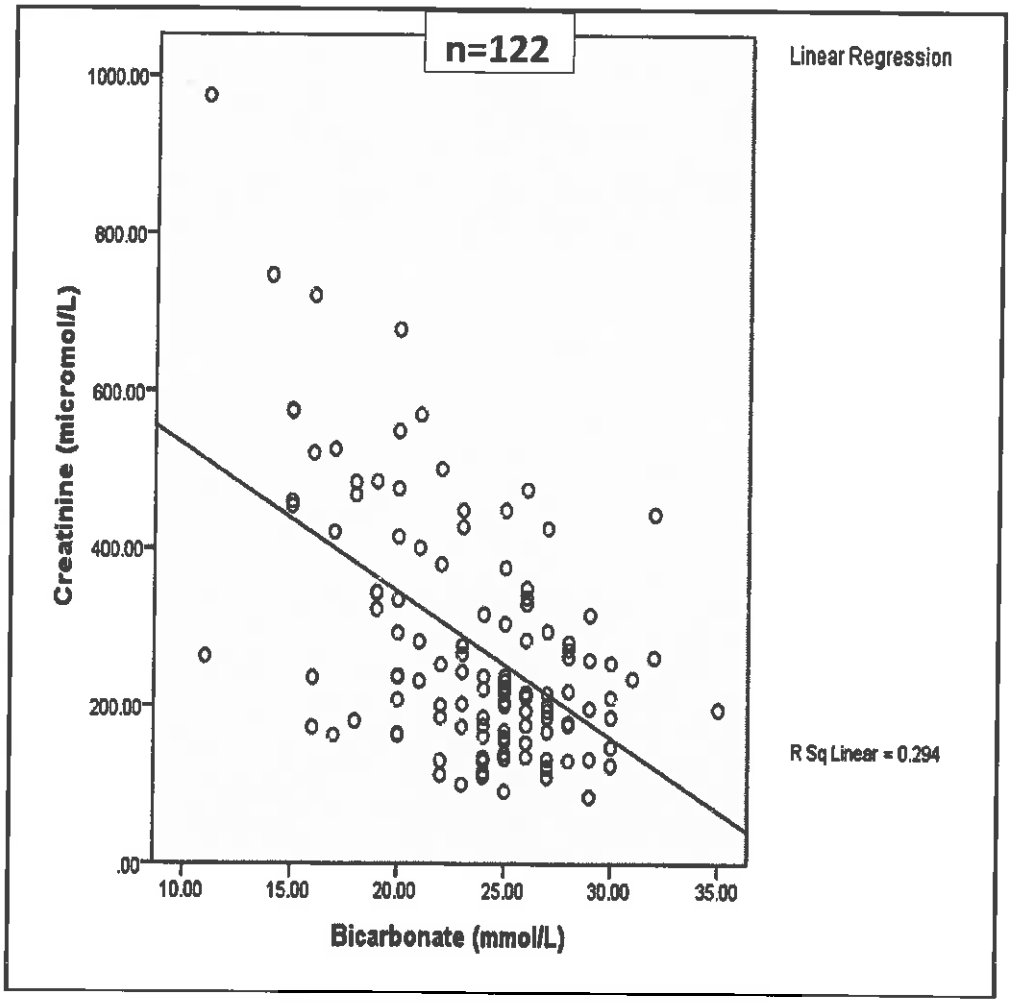

Figure 14: Correlation of serum creatinine to serum bicarbonate levels

(r=-0543; p<0.001; n=122)

There is a statistically significant correlation (p<0.001) between creatinine and bicarbonate levels (Pearson's correlation coefficient).

3.7 PROGRESSION OF CKD

Progression of CKD was defined as doubling of serum creatinine. Only 8.25% (n=10) of patients had doubling of serum creatinine. The aetiology of CKD in the patients that progressed was hypertension in 30% (n=3), diabetes mellitus in 50% (n=5) and the cause was unknown in 20% (n=2).

Table 9: Characteristics of patients that progressed (doubling of serum creatinine)

Variables	Median (IQR) Baseline	Median (IQR) End of study	p-value[a]
MAP (mmHg)	132 (110-130)	121 (115-132)	0.44
Proteinuria (g/24hr)	1.5 (0.6-1.5)	1.6 (0.3-3.2)	0.67
Haemoglobin (g/dL)	12.3 (10-13.7)	9.7 (8.6-11)	**0.005**
Bicarbonate (mmol/L)	22 (19-25)	19 (16-22)	**0.013**

a= Wilcoxon test for non-parametric, related samples

Table 10: Multiple regression analysis showing associations with CKD progression

Variable	B coefficient	Confidence interval	p- value
Haemoglobin	0.030	0.002 to 0.058	**0.036**
Bicarbonate	0.015	0.001 to 0.028	**0.034**
Albumin	0.001	-0.012 to 0.0014	0.904
Systolic BP	-0.004	-0.008 to 0	**0.036**
Diastolic BP	0.006	0 to 0.13	0.077
Proteinuria	0.165	-0.231 to 0.562	0.410

On multiple regression analysis, systolic blood pressure, acidosis and anaemia remained significantly correlated to doubling of serum creatinine levels.

3.8 BLOOD PRESSURE

The mean systolic BP at the beginning of the study was 143.9±25.6 mmHg. There was statistically significant improvement in the systolic BP at the end of the study, with a BP of 136.3±17.1 mmHg ($p<0.001$).

The mean diastolic BP at the beginning of the study was 83.3±13.9 mmHg. There was statistically significant improvement in the diastolic BP at the end of the study, with a BP of 75.9 mmHg ($p<0.001$).

