Estimating Glomerular Filtration Rate and the Effects of Acute Kidney Injury on Progression of Chronic Kidney Disease

MD Thesis

Dr. Hannah Speranza Kilbride

Department of Renal Medicine
East Kent Hospital University NHS Trust
Canterbury
Kent, UK

University of Kent

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Abstract

Chronic kidney disease (CKD) is a common health problem with a high prevalence in the elderly and is associated with high mortality rates and co-morbidity. CKD guidelines recommend that diagnosis and staging of CKD be based on estimated glomerular filtration rate (eGFR). Estimating GFR requires estimating equations using the variables gender, race and age and body surface area based on serum creatinine levels. The commonly recommended and used equations are the Modification of Diet in Renal Disease (MDRD) study and the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equations but these have not yet been validated in elderly people, who are at significant risk of developing CKD. The numbers of patients with progressive CKD is reportedly low with only a small proportion of patients reaching end-stage renal disease (ESRD). This study set out to find out why there is such a disproportion in the high prevalence of CKD and the low incidence of ESRD patients. Many patients die before they reach ESRD but prevalence studies have shown that mortality rates alone do not account for these numbers. I hypothesised that the methods used to estimate GFR underestimate renal function in elderly people causing an overestimate in CKD prevalence. This study firstly set out to assess the accuracy of the MDRD and CKD-EPI equations in an elderly Caucasian population against measured GFR across a wide range of renal function. The study demonstrated both equations perform fairly accurately in the elderly population with a tendency to slightly over-estimate GFR. This study has validated the use of these estimating equations in an elderly Caucasian population disproving my first hypothesis.

If the CKD prevalence data is a fair estimate and only a small proportion progress then the answer may lie in how CKD progresses. There are several known factors that influence CKD progression including GFR and albuminuria category, cause of renal disease and hypertension. Some of these risk factors are modifiable and need to be identified and managed in order to impact on long term outcomes including death, cardiovascular events and disease progression. Acute kidney injury (AKI) is

also rising in incidence and is complicated by high mortality rates, increased risk of cardiovascular events and more recently CKD progression. Little is known about the impact of more minor episodes occurring in the community on renal outcome.
The second part of this study examined the relationship of multiple episodes of community AKI with CKD progression in a population of patients with CKD stage 3-5 referred to renal services. In this observational study, patterns of CKD progression were assessed and multiple AKI events were recorded. This study demonstrated a clear relation between multiple AKI events and CKD progression however only low eGFR at referral, diabetes and albuminuria were independent risk factors associated with disease progression. During the study it emerged that there were two patterns of CKD progression. In comparison to the more commonly assumed linear decline, the more common pattern was a stepwise progressive pattern characterised by accelerated rates of decline followed by a period of stability. Multiple AKI events were significantly more common in the stepwise progressive group suggesting AKI may have an important role as a promoter of CKD progression. This study suggests that community AKI is a modifiable risk factor that needs identifying at early stages in order to minimise risk of poor outcomes including CKD progression.

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Abbreviations

AASK	African-American Study of Kidney Diseases and Hypertension
ACEI	Angiotensin Converting Enzyme Inhibitor
ACKD	Acute-on-Chronic Kidney Disorders
ACR	Albumin to Creatinine Ratio
AKD	Acute Kidney Disease
AKI	Acute Kidney Injury
AKIN	Acute Kidney Injury Network
AusDiab	Australia Diabetes Study
ARB	Angiotensin Receptor Blocker
ARIC	Atherosclerosis Risk In Communities
BMI	Body Mass Index
BP	Blood Pressure
BSA	Body Surface Area
CHS	Cardiovascular Health Study
CI	Confidence Interval
CKD	Chronic Kidney Disease
CKD-EPI	Chronic Kidney Disease Epidemiology Collaboration
CKiD	Chronic Kidney disease in Children
CV	Cardiovascular
eGFR	Estimated Glomerular Filtration Rate
EKHUFT	East Kent Hospitals University NHS Foundation Trust
ESRD	End Stage Renal Disease
FU	Follow Up
GFR	Glomerular Filtration Rate
GISEN	Gruppo Italiano di Studi Epidemiologica in Nephrologica
HOPE	Heart Outcomes and Prevention Evaluation
Hr	Hour
ID-MS	Isotope Dilution Mass Spectrometry
IL	Interleukin
IQR	Interquartile Range
KDIGO	Kidney Disease: Improving Global Outcomes
KDOQI	Kidney Disease Outcomes and Quality Initiative
KIM-1	Kidney-Injury Molecule-1
MAP	Mean Arterial Pressure
MDRD	Modification of Diet in Renal Disease
Min	Minute
N	Number
NCEPOD	National Confidential Enquiry into Patient Outcomes and Death
NEOERICA	NEw Opportunities for Early Renal Intervention by Computerised Assessment
NGAL	Neutrophil Gelatinase-Associated Lipocalin
NHANES	National Health and Nutrition Examination Surveys
NHS	National Health Service
NICE	National Institute for Health and Clinical Excellence
NIST	National Institute for Standardisation and Technologies

NKD	No known Kidney Disease
NKF	National Kidney Foundation
NSAID	Non-Steroidal Anti-Inflammatory Drug
NSF	National Service Framework
QOF	Quality and Outcomes Framework
P_{30}	Percentage of estimates with 30% of the reference test
PCR	Protein Creatinine Ratio
Pmp	Per million population
PREVEND	Prevention of REnal and Vascular ENd-stage Disease
RAS	Renin-Angiotensin System
REIN	Ramipril Efficacy In Nephropathy
RENAAL	Reduction of End-points in Non-insulin dependent diabetes mellitus with the Angiotensin II Antagonist Losartan
RIFLE	Risk, Injury, Failure, Loss and End-stage renal disease
Rpm	Revolutions per minute
RRT	Renal Replacement Therapy
sCr	Serum creatinine
SD	Standard Deviation
SDMA	Symmetric Di-Methyl Arginine
SHARP	Study of Heart And Renal Protection
SRM	Standardised Reference Material
UK	United Kingdom
UKPDS	United Kingdom Prospective Diabetes Study
US	United States
Yr	Year

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I acknowledge the support of the National Institute for Health Research (NIHR), through the Comprehensive Clinical Research Network (Research for Patient Benefit Programme, grant reference PB-PG-0107-12073) for the iohexol project.

Declaration

Results from the iohexol study have contributed to two publications in a peer reviewed journal of which I was first author of one. Results however were combined with work carried out by Dr Jo Carter also from Clinical Biochemistry department, EKHUFT who performed cystatin C measurements on all samples taken for the iohexol study and the estimating equations' performances were compared based on serum creatinine alone, serum cystatin C alone and creatinine and cystatin C combined. Dr Carter and I collaborated together with Dr Edmund Lamb and Dr Paul Stevens to publish these multi co-author papers (Appendix 1). Subsequent to the publications of the first paper, the BIS estimating equations were developed and published, claiming superior performance in the elderly population. The results of the iohexol study were then used to assess the performance of the BIS estimating equations and this work was carried out by Dr Inji Alshaer, EKHUFT who was first author of the second publication (Appendix 2).

All laboratory measurement of iohexol and serum creatinine using mass spectrometry were performed by Professor Neil Dalton and his laboratory staff at the Welchild Clinical Laboratory, Evelina Children's Hospitals, London. The analytical variations between iohexol and creatinine assays were also assessed by his staff. All urinary albumin, protein and creatinine measurements for the iohexol study were carried out by the Clinical Biochemistry staff, EKHUFT.

Publications and Presentations Arising From This Work

Publications

Accuracy of the MDRD and CKD-EPI equations for Estimation of GFR in the Elderly.

Kilbride H, Stevens P, Eaglestone G, Knight S, Carter J, Delaney M, Farmer C, O'Riordan, S, Dalton N, Lamb E

American Journal of Kidney Disease (2013) 61; 57-66

External Validation of the Berlin Equation for Estimation of GFR in the Elderly.

Alshaer I, **Kilbride H**, Stevens P, Eaglestone G, Knight S, Carter J, Delaney M, Farmer C, Irving J, O'Riordan S, Dalton N, Lamb E

American Journal of Kidney Disease (2014) 64 (5); 862-865

International Presentations

American Society of Nephrology (ASN), Denver 2010

Applicability of the MDRD and CKD-EPI Equations for Estimation of GFR in Older People.

Kilbride H, Stevens P, Eaglestone G, Knight S, Carter J, Delaney M, Farmer C, O'Riordan, S, Dalton N, Lamb E

American Society of Nephrology (ASN), San Diego 2009

Unrecognised Acute Kidney Injury and Chronic Kidney Disease: Chicken or Egg?

Kilbride H, Stevens PE, Hobbs H, John RI, Farmer C

World Congress of Nephrology (WCN), Rio de Janiero 2007

Discrete, 'Silent' Episodes of Acute Kidney Injury Contribute to Chronic Kidney Disease.

Kilbride H, Hobbs H, Klebe B, John R, Farmer C, Stevens P

National Presentations

South, West and East Kidney Society (SWEKS), London 2011

Applicability of the MDRD and CKD-EPI Equations for Estimation of GFR in Older People.

Kilbride H, Stevens P, Eaglestone G, Knight S, Carter J, Delaney M, Farmer C, O'Riordan, S, Dalton N, Lamb E

British Renal Society (BRS) and Renal Association (RA), Manchester 2010

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Chapter 1

Introduction

Chronic Kidney Disease and Acute Kidney Injury

1.1 Chronic Kidney Disease - Background

Chronic kidney disease (CKD) is a common condition that affects a diverse population and is a growing worldwide public health problem. It is estimated to affect approximately 8-11% of the population with a higher prevalence in the elderly (1, 2). CKD is often irreversible and a long-term condition that is detected by abnormalities in kidney function either in the urine or blood, or structural abnormalities identified by radiological methods. The term CKD covers a heterogeneous group of conditions that varies in severity, clinical presentation and progression. It can be caused by primary intrinsic renal conditions, predominantly glomerular and tubulo-interstitial disease, obstruction and cystic kidney disease, however, the most prevalent cause in the developed and developing world is diabetes (1). The prevalence of CKD is rising partly due to the better detection and increasing awareness of the condition but mostly due to the increasing prevalence of risk factors for developing CKD such as hypertension, diabetes, cardiovascular disease and obesity in an increasingly aged population. In the UK, the NEw Opportunities for Early Renal Intervention by Computerised Assessment (NEOERICA) project demonstrated the age-standardised prevalence of CKD was 10.6% for females and 5.8% for males however > 75% of patients with more advanced stages of CKD were ≥ 70 years (2).

CKD is often asymptomatic until its later stages and symptoms are often due to complications of reduced renal function which when severe may require renal replacement therapy (RRT). As a result, people are often referred late with advanced disease when treatments to reduce progression and potential for recovery are limited (3, 4). At earlier stages of CKD, patients are often asymptomatic and regular

monitoring of renal function is necessary to determine whether CKD is progressive or stable. The rate of progression of CKD can vary from months to decades. Earlier diagnosis allows time for appropriate referral to specialist centres, assessment and treatment of complications, reducing cardiovascular risk and mortality, and enables necessary preparation and education for patients requiring dialysis therapy or conservative management. Although CKD is irreversible in most cases, there is evidence that appropriate treatment at earlier stages can often prevent or delay disease progression. Several trials have demonstrated that therapeutic interventions focusing on targeted blood pressure control and proteinuria reduction not only reduce rate of CKD progression but also reduce cardiovascular events and mortality in people with CKD and these trials are discussed in more detail in Chapter 3 (5-7).

1.2 Definition and Staging of CKD

The best measure of renal function is direct measurement of glomerular filtration rate (GFR) using a filtration marker however these are impractical tests to perform in large populations on a frequent basis. In clinical practice, estimated GFR (eGFR) is obtained from serum creatinine measurements using estimating equations that include age, gender and ethnicity as variables to improve accuracy of the estimation. The Modification of Diet in Renal Disease (MDRD) Study equation is currently the most commonly used estimating equation and has been adopted by national and international guidelines for assessing renal function (8). There have been many other estimating equations developed which have claimed better performances in specific populations (9). Other markers of renal function may prove more accurate in estimating GFR from a single serum assay, such as cystatin C, but these not widely used. Methods for measuring renal function are discussed in more detail in Chapter 2.

The current accepted definition and classification of CKD is based on eGFR and the presence of proteinuria or albuminuria. A simplified CKD staging system was recommended by the National Kidney Foundation (NKF) - Kidney Disease Outcome

Quality Initiative (KDOQI) in 2002 to implement clear strategies and guidelines in the management of CKD (10). They defined CKD as the presence of kidney damage or GFR less than 60ml/min/1.73m^2 for three or more months irrespective of the cause. They also divided CKD into five stages according to severity of kidney damage as measured by GFR (Table 1.1). Stages 3-5 are defined by GFR alone and stages 1 and 2 require the presence of a structural abnormality detected by imaging or the presence of persistent proteinuria, haematuria or albuminuria. Persistent non-visible haematuria as detected on urine dipstick testing may indicate urinary tract malignancy and requires prompt investigation at appropriate age groups. In the absence of malignancy and albuminuria, non-visible haematuria may indicate early glomerular damage and these people need annual screening of renal function, blood pressure monitoring and albuminuria as some people may progress to more advanced stages of CKD and may require further diagnostic evaluation and treatment of complications. Stage 5 is described as established renal failure or End-Stage Renal Disease (ESRD) that has progressed for far that it often requires RRT to maintain life. Based on this classification system, the diagnosis of CKD can be made by a simple urine test to detect proteinuria/albuminuria or haematuria and a blood test to estimate GFR and these changes have facilitated a significant improvement in the detection and management of CKD in primary and secondary care.

More recently, amendments to the classification of CKD have been made in response to emerging evidence of the prognostic outcomes in CKD. In 2004, Go *et al* observed that there was an independent graded association between reduced eGFR and the risk of death and cardiovascular events and this risk rose sharply for subjects with eGFR < 45ml/min/1.73m^2 (11). This association led to the re-definition of stage 3 CKD into stage 3a and 3b as having an eGFR < 45ml/min/1.73m^2 is clearly associated with a higher risk of adverse cardiovascular outcomes and death (12).

Table 1.1 Stages of Chronic Kidney Disease

Stage	eGFR (ml/min/1.73m^2)	Description
1	≥ 90	Normal or increased GFR with other evidence of kidney damage: abnormal urine findings, structural abnormalities on imaging or histological abnormalities
2	60-89	Slight decrease in GFR with other evidence of kidney damage
3a 3b	45-59 30-44	Moderate decrease in GFR, with or without evidence of kidney damage
4	15-29	Severe decrease in GFR, with or without evidence of kidney damage
5	<15 or receiving RRT	Established or end-stage kidney disease

Abbreviations: GFR, glomerular filtration rate; eGFR, estimated glomerular filtration rate; RRT, renal replacement therapy; CKD, chronic kidney disease.

Table 1.1

This table shows the stages of CKD. The stages are differentiated according to level of eGFR. Stages 1 and 2 require other evidence of kidney damage whereas stages 3 to 5 are based on the eGFR category alone. Stage 3 is subdivided into 3a and 3b as there is clear evidence suggesting different prognostic outcomes between the two subgroups. This staging system was introduced by the Kidney Disease Outcome Quality Initiative in 2002 and later modified in 2012 to include the stages 3a and 3b (10, 12).

1.3 Risk Factors for CKD and Progression

CKD has become recognised as a significant public health problem and it is important to appreciate the characteristics of our population to understand why. CKD is increasingly prevalent in the elderly and we are facing an increasingly aged population with 16% of the UK population currently aged over 65 years (13). The population characteristics across most of the developed world suggest that the population will continue ageing for many years to come. It is projected that there will be a rise of 31% in the number of people reaching state pension age in the UK between 2012 and 2037 taking into account the future rises of the state pension age. Factors associated with CKD progression overlap to a large extent with factors associated with increased cardiovascular risk particularly between diabetes, hypertension, cardiovascular disease and CKD. Diabetic renal disease remains the single most common cause of ESRD accounting for 42% of those on dialysis in the US (14) and 25% in the UK (15). Over the past two decades there has been an increase in the incidence of ESRD mainly due to the rising prevalence of diabetes but this rise in incidence is also thought to be associated with the improved survival from cardiovascular events (16). Hypertension alone accounts for 7% of patients commencing RRT in the UK (15). Six per cent of the UK population have diabetes and 30% are hypertensive and are therefore at risk of developing CKD. (17, 18) The NEOERICA study found that across stages 3-5 CKD the odds ratio for hypertension was 2.1, diabetes 1.33 and cardiovascular disease 1.69 (2). Moreover people with both diabetes and hypertension have a 5-6 fold increased risk of developing ESRD. Obesity is also growing in incidence and is partly responsible for the rise in prevalence of diabetes and hypertension. Often these conditions co-exist and the development of CKD may be multi-factorial in nature and it therefore difficult to assign these cases of CKD to a single causative factor.

With an aging population and the growing prevalence of associated risk factors for CKD, in particular diabetes and hypertension, the incidence of CKD is set to rise and we have to prepare for the impact this will have on our health resources.

CKD in the Elderly

The prevalence of CKD stages 3 to 5 in people aged over 70 years is estimated to be as high as 25% in the UK and with our ageing population numbers are expected to rise significantly (2). In the USA, using data from the NHANES III, the highest prevalence of CKD (45%) was found among subjects aged 80 years and over (19). The overall population growth rate in the UK is 1.1% per year but it increases to 2.6% in those > 60 years and 3.9% in those > 80 years (20). There has been a great deal of controversy surrounding the decline of GFR with age which debates whether the decline is an inevitable consequence of senescence or whether it represents a disease process. Some researchers have shown that vascular changes in the renal vessels as in other systemic vessels occur with age and are often due to co-morbidity, but these changes have also been documented in the absence of co-morbid conditions (21). It is suggested that these changes eventually cause glomerulosclerosis, interstitial fibrosis and tubular atrophy in the cortex, with compensatory hypertrophy and hyperfiltration of the remaining functioning glomeruli, eventually manifesting as CKD. In the autopsy study by Neugarten *et al*, older age was associated with increased numbers of sclerotic glomeruli and interstitial fibrosis with a loss of about 20-30% of the glomeruli compared to younger patients (22). In other studies, ageing was associated with a loss of renal mass by 20-25% from the ages of 30 to 80 years and the length of the kidney was found to decrease by 15% from the age of 17 to 85 years of age (23-25). Significant loss of renal function however is not always an inevitable consequence of ageing.

In 1950, a cross-sectional study was published by Shock *et al*, which found that measured GFR using the gold standard inulin clearance, declined with advancing age in 'normal' individuals independent of hypertension or cardiovascular disease suggesting that there is a natural decline in GFR which occurs with age in males (26). Studies have shown that the co-morbid conditions in this population associated with an accelerated rate of GFR decline are hypertension, diabetes, atherosclerotic disease and cardiac insufficiency. Other cross-sectional studies have reported that the decline in GFR can start from ages 30-40 years (27). The major limitation of these

cross-sectional studies is that they introduce a bias as only the highly selected people who survive to reach old age are studied. This is known as selective mortality and as a result the cohort studied have little co-morbid burden. Longitudinal studies observing serial measurements of renal function overcome the bias of cross-sectional studies.

Perhaps the two most recognised longitudinal studies in ageing were performed by Rowe *et al* in 1976 (27) and later by Lindeman *et al* in 1985 (28) as part of the Baltimore Longitudinal Study of Ageing. The earlier study assessed creatinine clearance as a measure of renal function in 293 men aged 30 to 80 years with no history of diabetes. They found the average decline in creatinine clearance was 45 ml/min/1.73m^2 with a rate of decline in eGFR of 0.9 ml/min/1.73m^2/year. There are limitations however to longitudinal studies as the validity of the results can be compromised by a limited time of observation, too long or too short an interval between testing, and infrequent testing can cause inaccuracy in determining a statistical change in eGFR. Rowe's study required 3 or more serum creatinine clearance determinations over a mean interval of 6 years which is clearly inadequate to estimate rate of decline of eGFR. Creatinine clearance is also often criticised for its inaccuracies at estimating renal function and has been shown to overestimate GFR compared to measured GFR using inulin clearance by as much as 22% (29). The study concluded that in order to achieve a minimally acceptable accuracy of the slope of decline, a minimum of annual tests over 18 years would be required.

The Baltimore Longitudinal Study of Ageing measured serial creatinine clearance in 293 'normal' healthy males aged 22 to 97 years with no other co-morbidities and required a minimum of 5 creatinine clearance measurements over 8-14 years (28). Creatinine clearance was stable in healthy men < 35 years old and in men aged 35-60 years, approximately one third of healthy individuals showed no decline in GFR with ageing but the remaining two-thirds had a creatinine clearance decline of up to 8ml/min/1.73m^2/year and 21.6% had a statistically significant higher rate of decline. This decline in creatinine clearance declined more steeply after the age of 60 years.

Again this study was subject to the inaccuracies of creatinine clearance as a measure of renal function, limited to male subjects but also included diabetics in the 'normal' group confounding the results as glomerular hyperfiltration is known to occur in diabetic patients. These studies suggested that the age-related decline in GFR is likely to be both due to physiological and pathological consequences of ageing.

Guidelines now suggest that age-related decline in GFR is thought to decrease by no more than 1-2 ml/min/1.73 m^2/year after the age of 40 and a more accelerated decline is due to pathological damage due to progressive CKD (10). Using the KDOQI CKD staging system where CKD 3-5 is based on eGFR definition alone, many individuals over 65 years will be labelled as having CKD even though their eGFR is in the normal range for their age and gender grouping many at CKD stage 3a. A GFR of < 30 ml/min/1.73 m^2 in older people though does predict an increased mortality and a lower eGFR is more frequent in frailer subjects (30).

It has also been suggested that the increased prevalence of CKD in older people may be partly related to the equations used to estimate GFR from serum creatinine, especially in older females and there have been questions raised concerning the validity of the estimating equations used in GFR measurement in older people. Several publications have suggested that the current methods of estimating GFR may underestimate GFR in elderly patients leading to incorrect CKD staging and consequently inappropriate management of these patients (9, 31). With an ageing population prevalent with cardiovascular disease, diabetes and hypertension who are often exposed to complex poly-pharmacy, it is important to have accurate methods of measuring GFR so we can appropriately manage those with CKD stages 3-5.

Progression of CKD

There has been much debate about the definition of progressive CKD but it has been generally accepted as a decline in eGFR of > 5 ml/min/1.73m^2 in 1 year or > 10ml/min/1.73m^2 in 5 years and patients with progressive CKD would benefit from

referral for renal specialist care (12). There is a great variability in rate of CKD progression yet the majority of patients with CKD do not progress. Progression to ESRD in earlier stages of CKD is low and progression in those with stage 4-5 CKD is much higher bur despite this only a small proportion of patients with stages 4-5 CKD inexorably progress. Data from the third National Health and Nutrition Examination Survey (NHANES III) in 2004 estimated that 20 million adults in US have CKD yet only 2% of the CKD population had progressive CKD to ESRD requiring RRT (1). Keith *et al* performed a longitudinal study of 28,000 CKD patients and found the rate of RRT over a 5-year follow-up was 1.1%, 1.3% and 19.9% respectively for stages 2, 3 and 4 CKD however, in this study, progression was defined as developing ESRD (32).

Cause of CKD clearly has an influence on CKD progression to ESRD with certain conditions such as chronic glomerulonephritis, inherited renal disease and cystic kidney disease often displaying an inexorable decline in renal function to ESRD. Data from the prospective longitudinal trial, Chronic Kidney Disease in Children (CKiD), demonstrated a more rapid decline in renal function in children whose underlying cause of CKD was due to a glomerular cause with an annual rate of decline in measured GFR of 10.5% compared to those with a non-glomerular cause who had a rate of decline of only 3.9% (33). Population studies have identified other risk factors associated with the progression of CKD: atherosclerotic disease (34), diabetes (35), hypertension (36), proteinuria (37), chronic use of non-steroidal anti-inflammatory medications (NSAIDs) (38), and Black or Asian ethnicity (39) which will be discussed in Chapter 3. These risk factors are highly prevalent in our UK population. There are many patients with CKD stages 4 and 5 who do not progress but this raises the question of how they got there in the first place. Patients with stable CKD may have an isolated event causing an acute decline followed by a partial recovery with no further event and another period of stability.

Albuminuria and Proteinuria

Diagnosis and staging of CKD not only requires eGFR but also requires testing for proteinuria or albuminuria. Proteinuria is a general term for the presence of

increased amounts of protein in the urine that reflects abnormal loss of protein derived from the kidney or lower urinary tract. Proteinuria is a common finding in CKD and is an early marker of glomerular disease often presenting prior to a reduction in GFR. Albumin is a type of plasma protein found in the urine in small quantities in normal subjects and in larger quantities in patients with CKD. Albuminuria is a common finding in CKD but significant albuminuria is not always present in CKD. Proteinuria is an important factor linked with progression of CKD disease and poor outcomes (32).

More recently, CKD guidelines emphasize testing for albuminuria rather than proteinuria due to emerging evidence of its strong association with CKD progression, prediction of cardiovascular risk and increased sensitivity in detecting glomerular damage. I discuss these seminal studies in more detail in Chapter 3. Albuminuria is expressed as a urine albumin to creatinine ratio (ACR) and the persistent presence of an ACR ≥ 3mg/mmol for > 3 months indicates CKD.

The importance of albuminuria was highlighted in 2013 when KDIGO recommended that albuminuria strata, represented as ACR, be added within each GFR stage (ACR <30 mg/g, 30–299 mg/g, or ≥300 mg/g) in the CKD classification system (Table 1.2) (12). The recommendation for predicting outcome of CKD is to identify the following variables: Cause of CKD; GFR category; albuminuria category; other risk factors and co-morbid conditions. The risk associations of GFR and albuminuria categories appear to be largely independent of one another so neither the GFR or ACR category can fully capture the prognosis for a patient with CKD. These categories have been combined to identify those at greater risk of progression or poor outcomes in order to create a referral guide which is shown in Table 1.2.

Table 1.2 KDIGO Staging of CKD using GFR and Albuminuria Categories

GFR Category	Terms	GFR (ml/min/1.73m^2)
G1	Normal or high	≥ 90
G2	Mildly decreased	60-89
G3a	Mildly to moderately decreased	45-59
G3b	Moderately to severely decreased	30-44
G4	Severely decreased	15-29
G5	Kidney failure	<15

ACR Category	Terms	ACR (mg/mmol)
A1	Normal to mildly increased	< 3
A2	Moderately increased	3-30
A3	Severely increased	>30

Abbreviations: CKD, chronic kidney disease; GFR, glomerular filtration rate; ACR, albumin to creatinine ratio; Kidney Disease: Improving Global Outcomes, KDIGO.

Table 1.2

The most recent classification system of CKD adopted the subdivision of GFR categories and included 3 albuminuria categories for each GFR category. This was introduced to indicate risk of CKD progression and poor prognosis. The grades G1 – G5 indicate categorisation of CKD by GFR and are differentiated according to level of GFR. Categories A1-A3 differ according to urine ACR levels. This classification was introduced by the Kidney Disease: Improving Global Outcomes (KDIGO) CKD Work Group in 2013 (12).

1.4 CKD Outcomes

The major risk to the majority of people with CKD is not so much progression to ESRD but the significant association with increased mortality and morbidity predominantly from cardiovascular disease (40-41). A longitudinal follow-up study in 2004 showed that even among those with advanced stage 4 CKD, death prior to RRT is twice as likely as progression to ESRD (32). Death was far more common than dialysis at all stage with five-year mortality rates were 19.5%, 24.3% and 45.7% for CKD stages 2, 3 and 4 respectively. Even mild CKD is an independent risk factor for cardiovascular disease and death (42).

Hospitalisation among people with CKD is also high. Khan *et al* demonstrated that during a median follow-up of 11.4 months, 47% of subjects with CKD had at least one hospitalisation with an average of 0.96 hospital admissions, 6.6 in-patient days and 4 nephrology outpatient visits per person per year (43). Amongst the dialysis population this rose to 2.2 hospital admissions and a mean length of stay of 14.8 days.

CKD is also associated with numerous adverse health outcomes including anaemia renal bone disease, infections and impaired physical function. Even cognitive impairment has been associated with CKD not only in the elderly but in those aged 20 to 59 years with ESRD or with a GFR between 30-59 ml/min/1.73m^2 (44-45). These complications of CKD lead to an additional impact on the high morbidity, mortality and costs.

Only a minority of people with CKD will progress to develop more advanced disease and only 1-2% of patients will progress to develop end-stage renal disease (ESRD) which, without renal replacement therapy (RRT), in the form of dialysis or transplantation, leads to death (15). The financial burden of RRT on the health service is immense with over 2% of the total National Health Service (NHS) budget being spent on dialysis and transplantation alone. The total cost of CKD in England in 2009-2010 was £1.45 billion pounds (46).

CKD Management

Good management of patients with CKD requires strong links between primary and secondary care. Identification and management of CKD stages 1-3 usually takes place in primary care and often combines reduction of cardiovascular risk along with regular monitoring of renal function and management of the complications of CKD. Strategies to reduce blood pressure to recommended targets and reduce albuminuria are measures that have been consistently found to significantly reduce progression of CKD and its associated adverse outcomes, and this is discussed in Chapter 3. There is a growing body of evidence that treatment of other traditional cardiovascular risk factors in CKD patients are of benefit, which include smoking cessation and lipid lowering treatments. Halimi *et al* demonstrated that smoking is associated with CKD progression (47) however no studies have supported that smoking cessation delays progression of disease. The Study of Heart and Renal Protection (SHARP) was the largest randomised controlled trial of CKD patients and found lipid lowering resulted in a 17% reduction in atherosclerotic events (48). Avoidance of non-steroidal anti-inflammatories (NSAID) and other nephrotoxic agents have also been shown to reduce progression risk. Gooch *et al* determined that high cumulative NSAID exposure is associated with increased risk for rapid CKD progression in an elderly cohort, a population with a high prevalence of CKD and NSAID use (38).

Specialist management focuses on delaying progression of disease, treating complications of CKD, timely preparation for RRT or pre-emptive transplantation or if opting for conservative management, preparation for end of life care and symptom control. Late referrals to nephrology services often result in unplanned dialysis initiation and are associated with increased morbidity and mortality, increased hospitalisation rates and increased costs (49-50). There has been a variable definition of what constitutes late referral from < 1 month to < 1 year but is generally accepted as referrals made less than 3 months prior to dialysis initiation which is thought to be the minimum amount of time required for adequate assessment, education and preparation for RRT including creation of access for

dialysis. Late referrals are considered an adverse measure and although certain conditions can cause a rapid progressive decline in renal function where late commencement of renal replacement therapy is unavoidable, they account for only 25% of late referrals in the UK (15).

CKD Screening and Referral Guidelines

Studies have looked at whether the systematic monitoring of adults with CKD stages 1–3 for worsening kidney function or damage is actually of benefit in improving clinical outcomes. Undesirable consequences from CKD screening and monitoring may include misclassification of patients with CKD, unnecessary tests, the associated adverse psychological effects of being labelled with CKD, adverse effects associated with pharmacological treatments initiated or changed after a CKD diagnosis, and possible financial and insurance ramifications of a new CKD diagnosis.

In 2002, the European Best Group Practice published guidelines for CKD management and recommended patients with an eGFR < 30ml/min/1.73m^2 and declining eGFR be under nephrologist care (51). In 2002, NKF-KDOQI reiterated the recommendation that patients who reach CKD stage 4 (i.e. eGFR < 30 ml/min/m^2) receive timely education for RRT from renal specialists (10). In 2006, the Quality and Outcomes Framework (QOF) introduced renal incentives creating a register of people in primary care with an eGFR <60 ml/min/1.73m^2 (i.e. stages 3-5 CKD). In the same year, a national strategy was launched to report eGFRs automatically with every serum creatinine request from all clinical chemistry laboratories. In 2006, the Royal College of Physicians together with the Renal Association published and disseminated UK guidelines for identification and referral of CKD (52). In 2008, the National Institute for Health and Care Excellence (NICE) disseminated national guidelines for best practice in managing CKD in primary and secondary care (53). These initiatives were all developed to promote early identification of CKD and management with guidance for referral and have no doubt influenced referral patterns. The fundamental problem with all these guidelines that although they were designed to reduce late referral of people with advanced CKD, they do not take

into account the impact that following these guidelines would have in the short-term on referral rates to specialist centres (54), on the increased workload it would create in primary care or its cost implications. Guidelines have not been specific enough increasing unselected referrals and studies have shown that the number of patients referred to renal services significantly increased after April 2006 (55-56).

In July 2014, NICE updated their CKD guidelines and highlighted the importance of identifying patients with progressive CKD altering their referral criteria (57). The recommendations focussed on strategies aimed at earlier identification and prevention of progression to ESRD. They suggested re-defining accelerated progression of CKD as a sustained decreased in eGFR by 25% or more and a change in GFR category within 12 months, or a sustained decrease in GFR of 15ml/min/1.73m^2 per year. Calculating rate of CKD progression is recommended by obtaining a minimum of 3 GFR estimations within a minimum period of 90 days and planning intervention strategies in a timely fashion if they are predicted to require RRT in their lifetime.

These guidelines have improved the identification of patients with CKD and UK prevalence data has indicated there has been a recent steady national decline in late referrals with an overall rate of 20% with some centres achieving < 10% (15). We now need to focus on appropriately identifying and managing those patients in the earlier stages of CKD who are at risk of progression so they can benefit from earlier interventions to reduce progression and improve mortality and morbidity outcomes.

1.5 Acute Kidney Injury (AKI) - Background

AKI, previously termed acute renal failure, is a complex disorder with multiple aetiologies and outcomes. AKI is a common condition, which in contrast to CKD, is often reversible, often iatrogenic in aetiology and potentially preventable (58-59). AKI usually develops over hours to days and leads to an abrupt rise in serum urea and creatinine levels often causing electrolyte disturbances, metabolic acidosis and reduced urine output. The incidence of AKI is higher in hospitalised patients occurring up to 18% of all hospital admissions with reports of up to 65% in critically ill patients in the intensive care setting (60-61). AKI encompasses a wide spectrum of illnesses and varies in severity from minor increases in creatinine of only 26µmol/l (0.3mg/dl) through to critically unwell patients requiring RRT. The incidence of AKI in the UK ranges from 486-630 per million population (pmp) per year in published series (61-62). The incidence of AKI is increasing partly due to better recognition but also due to the rising prevalence of similar risk factors responsible for the rise in CKD such as an increasingly aged population and the increased incidence of diabetes, cardiovascular disease and other chronic diseases. AKI is potentially preventable for those at risk through appropriate medical and drug interventions and increased vigilance and testing.

Very few cases of AKI are actually due to intrinsic renal conditions and approximately 70% of cases of community-acquired AKI are due to pre-renal causes (63). More severe cases often require higher dependency care or renal specialist support in order to initiate appropriate treatment in a timely fashion in order to optimise renal recovery and outcomes. Several studies have demonstrated that non-specialist management of AKI is sub-optimal. Stevens *et al* demonstrated in a 12-month period, 18% of cases of AKI initially managed by non-specialists were preventable and approximately a third of cases were iatrogenic in aetiology (59).

1.6 Definition and Staging of AKI

The concept and understanding of AKI and advances in research, in comparison to CKD, is relatively new and this has been partially due to the lack of a universally accepted definition of AKI. Having a uniform standard for diagnosing and classifying AKI has enhanced our understanding and management of AKI. In 2002, the Acute Dialysis Quality Initiative (ADQI) was created with the primary goal of developing a consensus in the definition and evidence-based guidelines for the prevention and treatment of AKI. They introduced the RIFLE criteria, an acronym of Risk, Injury, Failure, Loss and End-stage renal failure, to define AKI (64). The RIFLE classification system was demonstrated to be suitable for assessing mortality and AKI particularly in intensive care units (65-67) but was criticised for including outcomes (Loss and End-stage renal failure) in the staging criteria (68). In 2004, the Acute Kidney Injury Network (AKIN) was formed who advised that the definition of AKI encompass the full spectrum of the disease, from mild to severe, and removed the outcomes loss of function and ESRD in the classification system (69). The AKIN staging system of severity of AKI is shown in Table 1.3 and is based on rises in serum creatinine and urine output criteria and differs from RIFLE as it is defined as changes within 48 hours while RIFLE defined AKI over 7 days. This new staging system reduced the need for a baseline creatinine but it does require at least two creatinine measurements within 48 hours. In 2012, KDIGO published clinical practice guidelines for AKI combining the RIFLE and AKIN definitions. KDIGO defined AKI as presence of any of the following; increased in serum creatinine by > 26µmol/l within 48 hours, or an increase in serum creatinine 1.5 times above baseline within 7 days and a urine output < 0.5ml/kg/hr over 6 hours (70). Since then it has become apparent that the incidence of AKI is significantly higher than previously thought and the true impact AKI has on mortality and morbidity is potentially catastrophic.

Table 1.3 Stages of Acute Kidney Injury

Stage	Serum Creatinine	Urine Output
1	≥ 1.5-1.9 times baseline or Increase in creatinine ≥ 0.3 mg/dl (26 µmol/l)	< 0.5 ml/kg/hr for 6-12 hrs
2	≥ 2.0-2.9 times baseline	< 0.5 ml/kg/hr for ≥ 12 hrs
3	≥ 3.0 times baseline or Increase in creatinine ≥ 4.0 mg/dl (353 µmol/l) or On renal replacement therapy	< 0.3 ml/kg/hr for ≥ 24 hrs or Anuria for ≥ 12 hrs

Abbreviations: hr, hours; AKI, acute kidney injury; KDIGO, Kidney Disease: Improving Global Outcomes

Table 1.3

This is the classification system for AKI adopted by KDIGO modified from the Risk, Injury, Failure, Loss of kidney function , End-stage kidney (RIFLE) and Acute Kidney Injury Network (AKIN) criteria based on rise in serum creatinine and/or urine output changes. AKI is defined as an abrupt reduction in renal function (within 48 hours) defined as an increase in serum creatinine ≥ 26 µmol/l (0.3mg/dl), ≥ 50% increase in serum creatinine (1.5-fold from baseline) or a reduction in urine output < 0.5ml/kg/hr for > 6 hours (70). The AKIN stages are based on severity of AKI.

Measuring Renal Function in AKI

Diagnosis of AKI is exceptionally easy and is obtained by a simple blood test measuring serum creatinine and/or measurement of urine output. In order to diagnose and assess the severity of an AKI episode, knowledge of baseline kidney function is required if known. A problem arises when the baseline creatinine is not known and estimating baseline function can be subject to a degree of interpreter variability. Inaccurate determination of the baseline renal function can sometimes lead to a misclassification of AKI. There has been a lot of debate about the implications of the choice of measurements of the baseline kidney function when defining the presence and severity of AKI. Several hospital-based studies have made assumptions on baseline serum creatinine in the absence of outpatient values when investigating acutely ill hospitalised patients (64-66). This approach ignores the association that pre-existing CKD often contributes to the development of AKI.

The KDIGO AKI guideline suggests that an estimated creatinine can be used, provided there is no evidence of CKD (70). However there remain many cases of CKD in the community that have not been previously appreciated, and hence estimating the baseline serum creatinine may lead to a diagnosis of AKI when in fact the patient has previously un-recognised CKD. Recently, a proposed national algorithm standardising AKI definition has been endorsed by NHS England with plans to introduce a wide-scale uptake of an automated computer software algorithm to detect AKI according to AKI stage (71-72). This algorithm defines the baseline creatinine as the lowest value if measured 0-7 days prior to the AKI episode or the median of results within 8-365 days. If no results are available within 365 days then results are compared to reference intervals related to age and gender.