The mean arterial pressure (MAP) improved from 131.1±21.1 mmHg at the beginning of the study to a MAP of 121.4±14.5 mmHg at the end of the study ($p<0.001$).

3.8.1 BLOOD PRESSURE CONTROL

Target blood pressure was defined as a mean arterial pressure of between 70-110mmHg.

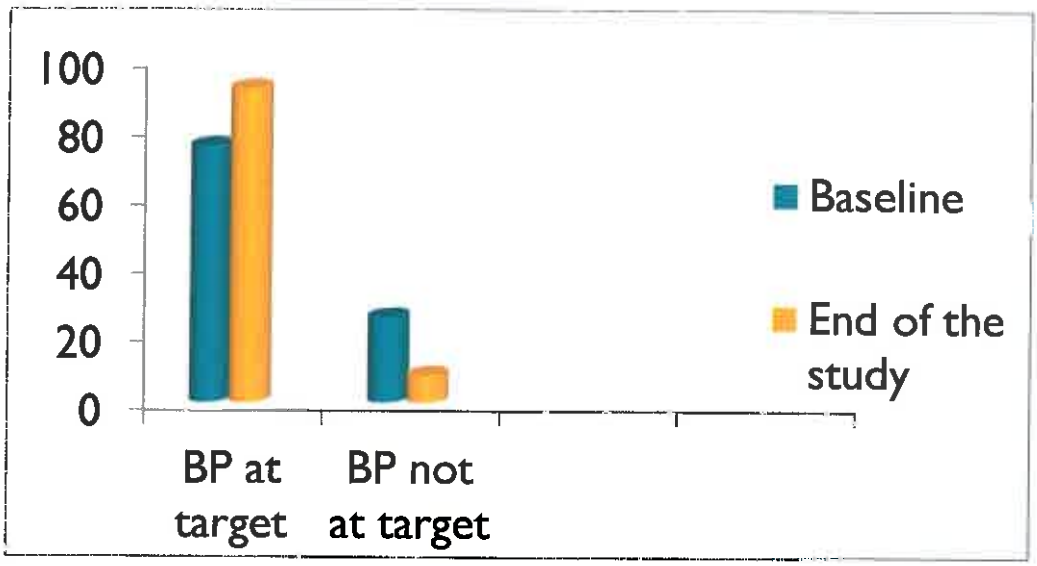

Figure 15: Blood pressure control

At the beginning of the study period, 74.6% (n=92) of patients had blood pressure controlled to target levels.

At the end of the study period, the BP control improved with 91.8% (n=112) of patient's blood pressure level being at target with only 8.2% (n=10) not at target levels.

3.8.1 ANTI-HYPERTENSIVE AGENTS USED

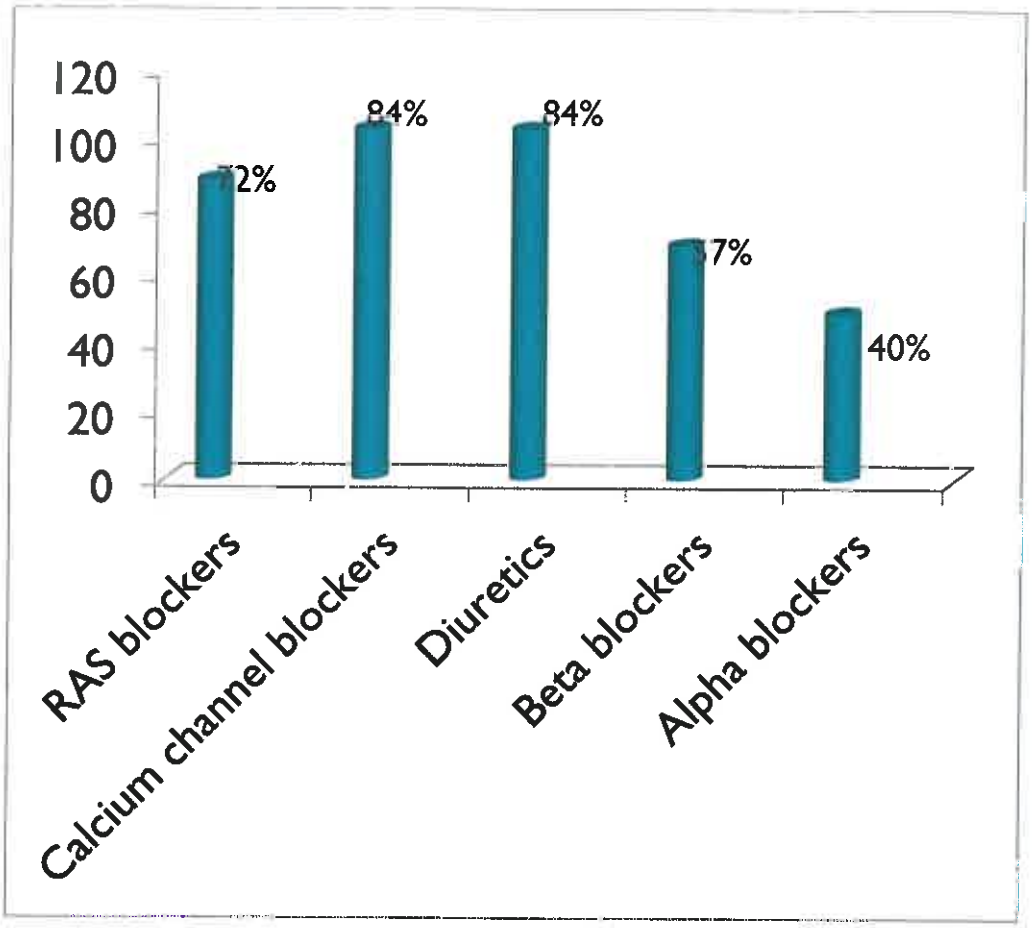

Figure 16: Pattern of anti-hypertensive agents used

The most frequently used anti-hypertensive agents were calcium channel blockers (CCB) and diuretics, at 84% each. The most frequently used diuretic was furosemide, with 92 patients (75.4%) being on this agent. Ten patients (8.2%) were on hydrochlorothiazide, with 20 patients (16.4%) not on any diuretic.