Other Markers of Acute Kidney Injury

Despite the fact that serum creatinine is affected by several non-renal factors (muscle bulk, gender, concomitant drugs) and takes 24-48 hours to rise after the initial renal insult, it remains the traditionally used parameter to diagnose AKI. It is a

reasonably cheap test and a fairly reliable and available assay. Estimated GFR does not accurately reflect true GFR in AKI as it does in CKD and in light of the inadequacies of conventional markers of renal damage in the acute setting, there have been numerous studies investigating newer AKI biomarkers including urinary Neutrophil Gelatinase-Associated Lipocalin (NGAL) (73), Kidney Injury Molecule (KIM)-1, IL-18 and cystatin C (74). These biomarkers aim to detect AKI before a detectable serum creatinine rise to aid diagnosis and improve management and outcomes. As the aetiology of AKI is not uniform, these biomarkers need to consistently perform accurately across a wide range of settings. So far few biomarkers of AKI have been adopted in routine clinical use as studies have reported variable results with inconsistent performances across specific conditions and they are neither practical nor cost-effective in clinical practice (75).

1.7 Risk Factors for AKI

Several risk factors have been identified for the development of AKI that occur across a wide range of settings which can be modifiable or non-modifiable and these are summarised in Table 1.4. Sepsis is the leading cause of hospital-acquired AKI and studies have shown that septic AKI confers a higher intensive care unit (ICU) and hospital mortality when compared to non-septic AKI (76). One of the most important risk factors for the development of AKI is the presence of co-morbidities. Diabetes, CKD, vascular diseases and cardiac insufficiency are frequently associated with development of AKI. The presence of pre-existing CKD dramatically increases the risk of development of AKI and the relationship of these two conditions is discussed in more detail in subsequent chapters (77).

The elderly population are at particularly high risk of community and hospital-acquired AKI (78-79). The segment of the population in which the incidence of AKI has been increasing the most rapidly is those with advanced age. It is suggested that background changes in the kidney, related to age and CKD, together with an increase in the prevalence of clinical situations such as dehydration, drug toxicity and co-

morbidities put this patient population at higher risk of developing AKI. Elderly patients are more likely to have chronic illnesses, cardiovascular disease or diabetes and are more likely to be on complex polypharmacy. Urinary tract obstruction is a major cause of AKI almost exclusively observed in the elderly and is responsible for 25% of AKI occurring in the elderly (80). With more co-morbidity, older people are likely to under-go more surgical procedures and require more interventions or imaging with exposure to nephrotoxic contrast media.

The most important modifiable risk factors for AKI are dehydration, hypovolaemia, toxicities related to medications or contrast media, surgery related complications, sepsis and cardio-renal syndrome. Hospital-acquired AKI following surgery is an important contributor to post-operative morbidity and mortality and is often associated with pre-renal causes of AKI. Pre-renal causes can represent true volume depletion as well occur in conditions which decrease effective arterial blood volume causing renal hypo-perfusion. The incidence of AKI can range from 0.1% in general surgery to 31% following cardiac surgery highlighting the variable nature of this condition (81). The high prevalence of co-morbidities in the older population often leads to an increase in demand for interventional invasive tests and surgical procedures with higher perioperative complications. The potential nephrotoxicity of commonly used drugs is often misunderstood and medication-related toxicity often leads to the development of AKI of which the common culprits are NSAIDs, diuretics, angiotensin blocking agents and contrast agents.

Several studies have identified that AKI almost always occurs due to multi-factorial risk factors. Some studies have developed and prospectively validated risk-stratification scores most notably after cardiac surgery and coronary angiography to categorise patients according to risk of developing AKI (82-83). Further work is needed to cross-validate these risk scores across different populations and in various settings due to the multiple aetiologies of AKI.

Table 1.4 Risk Factors for Development of Acute Kidney Injury

Co-morbidities/ Non-modifiable Risk Factors	**Clinical Conditions**	**Drugs**
Advanced age Diabetes CKD Heart Failure Liver Failure Female gender Genetic factors Low albumin Arterial disease Myeloma	Sepsis Hypotension/shock Volume depletion Rhabdomyolysis Cardiac/vascular surgery Non-renal solid organ transplant Hepatic/biliary surgery	Contrast media Antibiotics Chemotherapy Non-steroidal anti-inflammatories Angiotensin blocking medications

Abbreviations: CKD, chronic kidney disease; AKI, acute kidney injury

Table 1.4

This table lists the commonly recognised risk factors associated with the development of AKI. Risk factors are divided into pre-existing conditions, acute clinical conditions and the common medications often related to the development of AKI. Co-morbidities including CKD and vascular diseases and older age are common pre-existing non-modifiable risk factors. Certain clinical conditions with increased risk of reduced blood flow and medication-associated AKI are common modifiable risk factors. Many drugs contribute to the development of AKI and therefore should be avoided in patients with or at risk of AKI.

Incidence of AKI

Hsu *et al* examined a cohort of beneficiaries of the health-care delivery system Kaiser Permanante in North California and found the use of acute RRT in patients with AKI was 295 pmp per year (pmp/yr) and an increased incidence of AKI not requiring dialysis of 522 per 100,000 person-years (78). They also found that the incidence of community-acquired AKI increased with age until the ninth decade (1232 pmp/yr in patients aged 70-79 year; 625 pmp/yr in 80-89 years). Hospital-acquired AKI is thought to be approximately 5-10 times more frequent than community-acquired AKI (84) but this figure varies according to studies and local demographics. Community-acquired AKI occurring in more rural settings rarely reach hospital and therefore are not captured in accurate measurement of AKI incidence. Severe AKI requiring ICU admission occurs in 11 patients per 100,000 population per year and AKI has been reported to occur in 30-67% of all ICU admissions usually as a manifest of multi-organ failure (85).

1.8 AKI Outcomes

AKI is often reversible and early detection of AKI is vital as instituting appropriate management can lead to complete recovery. Many studies have addressed variable short-term and long-term outcomes of AKI most frequently assessing mortality. AKI can result in multi-organ failure and can accelerate cardiovascular disease and CKD.

AKI in hospitalised patients is associated with increased mortality and this risk is even higher in patients requiring renal replacement therapy and increases according to severity of AKI (60, 84). AKI is associated with a number of changes in the vascular endothelium and it has been suggested that these have an impact on cardiovascular health. There is emerging evidence that even minor increases in serum creatinine concentration of 26µmol/l (0.3 mg/dl) are associated with increased mortality, increased length of hospital stay and increased costs to the healthcare economy (86). Even in patients with complete renal recovery there is still a reduced survival rate. (87).

Despite important technical advances in clinical care, the prognosis for patients with AKI remains poor and mortality rates can exceed 40-80% in the intensive care setting (88). Furthermore, it is also suggested that an episode of AKI confers an increased risk of subsequent mortality following recovery and discharge from hospital. Studies have previously looked at 30 and 90-day survival and renal recovery at these time points (89). From our local data studying the incidence of AKI in a district general hospital, 15% of all admissions sustained an episode of AKI with increased subsequent short and long-term morbidity and mortality, even in those with AKIN1 (90). Only 56% of patients who experience an episode of severe AKI in hospital survived to be discharged, and only 28% survived to 3 years post discharge.
In a study that examined survival in both young and old patients, AKI and older age were independently associated with the risk of long-term death (91). Recent cohort studies have shown that despite new therapeutic interventions the incidence of AKI is increasing but mortality is decreasing (84).

Not all AKI is reversible and patients may be rendered RRT dependent or be left with a significant residual loss of renal function. Ishani *et al* demonstrated that AKI increased the risk of ESRD by 13-fold (87). The differences in rates of recovery from AKI between the young and old remain unclear but we know that the chances of renal recovery are greatly deceased in the presence of CKD. As CKD is more prevalent in elderly people and GFR decline occurs in ageing, it is very probable that the elderly population have a reduced likelihood of recovering renal function.

Very few studies have looked at the long-term quality of life in AKI survivors. One small study looked at a small cohort (n=16) of survivors of an AKI of a total original cohort of 117 patients with an AKI requiring RRT and found a significantly lower quality of life score than population norms (88).

Management of AKI

Strategies to reduce the incidence and severity of AKI involve identifying relevant risk factors with regular monitoring of renal function and immediate treatment when AKI occurs. Management of AKI usually involves supportive measures and most cases remain under the care of the admitting clinical team however the more severe cases of AKI (AKIN 3) often require renal specialist or ICU input. Supportive measures include fluid resuscitation, prompt recognition and treatment of sepsis, cessation and avoidance of nephrotoxic agents, managing complications of AKI and close haemodynamic monitoring. Management of AKI in the elderly can be more challenging particularly in extreme age or the presence of multiple co-morbidities.

Complications of AKI such as hyperkalaemia, acidosis, volume overload and encephalopathy can increase mortality and risk for persistent decline of kidney function and need treating promptly. Very few studies have shown a benefit from pharmacological interventions and even timing of initiation and type of renal replacement therapy have shown little in terms of prognostic benefit (92-94).

Early diagnosis of AKI and establishment of these supportive measures are vital in reducing AKI severity and preserving functioning nephrons, hence improving patient and renal outcome and there have been significant advances in the research and intervention in AKI since it has become evident what the true incidence of AKI is. It is now clear that AKI has a significant impact on morbidity and mortality increasing the burden on our health services. It has been known for some time that many cases of AKI are iatrogenic in origin and up to 30% are preventable (59). An example of this is contrast-induced nephropathy, a condition associated with up to a 6-fold increase in mortality (95). Simple precautions such as pre-hydration prior to contrast administration, discontinuation of drugs which reduce renal blood flow or reduce intravascular volume, using minimal volume of contrast and if possible avoidance of contrast by alternative diagnostic methods have been shown to minimise risk for developing contrast associated AKI (96). It is important that patients with AKI or at risk of developing AKI are recognised at the earliest opportunity and that instituting

supportive measures is directed at minimising further injury and development of its complications.

The importance of AKI on patient outcomes and overall poor care in the management of AKI was emphasised in 2009 by the National Confidential Enquiry into Patient Outcomes and Death (NCEPOD) report on hospitalised patients with AKI (97). This report investigated the management of patients who died as result of AKI. It identified significant failings in the recognition and management of AKI highlighting widespread deficiencies in the clinical care of these patients. The report included recommendations for the early diagnosis, prevention and management of AKI in acute hospitals and suggested organisational changes. Since then there has been a huge national drive in improving awareness with publication of national guidelines, development of computer AKI alert systems and formation of AKI networks all in order to aid better detection and management to improve clinical outcomes. The NCEPOD report has resulted in a significant increase in research in the field of AKI and has raised the awareness of AKI and led to the introduction of structured management guidelines. Much of the current focus in AKI research has been on identifying risk factors for AKI and developing strategies to prevent AKI in high-risk patients.

CKD is increasingly prevalent yet few people with CKD progress to ESRD. AKI is increasingly common and recently significant advances have been made to improve prevention, recognition and management of AKI. There is increasing evidence that AKI events may contribute to the progression of CKD yet there are multiple unanswered questions about their relationship. This study aims to answer ‘is the prevalence of CKD a true prevalence?’, and ‘does AKI contribute to CKD?’

CHAPTER 2

Measuring Renal Function

Publication of the second part of the Department of Health's National Service Framework (NSF) for Renal Services (10) in 2005 highlighted the importance of CKD but changed the focus of care delivery from treatment of established renal disease to early identification and prevention of CKD. The guidelines suggested identifying people at risk of developing CKD and initiating interventions to prevent progression of disease, minimise cardiovascular risk and identify those at risk of progression to ESRD. Delivering these quality requirements relies on accurate measures of kidney function in order to treat and screen those with or at risk of developing CKD appropriately.

Diagnosis of CKD and assessment of progression of disease is dependent on monitoring of renal function and the best measure of renal function is glomerular filtration rate. A reduction in GFR precedes kidney failure in all forms of progressive renal disease. In addition, estimated GFR is used in clinical practice to allow for proper drug dosing of medications that are excreted by glomerular filtration in order to avoid drug toxicity. Where a highly accurate measure of GFR is required such as monitoring during chemotherapy, a gold standard measure of GFR is usually recommended (12). Inaccuracies in methods used to estimate GFR would incur serious consequences and increase the risk of unsafe drug prescribing and may lead to the misdiagnosis and misclassification of patients with CKD.

2.1 Serum Creatinine Measurement

Serum creatinine levels were until recently used to assess level of kidney function in both the acute and chronic setting. Creatinine is a chemical waste product of creatine phosphate which is formed physiologically by muscle metabolism and is produced at a fairly constant rate. Creatinine is exclusively removed from the body by the kidneys primarily by glomerular filtration but also by proximal renal tubule secretion and higher levels of serum creatinine indicate worsening renal function. Little or no tubular reabsorption occurs. Each day approximately 1-2% of muscle creatine is converted into creatinine. A rise in serum creatinine is only observed after a significant number of functioning nephrons are lost and is therefore unsuitable at detecting early stages of kidney damage. Serum creatinine measurement is a simple obtainable investigation that requires a single blood sample and is available in all medical laboratories.

Biological Variability of Serum Creatinine Measurements

The use of serum creatinine level alone is considered inaccurate in measuring renal function and assessing CKD as it has several limitations. Many studies have documented that creatinine production varies substantially across sex, age and ethnicity (98). More so, there is considerable variability in the individual muscle mass of the population. The reference values associated with serum creatinine have been traditionally stratified by gender and age to reflect differences in muscle mass however they are often inadequate in estimating muscle mass and also do not take ethnicity into account. Differences in muscle metabolism and variable rate of secretion of creatinine in the proximal tubules will also affect serum levels. Certain medications such as trimethoprim and cimetidine reduce tubular creatinine secretion limiting the ability to accurately estimate GFR. Increased dietary intake of creatine or high ingestion of meat or fish can also increase creatinine levels (99-100). The NICE CKD guidelines now recommend avoidance of meat consumption 12 hours prior to serum creatinine measurement (57). The inability to relate creatinine to GFR was believed to have led to the under-diagnosis and misclassification of CKD stage in

patients who may well have benefited from earlier intervention. Despite its limitations, serum creatinine testing remains one of the most frequently used indicators of renal function and is a standard component of a panel of tests requested at most medical care centres.

Standardisation of Serum Creatinine Measurements

Prior to 2006, normal creatinine value ranges varied significantly among different laboratories due to variability in their analytical methods and assays. This raised concerns of a possible detrimental impact in the clinical care of patients and called for global public health efforts to highlight the importance of reliable serum creatinine measurements in identifying people with CKD. It is now an international priority that serum creatinine assays must be properly calibrated and traceable to high-order reference systems in order to eliminate or reduce variation among laboratories. Expert professional bodies recommend that clinical laboratories now align their creatinine measurements against a new standardised isotope dilution mass spectrometry (IDMS) method using a commutable IDMS-traceable reference material (Standardised Reference Material (SRM) 967) to measure serum creatinine in accordance with the NKF-KDOQI CKD guidelines (101-102). IDMS appears to give lower creatinine values compared to older methods when the serum creatinine levels are relatively low.

2.2 Measuring Glomerular Filtration Rate

Measured GFR is generally accepted as the best overall measure of renal function. It provides an accurate measure of the filtration capacity of the kidneys and, as the total GFR is equal to the total filtration rate of each of the functioning nephrons, this can be used to indicate the total functioning renal mass. Reduction in the GFR precedes CKD and persistent decline in function indicates progressive CKD. The GFR measured as clearance of creatinine can be a difficult test to accurately execute in clinical practice. Measuring glomerular clearance of creatinine requires accurate timed urine samples and is impractical and costly and hence unfeasible in assessing

CKD in a large cohort. GFR has not been demonstrated as providing an accurate assessment of renal function in AKI as it does not accurately reflect tubular damage.

The gold standard measurement of GFR uses reference procedures in which the clearance of an infused exogenous substance that is solely excreted by the kidneys is measured. This filtration marker must be exclusively eliminated in the urine by glomerular filtration of the functioning nephrons and not be affected by tubular secretion in order to be an accurate measurement of renal clearance. Inulin was traditionally used as the gold standard marker however this is not only expensive but measuring inulin levels is technically difficult (103). Measurement of GFR using the inulin clearance method is an impractical test partly due to the analytical problems associated with the measurement of inulin and due to requirements of bladder catheterisation in order to get accurate timed urine samples. ^{51}Cr-EDTA, ^{125}I-iothalamate and iohexol are more commonly used as alternative reference markers (104). The radio-labelled chelating agent ^{51}Cr-EDTA has been the most commonly used measured GFR marker in Europe for years however this test is not available in the US. This test is performed in nuclear medicine departments but is a difficult and time consuming test to perform and inappropriate for routine clinical use and dependent on availability of equipment for measuring radio-labelled markers.

Plasma Iohexol Clearance

Iohexol has become one of the most commonly used alternative marker for GFR measurement. It is a safe, non-radio-labelled and non-ionic contrast agent. It is not metabolised, is less than 2% protein-bound, and is eliminated without being metabolised exclusively by glomerular filtration. There is close agreement of GFR measured by iohexol renal clearance and iohexol plasma clearance however renal clearance measurements require urinary concentrations of iohexol with accurate timed urine collections and this is inconvenient in clinical practice. Instead, in order to simplify the procedure, plasma iohexol clearance has been developed for use clinically as the procedure requires only a single bolus of contrast and subsequent

timed serum sampling (105). This method avoids the need for intravenous infusions and timed urine collections.

A determination of GFR is obtained from the dose of iohexol administered and the area under the curve (AUC) serving as a function of time. Accuracy of the iohexol plasma clearance method is dependent on a mathematical model and the iohexol sampling times. When the iohexol marker is injected, there is diffusion of the marker both in plasma and in various compartments. The plasma concentration of iohexol is therefore dependent on 4 factors; the amount of marker administered, urinary excretion of the marker, the diffusion rate of the marker within various compartments and the retro-diffusion rate of the marker from the deeper compartments into plasma. There is thus an initial rapid change in plasma marker concentration during the distribution phase and later on, a slower reduction in plasma concentration during the excretion phase as the marker re-enters the plasma from the compartments. This multi-compartmental model would require several blood samples however Brochner-Mortenson *et al* (106) simplified this method by creating a model in which the accuracy of the plasma clearance values after the marker injection depends on the timing of the blood samples. The optimal time to sample serum is after the distribution phase. The time needed to complete equilibrium between plasma and extravascular compartments is inversely proportional to level of kidney function so it is recommended that later sampling is performed in individuals with reduced renal function to enhance accuracy. Characterising the iohexol disappearance curve based on samples only from early time points will result in an overestimation of GFR and it is recommended that iohexol GFR can be satisfactorily measured with a minimum of 3 measurement points within 4-5 hours of administration (105). The iohexol plasma clearance method however is not widely available and impractical for a large-scale application in the adult population where CKD is more prevalent.

2.3 Estimating GFR Equations

While GFR can be measured by clearance studies of exogenous markers such as iohexol, inulin, iothalamate and Cr^{51}-EDTA, they are costly, time consuming and not suitable for the routine detection of kidney disease. The accurate measurement of glomerular clearance of endogenous substances such as creatinine and urea requires serum and timed urine collections and again is impractical in screening large populations. It became apparent that the variable interpretation of serum creatinine levels often led to misdiagnosis of renal disease so estimating equations were then developed based on the serum creatinine level to provide an estimated GFR which take into account the effect that age, sex and race have on creatinine production. Numerous estimating equations have been developed to predict GFR in both adults and children in order to enable better identification of patients with early kidney disease who can be helped by therapeutic interventions. In principle, GFR estimating equations provide a more accurate estimate of measured GFR than serum creatinine levels. Until recently, variation in creatinine measurements between different laboratories was a major source of bias of these estimating equations. However since the creatinine-standardisation program has been implemented throughout the United States and United Kingdom, these equations are more accurate in estimating GFR (102). The national introduction of routine eGFR reporting with every serum creatinine request in 2006 has also helped to improve the identification of individuals with CKD.

The Cockcroft-Gault Equation

In 1976, Cockcroft and Gault proposed an estimating equation for GFR using weight, gender and age as variables based on serum creatinine levels (107). An important characteristic of the Cockcroft-Gault formula is the inclusion of total body weight as a reflection of the muscle mass, the main determinant of creatinine generation. The calculation of estimated creatinine clearance using the Cockcroft-Gault equation has been the most commonly used method to estimate kidney function for drug dosing

purposes for decades. This is based on prospective pharmacokinetic studies that were conducted on patients whose level of kidney function was determined by estimated or measured creatinine clearance. The Cockcroft-Gault equation however does not correct for race and a relative limitation of this equation is the need for a calculator. Another limitation is that with increasing age, the body composition changes with decreasing muscle mass and increasing fat tissue with a reduction in lean body mass in the elderly thus causing inaccuracies of this equation in estimating GFR in older people. The Cockcroft-Gault equation is no longer recommended by KDIGO guidelines in assessing eGFR as it was developed before the era of standardisation of creatinine assays (12).

The Modification of Diet in Renal Disease (MDRD) Study Equation

The MDRD study equation was developed in 1999 using data from a base population of 1628 out-patients with established CKD (8). Various MDRD equations have been published however the most widely used equation by the health care community is the abbreviated, four-variable MDRD equation, which has been reformulated to be used with a standardized serum creatinine assay (Table 2.1). It uses age, the inverse of serum creatinine, gender, and race (African-american versus non–african american) as variables. This equation directly relates the accounted variables (e.g. serum creatinine, age, gender and race) to GFR adjusted for body surface area. The set of equations developed from the data derived from the MDRD study aimed at estimating GFR compare to GFR measured by ^{125}I-iothalamate urinary clearance, the reference method used in this study. In contrast to the Cockcroft-Gault formula, the MDRD model accounts for the biological relation of creatinine metabolism observed in african-americans, but there is no adjustment for other ethnicities. The MDRD study equation has shown advantages over most other proposed equations and the NKF-KDOQI recommend using this equation, rather than the Cockcroft-Gault formula, to estimate kidney function.

Table 2.1 MDRD Study and CKD-EPI Equations for GFR Estimation

MDRD

GFR (ml/min/1.73m^2) = 175 x (sCr) $^{-1.154}$ x (Age)$^{-0.203}$ x (0.742 if female) x (1.21 if black)

CKD-EPI

Black

Female	sCr ≤ 61.9 µmol/l	GFR = 166 x (sCr/61.9)$^{-0.329}$ x (0.993)^{Age}
	sCr > 61.9 µmol/l	GFR = 166 x (sCr/61.9)$^{-1.209}$ x (0.993)^{Age}
Male	sCr ≤ 79.6 µmol/l	GFR = 163 x (sCr/79.6)$^{-0.411}$ x (0.993)^{Age}
	sCr > 79.6 µmol/l	GFR = 163 x (sCr/79.6)$^{-1.209}$ x (0.993)^{Age}

White or other

Female	sCr ≤ 61.9 µmol/l	GFR = 144 x (sCr/61.9)$^{-0.329}$ x (0.993)^{Age}
	sCr > 61.9 µmol/l	GFR = 144 x (sCr/61.9)$^{-1.209}$ x (0.993)^{Age}
Male	sCr ≤ 79.6 µmol/l	GFR = 141 x (sCr/79.6)$^{-0.411}$ x (0.993)^{Age}
	sCr > 79.6 µmol/l	GFR = 141 x (sCr/79.6)$^{-1.209}$ x (0.993)^{Age}

Abbreviations: MDRD, Modification of Diet in Renal Disease study equation; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration equation; GFR, glomerular filtration rate; Scr, serum creatinine.

Table 2.1

This table shows the MDRD study and CKD-EPI estimating formulae using gender, age, and ethnicity as variables. Age is represented in years. Both equations do not require weight or height variables but results are reported normalized to 1.73m^2 body surface area. The CKD-EPI equation uses a 2-slope spline to model the relationship between GFR and serum creatinine, age, sex and race. The CKD-EPI is reported to perform more accurately than the MDRD study equation at higher levels of eGFR.

The MDRD was then suggested to universally substitute the Cockcroft-Gault method to determine drug dosing in patients with impaired renal function. The findings of several retrospective studies suggest that although the MDRD and Cockcroft-Gault equations correlate with measured GFR, the MDRD often overestimated creatinine clearance leading to potential errors in drug dosing (108). The studies reported that drug dosages determined by the two equations did not agree in 10-40% of cases. As the Cockcroft-Gault equation typically yields a more conservative estimate compared to the other estimating equations, there is a need for a dose adjustment more frequently. Until safety concerns are adequately addressed with other estimating equations, the Cockcroft-Gault equation is still recommended for drug dosing in impaired renal function.

The MDRD study equation was developed in people with CKD with a mean ^{125}I-iothalamate GFR of 39.8±21.2, and as such its major limitations are imprecision and systematic underestimation of measured GFR (bias) at higher levels (8). A further confounder to the inaccuracy of the earlier derived MDRD equation was the lack of standardised serum creatinine assays that existed prior to 2006, with resultant lack of comparability of equation performance. With the advent of international creatinine standardisation, the MDRD equation was re-expressed for use with such assays (109). The MDRD equation, however, is not recommended for use when the GFR is > 60ml/min/1.73m^2 as estimates become progressively too low and have increased variability partly due to the poorer precision methods at lower versus higher creatinine concentrations.

The Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) Equation

Shortly after this, in 2009, the CKD-EPI equation was published, having been developed in a broader population and claiming to have superior bias compared to the MDRD equation at higher levels of GFR (110). It was developed and validated from a much larger cohort of 8,254 individuals compared to the MDRD study and included people with and without CKD and hence is more accurate at higher levels of

renal function. The CKD-EPI equation uses the same 4 variables as the MDRD equation but expresses the log serum creatinine modelled as a 2-slope linear spline with sex-specific knots at creatinine values of 62 µmol/l in females and 80 µmol/l in males (Table 2.1). This introduced a more complex equation and the limitations of using serum creatinine as a marker still apply. The CKD-EPI was derived from a population consisting of pre-dominantly young or middle aged people with an average GFR of 70 ml/min/1.73m^2. The CKD-EPI equation produces higher GFR estimates particularly in those aged < 60 years of age. In the CKD-EPI validation study, the CKD-EPI equation had slightly better accuracy than the MDRD equation at GFR < 60 ml/min/1.73m^2 but had good accuracy at GFR > 60 ml/min/1.73m^2 making it suitable for reporting eGFRs in this range (9).

Since the development of the CKD-EPI equation, several studies have evaluated the impact of using the CKD-EPI equation compared to the MDRD equation in various populations including transplant patients (111), patients undergoing nephrectomy (112-113), middle aged subjects without a history of cardiovascular disease (114) and non-institutionalised adult Australians (115). The CKD-EPI had a consistent performance across these study subgroups including race, gender, and varying body mass index. Overall these studies demonstrated that the CKD-EPI equation produced a more accurate estimate of GFR and a lower prevalence of CKD. The impact of using this equation for estimating GFR is it leads to the reclassification of CKD patients from stage 3a to stage 2 CKD. There have been recent proposals for the adoption of the CKD-EPI equation in place of the MDRD equation in the routine reporting of eGFR from serum creatinine measurements by laboratories worldwide. There is however, little known about the consequences of applying the CKD-EPI equation to older people.

The MDRD and CKD-EPI equations have limitations. Both were developed in a cohort of african-americans and caucasians and do not take other ethnicities into account. Both the MDRD and CKD-EPI equations have also not been validated in older people and there is little evidence to support their accuracy in this population group. The

MDRD equation for GFR estimation was developed amongst 1628 CKD patients with a mean age of 50.6±12.7 years and an underrepresentation of elderly patients (8). In a large study (n=2095), a sub-analysis in patients >65 years old and with GFR <60 demonstrated comparable performance of the MDRD equation to that amongst younger individuals (mean bias -1.0 ml/min/1.73 m^2) (116). However, only 57 subjects were aged ≥ 80 years. An evaluation of the MDRD equation in a pooled dataset of 5504 people found that the MDRD had a minimal negative bias in individuals aged over 65 years (n=586) compared to younger individuals but there were very few numbers of participants older than 80 years (n=23) (25). In the few studies of the MDRD performance in older people there have been inconsistencies in the direction of bias of the MDRD equation compared to the measured GFR either overestimating or underestimating GFR (117).

Standards for Estimating Equations

In 2002, the NKF-KDOQI published clinical practice guidelines that incorporated the evaluation of laboratory measurements for clinical assessment of kidney disease (10). They advised that serum creatinine alone should not be used for assessing the level of kidney function and that estimated of GFR is the best overall indicator of renal function. The MDRD and Cockcroft-Gault equations were recommended in estimating GFR in adults and the Swartz and Counahan-Barratt equations based on height and serum creatinine were recommended in children (118-119).

The guidelines called for all clinical laboratories to routinely report an estimate of GFR using a predicting equation when reporting a serum creatinine measurement and that all laboratories should calibrate serum creatinine assays using an international standard. Since the standardisation of commercially available creatinine assays, a revision of the MDRD equation offers traceability to a reference method (120). This modified IDMS-traceable MDRD study equation is now generally used to routinely report eGFR when a serum creatinine is requested from laboratories both nationally and worldwide.

The guidelines recommended that assessing the accuracy of a prediction equation to estimate GFR requires calculating bias, precision and accuracy.

- Bias expresses the systematic deviation from the gold standard measure of GFR. If an estimating equation consistently overestimates or underestimates the gold standard measure it will yield a positively or negatively biased estimate respectively. A negative bias indicates that the prediction equation underestimates GFR and a positive bias overestimates GFR compared to the reference GFR. Bias is defined as the median difference between the measured and estimated GFR. In some studies this is expressed as a percentage basis and in others it is expressed as ml/min/1.73 m^2.
- Precision expresses the variability or spread around the gold standard GFR result. Precision can be calculated by the *R*-square values or the interquartile range (IQR) for the differences.
- Accuracy combines precision and bias. Achieving a high level of accuracy of a test would require both a low bias and a high precision. A useful measure of accuracy is the percentage of estimates falling within 30% (P_{30}) or 50% (P_{50}) above or below the measured GFR. This takes into account greater errors at higher values.

The performance of the MDRD and CKD-EPI equations were assessed using these three measures in the two validation studies (31, 110). The NKF-KDOQI CKD guidelines also recommended that future studies should have at least 100 adults or 50 children to overcome the unreliable estimates of accuracy from smaller sample studies. Another initiative recommended to clinical laboratories to improve the performance of estimated GFR calculations was after recalibration of serum creatinine measurements to an IDMS method, to set a total error goal for creatinine measurements to a maximum 10% error in eGFR. This interprets that laboratories need to aim for an analytical bias of < 5% and analytical imprecision of < 8% (including between laboratory calibration variability) at serum creatinine concentrations of > 88 µmol/l (10).

The current existing estimating equations provide a cost effective method of estimating GFR but their precision is limited. The NKF-KDOQI guidelines called for newer accurate methods of estimating GFR in terms of bias, precision, accuracy and practicality particularly in mild to moderate kidney disease. Future estimating measures would have to perform substantially better than the 12.1% median difference achieved by the MDRD Study equation in the validation study (31). They would need also to achieve P_{30} values in excess of 90%. To date, across a wide range of studies in different settings and using different reference techniques, this has rarely been achieved (108, 119). This is partly due to the small sample sizes used in many studies. Since accuracy from smaller studies can be unreliable, recommendations have been made to re-evaluate them in larger validation studies.

In the MDRD validation study, the MDRD study equation had an overall bias of 2.7ml/min/1.73 m^2, a precision of 5.8% and a P_{30} value (accuracy) of 83%. When assessing the performance at GFR levels, the MDRD study equation had little bias for eGFR < 60 ml/min/1.73 m^2, underestimated GFR for levels of eGFR between 60 and 119 ml/min/1.73 m^2 and overestimated GFR at eGFR levels > 120 ml/min/1.73 m^2 (31). In the CKD-EPI validation study, the CKD–EPI equation performance was compared to the MDRD study equation and was found to have less bias (2.5 versus 5.5 ml/min/1.73 m^2), improved precision (IQR of the differences 16.6 versus 18.3 ml/min/1.73 m^2) and greater accuracy (P_{30} values of 84.1% versus 80.6%) in the total dataset (9).

Nevertheless the updated NICE CKD guidelines published in 2014 (57) recognised that inaccuracies of estimated GFR methods increased as true GFR increased and have recommended that the CKD-EPI creatinine equation be used to estimate GFR using creatinine assays with calibration traceable to standardised reference material.

Cystatin C

Generally studies have demonstrated advantages of the MDRD equation to other eGFR equations and its use has been endorsed, however, there remains a dearth of supportive evidence for its use in older people. Alternative markers of GFR have been proposed. Perhaps the most promising of these is serum cystatin C. Cystatin C is a low molecular weight protein which was introduced recently as a GFR estimate superior to creatinine. In particular, serum cystatin C is sensitive to detect mild GFR reduction between 60 and 90 ml/min/1.73 m^2 (121). However, no agreed reference method and no uniform calibration material exist for cystatin C yet and there are other limitations such as the effect of thyroid dysfunction, high glucocorticoid doses and potentially the presence of cardiovascular diseases on cystatin C levels. Cystatin C-based equations have been proposed to further improve GFR estimation, which seem to be superior to creatinine-based ones however there is again limited evaluation amongst older people.

One study by Eriksen *et al* contradicted these results and demonstrated that cystatin C was not a better estimator of GFR than plasma creatinine in the general population (122). However, there is now abundant evidence that GFR estimates based on cystatin C are more powerful predictors of clinical outcomes than creatinine-based eGFR. These findings are strongest for predicting mortality and cardiovascular events and the advantage is greater in individuals with GFR > 45 ml/min/1.73m^2 but without proteinuria (123). This group represented 3.6% of the US population and 41% of people estimated to have CKD based on creatinine-based eGFR and ACR alone. They indicate that use of cystatin C to estimate GFR in this population lead to a more accurate estimation of GFR and prediction for adverse outcomes. The 2014 CKD NICE guidelines (72) have incorporated cystatin C eGFR and recommend its use at initial diagnosis to confirm or rule out CKD in individuals with GFR 45-59 ml/min/1.73m^2 with an ACR < 3 mg/mmol or other marker of kidney disease.

With an ageing population and increasingly complex polypharmacy, the accurate estimation of GFR and detection and management of renal impairment is clearly an important issue in older people. Currently, the MDRD equation is being applied to this population amongst whom limited evaluation has been undertaken and the relationship between age and eGFR is being extended beyond that in which it was established. Other estimating equations have been developed which claim to be more accurate in specific populations but all have their limitations and there is limited data in the elderly. There is a need for a large-scale study validating use of these estimating equations in older people.

CHAPTER 3

CKD Progression and the Interaction of CKD and AKI

3.1 CKD Progression

Due to the controversies around the effect of ageing on GFR, there has been little consensus until recently on the definition of CKD progression. Definitions have varied from doubling of serum creatinine, increasing proteinuria and albuminuria, decline in GFR to commencement of RRT. Longitudinal studies following healthy kidney donors showed that there was a natural decline in GFR with age but no more than 2 ml/min/1.73m^2 (124). Rate of decline in eGFR is now used and a decline in eGFR of > 5ml/min/1.73m^2 in 1 year or > 10ml/min/1.73m^2 over 5 years defines CKD progression. As discussed earlier, there is a significant biological variability in serum creatinine quoted up to 5% and this must be taken into account when assessing GFR and rate of decline. Small fluctuations in GFR are common and not necessarily indicative of CKD progression and interpretation of baseline and rate of decline can be difficult particularly with limited numbers of GFR measurements and a short duration of follow-up. KDIGO guidelines (101) suggest that a minimum of three eGFR levels are obtained within 90 days to identify progressive CKD but the patterns of CKD progression can vary significantly even within one individual according to development of co-morbid conditions and too frequent testing of renal function is often unnecessary and impractical.

This definition of progressive CKD does not take cause of kidney disease into account and this can be of fundamental importance in predicting the outcome of CKD.
A large number of studies have investigated the clinical characteristics that are thought to influence CKD progression. Proteinuria and poorly controlled hypertension have been consistently found to be independent predictors of progressive CKD (35-36). In hypertensive patients with diabetes and CKD stages 1-4,

treatment guidelines recommend the use of an ACEI or ARB in combination with a diuretic to achieve a target blood pressure < 130/80 mmHg (101). The UK Prospective Diabetes Study Group (UKPDS) demonstrated that tight diastolic blood pressure control in diabetics not only reduced progression in terms of albuminuria but also reduced rate of cardiovascular events and death (125). Other studies have also shown that renal function declines more rapidly as the mean arterial pressure increases (7).

People with cardiovascular disease have been found to have a significantly increased risk of decline in GFR compared to those without (34, 126). Many other risk factors have been identified. The effect of smoking on renal function decline has been demonstrated in diabetic cohort studies and case-control studies with smokers having an increased odds ratio of a 20% decline in GFR compared to non-smokers (127-128). In cross-sectional studies, asian and black people with diabetes or hypertension had significantly higher rates of progression to ESRD compared to caucasians. In a US case series, african-caribbean people with neither diabetes nor hypertension at baseline were 3.7 times more likely to develop ESRD than caucasians (14). We know that NSAIDs reduce GFR and are associated with tubulo-interstitial inflammation and fibrosis and these medications are available without prescription. Studies have shown that even after one month of treatment with NSAIDs, there is a significant decline in creatinine clearance. Case-controlled studies have demonstrated there is a significant risk of progression to ESRD with chronic NSAID use (129). Other suggested risk factors for CKD progression include dyslipidaemia (48), persistent urinary outflow tract obstruction (130) and more recently AKI (131).

3.2 Albuminuria and Proteinuria

Proteinuria has long been recognised as an independent risk factor for CKD progression and is also an independent risk factor for cardiovascular mortality and morbidity (35). More recently urine ACR measurements have been recommended in preference to urine PCR as albumin measurements provide a more specific and sensitive measure of change in glomerular permeability than urinary protein levels

and there is substantial evidence linking increasing albuminuria to CKD outcomes. Proteinuria is used to define CKD stages 1 and 2 and has now been adopted in the classification of CKD staging not only as it is a marker of severity of disease but also as it is a powerful prognostic marker of disease progression and cardiovascular and all-cause mortality. Table 3.1 contains a summary of the key seminal studies investigating proteinuria and progression of CKD.

One of the earliest trials that demonstrated proteinuria was an independent risk factor for disease progression was the MDRD study, which grouped patients into higher and lower baseline GFR, and assigned them to either normal or low blood pressure (BP) targets (7). They found that the patients with a higher baseline proteinuria level experienced a faster rate of GFR decline and benefited from tighter BP control. More recently, Hemmelgarn *et al* showed that patients with severe proteinuria but without an overtly abnormal GFR had worse clinical outcomes than those with moderately reduced GFR and no proteinuria (132). Data from the Irbesartan Diabetic Nephropathy Trial (IDNT) also confirmed that baseline proteinuria was an important risk factor for renal failure in patients with type 2 diabetes and overt nephropathy (133). They demonstrated the cumulative incidence of ESRD was 7.7% for patients with < 1g/day proteinuria, 11.4% with 1-2 g/day proteinuria, 22.9% with 2-4 g/day proteinuria, 34.3% with 4-8 g/day proteinuria and 64.9% for those with > 8g/day. Doubling of proteinuria was also associated with doubling of risk for renal end point. Albuminuria emerged as a more sensitive marker of glomerular damage and studies demonstrated significant evidence of its associations with poor outcomes. In the PREVEND cohort study, the decline in GFR was significantly higher in people with albuminuria compared to the general population (-7.2 vs -2.3ml/min/1.73m^2, $p<0.01$) (134). Numerous studies have since identified that the severity of albuminuria is associated with adverse prognostic outcome irrespective of level of kidney function and underlying cause of CKD.

The relevance of finding a reduced eGFR level on its own in the absence of proteinuria or co-morbid factors should be interpreted differently when assessing the likelihood of progressing to ESRD or requirement of renal replacement therapy in

one's lifetime. This requires a degree of confidence in the accuracy of the prediction equations we use in clinical practice to estimate GFR. CKD progression can progress however in the absence of proteinuria and several studies have demonstrated this particularly in patients with diabetic nephropathy or advanced CKD (135-136). It is therefore important to monitor for proteinuria in assessing CKD progression but its presence does not clearly determine whether those patients will decline.

The presence of proteinuria also affects outcomes other than disease progression and this has been found at all levels of GFR. Studies have demonstrated that outcomes in those without proteinuria and GFRs of 30-59 ml/min/1.73m^2 are better than those with proteinuria and higher levels of GFR (137). A recent meta-analysis showed that the presence of an ACR of just > 10mg/g is associated with an increase in all-cause and cardiovascular mortality (138).

Studies began to focus on the renoprotective effect of angiotensin converting enzyme inhibitors (ACEI) and angiotensin II receptor blockers (ARB). Interventional studies began to investigate whether there was an anti-proteinuric beneficial effect of these drugs independent to their anti-hypertensive effect. The Heart Outcomes Prevention Evaluation (HOPE) Study investigated the impact of ACEI therapy on cardiovascular risk in patients with diabetes and found that ramipril lowered cardiovascular end points by 25% and reduced albuminuria and CKD progression (139). In 2004, the Reduction in End-points in Noninsulin dependent diabetes with the Angiontensin II Antagonist Losartan (RENAAL) study demonstrated that baseline albuminuria was almost linearly related to renal outcome and was the strongest predictor of a doubling of creatinine or progression to ESRD (140). In the Ramipril Efficacy in Nephropathy (REIN) Study, ramipril treatment prevented ESRD when used for three to four years in patients with CKD and proteinuria and this effect was independent of the effect of blood pressure changes (137). The African American Study of Kidney Disease (AASK) was the first trial to demonstrate a renoprotective effect of ACEI in african-americans and found a 50% reduction in proteinuria at six months was associated with a 72% reduction in risk for ESRD at 5 years in non-diabetic patients (141).

Table 3.1 Seminal Studies of Proteinuria and CKD Progression

Study	Cohort	Outcome Measures	Findings
Peterson *et al*, MDRD (Modification of Diet in Renal Disease) Study, 1995 (9)	Randomised interventional study N = 840 Patients with CKD, no diabetes	Normal mean arterial pressure (MAP) vs low BP goal and relationship with proteinuria	GFR decline associated with higher proteinuria levels and those with higher proteinuria levels had greatest benefit from low MAP goal
HOPE (Heart Outcomes Prevention Evaluation) Study, 2000 (130)	Randomised interventional study N = 3577 Patients with diabetes with 1 prior cardiovascular event or 1 risk factor	Cardiovascular event, cardiovascular mortality, all-cause mortality	Ramipril consistently lowered risk of combined primary outcome by 25% after adjustment for BP changes.
Hemmelgarn *et al*, Alberta Kidney Disease Network, 2010 (124)	Observational cohort study N = 1, 526 437 All people with a serum creatinine measurement	All-cause mortality, Myocardial infarction and progression to ESRD and relationship with proteinuria	Risk of outcomes and ESRD independently associated with higher proteinuria at given eGFR level
PREVEND (Prevention of Renal and Vascular End-stage Disease) study, (125)	Prospective cohort study N = 40, 856	Association with albuminuria and cardiovascular and non-cardiovascular mortality	Albuminuria was a predictor of all-cause mortality
RENAAL (Reduction in End-points in Noninsulin dependent diabetes with the Angiotenin II Antagonist Losartan) study, 2004 (131)	Double-blind randomised interventional study N =1513 Patients with type 2 diabetes with CKD	Doubling of serum creatinine or ESRD	Losartan reduced albuminuria by 25%. Baseline albuminuria was almost linearly related to renal outcome and the strongest risk predictor
REIN (Renoprotective Effect of Irbesartan in patients with Nephropathy) study, 2001 (132)	Randomised interventional study N = 1715 Hypertensive patients with type 2 diabetes	Doubling of serum creatinine, ESRD, all-cause mortality	Irbesartan reduced progression of nephropathy but sis not reduce mortality
AASK (African America Study of Kidney disease and hypertension) study, 2010 (133)	Randomised interventional study N = 1094 Black patients with hypertensive CKD	Doubling of serum creatinine, ESRD, all-cause mortality	Ramipril was better than other anti-hypertensive regimes in reducing progression of renal disease

Abbreviations: N, number; BP, blood pressure; MAP, mean arterial pressure; ESRD, end-stage renal disease; CKD, chronic kidney disease

Table 3.1 Table of the key observational and interventional studies that have demonstrated proteinuria is one of the strongest predictors of progression of CKD and associated with all-cause mortality and cardiovascular events. This association has been demonstrated in patients with and without diabetes and in different ethnic groups (7, 132, 134, 137, 139-141).

In a meta-analysis of randomized controlled trials of patients with CKD stages 1–3, several treatments involving ACEIs or ARBs reduced the risk of clinical outcomes, but the benefits appeared to be limited to specific CKD subgroups, some of which already had a clinical indication for the treatment studied (142). When compared to placebo, ACEIs and ARBs reduced the risk of ESRD overall, but this benefit appeared to be present only among patients with overt albuminuria, most of whom had diabetes and hypertension. In patients with CKD stages 1–3 with only albuminuria or impaired eGFR, ACEIs did not reduce the risk for ESRD when compared with a placebo. ESRD was also not significantly reduced in patients with CKD stages 1–3 who did not have proteinuria. Meta-analyses of these studies also found that taking an ACEI or an ARB did not reduce the risk of mortality, except when an ACEI was used for patients with microalbuminuria and cardiovascular disease or diabetes and other cardiovascular risk factors (142).

CKD guidelines suggest that patients with proteinuria, diabetes and hypertension may benefit from ACEI or ARB treatment and patients who have albuminuria and are at high risk for cardiovascular complications may benefit from ACEI treatment at adequate doses (70). In 2003, a randomised controlled trial of 336 patients with non-diabetic renal disease (Combination treatment of ARB and ACEI in non-diabetic renal disease study (COOPERATE)) published data that dual blockade of the renin-angiotensin system with an ACEI and ARB reduced the risk of primary end-points (143). This study had to be later withdrawn due to scientific misconduct. Since then a further study looking at renal outcomes with Telmisartan, Ramipril or both in people at high vascular risk (ONTARGET) data has argued against the benefit of dual RAS blockade and there now little use of combined ACEI and ARB therapy and there remains a distinct safety issue with regard to hyperkalaemia and elevated creatinine in dual use (144). More recently there have been concerns that use of ACEI and ARB in advanced CKD stages precipitates a more rapid decline to ESRD and we await data from the on-going STOP-ACE study which started recruiting in 2014.

A graded relationship seems to exist between the severity of proteinuria or albuminuria and adverse health outcomes, including mortality, ESRD and

cardiovascular disease. The 2008 NICE CKD guideline defined significant albuminuria as an ACR of 30mg/mmol or higher, equivalent to a urine PCR of 50 mg/mmol or higher (53). The risk for adverse outcomes conferred by reduced GFR and increased albuminuria (or proteinuria) appears to be independent and multiplicative.

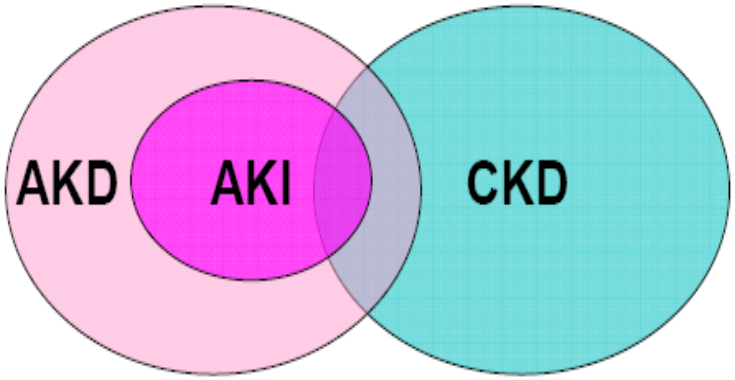
3.3 The Relationship of AKI and CKD

There is emerging evidence that there is a considerable overlap between the relationship between CKD, AKD and AKI (Figure 3.1). Both AKI and AKD can lead to CKD and CKD is a risk factor for AKI and AKD.

In order to diagnose AKI there must be an increase in serum creatinine over a period of two days but this definition may not capture all episodes of acute injury to the kidneys hence a new clinical approach to categorising these patient according to functional and structural criteria was proposed by the Kidney Disease: Improving Global Outcomes (KDIGO) AKI Work Group (70). In the new KDIGO AKI guidelines, AKI is defined as a syndrome, which includes direct injury to the kidney as well as acute impairment of kidney function. They introduced the term Acute Kidney Diseases and Disorders (AKD) as AKI is one of a number of conditions that acutely affects the kidney structure and function. AKD encompasses AKI and any event where GFR falls <60 ml/min/1.73m^2 for < 3 months or by ≥ 35% or where serum creatinine rises by > 50% for < 3 months. No known kidney disease (NKD) was also defined as a category and indicates no functional or structural renal abnormality using the criteria according to the definitions of AKI, AKD or CKD (Table 3.2).

Figure 3.1

Relationship Between Acute Kidney Injury, Acute Kidney Disease and Chronic Kidney Disease



Abbreviations: AKD, Acute Kidney Disease; AKI, Acute Kidney Injury; CKD, Chronic Kidney Disease

Figure 3.1

This model describes the likely considerable overlap between AKI, CKD and AKD. AKI is a subset of AKD. AKI and AKD can occur in patients with CKD. Adapted from Kidney Disease: Improving Global Outcomes (KDIGO) clinical practice guidelines for Acute Kidney Injury, 2012 (70).

Table 3.2 Definitions of Acute and Chronic Kidney Disorders

	Functional Criteria	**Structural Criteria**
AKI	Increase in sCr by 50% within 7d or Increase in sCr by 0.3 mg/dl within 2 days or Oliguria	No criteria
CKD	GFR < 60 for > 3 months	Damage for > 3 months
AKD	AKI or GFR <60 for < 3 months or Decrease in GFR by ≥ 35% or increase in sCr by > 50% for < 3 months	Kidney damage for < 3 months
NKD	GFR ≥ 60 Stable sCr	No damage

Abbreviations: AKI, acute kidney injury; CKD, chronic kidney disease, AKD, acute kidney diseases and disorders; NKD, no known kidney disease; GFR, glomerular filtration rate; sCr, serum creatinine.

Table 3.2

This table describes the functional and structural criteria for the acute and chronic kidney disorders: AKI, CKD, AKD and NKD. It incorporates GFR, serum creatinine, urine output changes and structural abnormalities as criteria. This was adapted from the KDIGO AKI Algorithm (70).

Both AKI and CKD represent a reduction in kidney function and both disorders can lead to ESRD and are associated with increased mortality (145). Both conditions share similar risk factors and studies have shown that CKD is an established risk factor for developing AKI and hospitalised episodes of AKI have been associated with worsening of pre-existing CKD but the extent to which these two conditions are related is not clearly understood (146). Although AKI is potentially reversible, several studies have shown that non-recovery of AKI leads to ESRD and prolonged hospitalised episodes of AKI can often lead to progression of CKD. It has been suggested that AKI may the causative factor of how patients who have CKD of undetermined aetiology may develop it in the first place – 'de novo' CKD.

CKD as a Risk Factor for AKI

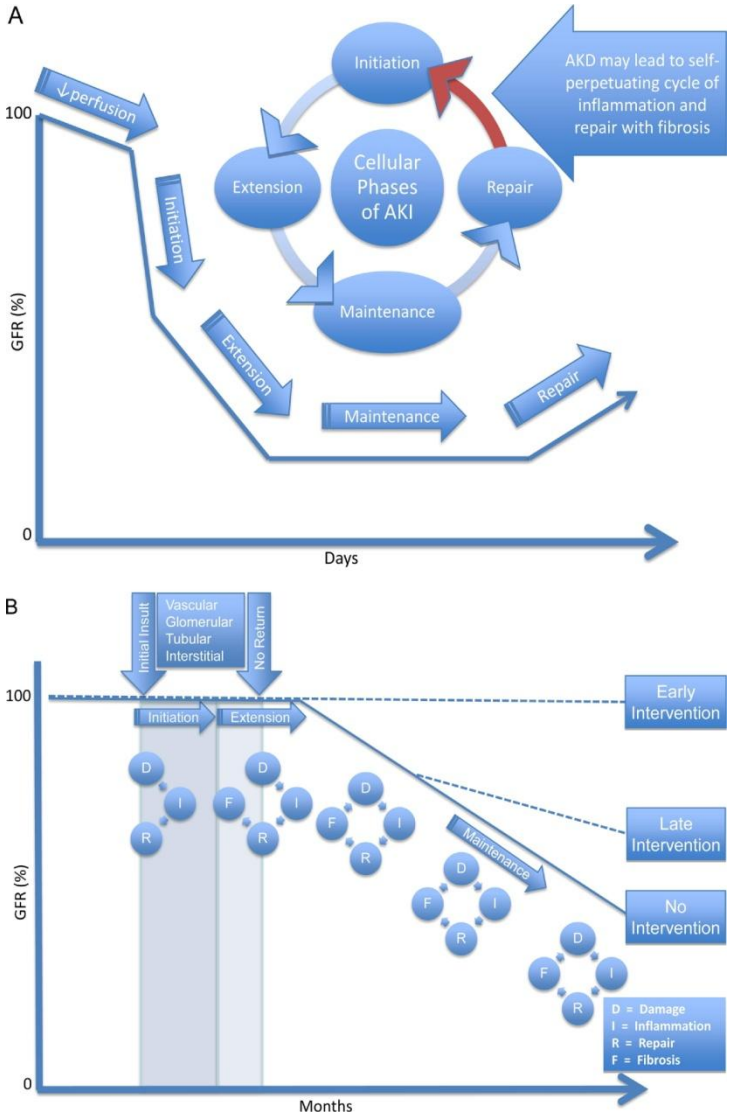
What makes the determination of the epidemiology of AKI and CKD and understanding of their relationship more difficult is the variation in definitions used and the different populations studied. In epidemiological studies of hospitalised AKI, CKD is found to be not only a risk factor but is a significant risk factor in the development of AKI (147-148). This seems to hold true when a correction is made for other confounding factors, such as the other co-morbidities associated with CKD. This has been consistently observed in the settings of AKI following radio-contrast administration, cardiac surgery and sepsis (149). Interestingly, some studies demonstrate lower in-hospital mortality rates in patients who develop AKI on a background of CKD compared with patients without background CKD (149). One explanation would be that patients with CKD require less of an insult to manifest as an AKI and are hence 'less sick' compared to those without CKD. It has also been suggested that these results may be confounded by malnutrition and lower serum creatinine values from low muscle mass. Another explanation is many patients without CKD experiencing 'discrete' AKI in the community, which do not lead to a hospital admission, will not be captured in most studies, and hence only the more seriously ill patients will be included in outcome data, impacting on mortality statistics.

The Effects of AKI on CKD Progression

There is mounting evidence that AKI contributes significantly to both CKD, ESRD and importantly leads to the progression of CKD. In terms of outcome, observational studies demonstrate that development of AKI on a background of CKD, leads to ESRD at a greater rate than AKI without a background of CKD (79). The mechanisms are unclear but it is hypothesised that this occurs either through ischaemic injury or hyperfiltration following sclerosis and loss of functioning nephrons. The theory is that a diseased kidney with reduced function and functional reserve has increased susceptibility to further injury. This theory is supported by previous experimental studies that demonstrated a compromise in structural recovery and worsening tubule-interstitial fibrosis after ischaemic injury following unilateral nephrectomy (150). There is a large amount of work studying the relationship between reduced renal mass, hypertension and hyperfiltration of functioning remaining nephrons and progressive interstitial fibrosis. The theory suggests that in a person with CKD or low functioning renal mass, an initial insult leads to inflammation and cell injury then repair (Initiation). The cell damage is further extended by renal vascular endothelial injury and dysfunction (Extension) and endothelial repair is vital for cell recovery. This can lead to fibrosis and further cell damage in a continuous cycle (Maintenance) leading to progressive linear decline. (Figure 3.2)

Okusa *et al* (151) using pathophysiological concepts from Sutton *et al* (150), suggested that following an episode of AKI, there appear to be four possible outcomes: (1) full recovery, (2) incomplete recovery resulting in CKD, (3) worsening of pre-existing CKD accelerating progression to ESRD, and (4) non-recovery of kidney function leading to ESRD. These outcomes are illustrated in Figure 3.3.

Figure 3.2 The Clinical Natural History of Acute Kidney Injury



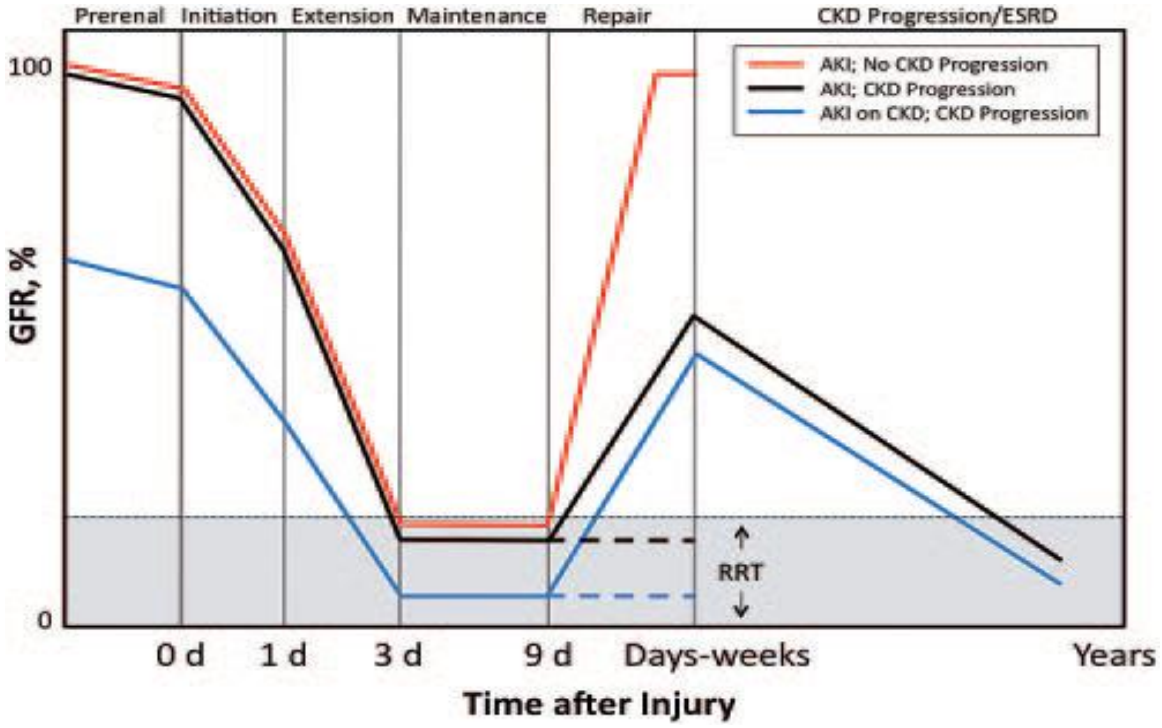
Abbreviations: GFR, glomerular filtration rate; AKI, acute kidney injury; AKD, acute kidney diseases or disorders.

Figure 3.2

This model illustrates the pathophysiological processes involved of the effect AKI has on cellular damage (A) and renal function (B). In patients with pre-existing chronic kidney damage, the initial insult of AKI may lead to inflammation and repair (Initiation) resulting in fibrosis (Extension) then further damage which can lead to a never-ending cycle of progression (Maintenance) eventually leading to end-stage disease. Intervening early at the Initiation phase may prevent the development or progression of CKD whereas later interventions may only delay progression.

Bedford *et al*, 2013 (152)

Figure 3.3 The Effect of Acute Kidney Injury on Chronic Kidney Disease Progression



Abbreviations: GFR, glomerular filtration rate; AKI, acute kidney injury; CKD, chronic kidney disease, RRT, renal replacement therapy; d, day, ESRD, end-stage renal disease.

Figure 3.3

This illustrates the time frame of the pathophysiological mechanisms in AKI. Initiation occurs when the initial insult leads to cellular damage and repair. This leads to extension of cellular damage through endothelial ischaemic injury (Extension) and repair with ongoing inflammation and fibrosis (Maintenance). The graph presents the different renal outcomes following the repair phase ranging from full recovery to ESRD.

Okusa *et al*, 2009 (151).

AKI in the Community

Most studies have observed AKI presenting to or occurring during hospitalisation however little is known about the incidence and outcomes of patients who have an acutely elevated creatinine in the primary care setting where patients are not admitted to hospital. A large proportion of AKI occurs in the community and many episodes may remain undetected and there are concerns that this is a previously unrecognised health issue. Studies have shown that up to 60% of hospital-based AKI is community acquired (62). Minor episodes of AKI often occur in the community however there is often a delay in its identification and hence in establishing appropriate management of this condition. This may be partly due to the lack of awareness of the deleterious effects that minor episodes of AKI may contribute to. There is a huge paucity of data regarding the prevalence of community-based AKI as most AKI data relates to in-patient studies.

Could it be that these episodes of AKI, which not associated with acute illness or hospital admission, are not appreciated to have occurred as renal function is either not tested or not properly assimilated? Are they contributing to the development and or progression of CKD? The effect of 'discrete' episodes of AKI in the community on the progression of CKD is at present unknown, but may prove to be an important factor. The literature clearly documents that there are people with CKD who do not progress and never develop ESRD, but there are also people who do progress. In the population with progressive CKD, it may also be that 'discrete' episodes of AKI in the community contribute to this progression.

These episodes of AKI in the community need to be captured. Even small isolated increases in serum creatinine have an associated increase in short-term morbidity and mortality and in longer-term outcomes including 1 year mortality in hospital-AKI. Does this risk extend to AKI managed by primary care? These 'discrete' episodes of AKI in the community remain to be well defined but may possibly relate to CKD progression therefore require further investigation as intervention in this group may have a significant beneficial effect on outcomes.

CKD is highly prevalent yet few people progress to ESRD suggesting that a large number of patients with CKD have stable disease. So how do they develop CKD initially and what other factors influence CKD progression aside from the recognised risk factors of proteinuria and cause of renal disease. There has been significant drive to improve recognition, prevention and management of AKI with emerging evidence of the strong overlap between CKD and AKI. Hospital-based AKI events have been suggested to contribute to CKD progression. Many episodes of AKI are not hospitalised but little is known about their outcomes on long term renal outcome. This study aims to study the patterns of progression and assess if these AKI events are associated with CKD progression.

CHAPTER 4

Validating Estimating GFR Equations in the Elderly

With an ageing population and the increased prevalence of impaired renal function in the elderly, the number of people with CKD is predicted to rise dramatically in the near future. Diagnosis and screening for CKD in at risk populations requires a quick, accessible and relatively cheap test which will provide an accurate measure of renal function. Estimating equations based on serum creatinine have been developed to help provide the physician an easy way of calculating kidney function with a reasonable degree of accuracy. Current CKD guidelines recommend that diagnosis, staging and progression of CKD are based on these estimating GFR equations and in the adult population the MDRD study and CKD-EPI equation are widely applied (12).

Epidemiological studies using the MDRD study equation suggests an overall non-institutionalised population burden of all stages (1-5) of CKD of approximately 11%, but this figure increases in older people with > 30% of the population > 80 years old with an eGFR < 60 ml/min/1.73m^2 (2). The MDRD study equation however loses accuracy in selected subgroups such as those with GFR > 60 ml/min/m^2, kidney transplant recipients and the elderly (31, 153).

The MDRD and CKD-EPI study equations were developed in a population with a very small number of elderly patients. The MDRD study equation originally was validated in 1,085 patients with CKD with a mean age of 51 ± 13 years with 22% older than 65 years (8). The authors subsequently developed the equation in a larger and more diverse population of 5,504 patients but again few people were older than 75 years (mean age 47 ± 15 years) and only 13% were > 65 years (31). In the CKD-EPI development cohort less than 1% was over the age of 75 years.

As both equations have not been validated in older people, there have been concerns that they do not reflect true GFR in older people.

4.1 Hypothesis

My first hypothesis is that the methods used for estimating GFR are inaccurate in assessing renal function in the elderly population who have the highest burden of CKD. I hypothesise that the estimating equations (the MDRD study equation and the CKD-EPI equation) underestimate GFR in older people particularly at higher GFR levels with consequential misdiagnosis and misclassification of CKD leading to a falsely high prevalence of CKD.

4.2 Aim

The aim of this first study is to assess if the estimating equations we use to assess renal function accurately reflect true GFR in older people. The MDRD study and CKD-EPI equations have both been assessed in light of the recommendations by the recent NICE CKD guidelines. The aim of this study was to assess the accuracy of the MDRD Study and CKD-EPI equations for estimating GFR compared to an iohexol reference GFR measurement in an elderly population. The null hypothesis was that both the MDRD and CKD-EPI equations perform accurately in terms of bias, precision and accuracy when compared to the measured GFR in an elderly population. The primary research question is 'do older patients identified as having CKD stage 3-5 really have GFRs below 60 ml/min/1.73m^2?'

4.3 Methods

Recruitment

This study took place in East Kent, a semi-rural area of South East England, with an estimated population of just over half a million adults with a high prevalence of elderly caucasians. Subjects were recruited and took part in the study from January 2008 to April 2011. Participants selected for this study were either patients known to the EKHUFT Department of Renal Medicine or residents of the local population. Advertisements were placed in the local outpatient departments and patients were approached in general nephrology clinics. The local population were recruited via a

variety of means including presentations at discussion groups in Age Concern centres, golf clubs, rotary clubs and residential care homes and through advertising the study via media briefings in hospital newsletters, local newspapers and radio stations. Overall 38% of participants were recruited via nephrology clinics and 62% via other methods. In line with the NKF-KDOQI 2002 guideline, a minimum of 100 adults were required to validate the study (10). A power calculation was also performed to assess the minimum number of patients needed to demonstrate a difference between median eGFR and median iohexol GFR where the true difference was a minimum of 2.0 ml/min/1.73m^2. I chose the conventional probabilities of < 0.05 significance to avoid a type 1 error and a power level of 0.8 to avoid a type 2 error and the number of patients required was 171 (154).

All subjects gave informed consent and the study had full ethical approval (East Kent REC number: 07/Q1803/37).

Cohort Description

All elderly patients aged 74 years or over were invited to participate in the study. Initially the inclusion criteria specified recruits aged 80 years and over, however due to a high initial refusal rate, recruitment was extended to individuals aged ≥ 74 years. Exclusion criteria included the following:

- Any history of untoward reactions to iodinated contrast media or allergy to shellfish
- Any known current active malignancy
- A predicted life expectancy of less than 3 months
- An inability to consent due to cognitive impairment or lack of capacity
- Any recent episode of acute kidney injury within 3 months of recruitment
- Patients receiving renal dialysis treatment
- Recipient of a functioning or non-functioning renal transplant graft.
- Hospitalisation at the time of the test

Patients who were unwell on the day of the test were invited to participate at a later date.

Initial Assessment

All patients were either assessed in their residential homes or invited to attend the hospital in the morning having avoided any meat consumption on the day of the test. Demographic data including gender, age and race were recorded and co-morbidity data and current prescription lists were documented with particular note of medications such as NSAIDs and those affecting the RAAS system and renal tubular secretion of creatinine. Blood pressure (mmHg), weight (kg) and height (m) of the individuals were measured prior to the procedure for body surface area (BSA) and body mass index (BMI) calculations.

Co-morbidity history was grouped into the following: hypertension, diabetes (type 1 and 2), vascular disease and history of malignancy. Vascular disease was considered present if there was a history of myocardial infarction, angina, cardiac arrhythmia, valvular disease, congestive cardiac failure or a requirement for coronary intervention (e.g. angioplasty, coronary artery bypass grafting or pacemaker/cardiac defibrillator insertion), cerebrovascular disease or peripheral vascular disease.

A morning random urine sample was collected for total protein, albumin and creatinine measurement. Presence of albuminuria was defined as an ACR ≥ 30mg/g and heavy albuminuria was recorded as a urine ACR > 300mg/g.

GFR Measurement

The reference GFR was measured using the 4-sample iohexol plasma clearance method (118). Iohexol was selected as the reference GFR in this study and not iothalamate clearance which was used in the MDRD and CKD-EPI development dataset as iohexol clearance measurements are available for our local paediatric population. Estimated GFR was calculated from serum creatinine using the MDRD study equation and the CKD-EPI equation.

Sampling

Blood samples were taken at time zero for baseline serum creatinine measurement and for iohexol measurement prior to administration for detection of any

background signal from the time 0 sample. A 5 ml bolus of Omnipaque 240 (518 g/L iohexol corresponding to 240 g/L of iodine, GE Healthcare www.gelifesciences.com) was slowly injected intravenously into the antecubital vein at time zero followed by a 10 ml normal saline flush either via a 21G cannula or butterfly needle. Patients were monitored for any haemodynamic instability or allergic symptoms. A blood sample was taken at 5 minutes from the opposite infusion arm to confirm that the iohexol had been administered intravenously without extravasation of contrast. The time 0 and 5 minute serum samples were also used to assess the analytical variability of the iohexol and creatinine assays. Extravasation of a large quantity of the administered contrast into subcutaneous tissues would affect the re-distribution phase of iohexol thus influencing measured iohexol values. Further blood samples were collected from the opposite arm to the infusion at approximately 120, 180 and 240 minutes after injection as recommended by the Brochner-Mortenson method as described in chapter 2 (106). Earlier samples were not taken due to the inaccuracies of interpreting iohexol levels during the redistribution phase. This protocol was adapted from the paediatric clinical protocol used by the Evelina's Children Hospital, Guys and St Thomas' NHS Foundation Trust. Exact timing of the sampling in relation to the bolus injection was accurately recorded. Haemolysed serum samples were discarded and repeat samples were taken.

All blood samples were collected in lithium heparin tubes, mixed, and then centrifuged at the end of each study at 3400 rpm for 4 min. Plasma was then extracted and stored at -80^{o}C prior to analysis. All the above sampling, extraction and storage were carried out by myself.

Analytical methods

Creatinine and Iohexol Measurement

All samples were transported on ice to the Wellchild Laboratories, London for the GFR measurement. Creatinine and iohexol measurements were performed by the accredited Wellchild clinical laboratory staff. Plasma creatinine and iohexol were measured simultaneously using a modified stable isotope dilution electrospray tandem mass spectrometric method reported for creatinine (155). The only

modifications were the inclusion of both 25 µmol/L $^{5}H_3$-iohexol and $^{2}H_3$-creatinine in the stable isotope reagent, addition of the precursor/product ion pairs (m/z 821.9/602.9, 826.9/607.9) for iohexol, and, because of the higher sensitivity of the Applied Biosystems SCIEX API5000 (Applied Biosystems, www.appliedbiosystems.com) instrument used, the precursor/product ion pairs (m/z 114.0/85.9, 117.0/88.9) were utilised for creatinine.

The NKF-KDOQI initiative to improve the performance of estimated GFR calculations was after recalibration of serum creatinine measurements to an IDMS method set a total error goal for creatinine measurements to a maximum 10% error in eGFR. The guidelines recommend that laboratories need to aim for an analytical bias of < 5% and analytical imprecision of < 8% at serum creatinine concentrations of > 88 µmol/l (10).

In order to assess whether analytical assay impression influenced these results, between assay imprecision for the plasma iohexol assay was assessed at three concentrations, covering the iohexol range of 10-400 µmol/L using timed samples taken at 5 minutes and 240 minutes. Plasma iohexol assay imprecision was <7% overall. Accuracy of the plasma creatinine assay was assessed using National Institute of Standardisation and Technologies (NIST) Standardised Reference Materials (SRM) 967 I and II in each assay as per national creatinine standardisation protocol (102). The accuracy of the serum creatinine assay was assessed in 31 samples: using SRM 967 I, the mean serum creatinine was 65.4 µmol/ml, and using SRM 967 II, the mean serum creatinine was 332.4 µmol/ml. The between-assay imprecision for serum creatinine was 4.3% and 3.8% respectively which is within the recommended target set by national clinical laboratory recommendations (10).

Iohexol GFR was calculated using a single compartment model,

GFR (ml/min) = 0.693 x iohexol volume of distribution (L) x 1000/half-life of iohexol (min)

The GFR (ml/min) was corrected for body surface area and the Brochner-Mortensen correction applied (106).

Urinary creatinine was measured using an enzymatic assay. Urinary albumin was measured using an immunoturbidimetric assays and albumin concentration was expressed relative to urinary creatinine concentration. Urinary assays were undertaken using an Abbott Architect analyser (Abbott Diagnostics Ltd, www.international.abbottdiagnostics.com) within 24 hr of sample collection.

All urine analyses were undertaken by accredited laboratory scientists registered with the Health Professions Council at EKHUFT.

Estimated GFR calculations

GFR was estimated using the simplified isotope dilution mass-spectrometric (ID-MS) traceable version of the MDRD study equation and the CKD-EPI equations. The two estimating equation formulae are shown in Table 2.1.

Estimating Equation Performance Measures

Iohexol GFR was accepted as the reference measure of GFR against which estimated GFR was compared. Performance of both the MDRD and CKD-EPI equations were assessed. Scatter plots comparing estimated and measured GFR were drawn for both estimating equations.

The KDOQI clinical practice guidelines for CKD recommend that when evaluating the accuracy of an estimating equation, one should consider bias, precision and accuracy (10).

Bias was measured as the median difference subtracting the measured GFR from the estimated GFR with positive values indicating higher estimated GFR than measured GFR (overestimation).

Precision was expressed as the interquartile range (IQR) for the differences.

Accuracy of the equations was expressed by the percentage of estimates within 30% above or below the measured GFR (P_{30}) and this has commonly been used as the benchmark for evaluation of estimating GFR equations in clinical practice.

Measured GFR and estimated GFR were compared for each patient graphically by plotting iohexol GFR and the difference (estimated GFR minus measured GFR) against estimated GFR (Difference Plots). These graphs depict the bias and variability of the estimating equations.

Secondary analysis was performed comparing individuals with GFR of < 60 and ≥ 60 ml/min/1.73 m^2 and individuals < 80 years and ≥ 80 years old.

Statistical Analysis

Bias precision and accuracy were measured to determine the performance of each equation. Measured GFR and estimated GFR datasets were checked to see if they were normally distributed using the Shapiro-Wilk test. Both were found to be not normally distributed ($p<0.001$, Shapiro-Wilk test). All data was therefore log transformed and non-parametric statistics were used throughout.

The statistical tests performed in this study were similar to the analytical methods originally used in the MDRD and CKD-EPI study design to ensure comparable results. Bootstrapping methods were used to calculate confidence intervals for bias and accuracy and again were employed in the original MDRD and CKD-EPI studies (156). Bootstrapping enables more accurate estimates of the population distribution by using the information based on a number of re-samples from the original sample. The 95% confidence intervals were calculated using 2000 bootstraps i.e. the sample results were re-analysed with 2000 randomly selected replacements of samples to give a more accurate representation of the cohort studied.

The Wilcoxon test is a nonparametric test that compares two paired groups and was used to compare the bias of each of the MDRD and CKD-EPI estimated GFR against

measured GFR as in the original MDRD and CKD-EPI studies (8, 9). A p value of < 0.05 was considered statistically significant.

The McNemar's Chi-squared test is a nonparametric test used to assess the significance of the difference between 2 correlated proportions which are based on the same sample of subjects. The Mcnemar's test was used to compare P_{30} values of the MDRD and CKD-EPI equations in comparison and was used in the CKD-EPI study when comparing performance against the MDRD study equation. A P value of <0.05 was considered statistically significant.

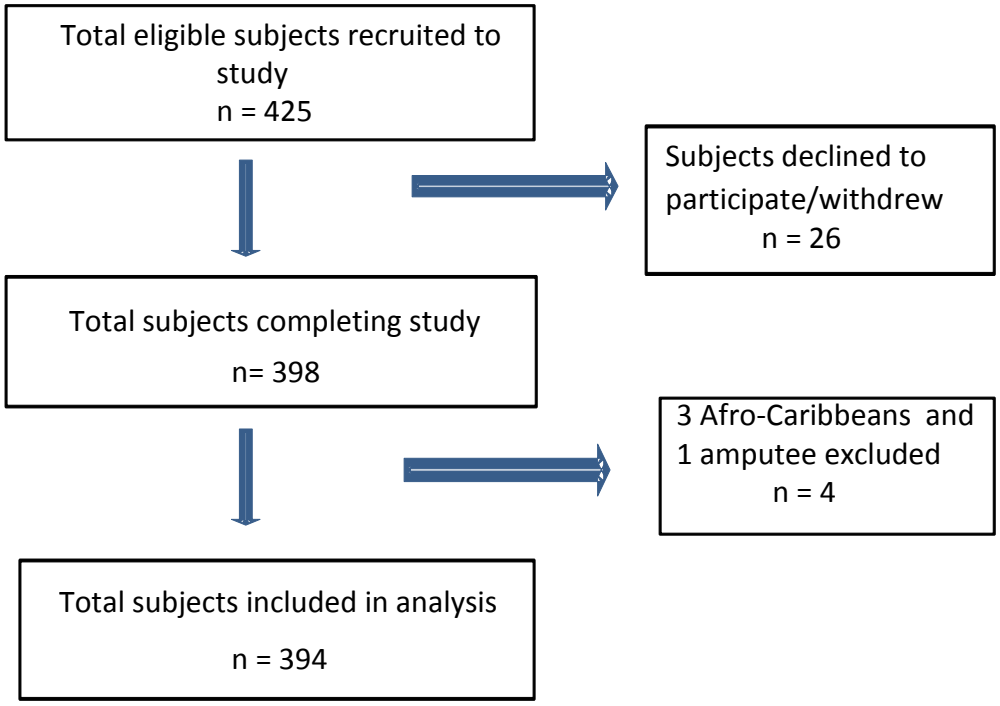
Percentage misclassification around the GFR threshold of 60 ml/min/1.73 m^2 was also assessed for each equation and percentage proportions were compared. Statistical analyses were not performed on the misclassification errors.
All the statistical analyses were performed using 2 computer programs: Analyse-it™ (Analyse-it™ Software Ltd, www.analyse-it.com) and Stata version 12.0 (StataCorp LP, www.stata.com).

4.4 Results

Study Cohort

A total of 398 subjects participated in the study. Initially 425 subjects volunteered however 26 individuals subsequently withdrew for reasons including inter-current illness at the time of the scheduled test, following discussion with family members and inability to provide alternative care arrangements for their dependents. One study participant withdrew from the study after suffering a vasovagal episode on initial cannulation and it was subsequently discovered he had haemophobia. Three

Figure 4.1 Accuracy of Estimating GFR Equations Study Selection Process



Abbreviations: n, number.