Agents that block the renin angiotensin system (RAS) were used in 72% of the patients.

3.8.3 NUMBER OF ANTI-HYPERTENSIVE AGENTS USED PER PATIENT

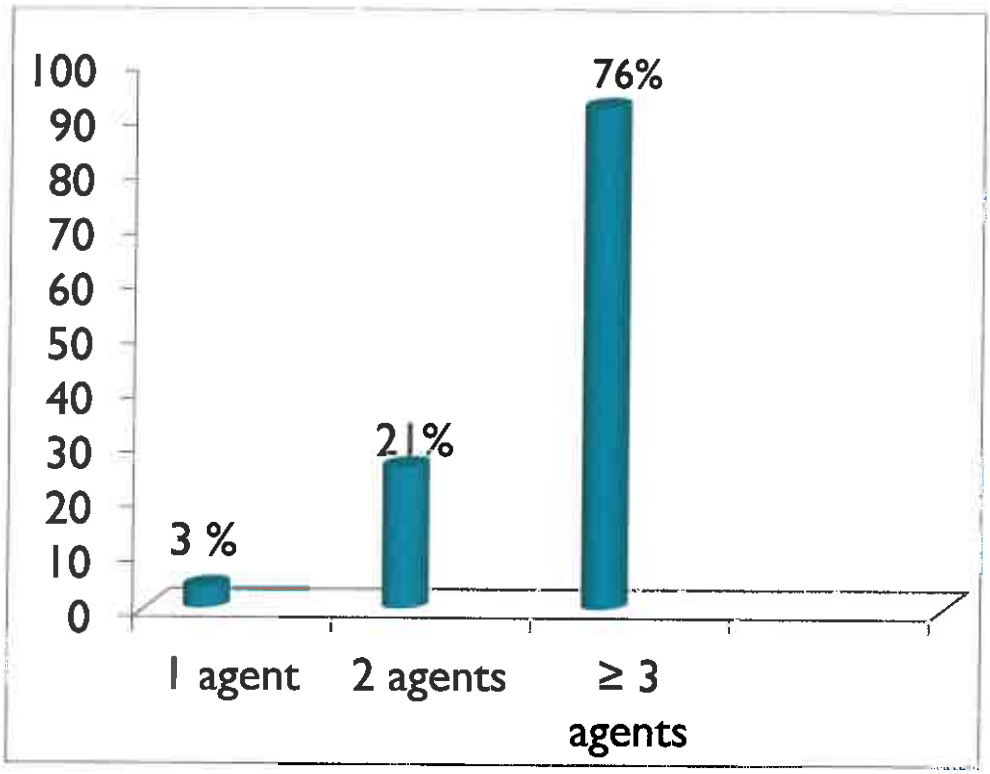

Figure 17: Number of anti-hypertensive agents per patient

Most patients (76%) were on three or more antihypertensive agents, inclusive of diuretics to achieve blood control. Only 3% of patients were taking only one antihypertensive agent.