Figure 4.1

Flow chart demonstrating the selection process of the study. Of a total 425 patients who initially volunteered, 26 withdrew and of the 398 subjects completing the study, 3 were excluded due to being of african-caribbean ethnicity and 1 was an amputee. A total of 394 subjects were examined.

individuals of african-caribbean ethnicity completed the study but were excluded from the final analyses. These exclusions were made after the study was completed on the basis that, as a subgroup they were too small to provide meaningful data as we know that the estimating equations vary significantly in african-caribbeans. One further subject was excluded as they were an amputee on the basis that the difference in total body muscle mass may skew GFR calculation. The final study cohort therefore consisted of 394 caucasian individuals and the study selection process is illustrated in Figure 4.1. Ten individuals lived in supported accommodation and completed the study at their residential care homes and the remaining participants were free-living and attended the hospital.

There were no reported adverse or allergic effects from the iohexol administration and no cases had to be excluded due to extravasation of iohexol. There was one recorded case of death in the study cohort 3 months subsequent to the test but the participant died from an unrelated cause (the cause of death was exacerbation of chronic obstructive airways disease).

Patient Characteristics

The characteristics of the study patients are summarised in Table 4.1. The median age of the study group was 80 years (range 74-97 years). There were of 193 subjects aged less than 80 years and 201 subjects aged 80 years or over of which 18 were over 90 years. There were similar numbers of males and female with a male to female ratio of 48% to 52% with a similar representation in the < 80 and ≥ 80 year subgroups.

The median body mass index of the cohort population was 26.1 kg/m^2 (range 13.7 - 47.6 kg/m^2). Median body surface area was 1.87 m^2 (range 1.24 – 2.59 m^2). Height, weight, BMI and BSA were similar in the < 80 and ≥ 80 year sub-groups.

Of the 374 (95%) with documented blood pressure measurements, the median BP was 140/74 mmHg (range 42-115 mmHg diastolic pressure and 76-203mmHg systolic pressure) and acceptable in this age group. Prescription data reflected the high incidence of complex poly-pharmacy typically associated with the elderly population

predominantly treating hypertension and vascular disease and is illustrated in Figure 4.2. A large proportion of the study population were on regular prescription medications known to reduce GFR: renin-angiotensin system blockers (39%), diuretics (37%) and non-steroidal anti-inflammatory drugs (9%).

There was a high prevalence of co-morbidities as expected in this population; (Figure 4.3) 55% had hypertension, 44% had vascular disease (including cerebrovascular disease, peripheral vascular disease and ischaemic heart disease) and 19% had diabetes. A total of 13% had survived and recovered from a previous episode of malignancy.

Albuminuria data was available in 368 subjects due to failure to obtain clean catch urinary samples in 26 individuals (Table 4.1). There was a high prevalence of albuminuria with 38% of the study cohort with a urine ACR ≥ 30mg/g and 8% with a urine ACR > 300mg/g. There was a higher prevalence of significant albuminuria (ACR ≥ 30mg/g) in the ≥ 80 year subgroup compared to the < 80 year subgroup.

Table 4.1 Characteristics of the Entire Study Population Subdivided by Age <80 Years or ≥ 80 Years

	Entire cohort	<80 y	≥80 y
N	394	193	201
Median age, years (range)	80 (74-97)	-	-
74-79, n (%)	193 (49)	-	-
80-84, n (%)	132 (34)	-	-
85-89, n (%)	51 (13)	-	-
≥90, n (%)	18 (5)	-	-
Male, n (%)	189 (48)	90 (47)	99 (49)
Median height, m (range)	1.67 (1.24–1.94)	1.67 (1.43–1.94)	1.67 (1.24–1.85)
Median weight, kg (range)	74 (32-126)	78 (46–126)	71 (32–109)
Median body surface area, m^2 (range)	1.87 (1.14–2.59)	1.92 (1.35–2.59)	1.82 (1.14-2.38)
Median body mass index, kg/m^2 (range)	26.1 (13.7-47.6)	27.2 (18.9-47.6)	25.5 (13.7-36.7)
[a]Median systolic blood pressure, mm Hg (range)	140 (78-203), n=374	140 (95-203), n=187	141 (78-198), n=187
[a]Median diastolic blood pressure, mm Hg (range)	74 (42-115), n=374	75 (50-115), n=187	74 (42-110), n=187
[b]Albuminuria data available, n (%)			
Urine ACR ≥30 mg/g	110 (30)	49 (28)	61 (32)
Urine ACR >300 mg/g	31 (8)	10 (6)	21 (11)

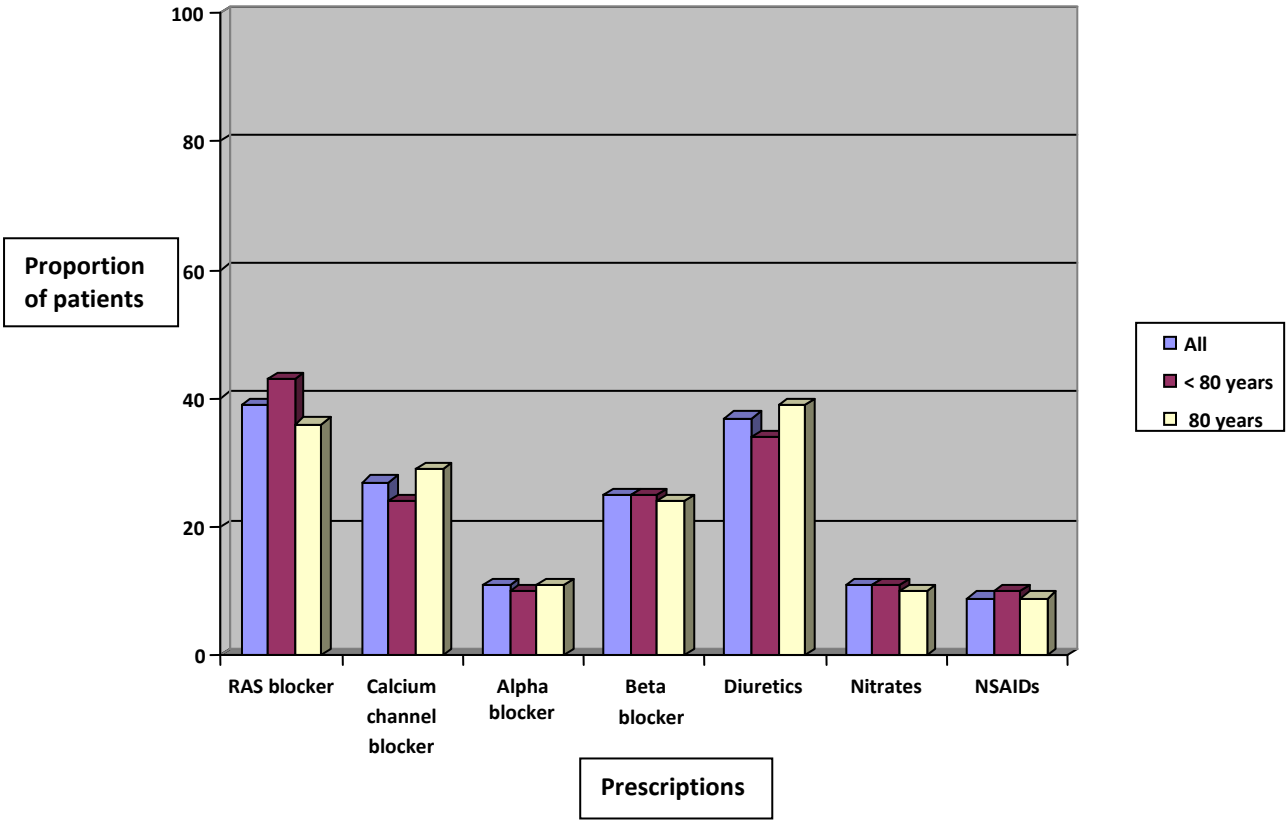
Abbreviations: n, number; y, year; ACR, albumin creatinine ratio

[a]Blood pressure data available for 374 (187 individuals <80 years and 187 ≥80 years).
[b]Albuminuria data available for 368 (177 individuals <80 years and 191 ≥ 80 years).

Table 4.1

Patient demographics of total study cohort and divided by age < 80 and ≥ 80 years. The median age was 80 years with an age range between 74-97 years. There was an equal distribution of gender among the subgroups. Although the median body surface of the population was 1.87 m^2 there was a wide range in body surface area between 1.14 – 2.59 m^2 and this was observed in the 2 age subgroups. Significant albuminuria (ACR ≥ 30mg/g) was present in 38% of patients overall. There were no significant differences in characteristics between < 80 and ≥ 80 year groups.

Figure 4.2 Prescription Patterns of the Entire Cohort Subdivided by Age < 80 Years or ≥ 80 Years

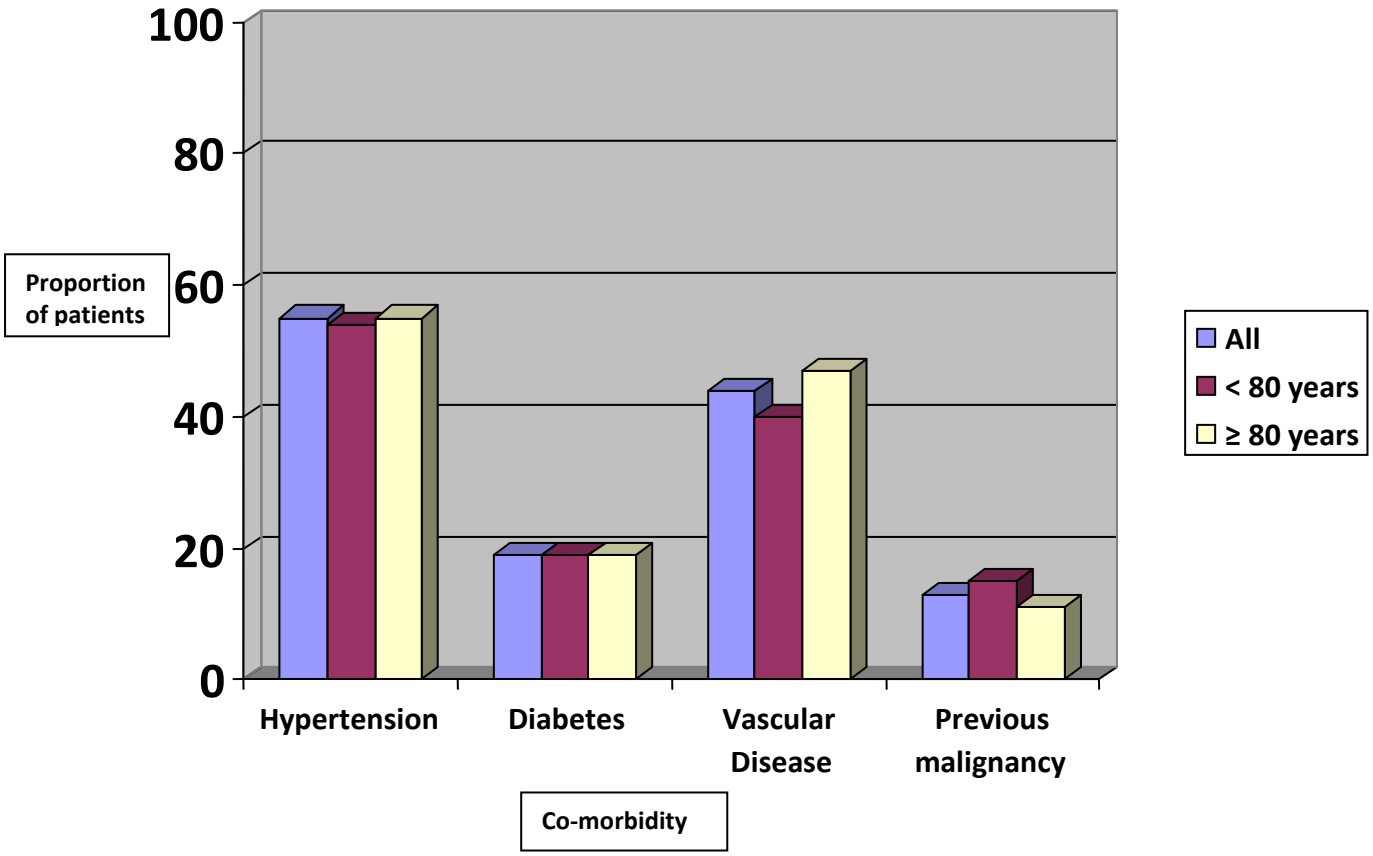


Abbreviations: n, number; RAS, Renin angiotensin system; NSAIDs, Non-steroidal anti-inflammatory drug.

Figure 4.2

This bar chart demonstrates the percentage proportion of study patients on particular prescription medicines. This illustrates the typical prescription patterns seen in elderly patients with 39% of the study cohort on RAS blockers, 37% on diuretics and 9% prescribed NSAIDs. Prescription patterns were similar in subgroups < 80 and ≥ 80 years.

Figure 4.3 Prevalence of Co-morbidities of the Entire Cohort Subdivided by Age < 80 Years or ≥ 80 Years



Abbreviations: n, number

Figure 4.3

This bar chart demonstrates the percentage proportion of study patients with certain co-morbidities and reflects the high co-morbidity burden of these elderly patients. Vascular disease was considered if there was a history of myocardial infarction, angina, arrhythmia, valvular disease, congestive cardiac failure, requirement for coronary intervention (angioplasty, coronary artery bypass graft, or pacemaker), or cerebrovascular or peripheral vascular disease. Co-morbid conditions were present in similar proportions among the < 80 and ≥ 80 year groups.

Level of Renal Function

Stratification of the cohort by level of renal function was based on the iohexol GFR results. The median measured GFR of the total cohort was 53.4ml/min/1.73 m^2 however the range of measured GFRs observed was 7.2 - 100.9ml/min/1.73 m^2 representing a variable spread of level of renal function. Median measured GFR was significantly lower in the ≥ 80 year group with a median GFR of 46.9 ml/min/1.73 m^2 (range 15 – 100.9 ml/min/1.73 m^2) compared to 60.6 ml/min/1.73 m^2 (range 7.2 – 96.3 ml/min/1.73 m^2) in the < 80 year group.

Table 4.2 shows subjects split between GFR categories. A wide range of GFRs was desired to establish an accurate assessment of the estimating equations' overall performances across a range of kidney function. Of the total study cohort, 40% had a GFR > 60ml/min/1.73m^2, 43% had a GFR between 30-60 ml/min/1.73m^2 (Stage 3a and 3b CKD) and 16% had a GFR below 30 ml/min/1.73m^2 (Stage 4-5 CKD). Only 1.5% had a GFR > 90 ml/min/1.73 m^2. There was almost double the proportion of patients in with CKD stage 3b-5 (i.e. GFR < 45 ml/min/1.73m^2) observed in the ≥ 80 year subgroup compared to those < 80 years of age.

Performance of Estimating Equations

Performance of both the MDRD and CKD-EPI equations were assessed compared to the measured (iohexol) GFR using bias (mean difference between estimated and measured GFR), precision (IQR of the difference) and accuracy (percentage of estimates within 30% of measured GFR, P_{30}).

A higher median estimated GFR was observed using the MDRD study and CKD-EPI estimating equations compared to measure GFR and this was also seen in both age subgroups. The median MDRD eGFR was 57.6 ml/min/1.73 m^2 (range 13.3-156 ml/min/1.73 m^2) and median CKD-EPI eGFR was 57 ml/min/1.73 m^2 (range 12-98.2 ml/min/1.73 m^2) compared to a median measured GFR of 53.4 ml/min/1.73 m^2 (range 7.2-100.9 ml/min/1.73 m^2). A wide range of estimated GFRs was observed

with a notably wider range using the MDRD study equation compared to the CKD-EPI equation with the highest MDRD eGFR calculated at 156 ml/min/1.73m^2 compared to 98.2 ml/min/1.73m^2 using CKD-EPI equation and a measured GFR of 100.9 ml/min/1.73m^{2}hence potentiating increased inaccuracy of the results.

Scatter plots of the two estimating equations against measured GFR are shown in Figure 4.4. These graphs visually represent the variability of estimated GFR to measured GFR when using both estimating equations to the identity line which are more notable in the MDRD equation at higher levels of GFR. Three particular outliers in the MDRD graph stand out which are not observed in the CKD-EPI graph supporting the evidence of better performance of the CKD-EPI equation at higher GFR when compared to the MDRD study equation. The CKD-EPI scatter plot demonstrates consistent performance of the equation at all levels of GFR.

Table 4.2 Measured and Estimated GFR of Entire Study Population Subdivided by Age < 80 and ≥ 80 Years

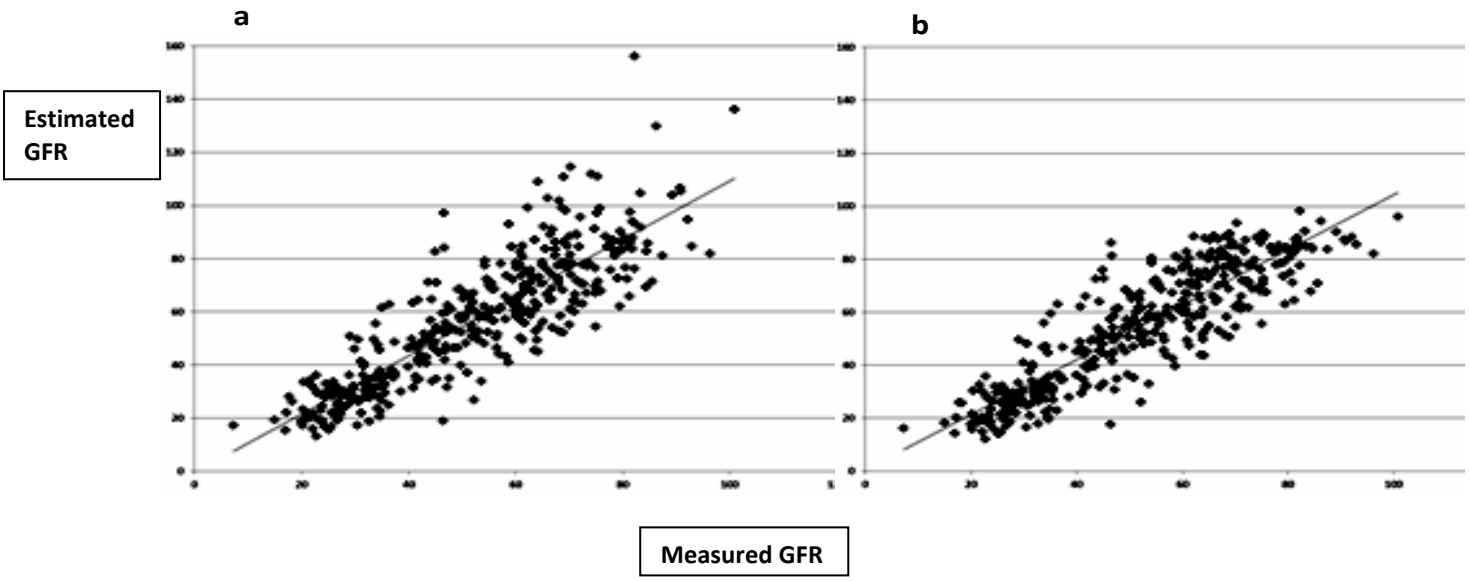
	Entire cohort	<80y	≥80y
N	394	193	201
Median serum creatinine[a] (range)	94.6 (34.5 – 380.1)	88.4 (40.7-328)	101.7 (34.5-380.1)
Median iohexol GFR[b] (range)	53.4 (7.2 – 100.9)	60.6 (7.2 – 96.3)	46.9 (15.0 – 100.9)
Measured iohexol GFR by GFR category, n (%)			
≤30	64 (16.2)	22 (11.4)	42 (20.9)
30-44	79 (20.1)	27 (14.0)	52 (25.9)
45-59	91 (23.1)	46 (23.8)	45 (22.4)
60-89	154 (39.1)	95 (49.2)	59 (29.4)
≥90	6 (1.5)	3 (1.6)	3 (1.5)
Median MDRD eGFR[b] (range)	57.6 (13.3 – 156.0)	62.0 (16.0 – 129.8)	52.3 (13.3 – 156.0)
Median CKD-EPI eGFR[b] (range)	57.0 (12.0 – 98.2)	63.1 (14.9 – 94.4)	50.3 (12.0 – 98.2)

Abbreviations: GFR, glomerular filtration rate; eGFR, estimated glomerular filtration rate; n, number; y, year; MDRD, Modification of Diet in Renal Disease study equation; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration equation.
[a]Serum creatinine expressed in µmol/l
[b] GFR expressed in ml/min/1.73m^2

Table 4.2

Measured and estimated GFR results of the study population. Measured GFR was determined by the reference iohexol method and eGFR was estimated using the MDRD study and CKD-EPI equations. Values for the continuous variables expressed as the median value with the ranges of values shown in brackets. Values for categorical values are expressed as a number with the percentage of the total study population shown in brackets. Median measured and estimated GFR results are shown and the proportion of patients in each GFR range is shown. There was a substantially lower median GFR in the ≥ 80 year subgroup with a median measured GFR of 46.9 ml/min/1.73 m^2 compared to 60.6 ml/min/1.73m^2 in the <80 year subgroup. A wide range of GFRs was observed however in both subgroups representing a variable spread between GFR categories. There were a higher proportion of patients with CKD stages 3b-5 in the older subgroup. A higher median estimated GFR is observed using the MDRD study and CKD-EPI estimating equations compared to measured GFR with a notably wider range of estimated GFR using the MDRD study equation.

Figure 4.4 Scatter Plots Examining the Correlation Between Estimating Equations and Measured GFR



Abbreviations: MDRD, Modification of Diet in Renal Disease study equation; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration equation; GFR, glomerular filtration rate.

Figure 4.4

Scatter plots of the estimated GFR (ml/min/1.73 m^2) plotted on the y axis versus the measured Iohexol GFR plotted on the x axis. Graph a represents the performance of the MDRD study equation and graph b represents the CKD-EPI estimating equation. An identity line of best fit helps to depict the increased variability of the MDRD study equation when compared to the CKD-EPI equation particularly at higher GFR levels with 3 significantly overestimated outliers observed in the MDRD study equation graph. The CKD-EPI scatter plot demonstrates consistent variability at all levels of GFR.

The performances of the MDRD Study and CKD-EPI equations overall are summarised in Table 4.3. The median differences between estimated and measured GFR were calculated using the two equations. The minimum difference of the MDRD study equation was 0 ml/min/1.73m^2 and maximum value was 73.8 ml/min/1.73m^2. The minimum difference of the CKD-EPI equation was 0 ml/min/1.73m^2 and maximum value was 39.6 ml/min/1.73m^2.

Bias

When assessing bias, both the MDRD and CKD-EPI equations showed positive bias compared to the reference test. The MDRD study equation had an overall positive bias of 3.5 (95% CI 1.9-4.8) and at GFR < 60 ml/min/1.73m^2 the bias was lower at 2.0 at GFR ≥ 60 ml/min/1.73m^2 the bias increased to 5.5. The CKD-EPI equation across the whole study population had a smaller positive bias of 1.7 (95% CI 0.3-3.2) with a lower bias in GFR < 60 ml/min/1.73m^2 (0.6) and a positive bias of 4.3 in GFR > 60 ml/min/1.73m^2. Using the Wilcoxon paired t-test to compare measured versus estimated GFR, bias achieved statistical significance overall and in GFR < 60 ml/min/1.73m^2 and in GFR ≥ 60 ml/min/1.73m^2 subgroups ($p < 0.05$). In individuals with GFR ≥ 60 ml/min/1.73m^2, bias was higher in both the MDRD and CKD-EPI equations suggesting poorer performance of both equations at higher levels of renal function.

Precision

Precision was assessed as the IQR of the difference of measured GFR from estimated GFR. Precision of the MDRD and CKD-EPI equations was similarly poor with an IQR of 13.7 and 13.1 respectively across the study group. Comparatively, the CKD-EPI equation had better precision at GFR ≥ 60 ml/min/1.73m^2 but overall both equations performed similarly and demonstrated that precision appears to decline at higher levels of renal function.

Accuracy

The overall P_{30} of the MDRD equation was 81% and it performed better at GFR ≥ 60 ml/min/1.73m^2 with a P_{30} of 86% compared to a P_{30} of 78% in GFR < 60

ml/min/1.73m^2. The CKD-EPI equation had a P_{30} of 83% and again accuracy was better in GFR ≥ 60 ml/min/1.73m^2 with a P_{30} of 93% compared to 76% in GFR < 60 ml/min/1.73m^2. The CKD-EPI equation appeared to be more accurate than the MDRD equation in all subjects however statistical significance was only achieved in those with GFR ≥ 60 ml/min/1.73m^2. No equation achieved a P_{30} of 90% or greater in the overall cohort except the CKD-EPI equation at GFR levels ≥ 60 ml/min/1.73 m^2.

Table 4.3 Performance of the MDRD and CKD-EPI Equations Compared to Measured GFR, Stratified by GFR < 60 or ≥ 60 ml/min/1.73 m^2

		GFR (ml/min/1.73 m^2)	
All participants	**Overall (n=394)**	**<60 (n=234)**	**≥60 (n=160)**
Bias (estimated minus measured GFR), median difference (95% CI)			
MDRD	3.5 (1.9 to 4.8)*	2.0 (0.8 to 3.9)*	5.5 (3.4 to 8.1)*
CKD-EPI	1.7 (0.3 to 3.2)*	0.6 (-0.7 to 2.3)*	4.3 (1.2 to 6.2)*
Precision, IQR of the difference (95% CI) {min, max difference (ml/min/1.73m^2}			
MDRD	13.7 (11.4, 16.0) {0, 73.8}	11.4 (9.5, 13.3) {0, 50.5}	18.3 (14.3, 22.3) {0.2, 73.8}
CKD-EPI	13.1 (11.7, 14.6) {0, 39.6}	11.7 (9.8, 13.6) {0, 34.6}	15.8 (13.0, 18.7) {0.1, 39.6}
Accuracy, percentage of estimates within 30% of measured GFR (P_{30}) (95% CI)			
MDRD	81 (77, 85)	78 (72, 83)	86 (79, 91)*
CKD-EPI	83 (79, 87)	76 (70, 81)	93 (88, 97)*

Abbreviations: n, number; CI, confidence interval; GFR, glomerular filtration rate; IQR, interquartile range; MDRD, Modification of Diet in Renal Disease study equation; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration equation.
*Indicates reached statistical significance ($p < 0.05$)

Table 4.3

Results are shown of the MDRD and CKD-EPI eGFR in terms of bias, precision and accuracy (percentage of estimates within 30% of the measured GFR (P_{30})) of the total cohort and split by subgroups GFR < 60 or ≥ 60 ml/min/1.73 m^2. Bias was calculated as the median difference of the estimated minus the measured GFR. The Wilcoxon matched-pairs signed rank test was used to compare the bias of each of the MDRD and CKD-EPI GFR estimates against measured GFR. Both equations demonstrated a statistically significant positive bias overestimating GFR with a more positive bias using the MDRD study equation and this bias increased at GFR ≥ 60 ml/min/1.73 m^2. Precision was calculated as the IQR of the median differences. Precision was similar in both equations overall but declined at GFR ≥ 60 ml/min/1.73 m^2. The McNemar test was used to compare P_{30} values of the MDRD and CKD-EPI equation GFR estimates. The CKD-EPI equation was only statistically better in terms of accuracy compared to the MDRD study equation at GFR ≥ 60 ml/min/1.73 m^2 and was the only subgroup to achieve a P_{30} of > 90%.

Performance of the estimating equations in the < 80 and ≥ 80 years of age and the GFR < 60 ml/min/1.73 m^2 and ≥ 60 ml/min/1.73 m^2 subgroups are presented in Tables 4.4 and 4.5 respectively.

In participants < 80 years old, the MDRD had a positive bias of 3.0 and the CKD-EPI had a positive bias of 3.1. The over estimation of both equations was more pronounced in those aged ≥ 80 years and the CKD-EPI equation performed slightly better than the MDRD equation. The CKD-EPI equation was found to be unbiased in people with a GFR < 60 ml/min/1.73m^2 ≥ 80 years. Again the bias observed was worse at GFR ≥ 60 ml/min/1.73m^2 for both equations. Similar trends in precision of the estimating equations were seen whether individuals were < 80 or ≥ 80 years of age with less precision at GFR ≥ 60 ml/min/1.73m^2.

The P_{30} of the MDRD study equation was 81% in individuals aged <80 years and 78% in aged ≥ 80 years old. The CKD-EPI P_{30} values were 83% in both the < 80 year and ≥ 80 year subgroups. The superiority of the CKD-EPI equation compared to the MDRD equation reached statistical significance only in the very elderly with a GFR ≥ 60 ml/min/1.73m^2 with 97% of all estimates falling within 30% of measured GFR in the ≥ 80 year subgroup.

Amongst males, the MDRD and CKD-EPI equations were unbiased; male MDRD bias was 1.6 (95% CI 0.0 to 3.9, P=0.07) and male CKD-EPI bias was 0.1 (95% CI -1.9 to 1.1), P=0.9). Amongst females the MDRD (4.8 (95% CI 3.0 to 6.6), P<0.001) and CKD-EPI (4.3 (95% CI 1.8 to 5.9) P<0.001) equations both overestimated measured GFR with a significantly positive bias.

Figure 4.5 shows the difference plots of the measured against estimated GFR for both estimating equations. In the MDRD graph, one observes 1 extreme outlier and we re-analysed data to see what effect this outlier would have on overall bias. Excluding this outlier, the MDRD bias reduced from 3.5 to 2.6 but it remained inferior to the CKD-EPI equation.

Table 4.4 Performance of the MDRD and CKD-EPI Equations Compared to Measured GFR, in Participants < 80 Years Stratified by GFR < 60 or ≥ 60 ml/min/1.73 m^2

Participants <80 years old	**Overall (n=193)**	**<60 (n=95)**	**>60 (n=98)**
Bias, median difference (95% CI)			
MDRD	3.0 (0.8 to 5.1)*	2.1 (-0.9 to 5.0)*	4.6 (0.8 to 7.2)*
CKD-EPI	3.1 (0.3 to 4.9)*	1.2 (-1.5 to 4.8)*	3.6 (0.3 to 6.6)*
Precision, IQR of the difference (95% CI) {min, max difference (ml/min/1.73 m^2)}			
MDRD	13.3 (10.4, 16.2) {0, 50.5}	11.9 (8.0, 15.7) {0, 50.5}	15.7 (10.9, 20.5) {0.2, 44.3}
CKD-EPI	13.0 (11.0, 15.0) {0.1, 39.6}	12.8 (8.9, 16.7) {0, 28.8}	13.2 (9.9, 16.5) {0.2, 39.6}
Accuracy, percentage of estimates within 30% of measured GFR (P_{30}) (95% CI)			
MDRD	84 (78, 89)	79 (69, 87)	89 (81, 94)
CKD-EPI	83 (77, 88)	75 (65, 83)	91 (83, 96)

Abbreviations: n, number; CI, confidence interval; GFR, glomerular filtration rate; IQR, interquartile range; MDRD, Modification of Diet in Renal Disease study equation; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration equation; IQR, interquartile range.
*Indicates reached statistical significance ($p < 0.05$)

Table 4.4

Results are shown of the MDRD and CKD-EPI eGFR in terms of bias, precision and accuracy (percentage of estimates within 30% of the measured GFR) of the total cohort and split by subgroups GFR < 60 or ≥ 60 ml/min/1.73 m^2 in subjects < 80 years. Bias was calculated as the median difference of the estimated minus the measured GFR. The Wilcoxon matched-pairs signed rank test was used to compare the bias of each of the MDRD and CKD-EPI GFR estimates against measured GFR. Both equations demonstrated a statistically significant positive bias overestimating GFR with a more positive bias using the MDRD study equation and this bias increased at GFR ≥ 60 ml/min/1.73 m^2. Precision was calculated as the IQR of the median differences. Precision was similar in both equations overall but declined at GFR ≥ 60 ml/min/1.73 m^2. The McNemar test was used to compare P30 values of the MDRD and CKD-EPI equation GFR estimates. Only the CKD-EPI equation at GFR ≥ 60 ml/min/1.73 m^2 achieved a P_{30} of > 90% but no equation was performed statistically significantly better than the other in all subgroups.

Table 4.5 Performance of the MDRD and CKD-EPI Equations Compared to Measured GFR, in Participants ≥ 80 Years Stratified by GFR < 60 or ≥ 60 ml/min/1.73 m^2

Participants ≥80 years old	**Overall (n=201)**	**<60 (n=139)**	**>60 (n=62)**
Bias, median difference (95% CI)			
MDRD	3.8 (1.6 to 5.2)*	2.0 (0.8 to 4.1)*	8.3 (3.8 to 12.9)*
CKD-EPI	1.2 (-0.1 to 2.6)*	0.5 (-1.2 to 2.2)	4.4 (-0.1 to 10.0)*
Precision, IQR of the difference (95% CI) {min, max difference (ml/min/1.73 m^2)}			
MDRD	13.8 (11.3, 16.3)	11.4 (9.0, 13.8) [53.2]	19.2 (13.7, 24.6) [89.3]
CKD-EPI	13.3 (10.9, 15.7) [51.4]	10.9 (8.4, 13.4) [50.9]	19.1 (15.3, 22.9) [40.7]
Accuracy, percentage of estimates within 30% of measured GFR (P_{30}) (95% CI)			
MDRD	78 (72, 84)	77 (69, 84)	81 (69, 90)*
CKD-EPI	83 (77, 88)	77 (69, 84)	97 (89, 100)*

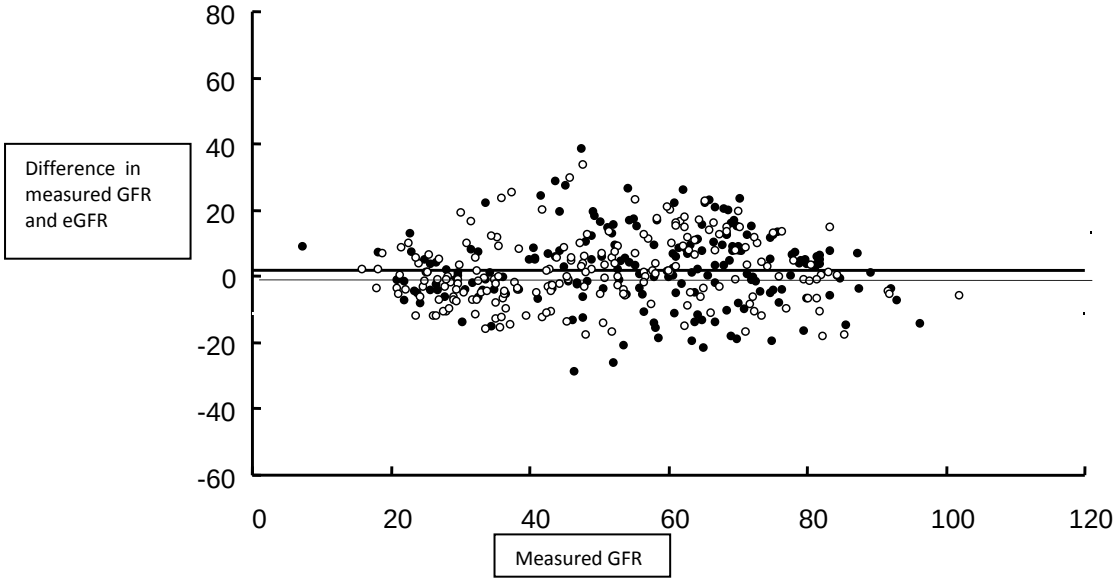
Abbreviations: n, number; CI, confidence interval; GFR, glomerular filtration rate; IQR, interquartile range; MDRD, Modification of Diet in Renal Disease study equation; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration equation; IQR, interquartile range.
*Indicates reached statistical significance ($p < 0.05$)

Table 4.5

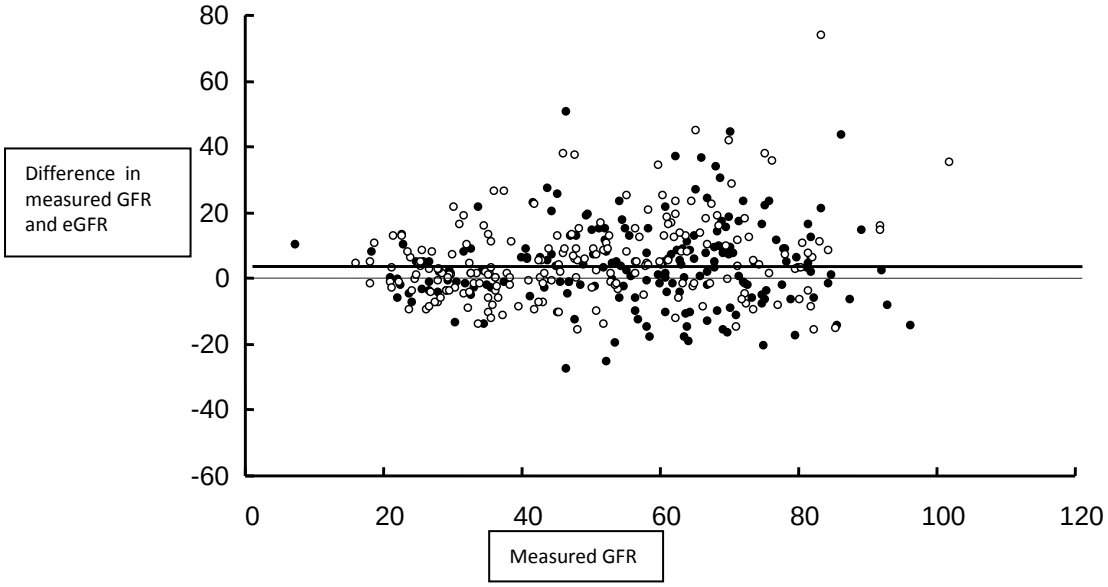
Results are shown of the MDRD and CKD-EPI eGFR in terms of bias, precision and accuracy (percentage of estimates within 30% of the measured GFR) of the total cohort and split by subgroups GFR < 60 or ≥ 60 ml/min/1.73 m^2 in subjects aged ≥ 80 years. Bias was calculated as the median difference of the estimated minus the measured GFR. The Wilcoxon matched-pairs signed rank test was used to compare the bias of each of the MDRD and CKD-EPI GFR estimates against measured GFR. Both equations demonstrated a statistically significant positive bias overall however the CKD-EPI did not have a significantly positive bias in GFR < 60 ml/min/1.73m^2. Bias increased at GFR ≥ 60 ml/min/1.73 m^2 in both estimating equations. Precision was calculated as the IQR of the median differences. Precision was similar in both equations overall but declined at GFR ≥ 60 ml/min/1.73 m^2. The McNemar test was used to compare P30 values of the MDRD and CKD-EPI equation GFR estimates. The CKD-EPI equation was only statistically better in terms of accuracy compared to the MDRD study equation at GFR ≥ 60 ml/min/1.73 m^2 and was the only subgroup to achieve a P_{30} of > 90%.

Figure 4.5 Bias Plots of the MDRD and CKD-EPI Equations Against Measured GFR

a



b



Abbreviations: MDRD, Modification of Diet in Renal Disease study equation; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration equation; eGFR, estimated glomerular filtration rate; mGFR, measured GFR.

Figure 4.5

Bias plots showing the difference of the estimated GFR (ml/min/1.73m^2) and measured GFR. Graph **a** displays the MDRD study equation and graph **b** displays the CKD-EPI equation. White circles are represented by subjects <80 years and black circles are subjects aged ≥ 80 years. The dotted line represents zero bias and solid line indicates the median bias of the estimating equation. There was an increased variability using the MDRD equation particularly at higher GFRs with significant outliers in the MDRD compared to the CKD-EPI graph. Both equations demonstrated a positive bias with a 3.5 positive bias using the MDRD equation and a 1.7 positive bias using the CKD-EPI equation overall.

Misclassification Errors

Misclassification errors followed the bias of the equations as would be expected. Both the MDRD and CKD-EPI equations were positively biased compared to reference GFR and were more likely to wrongly classified individuals as having an eGFR ≥ 60ml/min/1.73m^2. A total of 29 (14%) patients were misclassified according to GFR < or > 60ml/min/1.73m^2 s in the MDRD group and 24 (11.9%) in the CKD-EPI group and is shown in Table 5.7. Only 4 (2%) individuals using the MDRD equation and 6 (3%) individuals using the CKD-EPI equation were wrongly considered to have a GFR < 60ml/min/1.73m^2. In contrast, 25 (12.4%) people using the MDRD equation and 18% using the CKD-EPI equation were wrongly considered to have a GFR ≥ 60ml/min/1.73m^2. For both equations, misclassification errors appeared worse amongst individuals <80 years old compared to individuals aged 80 years and over.

In summary, this study has shown that the MDRD study and CKD-EPI equations are fairly accurate in assessing renal function in an elderly Caucasian population with a slightly better performance by the CKD-EPI equation particularly at higher GFRs. They are more likely to overestimate GFR which may have an effect on the misclassification or misdiagnosis of individuals with CKD.

Table 4.6 Misclassification Errors

Wrongly considered to have GFR <60 ml/min/1.73 m^2, n (%)	**All**	**< 80 years**	**≥ 80 years**
MDRD	20 (5.1)	16 (8.3)	4 (2)
CKD-EPI	21 (5.3)	15 (7.8)	6 (3)
Wrongly considered to have GFR ≥60 ml/min/1.73 m^2, n (%)			
MDRD	45 (11.4)	20 (10.4)	25 (12.4)
CKD-EPI	38 (9.6)	20 (10.4)	18 (9)
Total misclassified, n (%)			
MDRD	65 (16.5)	36 (18.6)	29 (14.4)
CKD-EPI	59 (15)	35 (18.1)	24 (11.9)

Abbreviations: n, number; CI, confidence interval; GFR, glomerular filtration rate; IQR, interquartile range; MDRD, Modification of Diet in Renal Disease study equation; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration equation

Table 4.6

This table shows the number and proportion of patients who were wrongly misclassified as having a GFR < 60ml/min/1.73m^2 or ≥ 60ml/min/1.73m^2 when estimating GFR using the MDRD and CKD-EPI equations. These results show that both equations were more likely to wrongly classify individuals as having GFR < 60ml/min/1.73m^2 and less likely to wrongly classify individuals as having GFR ≥ 60ml/min/1.73m^2. For both equations, misclassification errors appeared worse among individuals < 80 years old compared to individuals ≥ 80 years old.

This study has demonstrated that the MDRD equation performs reasonable well in estimating GFR when compared to measured GFR in older people. The MDRD study equation had a positive bias tending to overestimate GFR particularly in those with a GFR ≥ 60ml/min/1.73m^2 and there was little difference seen in those < 80 or ≥ 80 years old. In terms of accuracy the MDRD achieved P_{30} levels of 81% overall which falls short of the > 90% KDOQI guideline but accuracy improved at higher GFR levels. The CKD-EPI equation performed similarly but slightly better that the MDRD equation in terms of bias, precision and accuracy and this was also seen in the < 80 and ≥ 80 years subgroups. In the entire population studied, both the MDRD and CKD-EPI equations overestimated GFR. The MDRD and CKD-EPI equations also overestimated measured GFR when analysing the subgroups with a GFR < 60 ml/min/1.73 m2 and ≥ 60 ml/min/1.73 m2. The CKD-EPI showed no bias in people ≥ 80 years with GFR ≥ 60ml/min/1.73m^2 and achieved P_{30} values >90% in both age subgroups at GFR ≥ 60ml/min/1.73m^2. Inaccuracies of the MDRD and CKD-EPI estimating equations led to misclassification of 16% and 15.5% of the total study cohort respectively with a greater degree of misclassification in those aged < 80 years old.

CHAPTER 5

Validating Estimating GFR Equations in the Elderly

5.1 Discussion

The prevalence of CKD has risen not only in association with the rising incidence of diabetes, hypertension and vascular disease but also with our increasingly aged population. A very small minority of elderly patients with CKD will progress to end stage renal disease and the remaining majority with CKD will have the associated cardiovascular morbidity and mortality associated with CKD. Roderick *et al* have demonstrated that in the UK, in subjects aged 70 years and older, there is a graded and independent increase in all-cause mortality and cardiovascular risk particularly in those with eGFR < 45 ml/min/1.73m^2 (157). Hence it is important to ensure our methods of monitoring function and assessing eGFR are validated in this particularly susceptible group. Conversely, incorrectly labelling frail and elderly people with a condition like CKD can also lead to unnecessary referrals to specialist centres involving travelling long distances, subjecting them to complex polypharmacy and often significant concern and worry. Assessment of performance of these estimating GFR equations in the elderly was urgently needed to ensure our methods of diagnosing CKD are valid.

This study is the first large prospective study to evaluate the performance of the contemporary GFR estimating equations in an older Caucasian population. I hypothesised that the reported high incidence of CKD is partly due to the inaccuracies of the estimating equations used to assess renal function in older people. The aim was to assess the accuracy of the MDRD study and CKD-EPI equations estimated GFR in older people in comparison to measured GFR. The MDRD study equation was assessed because currently it is national practice to report eGFR derived from the MDRD study equation with every serum creatinine test

(10) into clinical practice due to its perceived increased accuracy. The CKD-EPI equation was also assessed as there have been recommendations to adopt it use in place of the MDRD study equation.

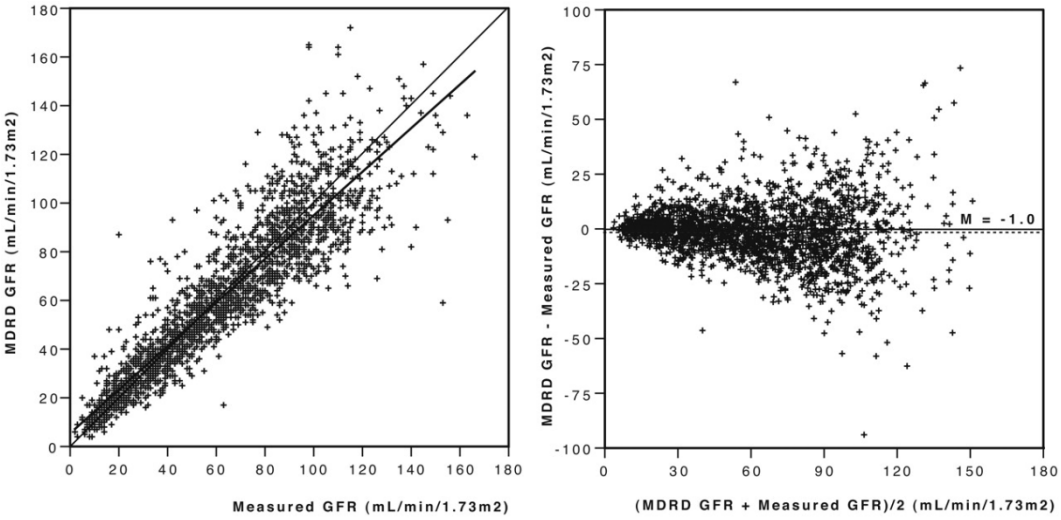
Overall this study has shown that the MDRD and CKD-EPI equations perform fairly well in an elderly Caucasian cohort. In comparison with previous studies, this study has demonstrated that the P_{30} values observed for both MDRD and CKD-EPI equations in older people appear reasonable and the inferiority of the MDRD equation was only significant amongst individuals with GFR ≥ 60 ml/min/1.73 m^2. The P_{30} values are slightly lower than those observed in the original MDRD and CKD-EPI validation cohorts, (8; 110) but consistent with other independent evaluations (108, 111-113, 115, 117, 122) Further, bias against the reference method, although significant for the MDRD and CKD-EPI equations, was small in clinical terms. Notably, the underestimation of GFR reported for the MDRD equation in some other studies (110-112, 117) was not seen in this study. There was in fact a small positive bias observed even, and especially, at higher levels of GFR. The CKD-EPI equation was more accurate than the MDRD equation at higher levels of GFR.

Since the publication of the MDRD equation in 1999 (8), there have been many evaluations of its performance. The MDRD equation was developed in a population with CKD and a general observation has been that the equation has not performed so well outside of that population. In particular, it has been found to report negatively biased estimates of GFR when GFR exceeds 60 ml/min/1.73m^2 (158). Several studies have shown that the MDRD equation performs less well at higher levels of renal function with observations of a divergence of the bias of the MDRD equation from the identity line as GFR increases, particularly in younger populations (116). This was well illustrated in the study by Froissart *et al*, which compared MDRD estimated GFR to measured GFR using renal clearance of ^{51}Cr-EDTA in a cohort of 2096 European subjects. Their findings raised caution regarding its use in stage 1 and 2 CKD and reported similar scatter plots and difference plots observed in this study with greater inaccuracy at higher GFRs (Figure 5.1). The inaccuracy of the equation was also noted in underweight individuals.

Several studies have undertaken direct comparison of the MDRD and CKD-EPI equations and a variety of statistical approaches have been used. Bias, precision and accuracy have been traditionally used to assess the performance of these estimating equations. Bias can often reflect systematic differences between the development datasets and the populations in which the equation is used. Typically, and understandably, equations have generally performed less well outside of the cohorts in whom they were developed (109, 159).

In the original MDRD validation cohort the MDRD equation achieved a P_{30} of 91% (8) whereas in the CKD-EPI cohort this fell to 81% (110). Amongst studies that have subsequently directly compared the CKD-EPI and MDRD equations in large (n > 100) adult Caucasian populations using standardized serum creatinine assays, the CKD-EPI equation has been superior to the MDRD equation with a lower bias particularly at higher eGFRs (>60 ml/min/1.73 m^2) whereas the MDRD equation performed better at lower eGFR values (114). Figure 5.2 illustrates the performance of both equations in the CKD-EPI validation set. Both equations' bias plots show increased scatter at higher eGFR levels but the CKD-EPI equation yielded improved median bias and was assessed as being as accurate as the MDRD study equation in those with an eGFR < 60 ml/min/1.73 m^2 and substantially more accurate in those with an eGFR ≥ 60 ml/min/1.73 m^2 consistent with the findings in this study (110). These results were consistent across subgroups defined by age, gender, race, presence of diabetes and body mass index.

Figure 5.1. MDRD Equation is Less Accurate at Higher GFR Levels



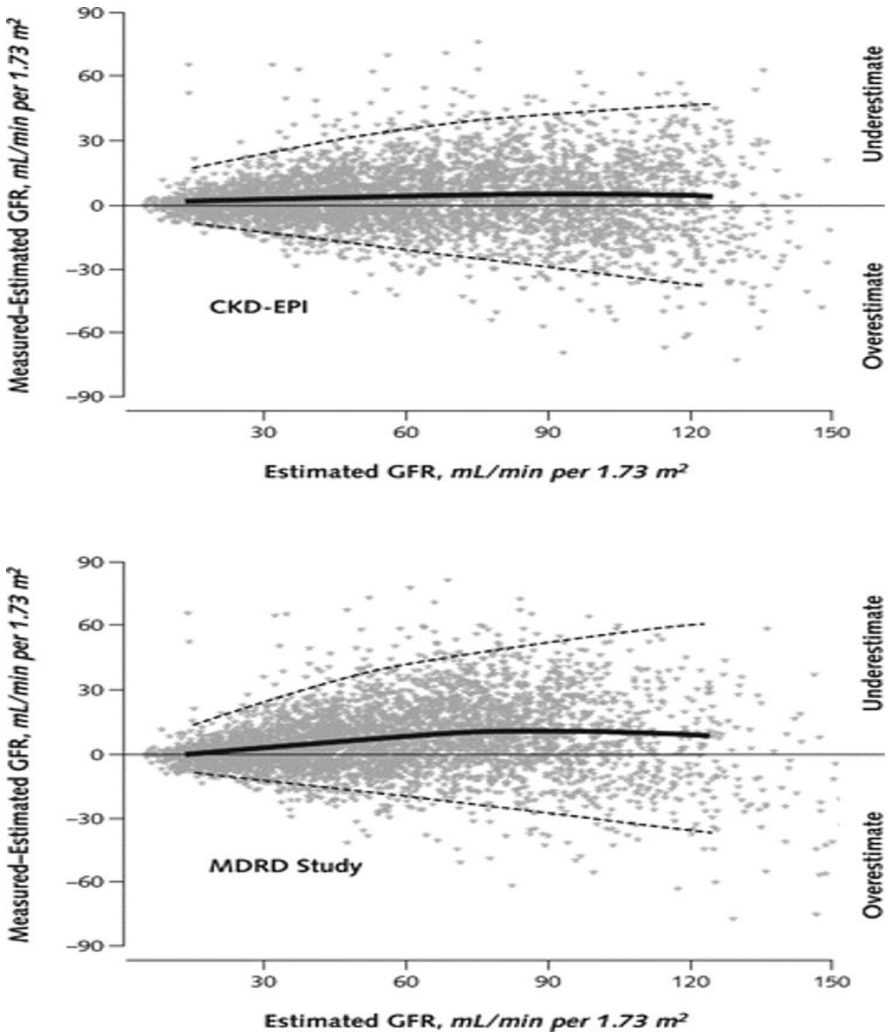
Abbreviations: GFR, glomerular filtration rate; eGFR, estimated glomerular filtration rate; MDRD, modification of diet in renal disease; M, mean difference.

Figure 5.1.

Froissart *et al*, demonstrated the relationship between measured GFR using renal clearance of ^{51}Cr-EDTA as the reference method and MDRD eGFR. Bland and Altman plots comparing measured GFR and MDRD eGFR is on the right with the mean difference (M) represented by the dashed line. This demonstrates the inaccuracy of the MDRD equation as it reaches higher levels of GFR.

Froissart *et al* 2005 (116).

Figure 5.2. Performance of the CKD-EPI and MDRD Study Equations in Estimating GFR in the CKD-EPI External Validation Study



Abbreviations: MDRD, Modification of Diet in Renal Disease study equation; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration equation; GFR, glomerular filtration rate

Figure 5.2

In 2009 Levey *et al,* assessed the performance of the MDRD and CKD-EPI equations in the CKD-EPI external validation set. Bland and Altman plots of the measured GFR and estimated GFR using the MDRD and CKD-EPI equations are shown. The mean difference (bias) is represented by the thick line. This demonstrated both equations underestimated GFR and bias increased as GFR increased. The CKD-EPI performed superior to the MDRD equation.

Levey *et al* 2009 (110)

Observed P_{30} values from studies evaluating the estimating equations have ranged from 73% to 93% for the MDRD equation and from 80% to 95% for the CKD-EPI equation (114). A recent large retrospective study from the Mayo clinic did not report P_{30} values but observed that the relative bias of the two equations differed depending on the clinical presentation, with the CKD-EPI being superior in kidney donors and inferior among CKD patients (160). In 2002, the NKF-KDOQI called for future GFR estimating equations to achieve P_{30} values in excess of 90% (10) yet across a wide range of studies, this has rarely been achieved to date (114). This study has demonstrated that both the MDRD and CKD-EPI equations fail to meet this standard in the elderly population overall with the exception of the CKD-EPI equation in the subgroup with GFR ≥ 60 ml/min/1.73 m^2.

So how do our study results compare with previous smaller studies in older people? With the exception of the study by Jones *et al*, who included a small number of older individuals (115), most but not all studies (108, 111-113, 117, 122) have included very few individuals over 75 years of age. The MDRD equation was originally validated in 1085 patients with CKD with a mean age of 51±13 years (22% >65 years) (8). The authors subsequently evaluated the equation in a larger (n=5504) and more diverse population, but again, few people were >75 years old (mean age 47 ± 15 years, 13% > 65 years) (31). Although very few elderly people were included, it is interesting to note that the bias they observed amongst the > 65 year subgroup was smaller than amongst younger individuals. The bias was minimal (-0.3 ml/min/1.73 m^2) even when GFR exceeded 60 ml/min/1.73 m^2, with a P_{30} of 88% being observed in this subgroup. It may be that the numbers in the study were too small as it is possible to end up with a smaller bias by chance. By contrast, MDRD underestimated measured GFR amongst younger individuals with higher GFR (by -6.4 and -10.6 ml/min/1.73 m^2 in individuals 40 to 65 years and <40 years respectively).

In the study by Froissart *et al*, a sub-analysis of patients aged > 65 years with GFR < 60ml/min/1.73m^2 showed that the performance of the MDRD equation was generally comparable to that amongst younger individuals tending to give a slightly negatively biased estimate of GFR (mean bias -1.0 ml/min/1.73m^2) (116). Only 57

subjects in this study, however, were aged 80 years or more. In a reanalysis of data from a small study performed by Lamb *et al*, eGFR using the MDRD equation showed minimal bias (mean bias -2.0 ml/min/1.73m^2) and reasonable precision (166). Data was from 46 patients with a mean age of 80 years (ranging from 69-92 years) and the median GFR was 55 ± 17 ml/min/1.73m^2. These two studies were relatively small in terms of numbers of older people and pre-date both internationalised standardisation of creatinine and the publication of the CKD-EPI equation. Nevertheless, they pointed towards good estimation of GFR in older people with the MDRD study equation.

In the CKD-EPI equation, the development and external validation datasets had mean ages of 47 ± 15 and 50 ± 15 years respectively and again included very few elderly people (110). Nevertheless, bias of the CKD-EPI and MDRD estimates in the external validation data set were minimal and broadly equivalent (-1.3 and -1.4 ml/min/1.73 m^2 for MDRD and CKD-EPI respectively) amongst individuals > 65 years, whereas larger biases were observed amongst younger individuals, in particular for the MDRD equation (e.g. -9.7 ml/min/1.73 m^2 for individuals < 40 years).

A study of Australian individuals also reported a similar age-related shift (115) and CKD-EPI eGFR was reported to be 3 ml/min/1.73 m^2 lower than MDRD eGFR in North American octogenarians (161). The recent large study of Murata *et al* (n=984) observed that both equations overestimated GFR (MDRD 9%; CKD-EPI 5%) in individuals >70 years with CKD and underestimated GFR in younger healthy individuals (e.g. amongst 40-69 year olds CKD-EPI -9%; MDRD -17%) (160).

Overall, a picture emerges of differences between the CKD-EPI and MDRD equations, and the differences between these equations and the reference methods being diminished amongst older compared to younger people. The MDRD equation was developed in a cohort of patients with CKD (mean GFR 40 ml/min/1.73 m^2). Its underperformance in healthy individuals with higher GFR has been attributed to the fact that few such individuals were included in the original development dataset. The CKD-EPI equation was developed in a broader cohort with better kidney function

(median GFR was 68 ml/min/1.73 m^2), purposefully including healthy individuals, thus explaining its superior performance at higher levels of GFR. Nevertheless, it was predominantly developed amongst cohorts of diseased individuals, in particular patients with CKD and/or diabetes (8).

As age increases, the body composition changes with an increase in body fat composition and a significant reduction in lean body mass particularly in those of extreme old age (24). These age-related changes may have an important effect on creatinine clearance and GFR measurements. Both the MDRD and CKD-EPI equations try to overcome these factors by incorporating body surface area in their calculations hence GFR per 1.73 m^2. One can speculate that the relationship between muscle mass, dietary protein intake (and thus creatinine) and GFR amongst older people is more akin to that of the diseased development cohorts of the MDRD and CKD-EPI equations than some of the various younger populations in which they have subsequently been tested. Consequently these equations work reasonably well and broadly equivalently in an elderly Caucasian population. This is a fortunate coincidence since the major burden of CKD in most populations resides amongst the elderly. Furthermore, whereas in middle-aged populations the CKD-EPI equation appears to better identify clinical risk than the MDRD equation, amongst older people, risk estimates based upon the two equations appear similar. This present study confirms acceptable performance of the MDRD and CKD-EPI equations in older people.

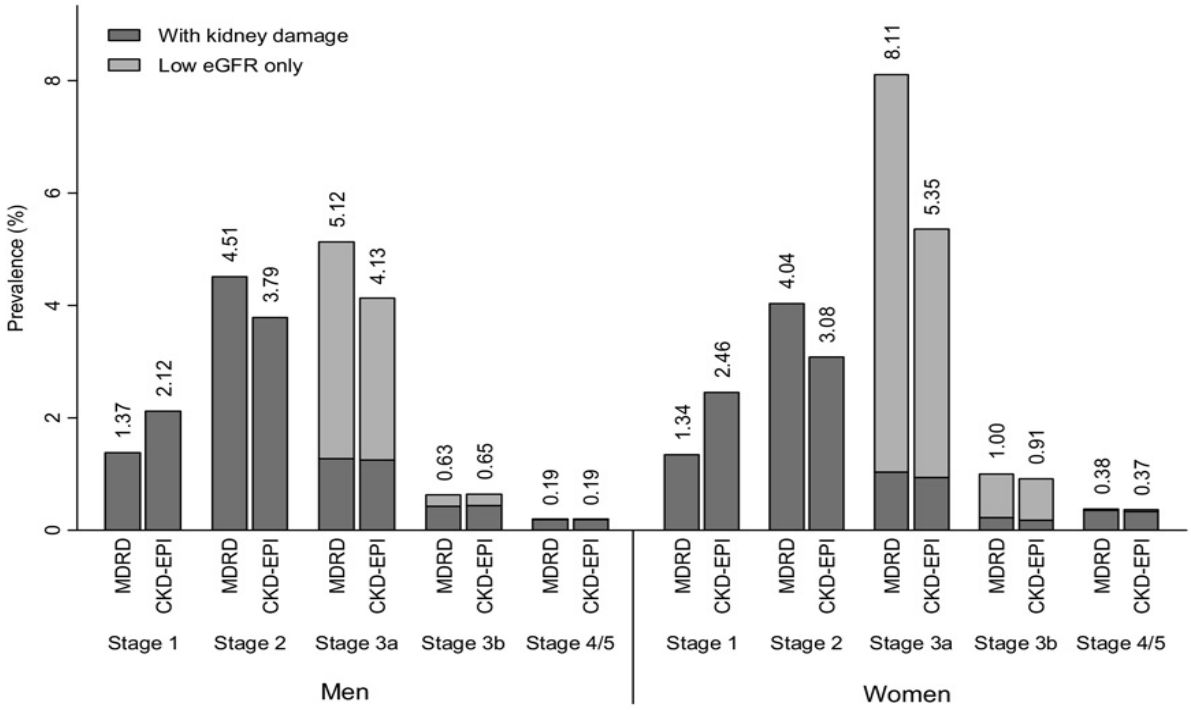
Of note, however, analysis of the characteristics of the outliers falling outside the P_{30} reference GFR revealed a higher proportion of females particularly in the CKD-EPI dataset (72 female vs 35 male). The extreme outliers were also associated with a low body surface area so the discordance between measured and estimated GFR using both equations may well be related to low muscle mass and hence low serum creatinine measurements. The NICE guidelines recommend that eGFR be interpreted with caution in cases of extremes of muscle mass such as amputees or muscle wasting disorders (57). For this reason, one participant was excluded from the study as he was an amputee on the basis that differences in muscle mass would affect the

study results. This highlighted that the original exclusion criteria were not robust enough. Studies have also shown the MDRD equation significantly underestimates GFR in obese subjects with GFR > 60 ml/min/1.73m^2 demonstrating the effect differing body habitus has on eGFR values (162).

Accumulating evidence suggests that the CKD-EPI equation better identifies clinical risk than the MDRD equation, (163-164) including amongst older people (165). It therefore seems likely that the CKD-EPI will be introduced routinely in clinical practice and it is important to understand how this will impact on the CKD prevalence rates. Reporting eGFR using the MDRD study equation is widespread nationally and internationally so a change in the estimating equation used by clinical laboratories would have significant implications. As the CKD-EPI equation has greater differences at higher GFRs, applying the CKD-EPI equation would lead to a higher estimated GFR in the population. It would be less sensitive but more specific in detecting people with eGFR < 60 ml/min/1.73m^2. This in turn would have public health implications leading to a lower prevalence estimate but higher risk profile for people within this range of GFR. It would enable better use of finance and resources in caring for patients with a reduced GFR predominantly in the primary care setting rather than nephrology services.

In a population based study in Australia, the estimated CKD prevalence decreased from 13.3% to 11.5% when the CKD-EPI equation was used (163). The prevalence was particularly lower in women but remained high in the elderly. The reclassification was mainly from stage 3a to 2 and mainly affected those in whom eGFR was the only diagnostic feature and those with kidney damage remained unchanged. Figure 5.3 illustrates the impact of using the CKD-EPI equation and MDRD study equation on CKD prevalence estimates in the AusDiab study.

Figure 5.3 Misclassifications of the MDRD Study and CKD-EPI Equations in the AusDiab Study



Abbreviations: MDRD, Modification of Diet in Renal Disease study equation; CKD-EPI, Chronic Kidney Disease Epidemiology Collaboration equation; GFR, glomerular filtration rate.

Figure 5.3

The AusDiab (Australian Diabetes, Obesity and Lifestyle) study was a large population based survey which compared the effect of utilising the MDRD study and CKD-EPI equations on prevalence of CKD. The greatest misclassification errors were observed in the KDOQI stage 3a. The estimated CKD prevalence decreased from 13.3% to 11.5% when the CKD-EPI equation was used.

White *et al*, 2010 (163)

As the MDRD study equation is inaccurate at higher levels of GFR, guidelines recommend that eGFR is reported as a range i.e. > 60 ml/min/1.73m^2, at higher levels of GFR. Differentiating between stages 1 and 2 using serum creatinine based equations would be risky using the MDRD study equation as it is unreliable in GFR > 60ml/min/1.73m^2. Moreover, the CKD staging system suggests that an eGFR of 90 ml/min/1.73m^2 is the lower limit of normal when in reality only a minority of the population have an eGFR > 90 ml/min/1.73m^2 most of whom are young men (1). Use of the CKD-EPI equation will enable reporting of an actual numerical value throughout the full range of GFR. Conversely, use of the CKD-EPI equation will overestimate GFR and slightly increase bias at lower GFR values so clinicians caring for these patients would have to be aware of this limitation.

This study reported only 2% individuals using the MDRD equation and 3% individuals using the CKD-EPI equation were wrongly considered to have a GFR < 60ml/min/1.73m^2 misclassifying them as having CKD stage 3a. There was, however, a higher proportion (12.4% MDRD and 18% CKD-EPI) of subjects wrongly considered to have a GFR ≥ 60ml/min/1.73m^2 which may have a significant impact on CKD prevalence estimates. Froissart *et al*, found that only 70.8% of subjects were classified in the proper KDOQI CKD category across all CKD stages and approximately 20% of subjects with measured GFR > 60ml/min/1.73m^2 were classified as having stage 3 CKD using the MDRD equation to estimate GFR (116).

This study's observations also fit well with data from several studies that have looked at the relative prevalence of CKD amongst populations and across age strata (110, 163, 166-177)). In the CKD-EPI validation studies, median eGFR was 9.5ml/min/1.73m^2 higher using the CKD-EPI equation reducing the prevalence estimate of CKD by 1.6%. In all cases, whilst a decrease in CKD prevalence when assessed using the CKD-EPI equation rather than the MDRD equation has been observed in middle-aged populations, the two equations give similar prevalence estimates due to virtual abolition of negative bias at GFR >60 ml/min/1.73 m^2 amongst older people. For example, in the East Kent population, one study observed the mean estimated GFR using the CKD-EPI equation to be 11.2% higher than the

MDRD eGFR amongst individuals aged 40 to 49 years: this difference gradually diminished to 0.7% amongst the 70-79 year olds (167). In people aged over 80 years the MDRD equation actually gave a lower CKD prevalence estimate than the CKD-EPI equation. The significance of this study's misclassification data on national CKD prevalence is unclear however as the study population is not a typically reflection of the UK population.

5.2 Limitations

Demonstrable improvements in the CKD-EPI equation to the MDRD study equation were marginal in this study but it is possible that the study was underpowered to detect additional benefits. East Kent is a peninsula and therefore may not be a representative population for reasons such as obesity prevalence and social deprivation. It is also predominantly of caucasian ethnicity. Local population data shows only 1.3% of the population to be black and a further 1.9% recorded as asian (168). Hence this study has only validated the use of these equations in the older caucasian population. The initial study cohort included three african-caribbean participants, however they were excluded from the final study analysis, as race is known to affect GFR and the number was too small to have any meaningful data.

The CKD-EPI and MDRD study equations were developed in North American and European populations that compromised mainly of african-caribbean and caucasian persons. There are known racial and ethnic differences in muscle mass and diet and further studies will need to be performed in other ethnically diverse groups. In studies examining the performance of the estimating equations in other ethnicities compared to North Americans, Europeans or Australians, both the MDRD and CKD-EPI equations were less accurate (P_{30} ranging from 29 to 94%)(169-170). Studies have shown that even in the population of North America, Europe and Australia (AusDiab study), the CKD-EPI equation does not meet the 2002 KDOQI benchmark of a P_{30} of greater than 90% (163). Zuo *et al* found the MDRD underestimated true GFR at normal function and overestimated GFR at lower levels in 684 Chinese individuals

achieving a P_{30} of 80% (169). Some studies have developed a coefficient to adjust for racial variation but these coefficients do not consistently improve accuracy in other ethnic populations. In Japan, the Japanese coefficient- modified CKD-EPI equation was found to be more accurate than the Japanese coefficient-modified MDRD equation leading to a lower estimated prevalence of CKD in Japan (7.9% vs 10%) (171). Incorporating locally derived coefficients to minimise bias in different ethnic populations can be used to improve accuracy of these estimating equations but would require increased resources and introduce complexity to a widely used screening and diagnostic test. We need better assessment and evaluation of these equations used to estimate GFR in other ethnically diverse groups many of whom have a high burden of CKD.

In order to reduce bias from the biological variability of serum creatinine levels, study participants were asked to avoid meat consumption prior to the procedure, a known confounder of serum creatinine measurement (99). Avoidance of meat in the study protocol could be viewed as a weakness as well as a strength. Although pre-test avoidance of meat was protocol, there were invariably participants who did not completely adhere to protocol. Lack of dietary regulation with a potential to affect serum creatinine measurements may have contributed to the inaccuracies of the prediction equations in this study. Physiologically, protein intake increases GFR and cooking meats converts creatine to creatinine. It is readily absorbed and causes increased serum creatinine levels and the effect persists for hours. Recent meat intake clearly has a significant impact on eGFR but the impact in the real world is unclear. Other foods high in protein content such as fish have also been proven to alter serum creatinine measurements. It is possible that a meat meal would have a differential effect in the elderly. The effect on eGFR has been largely ignored and no robust recommendations had been made regarding sampling until recently. There is probably variable adherence to meat avoidance for serum creatinine testing in the everyday setting.

In 2007, Preiss *et al* studied the effect of a meat meal on serum creatinine levels and eGFR and found the median eGFR fell by 25 ml/min/1.73m^2 1-2 hours post

consumption of meat and remained 20 ml/min/1.73m^2 lower than baseline up to 4 hours later (99). The MDRD study, NHANES data and AusDiab studies all used fasting samples to overcome these inaccuracies (1, 8, 163). Of note, cystatin C was robust in the face of meat ingestion. Recent NICE guidelines have recommended that ideally blood samples or eGFR should be obtained with the individual avoiding meat consumption 12 hours prior to blood sampling (57).

No study participants were receiving cimetidine, trimethoprim or co-trimoxazole prescriptions at the time of the test. These medications are known to inhibit creatinine secretion in renal tubules increasing serum creatinine levels. The study cohort however composed of a high proportion of patients prescribed medications known to reduce GFR such as ACEIs and ARBs and NSAIDs and almost 50% of the individuals in this study were on one or more anti-hypertensive agents including diuretics. Although these may have had an impact on GFR at the time of the test, the prescription patterns are typically representative of an elderly population.

Inaccuracies of the estimating equations around eGFR < or ≥ 60 ml/min/1.73m^2 may lead to misclassification of CKD with consequences on CKD prevalence data. Perfect measurement of GFR, however, is not so necessary at the earlier stages of CKD clinically. Decreased GFR is a well known risk factor for cardiovascular disease, mortality and progression to kidney failure so misclassification of patients at the more severe CKD stages (CKD 3b-5) may have important implications for clinicians and patients clinically. A limitation of this study is that it did not analyse the level of inaccuracy of the estimating equations in all the CKD stages GFR subcategories and analysis of misclassification was limited to CKD stage 2 and 3a with eGFR < 60 or ≥ 60 ml/min/1.73m^2, levels at which the 2 estimating equations perform more accurately. This study demonstrated less bias but better accuracy of both estimating equations at higher GFRs in both < 80 and ≥ 80 year subgroups. Misclassification of individuals and inaccuracies of the estimating equations at lower CKD stages has an important effect clinically as the clinician relies on eGFR for detecting disease, predicting prognosis, guiding therapy and drug dosing. Further analyses of their performance in each of the different CKD stages would have been of interest. Misclassification errors

need to be interpreted with caution however as misclassification can occur with a bias of only 1 ml/min/1.73m^2. It is contradictory that GFR can be estimated yet diagnosis of CKD cannot.

Another criticism of this study is the choice of iohexol GFR as the reference GFR method. Standard clearance of inulin, including urine collection, remains the 'gold-standard' method for GFR measurement but few studies use this. Most evaluations of GFR equations have used radio-labelled plasma clearance methods which are assumed to be closely related to inulin clearance, although it is increasingly appreciated that such methods are not all equivalent (172). Radio-labelled iothalamate plasma clearance was the method used for developing the MDRD (8) and CKD-EPI equations (9). The CKD-EPI equation validation dataset used a variety of reference GFR methods including iohexol with small differences in clearance compared with iothalamate (110). There has been much debate about which is the more accurate method for measuring GFR. Iohexol clearance is widely used in clinical and research practice and there is no convincing evidence that it is better or worse than other reference GFR procedures compared to urinary inulin clearance (118). Prior to clinical introduction of the iohexol GFR method used in this study, it was compared with our existing GFR method (plasma clearance of Inutest) primarily used in children and the results were not significantly different. Seegmiller *et al* found that renal clearance of iohexol was slightly lower than renal clearance of iothalamate across a wide range of GFRs (173). This difference may be due to a greater plasma binding property of iohexol compared to iothalamate. One study suggested that iothalamate clearance overestimated urinary inulin clearance by 20 ml/min/1.73 m^2 (174).

Due to problems of ensuring complete bladder emptying in this population, it was not possible to collect timed urine samples and plasma iohexol was sampled over a 4-hour period in this study. The study would be more robust if a 24 hour sample was taken to improve accuracies at lower GFRs but practical limitations meant that collecting a delayed 24 hour blood sample was also not feasible. Plasma clearance of iohexol is dependent on level of GFR and some studies recommended later sampling

of iohexol measurements in those with GFR < 30 ml/min/1.72m^2 at 6 hours, 8 hours and 24 hours (175). They argued that later sampling would be more representative of the final exponential phase in the iohexol disappearance curve. Plasma clearance of iohexol using a 4-hour procedure has been shown to overestimate, (176) underestimate (177) or accurately reflect reference urinary clearance measures (178). The variety of sampling protocols, compartmental models used, patient mix and GFR ranges studied in the literature makes interpretation of these various reports difficult.

More recently, the Berlin Initiative Study (BIS) group assessed correlations between 4 hour and 5 hour iohexol clearance measurements in an elderly population and found that GFR can be satisfactorily measured within 3 measurements points within 4 hours after iohexol administration (179). They found no benefit for GFR calculation by extending the measurement to 5 hours across the range of GFRs.

Furthermore, irrespective of how accurately this study has assessed 'true' GFR, the conclusions regarding the relative performance of the two GFR estimating equations remain valid. This data suggests that GFR estimation using the MDRD and CKD-EPI estimating equation in older Caucasian people is as accurate as it has been reported to be in younger individuals. Although calls for the adoption of the CKD-EPI equation into regular clinical practice have been made, it fails to consistently achieve the 2002 KDOQI recommendations of P_{30} values > 90%. Newer estimating equations more recently have been proposed which claim to have less bias and better accuracy than the MDRD and CKD-EPI equations. The BIS group proposed a new equation (BIS) which is reportedly unbiased against measured GFR and achieved P_{30} values of 95% in an older population (179). Further work from this study has gone on to evaluate the performance of the BIS equation in this study cohort (180). In contrast to Schaeffner *et al*, the BIS equation was negatively biased compared to measured GFR especially in those individuals with GFR ≥ 60ml/min/1.73m^2 and negligible in individuals with GFR < 60ml/min/1.73m^2. Both cohorts however were of northern European white origin with broadly similar age, gender, body habitus and co-morbidity characteristics and validation and experience of the BIS equation remains

restricted to older white populations. Further studies are required to confirm that the BIS equation performs well in a younger cohort and in other ethnic groups as adopting various estimating equations in different age groups may be outweighed by the practical difficulties applying them in clinical practice.

Many of the inaccuracies of these estimating equations are down to the biological variability of serum creatinine. Alternative filtration markers such as β-trace protein, cystatin C and symmetric dimethyl arginine (SDMA) have shown promise in improving estimating equation performance in. Perhaps the most studied is cystatin C which is thought to be more accurate a measure of renal function as is it not believed to be related to muscle mass or diet. In a pooled dataset of 3,134 people with CKD, cystatin C levels alone provided GFR estimates that were nearly as accurate as serum creatinine level adjusted for age, sex and race (181). Several equations based on serum cystatin C have been created to estimate GFR. Recently the CKD-EPI group published 2 additional CKD-EPI equations: one based on cystatin C concentrations (CKD-EPI$_{cys}$) and once using both cystatin C and serum creatinine (CKD-EPI$_{cr\text{-}cys}$) (121). They demonstrated better performance of the equations combining creatinine and cystatin C compared to the CKD-EPI equation. In the external validation datasets of Inker *et* al, the CKD-EPI$_{cys}$ and the CKD-EPI$_{cr\text{-}cys}$ equations achieved P_{30} values of 86% and 92% respectively. The BIS group developed the BIS2 equation which utilises both creatinine and cystatin C and reported P_{30} values of 96% in older people (179). Further work from this study performed by Carter *et al*, has evaluated these 2 cystatin C based estimating equations in older people using serum from the study time 0 samples (180, 182). They reported broadly equivalent bias, precision and accuracy in comparison to the creatinine based estimating equations. Recent studies have also confirmed the good performance of the cystatin C based equation in specific populations such as patients with diabetes and individuals with mild to moderately impaired kidney function (183). This better performance is also supported by studies that show a better estimate of mortality risk compared to creatinine based equations (184-185). These cystatin C equations have however been developed by the use of a limited sample of test subjects and have yet to be validated across a wide range of populations and

there have been concerns regarding the lack of standardisation of cystatin C assays and the unavailability of the assay in many centres. Nonetheless, these cystatin C based equations show promise and have been adopted in the 2014 NICE CKD guidelines as an additional diagnostic tool for patients with a borderline diagnosis of CKD which may result in a significant proportion of people reclassified as not having CKD.