3.8.2 CORRELATION OF BLOOD PRESSURE TO PROTEINURIA

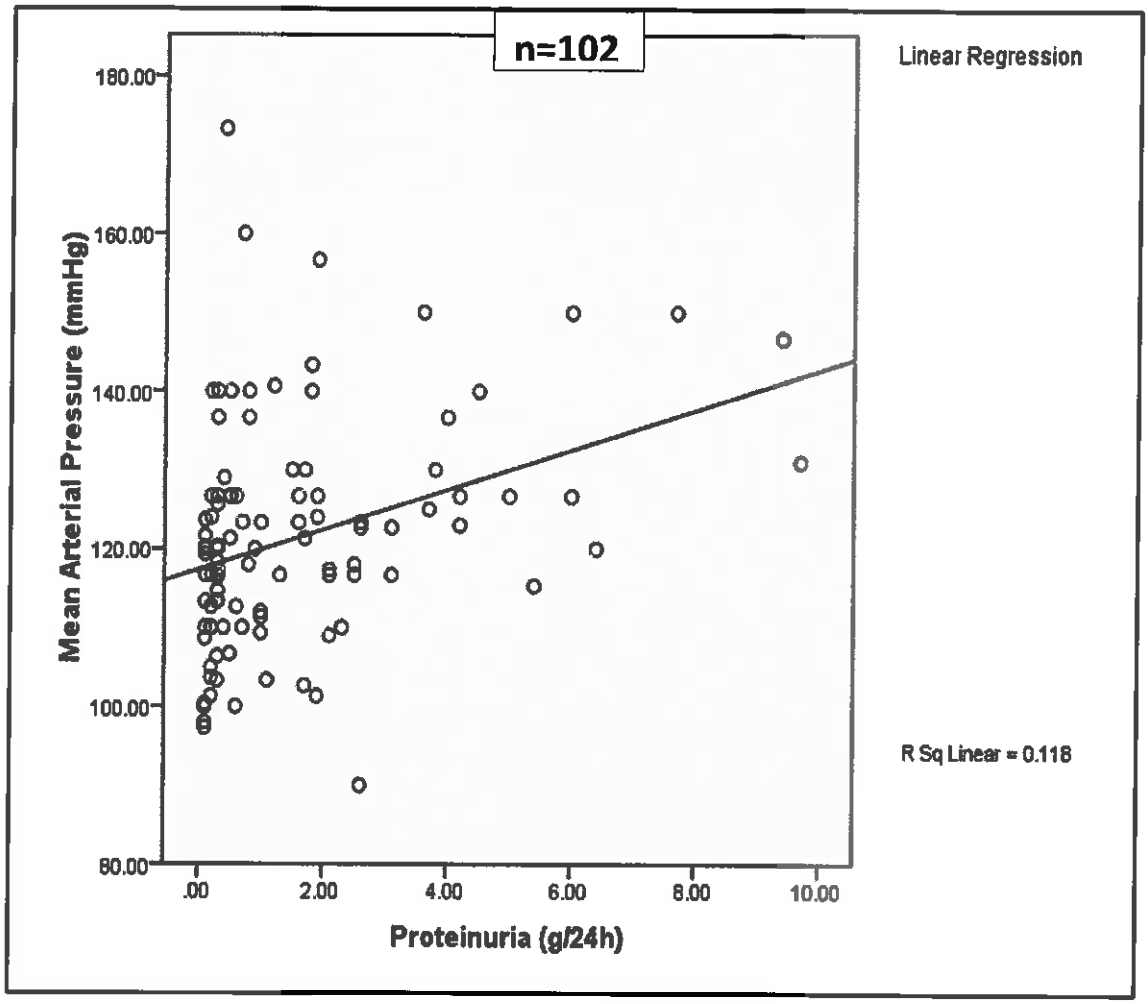

Figure 18: Correlation of Mean Arterial Pressure to Proteinuria

(r=0.255; p=0.01; n=102)

There was a statistically significant correlation between MAP and proteinuria with a p value of 0.01(Pearson's correlation coefficient)

3.9 Survival analysis

The Kaplan Meier renal survival curves for CKD patients demonstrate an overall 24- month survival rate of 82% (figure 17). There were no patients lost to follow-up.