In an ideal world, in order to overcome its limitations, improvement of this study would incorporate a larger dataset across a diverse ethnic population with a wide range of co-morbidities and of varying body surface areas. To silence critics of the selected reference method, both iothalamate and iohexol GFR would be measured to enable better comparison with other studies and also assess the performance of the 2 reference methods comparably. Patient abstinence from ingestion of meat and fish for at least 12 hours prior to sampling would need to be enforced to reduce biological variation of creatinine levels. Timed urine samples to measure urine creatinine and iohexol levels and a 24 hour serum sample collection would be included in the protocol to enhance accuracy of iohexol GFR measurement at lower GFR levels. The problem with this, however, is that in order or the equations to be applicable in clinical practice, one would have to enforce these measures in routine clinical practice. With a larger dataset, analysis of performance in more specific subgroups, particularly the very elderly, those with low body mass and in the different CKD stage GFR categories would highlight in which subjects caution is needed in interpreting GFR. Misclassification errors at all the stages of CKD would help to predict what the implications of adopting the CKD-EPI equation into routine practice in the future will be. The inaccuracies of the estimating equations fall largely down to the biological and, to a lesser degree, analytical variability of serum creatinine levels so measurement of other markers of renal function, such as cystatin C, in this study design would help to determine a better marker for measuring renal function that is accurate, quick, practical to apply in a large-scale basis and transferable across wide range of populations. The definition of CKD is however subject of debate most notably in the elderly (186-187). These aspirations however fall outside the remit of the study which set out to assess whether the prevalence of

CKD is overestimated due to the inaccuracies of the estimating equations in the elderly population.

5.3 Conclusion

In conclusion, this study has demonstrated good performance of the current estimating equations in an independent validation cohort inclusive of a large number of older people drawn from secondary care and the community with co-morbidity and pharmacotherapy typical of such populations in the western world. Although it falls short of the > 90% P_{30} aspiration of the 2002 KDOQI guideline (10), the GFR estimating equations appear to work just as well in older compared with younger populations. This study found no evidence that the MDRD study equation was underestimating GFR, irrespective of the level of GFR. The CKD-EPI equation performed marginally better than the MDRD study equation particularly at GFR ≥ 60 ml/min/1.73m^2. This study has found discrepancies in CKD prevalence in using both estimating equations with a tendency to misclassify individuals as not having CKD or stage 1-2 CKD when in fact measured GFR classified them as stage 3a CKD. More epidemiological studies using GFR measurements with a reference method are urgently required to evaluate the newer estimating equations across a broad population. This work has led on to the eGFR-C study which is currently recruiting and aims to compare the accuracy and precision of the CKD-EPI estimating equations based on creatinine and cystatin C over a 3 year period. Its aims are to assess its performance in people with CKD stage 3 according to ethnic groups (particularly caucasians, african-caribbeans and south-asians) and patients with diabetes and albuminuria and to establish which estimating equation better predicts CKD progression.

CHAPTER 6

Multiple AKI Episodes and CKD Progression

If the estimating equations we use to determine GFR are accurate, the question remains why the prevalence of CKD is so high when the numbers progressing to ESRD are so low. Epidemiological studies have shown an independent graded association with CKD, death and cardiovascular events with an increased risk ratio of death of 3.2 and 5.9 in CKD stages 4 and 5 respectively (188). Mortality rates alone do not account for the discrepancy in numbers. CKD progression is defined by as a sustained decline in eGFR of > 5ml/min/1.73m^2/year (12). The use of such a definition suggests that progression of disease occurs in a linear pattern. In clinical practice, the use of this definition may be limited by few GFR estimates or a limited time.

Albuminuria, underlying renal diagnosis, hypertension and low GFR are associated with progression of CKD (29-32), however some patients with these risk factors do not progress and conversely there are many patients who progress to ESRD without these risk factors. One possible explanation is that episodes of AKI lead to the development or progression of CKD. The integrated syndrome between AKI and CKD is still largely being discovered. CKD is a risk factor for AKI and studies have shown that incomplete recovery from a hospital-managed AKI event may result in new incident CKD and that severity of hospital-managed AKI predicts progression to chronic kidney disease. AKI can be hospital-acquired, community-acquired admitted to hospital and community-acquired and managed by primary care. Little is known about the characteristics and outcomes of the latter subgroup and clinicians may not pick up many of these events.

Perhaps a proportion of people with CKD have periods of stable renal function followed by an acute illness manifesting as an AKI event with incomplete recovery back to baseline but a subsequent period of stability in function. Many patients however who survive an AKI event eventually fully recover renal function. What determinants are associated with full renal recovery and are these factors modifiable?

6.1 Hypothesis

This second study tests the hypothesis that multiple community-AKI events are an independent risk factor for progression of CKD.

6.2 Aims

The main objective of this study was to determine whether multiple episodes of AKI occurring in the community are associated with progression of CKD in a cohort or patients with CKD referral to renal services. A secondary objective was to examine the different patterns of CKD progression that occurred in this CKD population leading to referral.

6.3 Methods

This was a retrospective observational cohort study examining the characteristics and CKD disease pattern in a selected group of patients referred to the Department of Renal Medicine within the catchment area of EKHUFT, United Kingdom.

Cohort Definition

All new adult patients referred to the Department of Renal Medicine, EKHUFT, outpatient services between 1st April 2005 and 31st March 2006 with stage 3-5 CKD

were included. Referrals were received from primary care providers or secondary care specialists.

All patients younger than 18 years at referral were excluded. Any patients residing outside Kent or from the West Kent catchment area were excluded due to lack of access to historical pathology data. Patients with eGFR measurements greater than 60ml/min/1.73m^2 at time of referral were removed to exclude any patients without a diagnosis of CKD. Any patients who had limited pathology data dating back less than one year, or with less than four serum creatinine measurements prior to referral date were removed from the study as it was deemed insufficient to determine progression of CKD. Patients coded as having a diagnosis of end-stage renal failure undergoing renal replacement therapy and renal transplant recipients were excluded.

Ethical approval was granted by the National Research Ethics Service (NRES) Committee South East Coast and the Research and Development department, EKHUFT supported this study.

Data Extraction

Demographic data were collected, which included age at referral, eGFR at referral, gender, and co-morbidity prevalence from referral letters, patient notes and hospital computer patient records. Primary cause of CKD (if known) and co-morbidity data were also obtained from the Department of Renal Medicine's renal patient database system (*RenalPlus*). Co-morbid conditions were grouped into the following categories; urological, malignant, cardiovascular, diabetes and hypertension. All biochemistry requests in the East Kent catchment requested in the community or in hospital are processed in the EKHUFT biochemistry laboratories and therefore available on local pathology databases for this study. The date and result of every serum creatinine measurement for each individual was recorded together with any clinical information provided at the time of the pathology request. No pathology data was available prior to 1998. Estimated GFR was estimated from serum

creatinine using the MDRD study equation (8). Although this study earlier demonstrated superiority of the CKD-EPI compared to the MDRD study equation, the MDRD study equation was used for this study as it was carried out prior to the CKD-EPI validation study was reported and it is currently still national practice to report eGFR using the MDRD study equation. To remove systematic bias in creatinine and eGFR levels, calibrated and standardised creatinine assays were used in the study as per KDIGO guidelines for all serum creatinine results after April 2006 (10). Creatinine assays used in Kent were directly calibrated to the method employed by the central laboratory used for the MDRD Study (Beckman Rate Jaffe/CX3 Synchron assay). This in turn enabled indirect calibration of the other creatinine assays used in the laboratories prior to April 2006 to ensure comparable creatinine and eGFR levels in the different time periods (155).

Pathology databases were interrogated for albuminuria and proteinuria data. Results and dates were recorded of the first measured urine ACR or urine PCR (mmol/ml) before and after referral. Levels of albuminuria and proteinuria were graded 1-3 according to the albuminuria categories recommended by KDIGO in 2012. Grade 1 was defined as no significant albuminuria i.e. a urine ACR < 3mg/mmol (< 30mg/g), Grade 2 was defined as an urine ACR 3-30 mg/mmol (30-300 mg/g) or urine protein creatinine ratio between 5-50mmol/ml, and Grade 3 was defined as an ACR > 30mg/mmol (> 300mg/g) or a urine PCR > 50 mg/mmol. Length of follow up was calculated from the date of the first serum creatinine measurement to the most recent recorded result as there were varying start dates.

Data Interpretation

Accumulated demographic and renal function data was entered into a *Microsoft Access* database and graphs of eGFR versus time were plotted for each patient.

Stable vs Progressive CKD

Patients were divided into stable or progressive CKD according to rate of decline of kidney function.

Stable CKD was defined as a decline in eGFR of < 10 ml/min/m^2 over 5 years or <2 ml/min/m^2 per year over the total observation period accordance with the KDIGO guidelines, 2012 (72).

Progressive CKD was defined as a decline in eGFR of > 2 ml/min/m^2 per year or 10 ml/min/m^2 over 5 years from the baseline over the total observation period.

Baseline creatinine was calculated as the median creatinine over previous 365 days prior to the recorded result but any creatinine values measured during AKI events were excluded from baseline calculations.

Linear regression lines were fitted to each eGFR trajectory to aid analysis of eGFR trajectories.

AKI Episodes

Any acute decline in kidney function from the predicted eGFR trajectory was assessed as to whether it fulfilled criteria for an AKI episode. We used serum creatinine levels to assess incidence of AKI. For each event we calculated baseline creatinine as the lowest serum creatinine in the 12 months prior to the acute rise to define AKI.

An episode of AKI was defined as either a rise in serum creatinine > 26µmol above baseline or an acute rise in creatinine > 1.5 times above baseline creatinine as per AKI network criteria introduced in 2007(69). This study was carried out prior to the publication of the KDIGO AKI guidelines (72).

AKI was treated as a binary variable, in other words the severity of AKI was not assessed because the frequency of AKIN 2 and AKIN 3 events were very low. The total number of AKI episodes was determined for each patient. 'Multiple AKI' was defined as 2 or more AKI events and patients were categorised into those with and those without multiple AKI events. Mean age at referral, mean eGFR at referral, gender and co-morbidity were compared between the 2 subgroups. The presence of

multiple AKI episodes were then compared between the stable and progressive CKD groups.

Linear and Stepwise Progression Subgroups

Two patterns of CKD progression emerged, therefore patients with CKD progression were further sub-divided into Linear and Stepwise (non-linear) subgroups according to their pattern of eGFR trajectory. Linear decline was defined as individuals with eGFR trajectories with close fit to the linear regression lines. Stepwise was identified as individuals with a fall in eGFR followed by a period of stability in kidney function without recovery of eGFR to the predicted baseline. The stepwise sub-group were characterised by deviation from the linear regression line with either a more rapid decline than that predicted or a period of stability. A period of stability was defined as a one year period with no decline in eGFR or improvement in eGFR from that predicted.

The Delphi technique is a method of obtaining a consensus amongst a panel of experts. Using the Delphi technique (189), five nephrologists independently analysed each eGFR vs time graph for all study patients. The panel were initially asked to assess the pattern of progression and individually categorise each patient into one of the three groups: stable, linear and stepwise. The Cronbach's alpha calculation is a test of reliability and a measure of internal consistency and this was applied to the panel's individual analyses of the progression patterns. A Chronbach's alpha reliability of 0.70 or higher indicates a reasonable level of agreement. The Crohnbach's alpha reliability for this study was 0.83 indicating a good level of agreement. Cases where agreement in classification of pattern was not initially obtained were re-discussed in a further meeting with all five nephrologists for a unanimous decision.

Statistical Analysis

Gender, cause of renal disease, co-morbidity prevalence and grade of albuminuria were compared between each category and the percentage prevalence within the study population was calculated. Skewness and kurtosis were determined for age at referral and eGFR at referral to determine whether data were normally distributed. Age at referral was not normally distributed whereas eGFR at referral was. Mean values and standard deviations were calculated accordingly. Data that was not normally distributed was log transformed to allow statistical analysis.

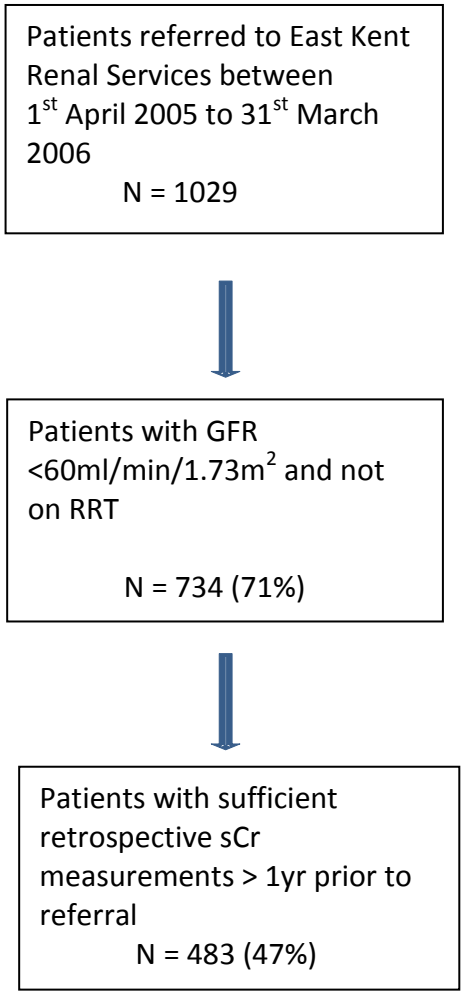
The Pearson Chi-squared test assesses the relationship between two categorical variables. The Pearson Chi-squared test was therefore used to test whether there was an association between each of the variables; co-morbidity, gender and grade of albuminuria, between the stable CKD and CKD progression groups. Statistically significance was considered with p values <0.05.

Analysis of Variance (ANOVA) was then applied to determine whether there was an association between each of the two continuous variables mean age and mean eGFR at referral, and CKD progression. A p value of < 0.05 was considered statistically significant.

Demographics of patients with multiple AKI episodes were compared with the stable and progressive group and the Chi-squared test was used to determine association between each variable and progression. Any significant categorical variables were then included in a logistic regression analysis to allow for any potential non-linearity in the risk relationships to CKD progression. This was performed to determine whether these categorical variables had a dependent or independent association with CKD progression. Again, a p value of < 0.05 was considered to show statistical significance. These statistical tests were repeated in the subgroup analyses to identify any variables associated with linear or stepwise patterns of CKD progression.

All statistical analyses were carried out using IBM *SPSS Statistics* program.

Figure 6.1 Flow Chart of Selection Process



Abbreviations: sCr, serum creatinine; N, number; GFR, glomerular filtration rate; yr, year; RRT, renal replacement therapy.

Figure 6.1

Flow chart illustrating the selection process of study subjects. Figure in brackets represents percentage of the total number of patients referred to the Department of Renal Medicine, East Kent within the study period of April 2006 - April 2007. A total of 1029 subjects referred to Kent renal services. 295 were excluded as they either did not have a GFR < 60ml/min/1.73m^2 or had end-stage renal failure. Only 483 subjects were included in the study. The remainder excluded were either from West Kent with out of area laboratory data or had insufficient retrospective biochemistry results dating back < 1 year prior to the study period or < 4 prior serum creatinine levels.

6.3 Results

Figure 6.1 illustrates the study selection process. A total of 1029 people were referred to the Department of Renal Medicine, EKHUFT's outpatient services between 1st April 2005 and 31st March 2006 as recorded on our CKD Referral database. There were 295 patients excluded as they either had ESRD and were receiving renal replacement or had an eGFR > 60ml/min/m^2 at the time of referral and may have been referred with structural abnormalities or hypertension. Of the 734 patients remaining, 164 were excluded as they resided out of the East Kent catchment area hence did not have pathology data recorded on our local pathology databases. A further 69 subjects were excluded due to pathology data dating back less than 1 year prior to referral and 18 were excluded as they had less than 4 measured creatinine values recorded prior to referral. After all exclusions, a total of 483 subjects were included in the study.

Patient Characteristics

Baseline characteristics of the patients are summarised in Table 6.1. There were 274 males (57%) and 209 females (43%) in total. As age and eGFR data at referral were normally distributed, mean and standard deviation were expressed. The mean age of patients at the time of referral was 73.2 years (SD 12.43) with a range from 23 to 96 years. The mean eGFR of patients at referral was 36.1 ml/min/m^2 (SD 12.1). The highest prevalent co-morbid disease was hypertension, this was present in 78.7% of study patients. The second most common co-morbid condition was cardiovascular disease (44.1%) followed by diabetes mellitus (38.9%) and 10.8% patients had a documented malignancy.

Coding of cause of CKD was obtained from our renal patient database and recording of diagnosis was often incomplete. Patients with incomplete records were categorised as having an unknown aetiology of CKD. As this group accounted for the majority of the patients (65%), statistical analysis was not performed on aetiology of CKD. The commonest coded cause of CKD was renovascular or hypertensive renal

disease accounting for 10.4% cases. Urological condition accounted for 9.3% patients and diabetic nephropathy was the primary aetiology in 7%. Very few (3.5%) had a histologically proven primary intrinsic renal pathology or inherited genetic renal condition as a primary renal diagnosis.

Albuminuria data was not available on 82 patients. Of the 401 with recorded albuminuria data, 26.7% subjects had A1 (ACR < 3 mg/mmol or PCR < 5 mg/mmol), 33.7% subjects had A2 (ACR 3-30 mg/mmol or PCR 5-50mg/mmol) and 22.6% patients had documented significant albuminuria (A3) at referral with an ACR >30 mg/mmol or PCR >50mmol/ml.

Stable vs Progressive CKD

Of the total 483 subjects studied, 309 (64%) had stable CKD whereas 174 (36%) individuals were classified as having progressive CKD. There were a higher proportion of men in both groups with a male to female ratio of 55:45 in the stable group and 59:41 in the progressive group. Gender was not found to be associated with CKD progression.

The mean age at referral was 73.1 years (SD 11.6) in the stable group and 74.2 (SD 10.8) in the progressive group and this did not reach statistical significance. There was a significant difference in mean eGFR at referral between the stable and progressive groups with mean eGFR of 39.5 ml/min/1.73m^2 (SD 11.7) and 30.3 ml/min/1.73m^2 (SD 11.2) respectively suggesting a lower eGFR at referral is associated with progression of CKD.

Table 6.1 Demographics of Patients

Demographics	Total (n=483)	Stable (n=309)	Progressive (n = 174) Total	Linear (n=58)	Stepwise (n=115)
Gender					
Male , %	56.7	55	59.2	60.3	59.1
Female, %	43.2	44.5	40.8	39.7	41.7
Mean Age at Referral, years (SD)	73.2 (11.3)	73.2 (11.6)	74.2 (10.8)	72.4 (12.4)	75.1 (9.8)
Mean eGFR at Referral, ml/min/m^2 (SD)	36.1 (12.1)	39.5 (11.7)	30.3 (11.2)*	31.5 (12.1)	29.7 (9.3)
Co-morbidity, %					
Hypertension	78.7	78.3	79.3	82.8	78.3
Diabetes	38.9	35.2	45.4*	41.4	47.8
Cardiovascular	44.1	69.5*	52.9	50	54.8
Malignancy	10.8	8.4	14.9*	13.8	15.7
Cause of CKD, %					
Urological	9.3	7.1	13.2	0	20
Drug related	4.1	5.5	1.7	1.7	1.7
Reno-vascular	10.4	9.1	12.6	8.6	14.8
Diabetes	7.0	5.5	9.8	10.3	9.6
AKI	0.6	1.0	0	0	0
Primary renal	3.5	2.3	5.2	8.6	3.5
Unknown	65	69.9	56.9	70.7	50.4
Albuminuria grade	n = 401	n = 257	n = 144	n = 48	n = 96
A1, ACR<3, %	32.2	38.9*	19.4	20.8	18.8
A2 ACR 3-30, %	42.1	40.5	41	39.6	41.7
A3 ACR>30, %	27.2	20.2	39.6*	39.6	39.6

Abbreviations: CKD, chronic kidney disease; ACR albumin to creatinine ratio; AKI, acute kidney injury; n, number; eGFR, estimated glomerular filtration rate; SD, standard deviation.
* Indicates p values reached statistical significance ($p < 0.05$).

Table 6.1 shows the characteristics of total study cohort. Patients were grouped into stable and progressive CKD according to rate of decline in eGFR and patients with progressive CKD were further subdivided into linear or stepwise progression according to their pattern of eGFR versus time trajectories. Pearson's Chi-squared test was used to determine association between the categorical variables gender, co-morbidity and albuminuria with progression whereas ANOVA was used to determine association of eGFR and age at referral with progression. Cause of renal disease was not analysed due to poor recording of data but is tabulated for interest. The variables associated with CKD progression were low eGFR at referral, presence of diabetes or malignancy and grade A3 albuminuria, whereas presence of hypertension and grade A1 albuminuria appear to be protective against progression. There were no significant variants between the linear and stepwise progressors.

The prevalence of hypertension was similar in both groups (stable 78.3% and progressive 79.3%) but there were a significantly higher proportion of diabetics in the progressive group compared to the stable group (45.4% vs 35.2%). Malignancy was also associated with progression with a prevalence of 14.9% in the progressive group and only 8.4% in those with stable CKD ($p = 0.03$). Cardiovascular disease appeared to be protective against progression and was more prevalent in the stable group (69.5%) compared to the progressive group (52.9%).

Of the 309 subjects with stable CKD, 32.7% had no significant albuminuria compared to 16.1% subjects with progressive CKD. Grade A2 albuminuria was present in equal proportions in the stable and progressive group (33.7% and 33.9%) however there was a statistically significantly higher proportion of patients with A3 albuminuria in the progressive group (32.8%) compared to stable CKD (16.8%). The results suggest that the presence of significant albuminuria (A3) is associated with CKD progression.

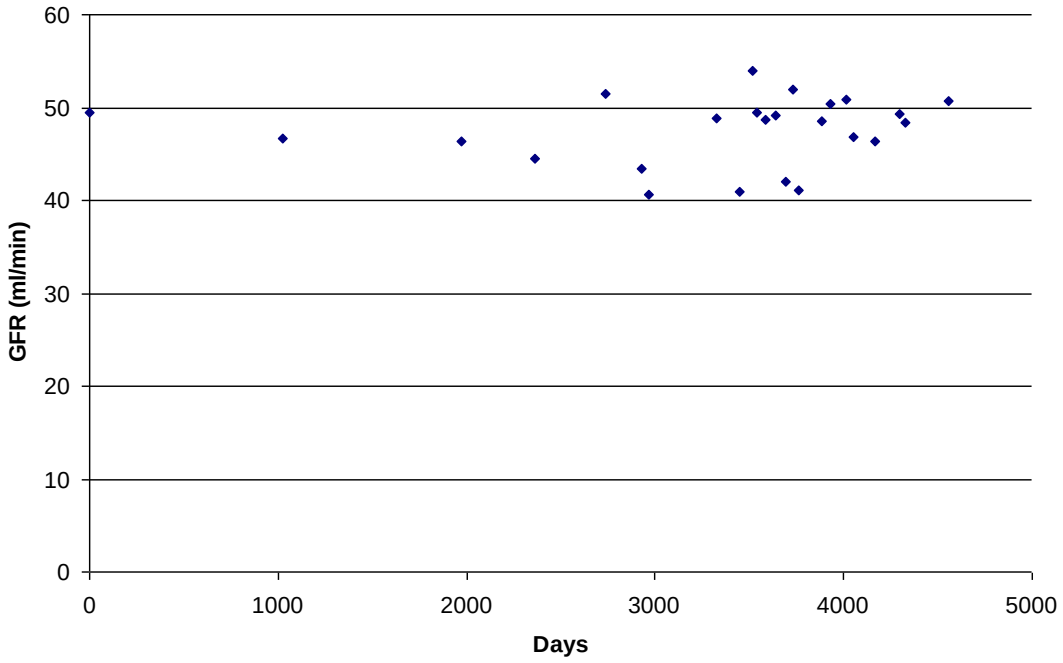
Linear and Stepwise Sub-groups

Graphs of eGFR versus time were plotted for each individual studied to aid analysis of progression pattern. On further analysis of the progressive group, there appeared to be two patterns of progression; linear and stepwise. The linear sub-group had eGFRs that appeared to decline in a linear pattern whereas the stepwise sub-group had a decline in baseline eGFR followed by a variable period of stability in renal function. Examples of each category; stable, linear and stepwise are illustrated in Figures 6.2-6.5. Of the 174 patients with progressive CKD, 66% were categorised as following a stepwise decline in function and 33% following a linear pattern of decline.

Patient characteristics between linear and stepwise sub-groups were analysed and found to be broadly comparable. Male to female ratios were similar between both the linear and stepwise groups and there was no significant difference in mean age at referral (75.1 (SD 9.8) years in the stepwise group compared to 72.4 (SD 12.4) years in the linear group) between the two subgroups. Mean eGFR at referral was

29.7 ml/min/1.73m^2 in the stepwise group and 31.5 ml/min/1.73m^2 in the linear group. Analysis of the co-morbidities between the two sub-groups did not reach statistical significance and albuminuria data was similar in both groups with 32.8% and 33% of the linear and stepwise groups respectively documented to have A3 albuminuria at referral.

Figure 6.2 Estimated GFR Versus Time Graph of a Representative Patient with Stable CKD and No AKI Events



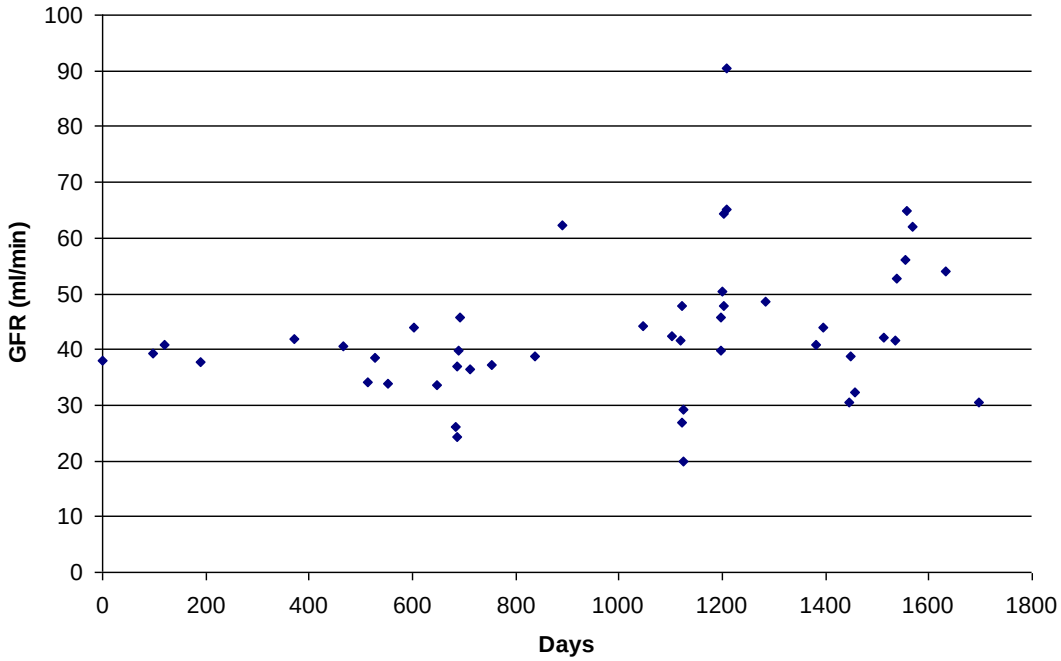
Abbreviations: GFR, glomerular filtration rate; eGFR, estimated glomerular filtration rate; CKD, chronic kidney disease; AKI, acute kidney injury

Figure 6.2

Estimated GFR (ml/min/1.73m^2) results are plotted against time from first recorded eGFR in this subject. Estimated GFR was calculated using the MDRD study equation based on serum creatinine levels. An AKI episode was defined as a rise in serum creatinine > 26µmol or > 1.5 x above baseline creatinine. Baseline creatinine was calculated as the median creatinine over the previous 365 days. Any creatinine values measured during AKI events were excluded from baseline calculations. Stable CKD was defined as a decline in eGFR of < 10 ml/min/1.73m^2 over 5 years or < 2 ml/min/1.73m^2 per year.

This scatter plot is a graphic example of a patient with stable CKD with no recorded episodes of AKI.

Figure 6.3 Estimated GFR Versus Time Graph of a Representative Patient with Stable CKD and AKI with Complete Recovery to Baseline eGFR



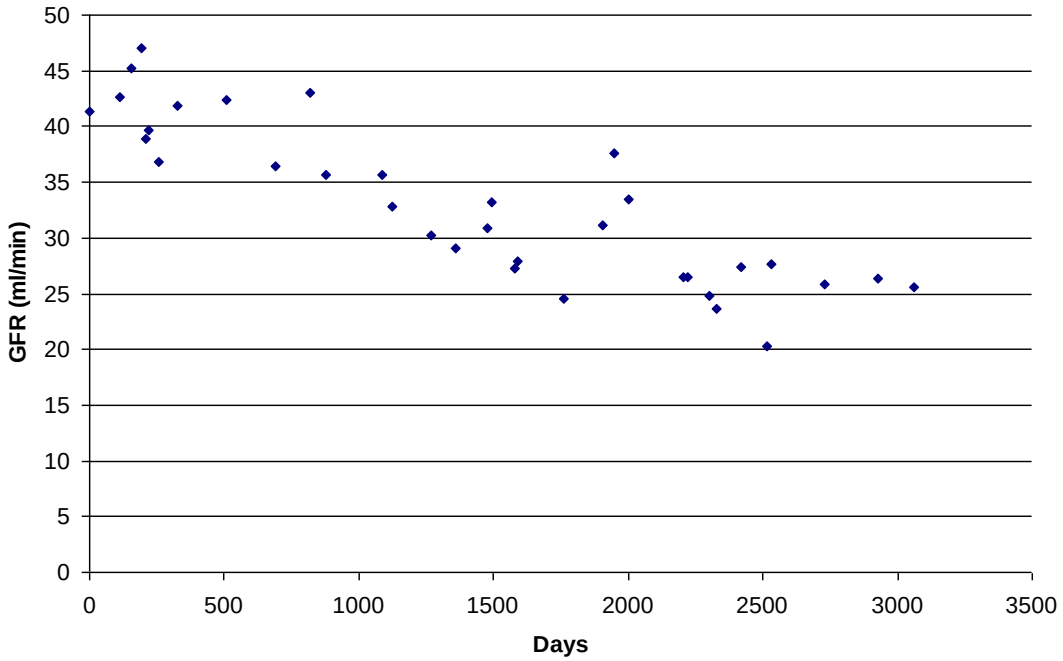
Abbreviations: GFR, glomerular filtration rate; eGFR, estimated glomerular filtration rate; CKD, chronic kidney disease; AKI, acute kidney injury

Figure 6.3

Estimated GFR (ml/min/1.73m^2) results are plotted against time from first recorded eGFR in a subject. Estimated GFR was calculated using the MDRD study equation based on serum creatinine levels. An AKI episode was defined as a rise in serum creatinine > 26μmol or > 1.5 x above baseline creatinine. Baseline creatinine was calculated as the median creatinine over the prior 365 days. Any creatinine values measured during AKI events were excluded from baseline calculations. Stable CKD was defined as a decline in eGFR of < 10 ml/min/1.73m^2 over 5 years or < 2 ml/min/1.73m^2 per year.

This scatter plot is a graphic example of a patient with stable CKD with multiple AKI events followed by full recovery to baseline.

Figure 6.4 Estimated GFR Versus Time Graph of a Representative Patient with Linear Progression with No AKI Events



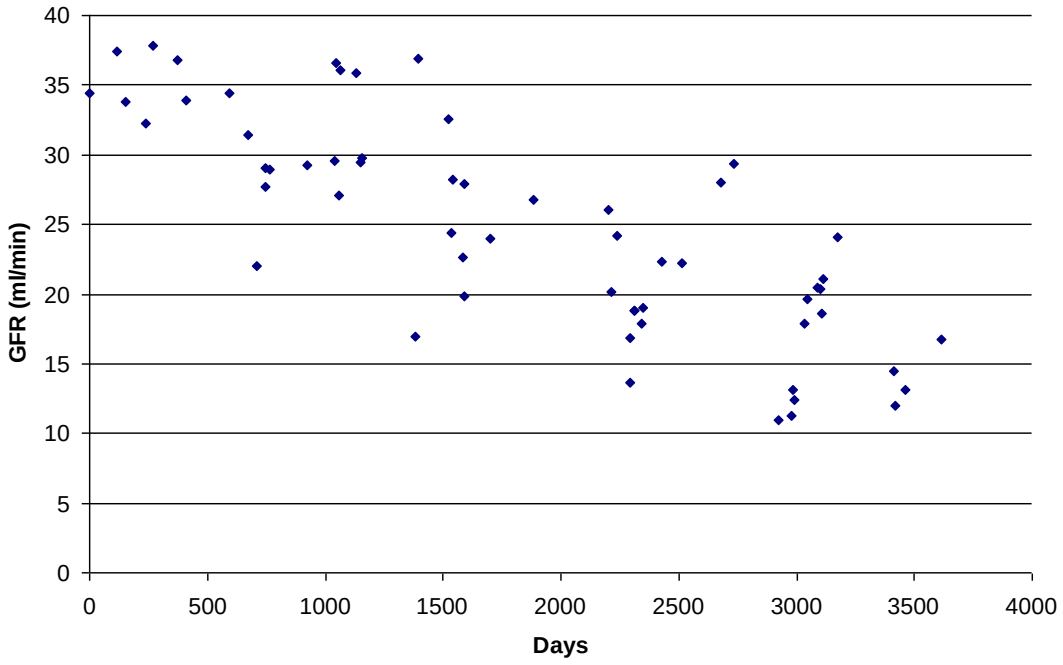
Abbreviations: GFR, glomerular filtration rate; eGFR, estimated glomerular filtration rate; CKD, chronic kidney disease; AKI, acute kidney injury

Figure 6.4

Estimated GFR (ml/min/1.73m^2) results are plotted against time from first recorded eGFR in a subject. Estimated GFR was calculated using the MDRD study equation based on serum creatinine levels. An AKI episode was defined as a rise in serum creatinine > 26µmol or > 1.5 x above baseline creatinine. Baseline creatinine was calculated as the median creatinine over the prior 365 days. Any creatinine values measured during AKI events were excluded from baseline calculations. Progressive CKD was defined as a decline in eGFR of > 10 ml/min/1.73m^2 over 5 years or > 2 ml/min/1.73m^2 per year.

This scatter plot is a graphic example of a patient with progressive CKD with a linear declining trajectory who had no episodes of AKI and represents the linear sub-group.

Figure 6.5 Estimated GFR Versus Time Graph of a Representative Patients with Stepwise Progression Associated with Multiple Episodes of AKI



Abbreviations: GFR, glomerular filtration rate; eGFR, estimated glomerular filtration rate; CKD, chronic kidney disease; AKI, acute kidney injury.

Figure 6.5

Estimated GFR (ml/min/1.73m^2) results are plotted against time from first recorded eGFR in a subject. Estimated GFR was calculated using the MDRD study equation based on serum creatinine levels. An AKI episode was defined as a rise in serum creatinine > 26µmol or > 1.5 x above baseline creatinine. Baseline creatinine was calculated as the median creatinine over the prior 365 days. Any creatinine values measured during AKI events were excluded from baseline calculations. Progressive CKD was defined as a decline in eGFR of > 10 ml/min/1.73m^2 over 5 years or > 2 ml/min/1.73m^2 per year.

This scatter plot is a graphic example of a patient with progressive CKD with multiple AKI events with partial recovery to a new baseline for a period of stability followed by a further AKI event. This represents a stepwise progression.

AKI Episodes

The number of AKI episodes in the Stable, Linear and Stepwise subgroups is shown in Table 6.2. Patients were further subdivided in those with no AKI events, 1 AKI event and multiple AKI events (≥ 2 AKI events). Recorded serum creatinine data varied significantly in length of time from first test to date of referral and mean years of follow up for each group is also listed in Table 6.3 with a mean follow up period of 5.94 years for each subject.

There were 389 recorded episodes of AKI in total with 162 episodes occurring in the stable subjects and 146 episodes in the progressive CKD subjects. A total 55.9% of the study group did not have an episode of AKI, 30.2% had 1 episode of AKI and 14.3% had 2 or more episodes of AKI. Of the 309 patients with stable CKD, 63.1% did not have a recorded episode of AKI whereas 43.1% of the 174 progressive subjects did not have an AKI episode. In the progressive CKD group, 35.1% had 1 AKI episode compared to 27.2% of the stable group. The presence of multiple AKI episodes were statistically associated with progression occurring in 20.7% of the progressive CKD group and only 10.7% of the stable group using the Chi-squared test. In the subgroup analysis of the progressive CKD group, there were fewer linear patients with an AKI episode with only 24.2% with 1 AKI event and 5.2% had multiple AKI events. In contrast, 40.9% of the stepwise group had 1 AKI episode and 28.7% had multiple AKI events. These results suggest that multiple AKI events are associated with CKD progression.

The characteristics of patients grouped into less than two or two or more AKI episodes are shown in Table 6.4. There was no significant association of male gender with multiple AKI episodes and mean age was similar of the two groups. There was however, a significant difference in mean eGFR at referral with a mean of 36.9 ml/min/1.73m^2 in those with less than two AKI episodes and 31.8 ml/min/1.73m^2 in those with two or more. This study suggests that lower eGFR at referral is associated with multiple AKI events.

Table 6.2 Table of the Number of AKI Events in Those with Stable CKD, Linear or Stepwise Progressive CKD

Number of AKI Events	Total, % (n=483)	Stable, % (n=309)	Progressors, %		
			Total (n=174)	Linear (n=58)	Stepwise (n=115)
0	55.9	63.1*	43.1	70.7*	29.6
1	30.2	27.2	35.1	24.1	40.9*
≥ 2	14.3	10.7	20.7*	5.2	28.7*
Total AKI episodes	309	162	146	20	126
Mean total yr of FU/pt	5.94	5.77	6.15	5.84	6.45

Abbreviations: n, number; AKI, acute kidney injury; yr, year; FU, follow up; pt, patient.
* Indicates reached statistical significance ($p < 0.05$)

Table 6.2

This table shows the number of AKI episodes in patients with stable and progressive CKD. Progressive CKD was defined as a decline in eGFR of > 10 ml/min/1.73m^2 over 5 years or > 2 ml/min/1.73m^2 per year and this group were further subdivided into linear and stepwise sub-groups according to their pattern of eGFR decline. The remaining patients had stable CKD. An AKI episode was defined as a rise in serum creatinine > 26µmol or > 1.5 x above baseline creatinine. Baseline creatinine was calculated as the median creatinine over the prior 365 days. Any creatinine values measured during AKI events were excluded from baseline calculations. Multiple AKI episodes were defined as ≥2 AKI events. Mean follow up periods for each category are shown. Multiple AKI events were compared between stable and progressive groups and between linear and stepwise subgroups using the Pearson Chi-squared test and p values < 0.05 were considered to be statistically significant. No AKI events were associated with stable CKD however in the subgroup analysis there was an association with no AKI events and the linear progressors. Multiple AKI events were significantly more prevalent in the progressive group more specifically within the stepwise subgroup.

When comparing co-morbidity between those with less than two or two or more AKI events, hypertension was statistically more prevalent in those with less than two AKI events compared to those with two or more AKI events (80.2% vs 69.5%). Diabetes was more prevalent in those with multiple AKI episodes versus less than 2 AKI events (55.1% vs 36.2%) as was cardiovascular disease (63.8% vs 40.8%). This study found no association between malignancy and albuminuria categories with multiple AKI events.

In the subgroup analysis of the progressive group, only 5.2% of linear progressors had multiple AKI episodes compared to 28.7% stepwise progressors which reached statistical signficance. Of the linear progressors, 70.7% had no AKI episodes compared to 29.6% of stepwise progressors. This study suggests that low eGFR at the time of referral, presence of diabetes and presence of cardiovascular disease are associated with multiple AKI events.

All variables found to be significantly associated with CKD progression were then included in a logistic regression analysis to determine any potential non-linearity in the risk relationships to CKD progression. Presence of cardiovascular disease, diabetes, malignancy, multiple AKI events, albuminuria and eGFR at referral were included as variables in the nominal regression. The likelihood ratio showed that low eGFR at the time of referral, presence of diabetes and A3 albuminuria were independently associated with CKD progression whereas malignancy, cardiovascular co-morbidity and multiple AKI episodes were not independent variables.

In summary, this study has demonstrated that in a selected population of people referred to nephrology services with CKD, a significant proportion progress. Of those who progress, the majority follow a stepwise pattern of decline compared to a linear decline. Low eGFR at presentation, diabetes, and severe albuminuria were independently associated with CKD progression. This study also found that multiple AKI events were also associated with CKD progression in particular the stepwise progressors and this factor was the only significant determinant between linear and stepwise progressors.

Table 6.3 Demographics of Patients With or Without Multiple AKI Episodes

Demographics	<2 AKI Episodes (n= 414)	≥2 AKI Episodes (n= 69)
Gender		
Male, %	56	60.9
Female, %	44	39.1
Mean age at referral, years (SD)	73.2 (10.8)	75.5 (11.1)
Mean eGFR at referral, ml/min/m^2 (SD)	36.9 (12.1)	31.8 (11.8)*
Co-morbidity, n (%)		
Hypertension	80.2*	69.5
Diabetes	36.2	55.1*
Cardiovascular	40.8	63.8*
Malignancy	10.9	10.1
Albuminuria category, %		
A1	27.5	21.7
A2	33.1	37.7
A3	22.5	23.2
Missing data	16.9	17.4

Abbreviations: ACR, albumin to creatinine ratio; eGFR, estimated glomerular filtration rate; SD, standard deviation; n, number; min, minute; AKI, acute kidney injury.
* indicates reached statistical significance ($p < 0.05$)

Table 6.3

This table shows the characteristics in patients with and without multiple AKI episodes. Multiple AKI events were defined as ≥ 2 AKI events in the observation period. An AKI episode was defined as a rise in serum creatinine > 26μmol or > 1.5 x above baseline creatinine. Baseline creatinine was calculated as the median creatinine over the prior 365 days. Any creatinine values measured during AKI events were excluded from baseline calculations. Age at referral, albuminuria and malignancy were not associated with multiple AKI episodes whereas low eGFR at referral, diabetes and cardiovascular disease were associated with multiple AKI episodes using the Pearson Chi-squared test where p values of < 0.05 were considered statistically significant.

CHAPTER 7

Multiple AKI Episodes and CKD Progression

7.1 Discussion

This study examined the characteristics of a cohort of patients with CKD with a high proportion with progressive disease to determine which factors influence progression. A high proportion (36%) of the study patients were classified as having progressive CKD. This is not unexpected in this selected cohort of patients as all patients had CKD and had been referred to renal services. The study cohort was also relatively old with an average age of 73.2 years and fairly representative of the local population of Kent which has a high prevalence of elderly people. Epidemiological studies have shown an increased prevalence of reduced eGFR in older people (1, 2) partially due to co-morbidity and partly due to age-related glomerulosclerosis (23). The eGFR at referral of the cohort was low at 36.1 ml/min/1.73m^2, which may explain the high proportion of subjects with progressive CKD in this study. Low eGFR at referral was found to be independently associated with progression. Epidemiological studies have shown an increased risk of progression in patients with more advanced CKD stages. In a prospective cohort study following 190 patients with CKD, 12 % reached ESRD and 6.5% died (187). Each 30% lower baseline eGFR was associated with a 3-fold higher ESRD rate and a 1.3-fold higher death rate.

There was a high prevalence of hypertension, cardiovascular disease, diabetes and significant albuminuria in the study cohort which is not surprising given the fact that many of these co-morbidities are associated with CKD. This study supports the evidence that there is a strong association with CKD progression and presence of albuminuria. An analysis of trials in patients with and without hypertension or diabetic nephropathy showed that initial changes in albuminuria showed a roughly inverse relationship to the degree of long-term renal function decline (141). Every

50% reduction in albuminuria during the first six months of treatment angiotensin blocking agents was associated with a 45% reduction in risk for ESRD. The GISEN (Gruppo Italiano si Studi Epidemiologica in Nefrologia) group demonstrated that early proteinuria predicted the long-term rate of renal decline (137).

Disappointingly, a high proportion (17%) of this study cohort lacked albuminuria data adding to the limitations of the analysis and highlights a need for more definitive assessment in primary care. The QOF renal indicators incentivising screening for albuminuria were introduced in 2006 which only came in shortly before the study period yet albuminuria recording remained at only 78-81% in patients registered with CKD stages 3-5 in 2008-2010 (190).

Proteinuria is not only a risk factor for CKD progression but has also been shown to be an independent risk factor of the development of AKI but this study did not observe a statistical association with albuminuria and multiple AKI events.
The Alberta Kidney Disease Network Study found that lower baseline eGFR and heavier proteinuria resulted in a significantly higher risk for hospitalisation with AKI (191). Proteinuria alone was associated with a 4.4 fold increased risk of AKI and 7.7 fold increased risk of AKI requiring renal replacement therapy. The definition of proteinuria used in this study however was the presence of 2+ protein on urine dipstick i.e. equivalent to category A2 used in this study.

Hypertension was not independently associated with CKD progression in this study. However, 78.7% of the total study cohort had hypertension resulting in a very small group without hypertension, limiting the ability to draw any conclusions from this result. Diabetes and malignancy were both associated with progression although diabetes was the only independent co-morbid condition associated with CKD progression. This supports several studies that have shown diabetes is a well-established risk factor that progression of CKD and is the leading cause of ESRD worldwide (14-15). Cardiovascular disease was highly prevalent in the study cohort (44.1%) however there was no association with progression of CKD. In contrast, other studies have shown associations between cardiovascular disease and

progression. Obemayr *et al* performed a large longitudinal cohort study with healthy volunteers from the general Viennese population (192). Their results showed that established cardiovascular risk factors predicted the development of new onset kidney disease. In a pooled analysis of the Atherosclerosis Risk in Communities (ARIC) Study and Cardiovascular Health Studies (CHS) of over 13,000 subjects, people with baseline cardiovascular risk had a significantly increased risk of a decline in renal function compared to people without cardiovascular disease (34).

Although cause of CKD was recorded in this study, the most common cause recorded was 'Unknown aetiology' which limits our use of including cause of CKD in the analysis. In clinical practice, CKD patients are often coded with an unknown aetiology as cases are often caused by a combination of multiple causes. In many cases the disease process is too advanced to obtain a true histological diagnosis or the risk of attaining a renal biopsy sample outweighs the potential benefits of a biopsy. This is particularly true for stable CKD but also true in some cases of progressive CKD. If a patient has CKD progressing in the way one would expect for example in a patient with diabetes, we would often not pursue a biopsy therefore cannot make the primary diagnosis diabetic glomerulosclerosis. Cause of CKD however is important in predicting risk for progression. The MDRD study demonstrated that polycystic kidney disease resulted in more rapid progression of CKD than other primary renal diseases. More recently the Study of Heart and Renal Protection (SHARP) study explored the relevance of cause of CKD to kidney progression in 6,245 non-dialysis participants and found cause had substantial prognostic implications not only in determining risk of progression but also in predicting risk of death prior to ESRD particularly in diabetics (48).

Pre-existing CKD, diabetes and albuminuria as well as age and hypertension are all independent predictors of decline in GFR (134, 192-193) yet many CKD patients with these risk factors do not progress. There must be other risk factors that influence CKD progression and emerging evidence suggests that AKI may have a role to play. This study suggests that multiple AKI events that are managed in the community often of less severity may be a significant risk factor leading to progression of CKD.

AKI and CKD were thought be two different entities but it is evident there is a complex interplay between the two condition. Determination of the epidemiology of AKI and CKD and their interaction has been difficult due to the variation in definitions used and the different populations studied. What we do know is that CKD is a recognised independent risk factor for AKI (194) and AKI may lead to the development of CKD or progression of pre-existing CKD (195).

What do we know about the relationship of AKI and CKD? AKI and CKD as separate entities are associated with high morbidity and mortality and one assumes that AKI in CKD has an even more significant effect on outcome but evidence in this area is debatable. This raises the question as to whether the outcome of AKI differs with the presence of background CKD. Some studies have shown AKI in the context of CKD show a lower mortality. It may be 'easier' to get AKI if a patient has CKD therefore people with AKI and no CKD may actually be more unwell thus conferring a greater mortality risk. Among critically ill patients with AKI, those with prior CKD experience a lower mortality rate but are more likely to be dialysis dependent at hospital discharge (196).

Existing CKD is one the strongest predictors of developing AKI following contrast exposure, surgical procedures and after certain medical illnesses (96, 197-199). These studies have suggested that underlying CKD maybe the single most important risk factor for AKI. However the question remains as to whether it is possible to accurately correct for all confounding variables in these patients, and how much of this risk is an increased susceptibility of a kidney with pre-existing disease to develop an acute kidney injury. Part of the increased risk is the fact that this population of patients with CKD is heavily burdened with co-morbidity. This population is more likely to be subject to nephrotoxic insults such as exposure to contrast agents and certain nephrotoxic treatments for co-existing co-morbid conditions. This population is also more likely to be taking medications such as ACEIs or ARBs (200), which gives the patient increased susceptibility for developing AKI. These patients then experience an additional insult, such as ascending urinary tract infection or other inter-current illness, sustaining an overt AKI requiring admission, an event included

in the epidemiological analyses of AKI. A patient without pre-existing CKD, who is less likely to have these susceptibility factors, may well have an episode of AKI linked to their clinical episode, but of reduced severity, not requiring admission and possibly go unrecognised and may therefore not be captured.

The difficulty in defining CKD as a risk factor for AKI is also confounded in studies by the actual definition of CKD and AKI used. These definitions also have substantially different effects at different levels of renal function. This study observed that lower eGFR at referral was associated with multiple AKI episodes. There have been few studies examining the relationship between the risk of AKI and severity of CKD and this is partly due to some AKI trials excluding patients with mild or moderately elevated baseline creatinine. A study by Hsu *et al* published in 2008, examined the risk of AKI in patients with chronic kidney disease, and how that risk varied with level of eGFR and hence stage of CKD (77). They assessed 1,746 hospitalized adults from the Kaiser Permanente integrated management care consortium in Northern California who developed dialysis-requiring AKI. Seventy-four per cent of these patients had CKD stage 3A or above at baseline. The 1,746 patients were then compared with 600,820 hospitalised members who did not develop AKI. The adjusted odds ratio for the development of dialysis-requiring AKI was 1.95 for stage 3A, 3.54 for stage 3B, 28.50 for stage 4 and 40.07 for stage 5 patients not yet on maintenance dialysis compared to patients with stage 1 and 2. Similar associations were seen after controlling for inpatient risk factors, again highlighting the importance of CKD severity as a risk factor for the development of AKI.

We are also unclear on how a large majority of patients develop CKD in the first place. An outcome of paramount interest is recovery of renal function and there is no agreed accurate and standardised definition of renal recovery. Patients with complete renal recovery following an AKI episode still have a higher incidence of CKD in the years following recovery (201). Patients without background CKD who develop AKI may already have an element of renal disease and reduced functional reserve, but have not yet manifested as a fall in GFR to define CKD. These patients are then more likely to develop CKD in the future following an episode of AKI and it may

speed up the process to development of overt CKD. This work has earlier demonstrated reduced accuracy of the MDRD equation at GFR > 60ml/min/1.73m^2 in the elderly with a tendency to overestimate GFR. As the MDRD study eGFR is less accurate at higher levels, should we be assessing rate of change in this group more closely or more accurately? The 2014 NICE CKD guidelines acknowledged this limitation and recommended that more accurate methods to estimate GFR such as Cystatin C or the CKD-EPI equation are used in patients with CKD stage 3a (57). This study suggests that a proportion of patients who have established CKD of uncertain aetiology may be attributed to a prior occurring AKI event.

If AKI is a cause of CKD, it seems logical that AKI may occur before the onset of CKD. There are however difficulties in testing this hypothesis. A few studies have suggested that development of CKD occurs following an episode of AKI and in some cases can progress to requiring chronic dialysis treatment. Ishani *et al* reported that of the patients who had suffered an episode of AKI and did not have a background of CKD, 72.1% had CKD documented within 2 years of the occurrence of AKI (87). Triverio *et al* demonstrated that following AKI 50% of patients without background CKD progressed to CKD within 3 years (202). Previous follow-up studies in children have suggested that a significant proportion of patients successfully discharged from hospital following an episode of AKI went on to show features of CKD (203). More recently, Bucaloiu *et al* performed a longitudinal study of patients with no evidence of overt CKD and studied the effect of AKI on residual renal function (204). They found that despite fairly rapid recovery of renal function following an AKI episode, even a minor rise in creatinine was associated with a 90% increased risk of developing CKD. In addition to this, they found that those who subsequently developed CKD after a resolved AKI had a significant increased mortality rate. There is no doubt that the mortality from AKI is high, and of those that survive, there may be decline in renal function, in some cases leading to the development of ESRD, either at the time of AKI, or in the future. Mortality may well be a confounding factor in terms of progression. For example, a severe insult resulting in AKI may result in death rather than result in worsening renal function.

Studies suggest that approximately 40% of patients commencing dialysis for the first time start due to AKI (60, 205). These patients may however recover function with time so they are not reflective of the proportion of patients on chronic dialysis. From the annual report of the United States Renal Disease Survey 2006, approximately 6% of patients with an episode of AKI progressed to ESRD within 2 years, and two thirds of hospitalised patients who had an episode of AKI and progressed to ESRD, had a background of CKD (206). Thakkar *et al* looked at the effect of AKI on CKD in hospitalised patients with diabetes and found 23% reached CKD stage 4 at the time of discharge (207).

Two studies, both prospective and observational, which were published in 2002, by Metcalfe *et al* and Robertson *et al* (60, 205), looked at patients starting dialysis for the first time, and split them into the following groups: CKD in which the patients started RRT in a planned manner for ESRD, AKI, and ACKD. The percentage of patients in each group was approximately, 40% due to CKD, 40% due to AKI and 20% due to ACKD. Importantly, though these patients started renal replacement therapy for the first time, this does not correspond to requiring on-going chronic dialysis. From the study by Metcalfe *et al*, 23.5% of the AKI group, and 16.5% of the ACKD group had recovered at 90 days (60).

Wald *et al* looked at the outcomes of chronic dialysis and death in patients with acute kidney injury requiring dialysis (208). This was a ten year cohort study of adult patients in Canada who survived a hospital associated AKI requiring in-patient dialysis who were free of dialysis for at least 30 days after discharge. These patients with AKI were three times more likely to require chronic dialysis compared to those without AKI. They found that individuals with pre-existing CKD, who had an episode of AKI requiring dialysis, had a two-fold higher risk of requiring chronic dialysis compared to patients with CKD without an episode of AKI requiring dialysis. They also interestingly reported that in patients without pre-existing CKD, an episode of AKI requiring dialysis conferred a fifteen fold higher risk of chronic dialysis than patients with CKD without an episode of AKI. This comes as a surprise as one would

expect patients with pre-existing CKD to have an increased risk of developing ESRD following an episode of AKI, compared to patients without pre-existing CKD.
In fact observational and database studies demonstrate that AKI on a background of CKD leads to ESRD at a higher frequency than does AKI alone. A study by Ishani *et al* looked at a random cohort of 233,803 patients hospitalised in the year 2000 based on Medicare claims, aged ≥ 67 years on discharge and with no previous ESRD or AKI (87). They reported that patients with concomitant AKI and CKD were far more likely to develop ESRD, indicating a strong causative effect of the interaction on ESRD development. In a population-based study by Ali *et al*, when comparing patients with ACKD to those with AKI alone, patients with ACKD were older and had less chance of renal recovery (62). These studies however again, all varied in definitions of both CKD and AKI.

This study demonstrated an association of multiple AKI episodes with CKD progression and this has also been observed in other studies. Both frequency and duration of AKI have been found to be associated with CKD progression. In the study by Thakkar *et al,* which looked at the effect of AKI in a cohort of US veterans with diabetes, there was a 30% increased risk of recurrence of AKI after the first episode and they suggested that each episode of hospital-acquired AKI doubled the risk of their CKD progression (207). Studies have also demonstrated that severity of AKI is linked to CKD progression (209) and longer duration of AKI is associated increased mortality (210) but there has been no association with duration of AKI and CKD progression. Most studies on AKI outcomes have focused on mortality and subsequent development of ESRD following severe and hospital-based AKI, however little is known of these more minor episodes of AKI that occur in the community and the effect they have on progression of CKD.

AKI frequently develops in the community and studies have shown that up to 60% of hospitalised-AKI is community-acquired (62), however, a substantial proportion of AKI events do not result in hospitalisation and are managed in the community. It is possible that some of these community-based AKI events may not have been identified through failure to seek medical help or lack of blood sampling at the time.

These ‘discrete’ episodes of AKI warrant further investigation as intervention in this group may have a significant effect on outcomes.

Although this study demonstrated a clear relationship between multiple AKI events and CKD progression, this association was not independent of other variables. By experiencing an episode of AKI these patients are more likely to have risk factors for AKI and a large number of these risk factors are common for CKD. AKI also occurs more frequently in an older population group which has a greater burden of co-morbidity, and in which there is a greater risk of progression of CKD anyway. AKI events however were also observed in patients with stable CKD, so what makes the progressive group more susceptible to non-recovery of function? Not all AKI is the same and this study did not look at aetiology of AKI which may well be the key. Further studies need to determine what factors influence the outcomes of progression or full recovery.

What this study does show is that CKD progression does not always follow a linear pattern. The slope in decline of GFR is a measure of progression rate and renal specialists often use this linear model of progression to predict when a patient will develop ESRD in order to initiate timely preparation for dialysis or transplantation. One advantage of this study was that the multitude of outpatient pathology results over several years follow up not only enabled a more accurate estimation of baseline kidney function but also enabled better interpretation of progression patterns. Most other studies in CKD progression have relied on follow-up data of less than five years and it is not always easy to interpret progression patterns in a short observation period with few GFR measurements.

This study demonstrated that progression patterns can be either linear or stepwise in trajectory. While the familiar linear progressive pattern occurred in 34% of people whose GFR declined, stepwise progression was a far more commonly observed pattern in the progressive group in this study (66%). The stepwise subgroup were characterised by a decline in kidney function that deviated from a linear progression trajectory representing periods of deceleration or acceleration of disease

progression. Periods of non-progression varied in length among individuals and in some subjects GFR increased with time. This study suggests that the determinants for progression of disease may vary over time within the same individual influencing their trajectory of decline in function. If we can identify these risk factors and intervene with the modifiable determinants, we may be able to slow CKD progression. The only significant variable differentiating the linear and stepwise subgroups were multiple AKI episodes suggesting an association of multiple AKI events and stepwise progressors.

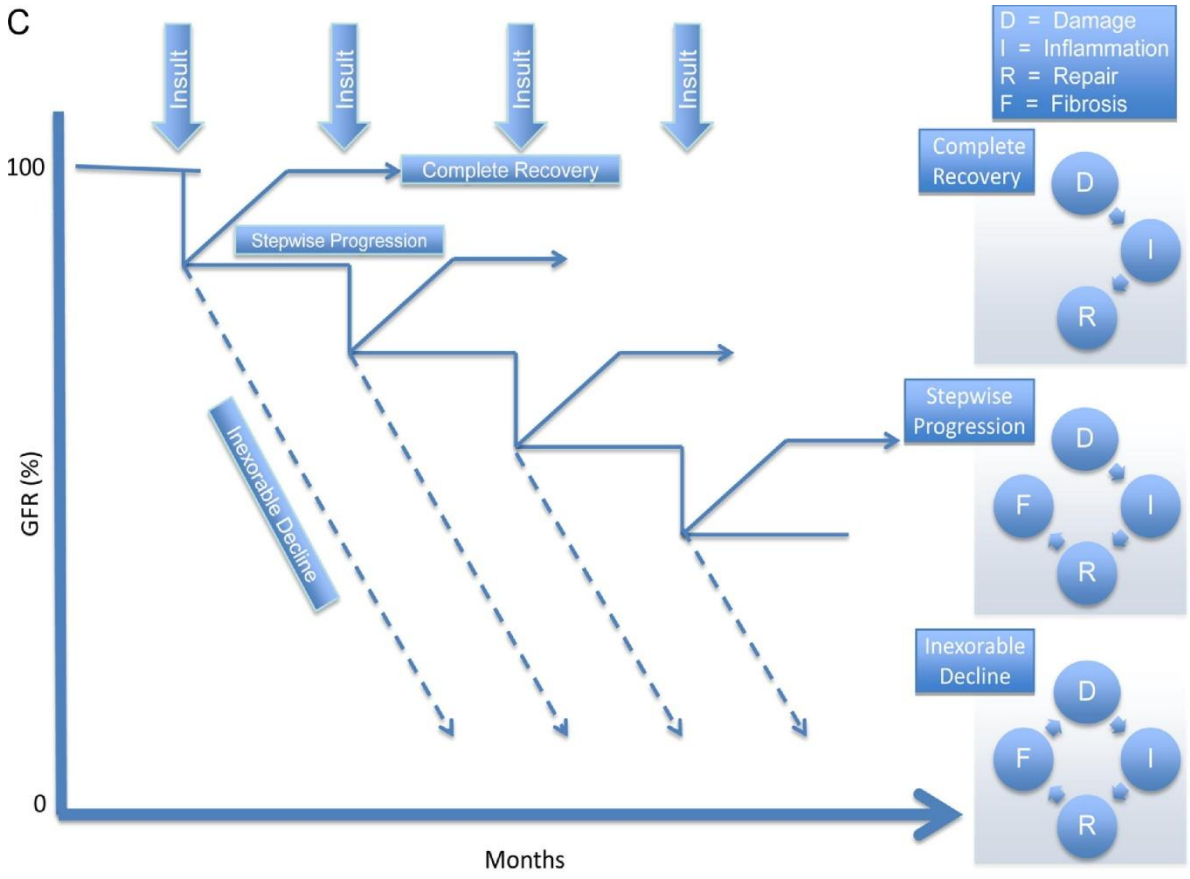
The potential effects of AKI events on CKD pattern progression is illustrated in Figures 7.1 which suggests the possible different outcomes following AKI. Complete recovery following an AKI would represent and initial insult leading to cellular injury and repair. A patient with non-progressive CKD may suffer an AKI leading to an acute drop from the prediction linear line. This may be followed by a new baseline GFR due to loss of functioning nephron mass subsequent to the AKI and the patient may have an altered pattern of decline in the future. In contrast, another individual may experience periods of rapid decline in GFR followed by prolonged periods of stability of varying length and that GFR trajectory pattern would represent stepwise progression. It is easy to see the potential to misinterpret CKD patterns if the GFR observations are limited to certain time periods during the deceleration phase or stable phase. Could it be that if rate of change was assessed over longer periods of observation, patterns of GFR decline would show a stepwise trajectory of decline whereby an accelerant phase is precipitated by a 'promoter' of progression such as an AKI event? Many studies have investigated risk factors for progression to ESRD but little is known about predictors of change in renal function in the community (211).

Many studies have not assessed risk factors for decline in renal function in populations without signs of kidney disease. The Tromso study was a prospective population study following individuals with no signs of kidney disease at baseline for 7 years with a primary outcome of decline in GFR and predictors of change in GFR were assessed (193). They demonstrated both high systolic BP and high fibrinogen

levels contributed to a more rapid decline in GFR for men and women. Obermayr *et al* looked at 17,375 healthy volunteers over 7 years with an age range 20-89 years (192). The primary outcome was development of CKD and they showed that cardiovascular risk factors, pre-existing NKF-CKD stages 1 and 2, proteinuria and surprisingly, doing no sports, predicted new-onset kidney disease.

Are these 'promoters' of progression modifiable and can we identify and prevent those at risk from developing or acquiring this risk factor? Can we identify those at risk of AKI and intervene to reduce incidence of community-AKI? We need to investigate these AKI events that occur in the community in more depth and understand what factors influence renal recovery following an AKI and what characteristics differentiate them from those whose renal function declines following an AKI. Although the study has failed to disprove the null hypothesis of an independent association of multiple AKI events and CKD progression, it may be that the study was not adequately powered to demonstrate significance. The study does suggest that multiple community-AKI is an important 'promotor' in the progression of CKD. It also suggests that GFR decline may not always follow a non-progressive or linear decline and 'random' events such as an AKI may affect the course of disease.

Figure 7.1 Possible Chronic Kidney Disease Outcomes Following Acute Kidney Injury



Abbreviations: GFR, glomerular filtration rate; AKI, acute kidney injury; CKD, chronic kidney disease; ACKD, acute-on-chronic kidney disease; AKD, acute kidney disease or disorders.

Figure 7.1.

This model demonstrates the possible outcomes in GFR following an AKI event and illustrates the relationship between AKI, AKD, ACKD and CKD. Following an initial insult (AKI) renal function may either fully recover, partially recover and develop *de novo* CKD, progress in a linear inexorable decline towards ESRD or progress in a stepwise pattern with periods of stability followed by accelerated decline precipitated by a further insult. The phases of cellular injury in CKD are represented to temporally relate to disease patterns of progression.

Bedford *et al,* 2012 (152).

7.2 Limitations

The results of this study need to be interpreted with caution as there are notable limitations. Firstly, demographic, clinical and pathology data collection was dependent on the quality of information provided on referral letters, hospital records and clinical information provided at the time of blood tests which may have led to missing data. The study subjects were selected from patients referred to nephrology services therefore not entirely representative of the general population with less co-morbid burden. Patients were recruited from a single county in the U.K. and the population demographics of East Kent is not entirely representative of the U.K. with more older patients with fewer ethnic minorities than the national average. A significant proportion of patients originally referred (24%) had to be excluded from the final cohort due to shorter follow-up time or lack of pathology data so interpretation of these results may not be truly representative of all those referred. All creatinine measurements however were measured at a single centre reducing the potential for GFR estimation error.

One of the major limitations of this study is the definition used to determine an AKI event. Definitions of AKI, as I have described earlier, have rarely been consistent across studies. The definition of AKI has changed in recent years in line with the RIFLE criteria, then the AKIN criteria and more recently the KDIGO definition. This study was carried out prior to the publication of the 2012 KDIGO AKI clinical guidelines (70). Although the study design defined an AKI event as a rise in serum creatinine of 26 µmol above the baseline creatinine, the criteria does not truly meet the AKIN criteria as it was not possible to fulfil the time constraints for either the AKIN or RIFLE definitions of AKI. Also, the baseline creatinine was not derived from the median value of multiple creatinine measurements over a 365 day period as suggested by the KDIGO guidelines (and subsequently adopted by NHS England), but by the lowest serum creatinine in the 365 days prior to the event. Only results that deviated from the linear regression line applied to each graph were assessed as to the whether they were defined as an AKI event. Using the study definition of AKI, a rapidly declining linear progressor may have several creatinines over-called as AKI

episodes. Many AKI events in the study may well have been an overestimation and may well have not taken the biological and analytical variation of serum creatinine into account. It is unlikely that the approach to the definition of AKI would have dramatically altered the results of the study. Redesigning the study to use the methodology described by La France and Miller, which extends the time reference creatinine to 12 months (212) prior to the event as suggested by the NHS England AKI algorithm (71), would overcome this limitation. Only serum creatinine samples were analysed in the assessment of an AKI episode and this study's methodology did not assess AKI using urine output criteria to define AKI. Assessing AKI using urine output criteria is difficult in clinical practice and often involves invasive procedures to accurately monitor output which would be impractical in the assessment of community-based AKI.

The study aimed to determine the effects of these minor community-managed AKI events however hospital admission records were not cross-referenced with the AKI events. Although the majority of the pathology tests at the time of the AKI events were requested from community healthcare, one cannot exclude that many of these AKI events did not lead to hospitalisation. We have limited data on how community-managed AKI and hospital-managed AKI differ in terms of aetiology and outcomes. It is clear AKI is more prevalent in certain at risk populations but AKI is still a random event and whilst people may be at a higher risk of developing AKI, we still cannot always predict when they are going to have an episode of sepsis or require surgery or a contrast-scan which may then precipitate an AKI. The difficulty comes in capturing all these AKI events when they do occur in the community particularly as when they occur at random. It is far easy to identify AKI in hospitalised patients and in at risk populations when you are screening for it. It is possible that many of these random events also occur in the community and are not picked and these AKI events go undetected. This has important implications for physicians when counselling patients about their projected renal outcome. AKI events can also occurring in a non-random fashion and are more likely to be picked up. For example, a patient is more likely to have renal function monitoring following initiation of a nephrotoxic agent.

One is more likely to detect an AKI event when susceptible individuals are screened. This highlights the difficulties in studying these events.

Severity of AKI is known to increase risk for progression to ESRD (209) and a patient is more likely to be admitted to hospital with AKIN 2 or 3. Many AKI events are often acquired and managed in the community and are more likely to be AKIN 1. This study aimed to determine the renal outcomes of community-managed AKI however it did not assess severity of AKI according to the AKIN stages and, as mentioned earlier, one cannot assume all these AKI events were managed in the community. Severity was excluded from the study as the numbers of AKIN 2 or AKIN 3 events were considered to be too small to allow meaningful analysis.

What is of significance is the lack of information regarding the aetiology, prescription data and management of these AKI events. AKI is multi-factorial and cause of AKI has a significant effect on outcome of AKI. Cause of AKI was not assessed as, although clinical information provided at the time of the blood request was extracted, information was not consistently provided or had little relevance to the AKI event so this was not included this in the analysis. Although prescription data was available on several patients at referral, prescription patterns at the time of the AKI event were not available hence analysis of the aetiology of AKI was not possible. It would be interesting to determine whether recovery from certain AKI events is determined by aetiology of AKI. One would assume that drug-related AKI events would expect a better prognosis compared to, for example, a sepsis related AKI.

Sepsis is a common problem with mortality rates as high as 36% even in those who reach intensive care (213). A recent large NHS survey documented mortality rates of 30-40% consistent across hospitals in the U.K. amongst severe AKI (AKIN 3) (214). Development of AKI during sepsis increases patient morbidity with significant effect on multiple organ functions, increases length of stay and predicts a higher mortality. Septic-AKI is the most common precipitating cause of AKI accounting for at least 50% of AKI in ICU patients. The combination of AKI and sepsis is associated with a mortality rate of up to 70% (215). The pathophysiology of septic-AKI is complex and

can be due to a combination of endothelial dysfunction, inflammatory cell activation and infiltration in the renal parenchyma (215), renal blood flow changes and tubular injury and necrosis.

The Surviving Sepsis campaign was launched in 2004 and after several updates and simplification of recommendations combined with sepsis care bundles, there has been reduction in hospital mortality from 44.1% to 20.0% (217) demonstrated since its implementation. In 2010, NCEPOD (97) reported that only 50% of care of patients who died from AKI was 'good' and subsequently KDIGO (70) and then NICE (72) published AKI guidelines, as clearly a strategy is required to improve the prevention and management of AKI. These guidelines and algorithms have the potential to reduce the morbidity and mortality associated with AKI as the Surviving Sepsis campaign has had for sepsis.

The methodology of describing CKD progression pattern used the Delphi technique with a panel of experts assessing eGFR versus time trajectory graphs and came to high levels of agreement. Any graphs with disagreement were further discussed to form a unanimous decision of linear or stepwise decline. This method could be subject to bias particularly if the group were influenced by a strong individual opinion when assessing stepwise or linear patterns. Interpreting progression patterns is more accurate with multiple eGFR measurements over a longer observation period and some individuals lacking in pathology data may not have been grouped appropriately. Differentiation between the linear and stepwise progressive subgroups using this methodology could also be open to criticism as interpretation of the different patterns was subjective and descriptive modelling was not applied to each pattern.

Overcoming these limitations would be difficult particularly in a retrospective setting. Capturing these AKI events as they occur randomly in the community would be challenging and information regarding the events would be lacking. In order to better assess CKD progression patterns and the effect of promoters of progression such as AKI events have on eGFR decline, a long-term prospective follow-up study of

a large cohort with and without CKD is needed. In an ideal world, selecting a study cohort from patients undergoing regular blood test monitoring, such as those on immunosuppression requiring monthly bloods tests, would hopefully capture many more of these random events. Conversely, such a select cohort of patients would only represent a specific population.

I would use the NHS England AKI algorithm defining AKI a serum creatinine ≥ 1.5 times higher than the median of all creatinine measurements 8-365 days prior to the test to ensure a more accurate estimate of AKI incidence. Hospital admission data would need to be cross-referenced against these AKI events to identify community-managed and hospital-managed. Each AKI episode would require extraction of clinical information at the time of the event in order to identify aetiology, duration, severity, prescription data and interventions. This would allow comparison of outcomes between hospital-managed and community-managed AKI on CKD progression and mortality rates and identify the determinants in renal recovery.

7.3 Conclusion

In conclusion, this study has suggested that the prevalence of progressive CKD may be higher than originally thought and the trajectory of GFR decline more commonly follows a non-linear stepwise pattern. Factors associated with progression were diabetes, malignancy, albuminuria and, although not an independent risk factor, multiple AKI episodes were significantly higher in those with progressive CKD particularly in the stepwise group. This suggests the incidence of community-AKI is more common than estimated and has a significant impact on progression of CKD. This observation has a significant implication both clinically and in research on CKD. Further analysis of these patterns could be used for predictive modelling to assess risk for CKD progression. We also need to determine why some individuals recover from an AKI event whereas others do not. Most risk scores predicting risk of AKI in order to help improve outcomes have been developed where the timing of the AKI insult can be predicted, such as in the setting of cardiac surgery (218) or coronary angiography (219). Further knowledge of these community-managed AKI events and

how they differ from hospital-managed AKI in terms of outcome is necessary. Multiple AKI events, together with other known promoters of progression, could then be included to develop prediction models determining progression of CKD.

CHAPTER 8

Concluding Discussion

The NEOERICA project demonstrated that the UK prevalence of people with an eGFR < 60 ml/min/1.73m^2 was 8.5% and observed an exponential increase with age with a prevalence of 25% in people aged > 70 years (2). Early identification of these patients with CKD 3-5 may allow implementation of multiple risk factor intervention strategies aimed at reducing morbidity, mortality and disease progression. KDOQI and NICE CKD guidelines highlight the importance of early identification of these patients with diagnosis and management recommendations to improve outcomes and improve appropriate referral to specialists (10, 67). The internationally adopted KDIGO CKD stages are defined by eGFR based on serum creatinine levels using the MDRD study equation. Not only do we base diagnosis and staging of CKD on eGFR but also drug dosing and important management decisions. The MDRD study equation was developed in a relatively young population with CKD and hence is inaccurate at higher GFR levels and in older populations. The CKD-EPI equation has been suggested to replace the MDRD study equation as it performs better particularly at higher GFRs which would have implications in population prevalence data. Neither equation has been validated in the elderly population.

Although CKD prevalence is so high, only 1-2% of these patients with CKD 3-5 progress to ESRD (15). Why is there such a disparity in numbers? Many patients with CKD 3-5 have increased risk of death particular from cardiovascular events but reported mortality rates do not account for the low rate of progression. The first of this study's hypothesis was that the high population prevalence estimates of CKD 3-5 are due to the inaccuracies of the MDRD study equation used to estimate GFR in older people. The hypothesis states that both equations underestimate GFR in older people and the MDRD study equation particularly underestimates GFR at higher levels where individuals are more likely to be misclassified as having CKD 3a when

they have a measured GFR ≥ 60 ml/min/1.73m^2. This would have a significant effect on CKD prevalence data.

This study assessed the performance of the two estimating equations compared to the measured GFR using iohexol GFR as the reference method in an elderly population. This study is the first study to endorse the validity of the current estimating GFR equations in an elderly population who have a high burden of CKD. Whilst falling short of the >90% P_{30} aspiration of the 2002 KDOQI guideline (10), the MDRD and CKD-EPI GFR estimating equations appear to work just as well in older compared to younger populations. There was no evidence that the MDRD equation underestimated GFR, irrespective of the level of GFR and this may reflect similar characteristics between this study cohort and the cohort the MDRD study equation was developed in. The CKD-EPI equation performed marginally better than the MDRD equation, particularly at GFR ≥ 60 ml/min/1.73 m^2 consistent with its performance in younger populations. This study has validated the accuracy and applicability of both the MDRD and CKD-EPI equations in estimating GFR in elderly caucasians and supports the KDIGO and NICE CKD guidelines recommending their use to assess renal function. These results will improve patient and physician confidence in the use of eGFR to accurately reflect renal function in older patients and its ensuing impact on CKD management and safer prescribing practice.

This study has proved that the prevalence estimates of CKD are fairly accurate so this still leaves us with a question of what factors influence CKD progression. Although CKD is highly prevalent, it does not inexorably progress. Progression to ESRD in those with earlier stages of CKD is low whereas progression in those with stage 4 is much higher. There has been much debate around the definition of progression and rate of change of GFR is currently used. Albuminuria has emerged as an important predictor of progression along with cause of disease and the presence of diabetes, cardiovascular disease and poorly controlled hypertension (34-36, 192-193). The KGIDO guidelines have included the category of albuminuria in the CKD staging and have included guidelines on how to identify individuals at risk for progression (12). Although the presence of risk factors such as albuminuria and hypertension

increases the risk of progression to ESRD, many patients with these risk factors do not progress. Another question that arises is how patients develop CKD in the first place, one potential explanation is AKI.

The incidence of AKI has dramatically increased over the past few decades. Pre-existing CKD is one of the most important risk factors for developing AKI. AKI is becoming increasingly recognised as an important determinant in the development and progression of CKD and long-term mortality. There has been a significant drive to improve the recognition and care of AKI patients particularly in hospitalised patients in order to improve survival. Several studies have helped our understanding of the complex relationship between AKI and CKD and the clinical consequences that occur with their co-existence, however, the majority of studies have focused on hospital based-AKI. We know that even minor AKI episodes in the hospital setting is becoming increasing common incurring a significant risk of progression to ESRD and reduced survival even after discharge. Community-acquired AKI is common, often less severe and most cases are not referred to nephrologists. Little is known about the effect that these community-based AKIs have on mortality and cardiovascular outcomes and on risk of progression to CKD.

The second hypothesis was that multiple episodes of community-based AKI have an independent association with CKD progression, influencing the pattern of progression and the development of 'de novo' CKD. The second study was a retrospective observational study of patients with CKD 3-5 referral to renal specialists assessing rate of decline of eGFR and what characteristics influence progression. The incidence of community-based AKI events were recorded and compared between stable and progressive CKD groups. This study suggests that a large majority of patients with progressive CKD experience a decline in GFR in a non-linear pattern with variable periods of non-progression and accelerated progression resulting in a stepwise decline. This study found a significant association with multiple AKI and CKD progression particularly those following a stepwise decline but this association is not independent of other risk factors for CKD progression and AKI. It may be that the definitions used and methodology of this study was not

appropriate to demonstrate a significant association but it does suggest that AKI plays an important part as a promoter of CKD progression.

This study suggests that the consequences of these small rises in serum creatinine occurring in primary care may have similar effects on disease progression as hospital-based AKI and merit further investigation for better understanding. It will be important to fully ascertain why the tests were requested at the time of the acute decline, whether these episodes of acute rise in serum creatinine are actually recognised with prompt appropriate management, or whether they go unrecognised by the healthcare system. Further studies are required to examine if there is a potential causal association between these events in primary care and poor outcomes other than CKD progression.