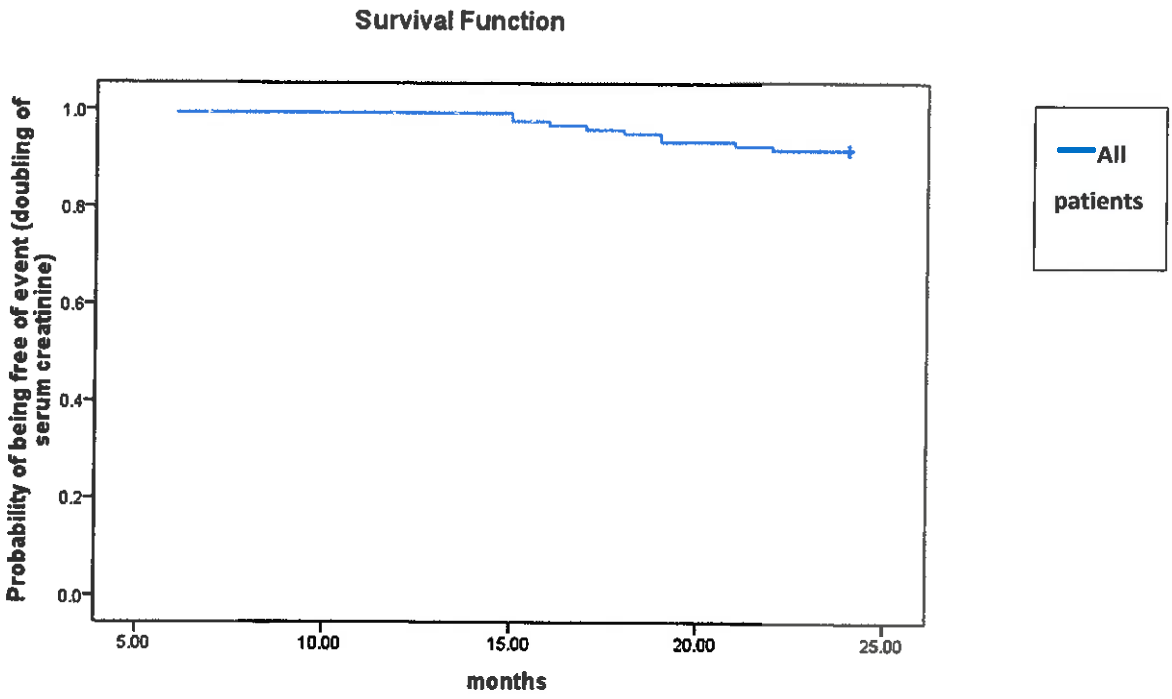

Figure 19: Renal survival of CKD patients

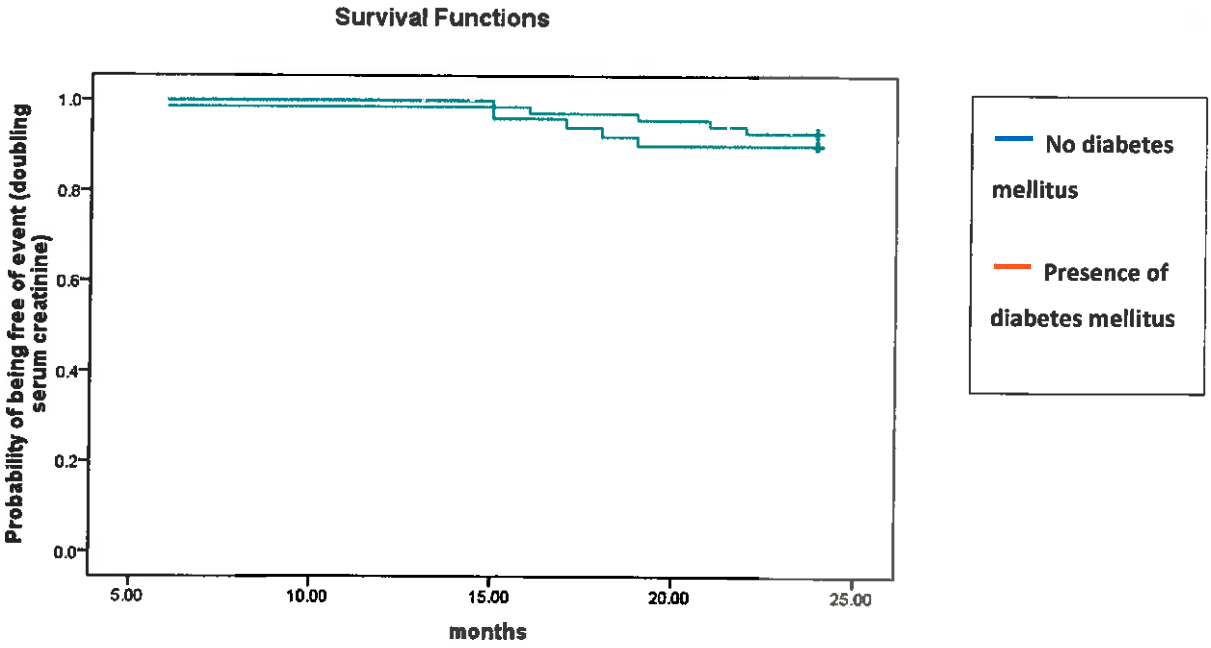

Figure 20: Survival curve for CKD patients based on status of diabetes mellitus

The estimated mean survival time for patients with diabetes mellitus was 23.3 months (22.6 to 23.9).

The estimated survival time for patients without diabetes mellitus was 23.5 months (23-24).

This difference was not statistically significant, with a p value of 0.321 and a log rank (Mantel Cox test) Chi square of 0.321.

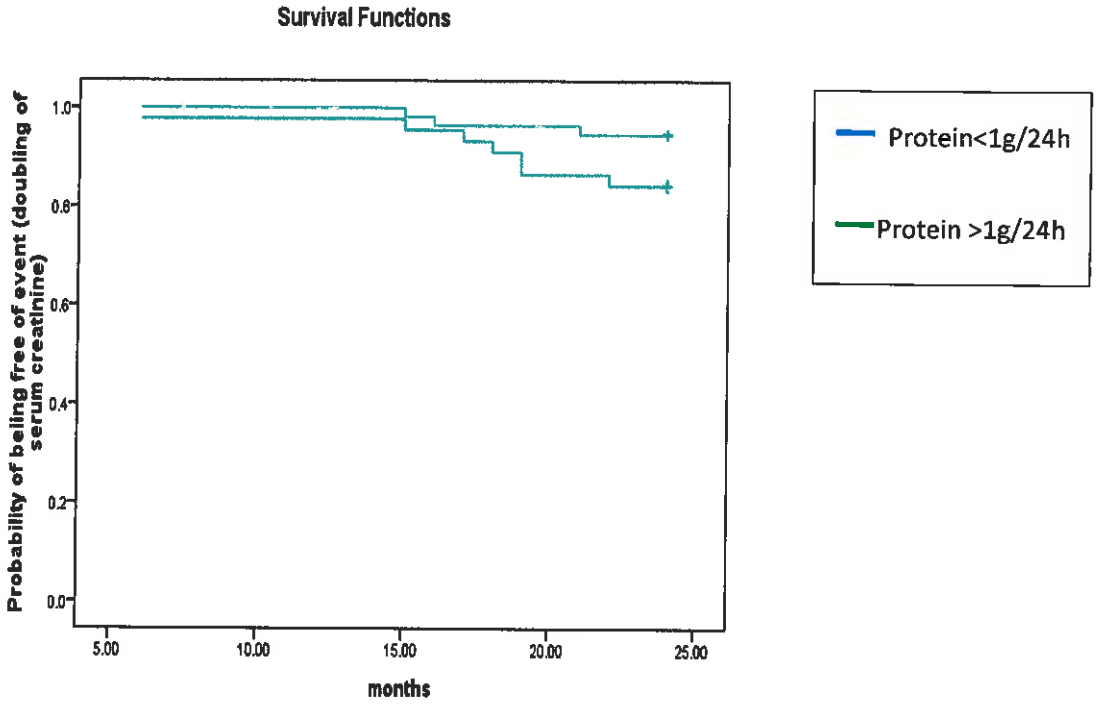

Figure 21: Survival curve for CKD patients based on level of proteinuria

The estimated mean survival for patients with proteinuria >1g was 22.8 months (confidence interval 23.2 to 24); whereas the estimated survival for patients with <1g/24 hours was 23.6 months (confidence interval 23.2 to 24). This difference was not statistically significant with a p value of 0.0085 and a log rank (Mantel Cox test) Chi square of 3.

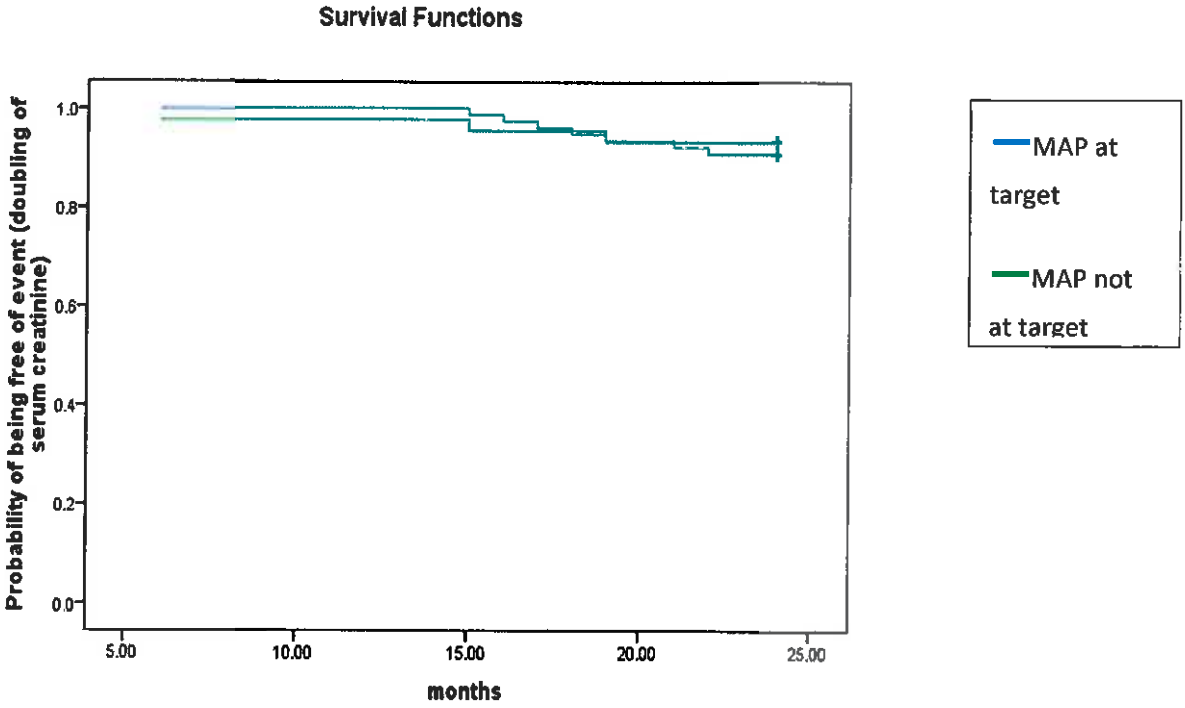

Figure 22: Survival curve for CKD patients based on blood pressure levels

There was no difference in the estimated mean renal survival rate between patients whose blood pressure levels were controlled to target compared to those whose blood pressure was not at target (23.4 months and 23.3 months respectively); p value 0.6 and log rank Chi square test (Mantel Cox) of 0.2.

CHAPTER 4.0 DISCUSSION

This is a review from Charlotte Maxeke Johannesburg Academic Hospital, South Africa. It is an analysis of 122 patients with chronic kidney disease attending the renal clinic. The analysis was done over a two year period.

4.1 DEMOGRAPHIC FACTORS

The mean age of the patients was 54.1±1.3 years. This is similar to mean age for CKD patients that were enrolled onto the REIN study in Europe (49.3±13.6 years). The mean age of patients initiated on chronic haemodialysis tends to be older; in Europe (60.2±15.2 years) and in the USA (60.5±15.5 years) (Goodkin *et al.*, 2004).

There was an equal sex distribution. Statistics from the 2011 Census Report from Statistics South Africa reveal that the population of South Africa is made up of females (48.7%) and males (51.3%), which is similar to our study population.

The majority of patients were Black African (55.7%). Again, the 2011 Census Report shows a similar pattern, with the majority of the population in South Africa being Black African (79.2%).

4.2 AETIOLOGY OF CKD

Diabetes mellitus was the commonest cause of CKD (41.8%), followed by hypertension (33.6%) in this study. It is noteworthy that patients with glomerulonephritides were excluded in this study. This is similar to the situation in the USA where diabetic nephropathy is the cause of ESRD in 40-42% of patients (Ruggenenti *et al.,* 2001. However, statistics from the South

African Dialysis and Transplant Registry (SADTR) of 1998 reveal that hypertension was the cause of end stage kidney disease in only 20% of patients.

The cause of CKD could not be ascertained in 6.6% of our patients. In the SADTR report, the cause of ESRD could not ascertained in 19.5% of patients. However it should be noted that this registry is for patients with ESRD and it does not accurately reflect the aetiology of chronic kidney disease in its entirety. In South Africa, there is rationing of renal replacement therapy (RRT) and many patients with diabetic end-stage renal disease are often not offered RRT due to associated co-morbidities such as ischaemic heart disease, cerebro-vascular diseases, peripheral vascular diseases etc. In the current study, patients with ESRD were excluded. There are currently no registries in South Africa that have looked at the aetiology of CKD in non-dialysis requiring patients.

4.3 PROTEINURIA

There was no difference in the amount of proteinuria from baseline compared to the end of the observation period; 1.5±1.98g/24h and 1.37±1.94g/24h respectively. The low level of proteinuria could have contributed to the low number of patients that had progression of kidney disease, as evidenced by a doubling of the serum creatinine. Indeed, in the REIN study, the higher the rate of baseline of proteinuria, the higher the risk of reaching the endpoint of doubling of serum creatinine. In the REIN study, patients with baseline proteinuria of less than 3g/24h had a slow rate of renal function deterioration, whereas those patients with baseline proteinuria of 3g/24h or more had a rapid decline in GFR (The GISEN group). The renoprotective effect of proteinuria reduction was also shown in the MDRD, where a reduction in proteinuria was associated with a

reduction in the rate of GFR decline (Peterson *et al.*, 1995). Patients with baseline proteinuria of > 0.25g/d had a faster mean rate of decline in GFR (5.7 to 6 mL.min.y). Assignment of these patients to a low blood pressure target slowed the mean decline in GFR. In patients with proteinuria of <0.25g/d, GFR declined slowly and assignment to a low blood pressure target did not affect the decline in GFR. Based on the above results, it was suggested that proteinuria levels should be considered when blood pressure targets are set for patients with CKD. When there is proteinuria of ≥1g/d, a MAP of 92mmHg (equivalent to a BP of 125/75 or less) was recommended. In patients with proteinuria of <1g/d, a MAP of 98mmHg (equivalent to a BP of 130/80 or less) was recommended.

4.3.1 NEPHROTIC RANGE PROTEINURIA

Nephrotic range proteinuria was present in 16.7% (n=17) patients. The cause of CKD in eleven of these patients was diabetic nephropathy. In the United Kingdom Prospective Diabetes Study (UKPDS), approximately one quarter of newly diagnosed patient with diabetes mellitus develop microalbuminuria or nepropathy by ten years (Adler *et al.*, 2003). At ten years, the prevalence of microalbuminuria was 24.9%, of macroalbuminuria was 5.3% and of elevated plasma creatinine or the institution of renal replacement therapy was 0.8% (Adler *et al.*, 2003).

None of the patients with proteinuria in the nephrotic ranges had tubulointerstitial disease as a cause of the CKD; which is in keeping with well-established fact that patients with tubulointerstitial diseases usually do not have significant proteinuria. Nephrotic range can occur is less that 1% of patients with acute tubulointestitial disease, usually secondary to the use on non-steroidal anti-inflammatory agents (NSAIDS).

It is unusual that 3 patients with a clinical diagnosis of hypertension had nephrotic- range proteinuria, as protein excretion is usually mildly elevated (less than 1g/day) in patients with hypertensive nephrosclerosis. Higher levels of proteinuria can occur; usually in patients with malignant hypertension, where the less severely affected glomeruli undergo compensatory hypertrophy with higher intraglomerular pressures and thus higher levels of proteinuria. In some instances, patients present quite late with an underlying chronic glomerular disease and thus a detailed workup is not possible. As hypertension and CKD frequently coexist, in these patients a clinical diagnosis of hypertension is sometimes made. None of the patients in this study had renal biopsies.

Fifteen of these patients with nephrotic range proteinuria were on RAS blockers, which is in keeping with current KDIGO Clinical Practice guidelines for the management of patients with chronic kidney disease. There were two patients with nephrotic range proteinuria who were not on RAS blockers. The reason why they were not on these agents could not be ascertained.

4.4 BLOOD PRESSURE

There was an improvement in the mean arterial pressure; from a MAP of 131±21mmHg at the beginning of the study to a MAP of 121±14 at the end of the study. There was also an increase in the number of patients who had blood pressure controlled to target levels; from 92 patients at the beginning of the study to 112 patients at the end of the study period. The association between CKD and hypertension is very strong; with each able to cause or aggravate the other. Hypertension is one of the major factors contributing of progression of CKD (Locatelli *et al*; 2005; Alvestrand *et al* 1988, Klag *et al* 1996).The good control of blood pressure in our study

group, together with the low levels of proteinuria are probably the main contributors to the low number of patients that progressed to doubling of serum creatinine. However, it is important to note that most patients required three or more antihypertensive agents to achieve good blood pressure control. This results in increased cost of treatment and a high pill-count, which can result in decreased compliance.

4.4.1 BLOOD PRESSURE CONTROL AND PROGRESSION OF RENAL FUNCTION

The MDRD study included patients with chronic kidney disease from a variety of causes. The authors were able to demonstrate that the patients who had a slower rate of progression were those with the lowest blood pressure (Peterson *et al.*, 1995). The low rate of progression in our study is probably related to the large number of patients whose blood pressure was controlled to target.

4.5 PROGRESSION OF KIDNEY DIESEASE

The primary outcome was doubling of serum creatinine. This occurred in 10 patients (8.2%). Further analysis of these ten patients revealed that systolic blood pressure, acidosis and anaemia remained significantly correlated to serum creatinine levels. The low number of patients that progressed (doubling of serum creatinine) could be attributed to the good control of blood pressure, low levels of proteinuria and the high proportion of patients that were on agents that block the renin-angiotensin system. In a study by Brenner *et al* on the effects of the ARB lorsartan on renal outcome in patients with type 2 diabetes mellitus, the authors were able to

demonstrate a 25% reduction in the risk of doubling of serum creatinine in patients on losartan compared to placebo (Brenner *et al.*, 2001).

However, in our study, the most commonly used blockers of the RAS are ACEI and not ARBs. The beneficial effects of ACEI on renal survival was demonstrated in the REIN study, where significantly less patients (n=18) on ramipril, compared to 40 patients in the placebo group reached the primary endpoint of doubling of serum creatinine (p=0.02).

4.6 RENAL SURVIVAL

The two year renal survival at the clinic is quite good; with an overall 24- month survival rate of 82%. It is intriguing to note that the renal survival was similar regardless of level of proteinuria or the use of agents that block the renin-angiotensin system. This is probably because a very small numbers of patients reached the primary endpoint of doubling of serum creatinine (n=10). This is in contrast to the results from the REIN study where the authors demonstrated better survival for patients on the RAS blocker ramipril than placebo (The GISEN group). In their study, a higher rate of baseline urinary protein excretion was associated with an increased risk of doubling of serum creatinine. This is not demonstrated in our study, probably due to the low baseline levels of proteinuria. The rate of renal function deterioration was shown to be slower in patients with baseline proteinuria of less than 3g/24h, compared to the faster rate of deterioration in patients with baseline proteinuria of more than 3 g/24h (The GISEN group). In our study, only 17 patients have levels of proteinuria that were >3 g/24h, with most patients (n=57) having levels of proteinuria < 1g/24h. Though we had a significant number of patients on agents that block the

RAS, the effect of these drugs was not pronounced due to the low levels of proteinuria in most of the patients.

4.7 RAS BLOCKERS

Approximately 72% of patients were using ACEI or ARBs in our study population. In a report by the US Renal Data System, only 60% of patients were on ACEI or ARB two months before end stage renal disease. This low reported use of ACEI/ ARBs is probably a reflection of the concern that most practitioners have of the use of these agents in patients with advanced kidney disease due to concerns about increases in serum creatinine and potassium levels. It is important to note that in our study, the potassium levels were not statistically different at the end of the study period compared to baseline. In an analysis of trials on diabetic and non-diabetic patients with chronic nephropathy, hyperkalaemia was found to be the reason for drop outs in less than 2% of patients (Ruggenenti *et al.*, 2001).

4.8 HAEMOGLOBIN LEVELS

The median haemoglobin level at baseline was 13 (11-14.6) g/dL and at the end of the study period, the haemoglobin level was 12.5 (11-13.9) g/dL. Erythropoietin Stimulating Agents (ESA) were not used in any of these patients. In our setting, due to resource constraints, these agents are reserved for use in patients on renal replacement therapy.

4.9 METABOLIC ACIDOSIS

The mean bicarbonate level at baseline was similar to that at the end of the study period, at 23.6±3.9. The NKF/KDOQI guidelines recommend maintenance of bicarbonate levels at 22mEq/L. Unfortunately, it was not possible to retrieve meaningful information on the use of oral bicarbonate supplementation in our patients. Bicarbonate supplementation is affordable and can be easily implemented as an additional treatment strategy which could retard progression of kidney disease.

4.10 SMOKING STATUS

Smoking is associated with progression of CKD. Unfortunately, the smoking status of the patients could not be adequately assessed from the clinical records. With an estimated prevalence of 14% of South Africans (21.2% males and 7% females) being current daily tobacco smokers, the impact of smoking on the progression of CKD needs to be assessed (WHO report, 2011).

4.11 LIMITATIONS

As this was a retrospective review, this study is prone to a number of errors. The most obvious is that there is likely to be a problem with the data recorded. There was some information that was missing from the files reviewed. There were inconsistencies in the manner in which different clinicians entered some of the data.

The length of follow up was 24 months. This is a relatively short follow-up period as progression of chronic kidney disease is slowly progressive.

The cause of chronic kidney disease was a clinical diagnosis based on the diagnosis of the attending clinician. No renal biopsies were done on this group of patients.

The treatment that was used for the analysis is the medication that was recorded in the renal clinic files. This is particularly important as some of the patients receive treatment from other departments. Unfortunately, in our hospital, there is no central database that records all the patients' medications.

During the follow up period, some of the patients were admitted and treated as inpatients. As far as possible, I have tried to eliminate the results that were taken during hospital admissions. The impact of these admissions on the progression of renal failure could not be ascertained. Moreover, the treatment given during these periods of inpatient care was not available.

4.12 CONCLUSION

Some important conclusions can be drawn for this study despite the above limitations:

1. Diabetes mellitus and hypertension are the leading causes of chronic kidney disease. However, all patients with glomerulonephritides were excluded from the study.
2. Target levels of blood pressure control can be achieved, though with the use of three or more agents.
3. Renal function progressed in a few patients (n=10). On multiple regression analysis, systolic BP, acidosis and anaemia are independent risk factors for progression of CKD.
4. Renal survival over a 2-year period in our patients is good (82% in 24 months).

Further research in this area that is needed includes:

1. Population-based studies to assess the incidence of chronic kidney disease in South Africa. This will assist in the assessment of the burden that CKD places in the society at large.
2. Prospective studies to assess the various factors that contribute to the progression in chronic kidney disease.
3. Prospective studies to assess the impact of various treatment modalities that can be used to prevent the progression of kidney disease

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APPENDICES

APPENDIX 1. ETHICS APPROVAL CERTIFICATE

UNIVERSITY OF THE WITWATERSRAND, JOHANNESBURG
Division of the Deputy Registrar (Research)

HUMAN RESEARCH ETHICS COMMITTEE (MEDICAL)
R14/49 Dr Nolubabalo U Nqebelele

CLEARANCE CERTIFICATE	**M111168**
PROJECT	**Prevention of Progression and Remission Strategies for Chronic Renal Failure: A Single Centre South African Perspective**
INVESTIGATORS	**Dr Nolubabalo U Nqebelele.**
DEPARTMENT	**Department of Medicine/Nephrology**
DATE CONSIDERED	**25/11/2011**
M1111680DECISION OF THE COMMITTEE*	**Approved unconditionally**

Unless otherwise specified this ethical clearance is valid for 5 years and may be renewed upon application.

DATE 25/11/2011 **CHAIRPERSON** .....
(Professor PE Cleaton-Jones)

*Guidelines for written 'informed consent' attached where applicable
cc: Supervisor: Prof S Naicker

DECLARATION OF INVESTIGATOR(S)

To be completed in duplicate and ONE COPY returned to the Secretary at Room 10004, 10th Floor, Senate House, University.
I/We fully understand the conditions under which I am/we are authorized to carry out the abovementioned research and I/we guarantee to ensure compliance with these conditions. Should any departure to be contemplated from the research procedure as approved I/we undertake to resubmit the protocol to the Committee. I agree to a completion of a yearly progress report.

PLEASE QUOTE THE PROTOCOL NUMBER IN ALL ENQUIRIES...