There needs to be an increased awareness of the risk factors for AKI both in primary and secondary care with particular vigilance in subjects with CKD in order to develop strategies to prevent AKI occurrences and reduce risk of CKD progression. Interventions and identification of modifiable risk factors may lead to longer periods of non-progression in CKD patients and reduce progression risk in the future. This work suggests that further research is needed to assess the impact AKI events have in the progression of CKD and in particular analyse what time related risk factors determine a patient's non-linear decline in GFR. This data can be used to develop a risk-model which could in turn be developed into a clinical tool used for determining an individual risk for patients at risk of both CKD progression and at risk of developing AKI. More recently studies have focused on the development of risk scores for identifying progressive decline in GFR and progressive increase in albuminuria. Some have studies have focussed on clinical risk factors including age, gender and blood pressure level and others have combined this with laboratory data such as eGFR level, albuminuria, C-reactive protein levels and serum albumin levels (220). These proposed predictive models have only been developed and validated in specific cohorts and further evaluation of these models is needed in different populations. The rationale for predicting risk of progression CKD is that it enables

determination of referral, care plans, frequency of monitoring and instituting appropriate treatment strategies.

Both AKI and CKD confer a significant morbidity and mortality. With an ageing population and increasing co-morbidity burden, AKI and CKD will continue to have a significant impact on the healthcare economy across the world. We should aim to prevent susceptible patients developing AKI following an exposure that places them at increased risk and at the very least aim to prevent patients in whom AKI is apparent from developing the complications that may result in increased mortality, ESRD and development or progression of CKD. AKI in the community and the concept of 'discrete' AKIs require a prospective evaluation to establish the role of AKI in the natural history of CKD.

References

1. Coresh J, Astor BC, Greene T, Eknoyan G, Levey AS. Prevalence of chronic kidney disease and decreased kidney function in the adult US population: Third National Health and Nutrition Examination Survey. Am J Kidney Dis 2003; 41: 1–12.

2. Stevens PE, O'Donoghue DJ, de Luisignan S, Van Vlymen J, Klebe B, Middleton R, Hague N, New J, Farmer CKT. Chronic kidney disease management in the United Kingdom: NEOERICA project results. Kidney Int 2007; 72: 92-99.

3. Roderick P, Jones C, Tomson C, Mason J. Late referral for dialysis: improving the management of chronic renal disease. QJM 2002; 95: 363–379.

4. Jungers P, Zingraff J, Albouze G et al. Late referral to maintenance dialysis: detrimental consequences Nephrol Dial Transplant 1993; 8 (10): 1089-1093.

5. Bakris GL, Williams M, Dworkin L, Elliot WJ, Epstein M, Toto R, Tuttle K, Douglas J, Hsueh W, Sowers J. Preserving renal function in patients with hypertension and diabetes: a consensus approach. National Kidney Foundation Hypertension and Diabetes Executive Committees Working Group. Am J Kidney Dis 2000; 36 (3): 646-661.

6. Jafar TH, Stark PC, Schmid CH, Landa M, Maschio G, de Jong PE. Progression of chronic kidney disease: the role of blood pressure control, proteinuria and angiotensin converting enzyme inhibition; a patient level meta-analysis. Ann Intern Med 2003; 139 (4): 244-252.

7. Peterson JC, Addler S, Burkart JM, Greene T, Herbert LA, Hunsicker LG. Blood pressure control, proteinuria and the progression of renal disease. The

Modification or Diet in Renal Disease Study. Ann Intern Med 1995; 123 (10): 754-761.

8. Levey AS, Bosch JP, Lewis JB, Greene T, Rogers N, Roth D. A more accurate method to estimate glomerular filtration rate from serum creatinine: a new prediction equation. Modification of Diet in Renal Disease Study Group. Ann Intern Med 1999; 130(6): 461-70.

9. Levey AS, Stevens LA, Schmid CH, Zhang Y, Castro AF, Feldman HI, Kusek JW, Eggers P, Van Lente FV, Greene T, Coresh J. A new equation to estimate glomerular filtration rate. Ann Intern Med 2009; 150: 604-612.

10. National Kidney Foundation. K/DOQI clinical practice guidelines for chronic kidney disease: evaluation, classification, and stratification. Am J Kidney Dis 2002; 3: S1-266.

11. Go AS, Chertow GM, Fan D, McCulloch CE, Hsu CY. Chronic kidney disease and the risks of death, cardiovascular events and hospitalisation. N Engl J Med 2004; 351 (13): 1296-1305.

12. Kidney Disease: Improving Global Outcomes (KDIGO) CKD Work Group. KDIGO 2012 Clinical Practice Guideline for the Evaluation and Management of Chronic Kidney Disease: Kidney Int Suppl 2013; 3: 1-150.

13. Office for National Statistics; Office for National Statistics population and Vital Statistics, England and Wales 2012; 3-12.

14. Foley RN, Collins AJ. End-stage renal disease in the United States: an update from the United States Renal Data System. J Am Soc Nephrol 2007; 18 (10): 2644-2648.

15. Fogarty D, Cullen R. UK Renal Registry, Renal Association, The Sixteenth Annual Report. Bristol UK; UK Renal Registry, 2012.

16. Hsu C, Vittinghoff E, Lin F Shiplak MG. The incidence of end-stage renal disease is increasing faster than the prevalence of chronic renal insufficiency. Ann Intern Med 2004; 141 (2): 95-101.

17. British Heart Foundation; Coronary heart disease statistics: A compendium of health statistics. British Heart Foundation Promotion Research Group. Dept Public Health 2012.

18. Stevens LA, Li S, Wang C, Huang C, Becker BN, Bombakc AS, Brown WW, Burrows NR, Jurkovitz CT, McFarlane SI, Norris KC, Shiplak M, Whaley-Connell AT, Chen SC, Bakris GL, McCullough PA. Prevalence of CKD and co-morbid illness in elderly patients in the United States: results from the Kidney Early Evaluation Program (KEEP). Am J Kidney Dis 2010; 55 (3 S2): S23-33.

19. Coresh J, Selvin E, Stevens A et al. Prevalence of chronic kidney disease in the United States: J Am Med Assoc 2007; 298 (17): 2038-2047.

20. World Population Ageing Report. The Department of Economic and Social Affairs, United Nations, 2013.

21. McLachlan MS, Guthrie JC, Anderson CK, Fulkner MJ. Vascular and glomerular changes in the ageing kidney. J Pathol 1977; 121 (2): 65-78.

22. Neugarten J, Gallo G, Silbiger S, Kasisike B. Glomerulosclerosis in aging humans is not influenced by gender. Am J Kidney Dis 1999; 34 (5): 884-888.

23. Beck LH. Changes in renal function with aging. Clinics in Geriatric Med 1998; 14 (2): 199-209.

24. Lindeman RD, Goldman R. Anatomic and physiological age changes in the kidney. Exp Gereontol 1986; 21 (4-5): 379-406.

25. Miletic D, Fuckar Z, Sustic A, Mozetic V, Stimac D, Zauhar G. Sonographic measurement of absolute and relative renal length in adults. J Clinical Ultrasound 1998; 26; 185-189.

26. Davies DF, Shock NW. Age changes in glomerular filtration rate, effective renal plasma blood flow, and tubular excretory capacity in adult males. Journal Clinical Investigation 1950; 29: 496-507.

27. Rowe JW, Andres R, Tobin JD, Norris AH, Shock NW. The effect of age on creatinine clearance in men: a cross-sectional and longitudinal study. J Gereontol 1976; 31 (2): 155-163.

28. Lindeman RD, Tobin J, Shock NW. Longitudinal studies on the rate of decline in renal function with age. J Am Geriatr Soc 1985; 33 (4): 278-285.

29. Botev R, Mallie JP, Cirillo M. Estimating glomerular filtration rate: Cockcroft-Gault and Modification of Diet in Renal Disease formulas compared to renal inulin clearance. Clin J Am Soc Nephrol 2009; 4 (5): 899-906.

30. Montesanto A, De Rango F, Beradelli M, Lattanzio F, Passarino G, Corsonello A. Glomerular filtration rate in the elderly and in the oldest old: correlation with frailty and mortality. Age (Dordr) 2014; 36 (3): 9641-9644.

31. Stevens LA, Coresh J, Feldman HI, Greene T, Lash JP, Nelson RG. Evaluation of the modification of diet in renal disease study equation in a large diverse population. J Am Soc Nephrol 2007; 18 (10): 2749-2757.

32. Keith DS, Nichols GA, Gullion CM, Brown JB, Smith DH. Longitudinal follow-up and outcomes among a population with chronic kidney disease in a large managed care organisation. Arch Intern Med 2004; 164 (6): 659-636.

33. Wong C, Moxey-Mims M, Jerry-Flucker J, Warady B, Furth S. CKiD (CKD in children) prospective cohort study: A review of current findings. Am J Kidney Dis 2012; 60 (6): 1002-1011.

34. Hiu X, Matsushita K, Sang Y, Ballew SH, Fulop T, Coresh J. CAK and cardiovascular disease in the Atherosclerosis Risk in Communities (ARIC) study: interactions with age, sex, and race. Am J Kidney Dis 2013; 62 (4): 691-702.

35. Keane WF, Zhang Z, Lyle PA, Cooper ME, de Zeeuw D, Grunfeld JP, Lash JP, McGill JB, Mitch WE, Remuzzi G, Shahinfar S, Snapinn SM, Toto R, Brenner BM. Risk scores for predicting outcomes in patients with type 2 diabetes and nephropathy: the RENAAL study. Clin Am J Soc Nephrol 2006; 1 (4): 761-767.

36. Klahr S, Levey AS, Beck GJ, Caggiula AW, Hunsicker L, Kusek JW, Striker G. The effects of dietary protein restriction and blood pressure control on the progression of chronic renal disease. N Engl J Med 1994; 330: 877-884.

37. Levey AS, de Jong PE, Coresh J, El Nahas M, Astor BC, Matsushita K, Gansevoort RT, Kasiske BL, Eckardt KU. The definition, classification and prognosis of chronic kidney disease: a KDIGO Controversies Conference report. Kidney Int 2011; 80 (1): 17-28.

38. Gooch K, Culleton BF, Manns BJ, Zhang J, Alfonson H, Tonelli M, Frank C, Klarenbach S, Hemmelgarn BR. NSAID use and progression of chronic kidney disease. Am J Med 2007; 120 (3): 280-287.

39. Dreyer G, Hull S, Mathur R, Cheser A, Yaqoob MM. Progression of chronic kidney disease in a multi-ethnic community cohort of patients with diabetes mellitus. Diabet Med 2013; 30 (8): 956-963.

40. Drey N, Roderick P, Mullee M, Rogerson M. A population-based study of the incidence and outcomes of diagnosed chronic kidney disease. Am J Kidney Dis 2003; 42: 677–684.

41. Collins AJ, Li S, Gilbertson DT, Liu J, Chen S-C, Herzog CA. Chronic kidney disease and cardiovascular disease in the Medicare population. Kidney Int 2003; 64(S 87): S24–S31.

42. Ritz E, McClellan WM. Overview: Increased cardiovascular risk in patients with minor renal dysfunction: An emerging issue with far-reaching consequences. J Am Soc Nephrol 2004; 15: 513–516.

43. Khan S, Kamzi WH, Abichandani R, Tighiouart H, Pereira BJG Kausz AT. Health care utilisation among patients with chronic kidney disease. Kidney Int 2002; 62: 229-236.

44. Sehgal AR, Grey SF, DeOreo PB, Whitehouse PJ. Prevalence, recognition, and implications of mental impairment among haemodialysis patients. Am J Kidney Dis 1997; 30: 41-49.

45. Hailpern SM, Melamed ML, Cohen HW, Hostetter TH. Moderate chronic kidney disease and cognitive function in adults 20 to 29 years of age: Third National Health and Nutrition Examination Survey (NHANES III). J Am Soc Nephrol 2007; 18 (7): 2205-2213.

46. Kerr M, Bray B, Medcalf. Estimating the financial cost of chronic kidney disease. Nephrol Dial Transplant 2012; 27(S3): S73-80.

47. Halimi JM, Giraudeau B, Vol C, Caces E, Nivet H, Lebranchu Y, Tichet J. Effects of current smoking and smoking discontinuation on renal function and proteinuria in the general population. Kidney Int 2000; 58: 1285-1292.

48. Baignet C, Landravy MJ, Reith C, Emberson J, Wheeler DC, Tomson C, Wanner C, Krane V, Cass A, Craig J, Neal B, Jiang L, Hooi LS, Levin A, Agodoa L, Gaziano M, Kasiske B, Walker R, Massy ZA, Feldt-Rasmussen B, Krairittichai U, Ophascharoensuk V, Fellstrom B, Holdass H, Tesar V, Wjecek A, Grobbee D, de Zeeuw D, Gronhagen-Riska C, Dasgupta T, Lewis D, Herrington W, Mafham M, Majoni W, Wallendszus K, Grimm R, Pederdon T, Tobert J, Young A, Collins R. The effects of lowering LDL cholesterol with simvastatin plus ezetimibe in patients with chronic kidney disease (Study of Heart and Renal Protection): a randomised placebo-controlled trial. Lancet 2011; 377 (9784): 2181-2192.

49. Innes A, Rowe, PA Burdern RP et al. Early deaths on renal replacement therapy: the need for early nephrological referral Nephrol Dial Transplant 1992; 7(6):467-471.

50. John RI, Webb MC, Young A, Stevens PE. Unreferred chronic kidney disease: A longitudinal study. Am J Kidney Dis 2004; 43 (5): 825-835.

51. The National Service Framework for Renal Services Part Two: Chronic kidney disease, acute renal failure and end of life care. Department of Health, 2005.

52. Chronic Kidney Disease (CKD) in Adults UK Guidelines for identification, management and referral. Royal College of Physicians, London and the Renal Association, Joint Speciality Committee on Renal Medicine, 2006.

53. Early identification and management of chronic kidney disease in adults in primary and secondary care. National Institute for Health and Care Excellence (NICE), 2008.

54. Klebe B, Farmer C, Cooley R, de Lusignan S, Middleton R, O'Donoghue, D, New J, Stevens PE. Kidney disease management in UK primary care: guidelines, incentives and Information Technology. Family Practice 2007; 24: 330-335.

55. Hobbs H, Stevens PE, Klebe B, Irving J, Cooley R, O'Donoghue D, Greene S, Farmer C. Referral patterns to renal services: What has changed in the past four years? Nephrol Dial Transplant 2009; 24 (11): 3411-3419.

56. Aghaie-Jaladerany H, Cowell D, Geddes CC. The early impact of the United Kingdom Chronic Kidney Disease (CKD) guidelines on the number of new attendances at renal clinics. Scot Med J 2007; 52 (4): 28-31.

57. Early identification and management of chronic kidney disease in adults in primary and secondary care. National Institute for Health and Care Excellence (NICE), 2014.

58. Leblanc MA, Kellum JA, Gibney RT, Liberthal W, Tumlin J, Mehta R: Risk factors for acute renal failure: inherent and modifiable risks. Curr Opin Crit Care 2005; 11 (6): 533-536.

59. Stevens PE, Tamimi NA, Al Hasani MK, Mikhail AI, Kearney E, Lapworth R, Prosser DI, Carmichael P. Non-specialist management of acute renal failure. QJM 2001; 94: 533–540.

60. Metcalfe W, Simpson M, Khan IH, Prescott GJ, Simpson K, Smith WC, MacLeod AM. Acute renal failure requiring renal replacement therapy: incidence and outcome. QJM 2002; 95: 579–583.

61. Hegarty J, Middleton R, Krebs M, Hussain H, Cheung C, Ledson T, Hutchison AJ, Kalra PA, Rayner HC, Stevens PE, O'Donoghue DJ. Severe acute renal failure. Place of care, incidence and outcomes. QJM 2005; 98: 661-666.

62. Ali T, Khan I, Simpson W, Prescott G, Townend JT, Smith W, MacLeod A. Incidence and Outcomes in Acute Kidney Injury: a comprehensive population-based study. J Am Soc Nephrol 2007; 18 (4): 1292-1298.

63. Kaufman J, Dhakal M, Patel B, Hamburger R. Community-acquired acute renal failure. Am J Kidney Dis 1991; 17 (2): 191-198.

64. Bellomo R, Ronco C, Kellum JA, Mehta RL, Palevsky P. Acute Dialysis Quality Initiative workgroup: Acute renal failure – definition, outcome measures, animal models, fluid therapy and information technology needs: the Second International Consensus Conference of the Acute Dialysis Quality Initiative (ADQI) Group. Crit Care 2004; 8: R204-R212.

65. Uchino S, Bellomo R, Goldsmith D. An assessment of the RIFLE criteria for acute renal failure in hospitalised patients. Crit Care Med 2006; 34: 1913-1917.

66. Bagshaw SM, George C, Dinu I, Bellomo R. A multi-centre evaluation of the RIFLE criteria for early acute kidney injury in critically ill patients. Nephrol Dial Transplant 2008; 23: 1203-1210.

67. Ostermann M, Chang RW. Acute kidney injury in the intensive care unit according to RIFLE. Crit Care Med 2007; 35: 1837-1843.

68. Kellum JA, Bellomo R, Ronco C. The concept of acute kidney injury and the RIFLE criteria. Acute Kidney Injury. Contrib Nephrol, Karger 2007; 156: 10-16.

69. Mehta Rl, Kellum Ja, Shah SV, Molitoris BA, Ronco C, Warnock DG, Levin A. Acute Kidney Injury Network: Report of an initiative to improve outcomes in acute kidney injury. Crit Care 2007; 11: R31.

70. Kidney Disease: Improving Global Outcomes: Clinical practice guidelines for acute kidney injury, 2012.

71. NHS England. Patient safety alert on standardising the early identification of acute kidney injury 2014. http://www.england.nhs.uk/ourwork/ patientsafety/akiprogramme/aki-algorithm/

72. Acute Kidney Injury Guidelines. National Institute for Health and Care Excellence (NICE), 2013.

73. Devarajan P. Neutrophil gelatinase-associated lipocalin: a promising biomarker for human acute kidney injury. Biomark Med 2010; 4 (2): 265-280.

74. Devarajan P. Biomarkers in acute kidney injury. Curr Opin Paediatr 2011; 23 (2): 194-200.

75. Ostermann M, Forni LG. Measuring biomarkers of acute kidney injury during renal replacement therapy: wisdom or folly? Crit Care 2014; 18 (3): 155.

76. Bagshaw SM, George C, Bellomo R. Early acute kidney injury and sepsis: a multi-centre. Crit Care 2008; 12 (2): R47.

77. Hsu CY, Ordonez JD, Chertow CM, Go AS. The risk of acute renal failure in patients with chronic kidney disease. Kidney Int 2008; 74: 101-107.

78. Hsu CY, McCulloch CE, Fan D, Ordonez JD, Chertow CM, Go AS. Community-based incidence of acute renal failure. Kidney Int 2007; 72 (2): 208-212.

79. Xue JL, Daniels F, Star RA, Kimmel PL, Eggers PW, Molitoris BA, Himmelfarb J, Collins AJ. Incidence and mortality of acute renal failure in Medicare beneficiaries, 1992-2001. J Am Soc Nephrol 2006; 17 (4): 1135-1142.

80. Akposso K, Hertig A, Couprie R, Flahaut A, Alberti C, Karras GA, Haymann JP, Costa De Beauregard MA, Lahlou A, Rondeau E, Sraer JD. Acute renal failure in patients over 80 years old: 25-years' experience. Intensive Care Med 2000; 26 (4): 400-406.

81. Noor S, Usmani A. Postoperative renal failure. Clin Geriatr Med 2008; 24 (4): 721-729

82. Thakar CV, Arrigain S, Worley S, Yare JP, Paganini EP. A clinical score to predict acute renal failure after cardiac surgery. J Am Soc Nephrol 2005; 16 (1): 162-168.

83. Fortescue EB, Bates DW, Chertow GM: Predicting acute renal failure after coronary bypass surgery: Cross-validation of two risk-stratification algorithms. Kidney Int 2000; 57: 2594–2602.

84. Liangos O, Wald R, O'Bell JW. Epidemiology and outcomes of acute renal failure in hospitalised patients: a national survey. Clin J Am Soc Nephrol 1: 43-51, 2006.

85. Hoste EA, Clermont G, Kersten A, Venkataraman R, Angus DC, de Bacquer D, Kellum JA. RIFLE criteria for acute kidney injury are associated with hospital mortality in critically ill patients: a cohort analysis. Crit Care 2006; 10: R73.

86. Praught M, Shlipak MG. Are small changes in serum creatinine an important risk factor? Curr Opinion Nephrol Hypertension 2005; 14: 265-270.

87. Ishani A, Xue JL, Himmelfarb J, Eggers PW, Kimmel PL, Molitoris BA, Collins AJ. Acute kidney injury increases risk of ESRD among elderly. J Am Soc Nephrol 2009; 20: 223-228.

88. Chertow GM, Christiansen CL, Cleary PD, Munro C, Lazarus JM. Prognostic stratification in critically ill patients with acute renal failure requiring dialysis. Arch Intern Med 1995; 155: 1505-1511.

89. Macedo E, Bouchard J, Mehta RL. Renal recovery following acute kidney injury. Curr Opin Crit Care 2008; 14: 660-666.

90. Bedford M, Stevens PE, Wheeler TWK, Farmer C. What is the real impact of acute kidney injury? BMC Nephrology 2014; 15: 95.

91. Bihorac A, Yavas S, Sunniah S, Hobson CE, Schold JD, Gabrielli A, Layon AJ, Segal MS. Long-term risk of mortality and acute kidney injury during hospitalisation after major surgery. Ann Surg 2009; 249 (5): 851-858.

92. Noble JS, Simpson K, Allison ME. Long-term quality of life and hospital mortality in patients treated with intermittent or continuous haemodialysis for acute renal and respiratory failure. Ren Fail 2006; 28: 323-330.

93. Bagshaw SM, Uchino S, Bellomo R, Morimatsu H, Morgera S, Schetz M, Tan I, Bouman C, Macedo E, Gibney N, Tolwani A, Oudemans-van Straaten HM, Ronco C, Kellum JA. Timing of renal replacement therapy and clinical outcomes in critically ill patients with severe acute kidney injury. J Crit Care 2009; 24 (1): 129-140.

94. Ostermann M, Dickie H, Barrett NA. Renal replacement therapy in critically ill patients with acute kidney – when to start. Nephrol Dial Transplant 2012; 27 (6): 2242-2248.

95. McCullough PA, Soman SS. Contrast-induced nephropathy. Crit Care Clin 2005; 21: 261-280.

96. Mehran R, Aymong ED, Nikolsky E, et al. A simple risk-score for prediction of contrast-induced nephropathy after percutaneous coronary intervention: development and initial validation. J Am Cardio 2004; 44: 1393-1399.

97. Stewart J, Finlay G, Smith N, et al. Adding Insult to Injury: A review of the care of patients who died in hospital with a primary diagnosis of acute kidney injury (acute renal failure). National Confidential Enquiry into Patient Outcome and Death (NCEPOD) report. 2009.

98. James GD, Sealey JE, Alderman M, Ljungman S, Mueller FB, Pecker MS, Laragh JH. A longitudinal study of urinary creatinine and creatinine clearance in normal subjects. Race, sex and age differences. Am J Hypertens 1988; 1(2): 124-131.

99. Preiss DJ, Godber IM, Lamb EJ, Dalton RN, Gunn IR. The influence of a cooked-meat meal on estimated glomerular filtration rate. Ann Clin Biochem 2007; 44(1): 35-42.

100. Nair S, O'Brien SV, Hayden K, Pandya B, Lisboa PJ, Hardy KJ, Wilding JP. Effect of a cooked meat meal on serum creatinine and estimated glomerular filtration rate in diabetes-related kidney disease. Diabetes Care 2014; 37 (2): 483-487.

101. National Kidney Foundation. K/DOQI clinical practice guidelines for chronic kidney disease: evaluation, classification, and stratification. 2012.

102. Myers GL, Miller GW, Coresh J, Fleming J, Greenberg N, Greene T, Hostetter T, Lever AS, Panteghini M, Welch M, Eckfeldt JH. Recommendations for improving serum creatinine measurement: A report from the Laboratory Working Group of National Kidney Disease Education Program. Clinical Chem 2006; 52 (1): 5-18.

103. Stevens LA, Levey AS. Measured GFR as a confirmatory test for estimated GFR J Am Soc Nephrol 2009; 20: 2305-2313.

104. Perrone RD, Steinman TI, Beck GJ, et al. Utility of radioisotopic filtration markers in chronic renal insufficiency: simultaneous comparison of 125I-iothalamate, 169Yb-DTPA, 99mTc-DTPA, and inulin. The Modification of Diet in Renal Disease Study. Am J Kidney Dis. 1990; 16 (3): 224-35.

105. Sterner G, Frennby B, Hultberg B, Almen T. Iohexol clearance for GFR-determination in renal failure – single or multiple plasma sampling? Nephrol Dial Transplant 1996; 11: 521-525.

106. Brochner-Mortensen J. A simple method for the determination of glomerular filtration rate. Scand J Clin Lab Invest. 1972; 30 (3): 271-4.

107. Cockcroft DW, Gault MH. Prediction of creatinine clearance from serum creatinine. Nephron 1976; 16 (1): 31-41.

108. Michels WM, Grootendorst DC, Verduijn M, Elliott EG, Dekker FW, Krediet RT. Performance of the Cockcroft-Gault, MDRD, and new CKD-EPI formulas in relation to GFR, age, and body size. Clin J Am Soc Nephrol 2011; 5 (6): 1003-1009.

109. Levey AS, Coresh J, Greene T, et al. Using standardised serum creatinine values in the modification of diet in renal disease study equation for estimating glomerular filtration rate. Ann Intern med 2006; 145 (4): 247-254.

110. Levey AS, Stevens LA, Schmid CH, et al. A new equation to estimate glomerular filtration rate. Ann Intern Med 2009; 150 (9): 604-612.

111. White CA, Akbari A, Doucette S, Fergusson D, Knoll GA. Estimating glomerular filtration rate in kidney transplantation: is the new chronic kidney disease epidemiology collaboration equation any better? Clin Chem. 2010; 56 (3): 474-477.

112. Tent H, Rook M, Stevens LA, et al. Renal function equations before and after living kidney donation: a within-individual comparison of performance at different levels of renal function. Clin J Am Soc Nephrol. 2010; 5 (11): 1960 1968.

113. Lane BR, Demirjian S, Weight CJ, Larson BT, Poggio ED, Campbell SC. Performance of the chronic kidney disease-epidemiology study equations for estimating glomerular filtration rate before and after nephrectomy. J Urol 2010; 183 (3): 896-901.

114. Earley A, Miskulin D, Lamb EJ, Levey AS, Uhlig K. Estimating Equations for Glomerular Filtration Rate in the Era of Creatinine Standardization: A Systematic Review. Ann Intern Med 2012; 156: 785-795.

115. Jones GR. Use of the CKD-EPI equation for estimation of GFR in an Australian cohort. Pathology. 2010; 42(5): 487-488.

116. Froissart M, Rossert J, Jacquot C, Paillard M, Houillier P: Predictive performance of the modification of diet in renal disease and Cockcroft-Gault equations for estimating renal function. J Am Soc Nephrol 2005; 16: 763-773.

117. Cirillo M, Lombardi C, Luciano MG, Bilancio G, Anastasio P, De Santo NG. Estimation of GFR: a comparison of new and established equations. Am J Kidney Dis 2010; 56 (4): 802-804.

118. Schwartz GJ, Furth S, Cole SR, Warady B, Munoz A. Glomerular filtration rate via plasma iohexol disappearance: Pilot study for chronic kidney disease in children. Kidney Int 2006; 69 (11): 2070-2077.

119. Lee CK, Swinford RD, Cerda RD, Portman RJ, Hwang W, Furth SL. Evaluation of serum creatinine concentration-based glomerular filtration rate equations in paediatric patients with chronic kidney disease. Pharmacotherapy 2012; 32 (7): 642-648.

120. Vickery S, Stevens P, Dalton N, van Lente F, Lamb E. Does the ID-MS traceable MDRD equation work and is it suitable for use with compensated Jaffe and enzymatic creatinine assays? Nephrol Dial transplant 2006; 21 (9): 2439-2445.

121. Inker L, Schmid CH, Tighiouart H, Eckfeldt JH, Feldman MD, Greene T, Kusek JW, Manzi J, Van Lente F, Zhang YL, Coresh J, Level AS. A new equation to estimate glomerular filtration rate from standardized creatinine and cystatin C. N Engl J Med 2012; 367: 20-29.

122. Eriksen BO, Mathisen UD, Melsom T, Ingebretson C, Jenssen TG, Njolstad I, Solbu MD, Toft I. Cystatin C is not a better estimator of GFR than plasma creatinine in the general population. Kidney Int. 2010; 78(12): 1305-1311.

123. Ix JH, Shlipak MG, Chertow GM, Whooley MA. Association of cystatin C with mortality, cardiovascular events, and incident heart failure among persons with coronary heart disease: data from the Heart and Soul Study. Circulation 2007; 115 (2): 173-179.

124. Rule AD, Gussak HM, Pond GR, Bergstralh EJ, Stegall MD, Cosio FG, Larson TS. Measured and estimated GFR in healthy potential kidney donors. Am J Kidney Dis 2004; 43 (1): 112-119.

125. Retnakaran R, Cull CA, Thorne KI, Adler AI, Holman RR. United Kingdom Prospective Diabetes Study (UKPDS) group: Risk factors for renal dysfunction in type 2 diabetes: UK Prospective Diabetes Study 74. Diabetes 2006; 55: 1832-1839.

126. Bang H, Mazumdar M, Newman G. Et al Screening for kidney disease in vascular patients: SCreening for Occult REnal Disease (SCORED) experience. Nephrol Dial Transplant 2009; 24: 2452-2457.

127. Sawicki PT, Didjurgiet U, Muhlhauser I, Bender R, Heinemann L, Berger M. Smoking is associated with progression of diabetic nephropathy. Diabetes Care 1994; 17: 126-131.

128. Orth SR, Stockmann A, Conradt C, Ritz E, Ferro M, Kreusser W et al. Smoking as a risk factor for end-stage renal failure in men with primary renal disease. Kidney Int 1998; 54: 926-931.

129. Ibanez L, Morlans M, Vidal X, Martinez MJ, Laporte JR. Case-control study of regular nonsteroidal anti-inflammatory use an end-stage renal disease. Kidney Int 2005; 67: 2393-2398.

130. Roth KS, Carter WH, Chan JC. Obstructive nephropathy in children: Long-term progression after relief of posterior urethral valve. Paediatrics 2001; 107 (5): 1004-1010.

131. Rifkin DE, Coca SG, Kalanta-Zadeh k. Does AKI truly lead to CKD? J Am Soc Nephrol 2012; 23 (6): 979-984.

132. Hemmelgarn BR, Manns BJ, Lloyds A, James MT, Klarenbach S, Quinn RR, Wiebe N, Tonelli M: Alberta Kidney Disease Network. Relation between kidney function, proteinuria, and adverse outcomes. JAMA 2010; 303 (5): 423-429.

133. Pohl MA, Blumenthal S, Cordonnier DJ, De Alvaro F, Deferrari G, Eisner G, Esmatjes E, Gilbert RE, Hunsicker LG, de Faria JB, Mangili R, Moore J Jr, Reisin E, Ritz E, Schernthaner G, Spitalewitz S, Tindall H, Rodby RA, Lewis EJ. Independent and additive impact of blood pressure control and angiotensin II receptor blockade on renal outcomes in the Irbesartan Diabetic Nephropathy Trial: Clinical implications and limitations. J Am Soc Nephrol 2005; 16: 3027–3037.

134. De Jong PE, Hileege HL, Pinto-Sietsma SJ, de Zeeuw D. Screening for microalbuminuria in the general population: a tool to detect subjects at risk for progressive renal failure in an early phase? Prevention of Renal and Vascular End-stage Disease (PREVEND) study. Nephrol Dial Transplant 2003; 18 (1): 10-13.

135. Penno G, Solini A, Bonora E, Fondelli C, Orsi E, Zerbini G. Clinical significance of nonalbuminuric renal impairment in Type 2 diabetic patients and in the general population (National Frequency of Renal Impairment coexisting with NIDDM (NEFRON) 11). Diabetes Care 2009; 32: 1497-1592.

136. Halimi JM. The emerging concept of chronic kidney disease without clinical proteinuria in diabetic patients. Diabetes Metab 2012; 38 (4): 291-297.

137. Lewis EJ, Hunsicker LG, Clarke WR, Berl T, Pohl MA, Lewis JB, Ritz E, Atkins RC, Rohde R, Raz I: Collaborative Study Group. Renoprotective effect of the angiotensin-receptor antagonist irbesartan in patients with nephropathy (REIN) due to type 2 diabetes. N Engl J Med 2001; 345: 851 – 860.

138. Fink HA, Ishani A, Tatlor BC, Greer NL, MacDonald R, Rossini D, Sadiq S, Lanrireddy S, Kane RL, Wilt TJ. Screening for monitoring and treatment of chronic kidney disease stages 1 to 3: a systematic review for the U.S. Preventative Services Task Force and Americal College of Physicians Clinical Practice Guideline. Ann Intern Med 2012; 156 (8): 570.

139. Heart Outcomes Prevention Evaluation Study Investigators (HOPE): Effects of ramipril on cardiovascular and microvascular outcomes in people with diabetes mellitus: Results of the HOPE study and MICRO-HOPE substudy. Lancet 2000; 355: 253 –259.

140. de Zeeuw D, Remuzzi G, Parving HH, Keane WF, Zhang Z, Shahinfar S, Snapinn S, Cooper ME, Mitch WE, Brenner BM: Proteinuria, a target for renoprotection in patients with type 2 diabetic nephropathy: Lessons from Reduction in End-points in Non-insulin Dependent Diabetes with the Angiotensin II Antagonist Losartan (RENAAL). Kidney Int 2004; 65: 2309 – 2320.

141. Matsushita K, Van der Velde M, Astor BC, Woodward M, Levey AS, de Joong PE, Coresh J, Gansevoort RT. Association of estimated glomerular filtration rate and albuminuria with all-cause mortality and cardiovascular mortality in general population cohorts: a collaborative meta-analysis. Lancet 2010; 375 (9731): 2073-2081.

142. Bakris GL. Slowing nephropathy progression: focus on proteinuria reduction. Clin J Am Soc Nephrol 2008; 3: S3-S10.

143. Vogt L, Laverman GD, de Zeeuw D, Navis N. The COOPERATE trial. Lancet 2003; 361 (9362): 1055-1056.

144. Mann JFE, Schmieder RE, McQueen M, et al on behalf of the ONTARGET investigators. Renal outcome with telmisartan, ramipril, or both, in people at high vascular risk (the ONTARGET study): a multicentre, randomised, double blind, controlled trial. Lancet 2008; 372: 547-553.

145. Palevsky PM. Chronic-on acute kidney injury. Kidney Int 2012; 81: 430-431.

146. Bagshaw SM, Laupland KB, Doig CJ, et al. Prognosis for long-term survival and renal recovery in critically ill patients with severe acute renal failure: a population-based study. Crit Care 2005; 9**:** R700-709.

147. Drawz PE, Miller RT, Sehgal AR. Predicting hospital-acquired acute kidney injury - a case-controlled study. Ren Fail 2008; 30**:** 848-855.

148. Waikar SS, Curhan GC, Wald R, McCarthy EP, Chertow GM. Declining mortality in patients with acute renal failure, 1988 to 2002. J Am Soc Nephrol 2006; 17: 1143-1150.

149. Bonventre JV, Weinberg JM. Recent advances in the pathophysiology of ischaemic acute kidney injury. J Am Soc Nephrol 2003; 14: 2199-22.

150. Sutton TA, Risher CJ, Molitoris BA. Microvascular endothelial injury and dysfunction during ischaemic acute renal failure. Kidney Int 2002; 62: 1539-1549.

151. Okusa MD, Chertow GM, Portilla D. The nexus of acute kidney injury, chronic kidney disease and world kidney day. Clin Am J Soc Nephrol 2009; 4: 520-522.

152. Bedford M, Farmer C, Levin A, Ali T, Stevens PE. Acute kidney injury and CKD: chicken or egg? Am J Kidney Dis 2012; 59 (4): 485-491.

153. Stoves J, Lindley EJ, Barnfield MC, Burniston M, Newstead C. MDRD equation estimates of glomerular filtration rate in potential living kidney donors and renal transplant recipients with impaired graft function. Nephrol Dial Transplant 2002: 17 (11); 2036-2037.

154. Jones SR, Carley S, Harrison M. An introduction to power and sample size estimation: Emerg Med J 2003; 20: 450-458.

155. Greenberg N, Roberts WL, Bachmann LM, et al. Specificity characteristics of 7 commercial creatinine measurement procedures by enzymatic and jaffe method principles. Clin Chem 2012; 58(2): 391-401.

156. Davison AC, Hinkle DV. Bootstrap methods and their application: Cambridge University Press (2006).

157. Roderick PJ. Chronic kidney disease in older people: a cause for concern? Nephrol Dial Transplant 2011; Editoral comment: 1-4.

158. Stevens LA, Schmid CH, Greene T, et al. Comparative performance of the CKD Epidemiology Collaboration (CKD-EPI) and the Modification of Diet in Renal Disease (MDRD) Study equations for estimating GFR levels above 60 ml/min/1.73 m2. Am J Kidney Dis 2010; 56 (3): 486-495.

159. Lamb EJ, Tomson CR, Roderick PJ. Estimating kidney function in adults using formulae. Ann Clin Biochem 2005; 42 (5): 321-345.

160. Murata K, Baumann NA, Saenger AK, Larson TS, Rule AD, Lieske JC. Relative performance of the MDRD and CKD-EPI equations for estimating glomerular filtration rate among patients with varied clinical presentations. Clin J Am Soc Nephrol 2011; 6 (8): 1963-1972.

161. Shastri S, Tighiouart H, Katz R, et al. Chronic kidney disease in octogenarians. Clin J Am Soc Nephrol 2011; 6 (6): 1410-1417.

162. Nair S, Mishra V, Hayden K, Lisboa PJ, Pandya B, Vinjamuri S, Hardy KJ, Wilding JP. The four-variable modification of diet in renal disease formula underestimates glomerular filtration rate in obese type 2 diabetic individuals with chronic kidney disease. Diabetes 2011; 54 (6): 1304-1307.

163. White SL, Polkinghorne KR, Atkins RC, Chadban SJ. Comparison of the prevalence and mortality risk of CKD in Australia using the CKD Epidemiology Collaboration (CKD-EPI) and Modification of Diet in Renal Disease (MDRD) Study GFR estimating equations: the AusDiab (Australian Diabetes, Obesity and Lifestyle) Study. Am J Kidney Dis. 2010; 55 (4): 660-670.

164. Matsushita K, Selvin E, Bash LD, Astor BC, Coresh J. Risk implications of the new CKD Epidemiology Collaboration (CKD-EPI) equation compared with the MDRD Study equation for estimated GFR: the Atherosclerosis Risk in Communities (ARIC) Study. Am J Kidney Dis 2010; 55 (4): 648-659.

165. Matsushita K, Mahmoodi BK, Woodward M, et al. Comparison of risk prediction using the CKD-EPI equation and the MDRD study equation for estimated glomerular filtration rate. JAMA 2012; 307 (18): 1941-1951.

166. Lamb EK, Wood J, Stowe HJ et al. Susceptibility of glomerular filtration rate estimations to variations in creatinine methodology: A study in older patients. Ann Clin Biochem 2005; 42: 11-18.

167. Carter JL, Stevens PE, Irving JE, Lamb EJ. Estimating glomerular filtration rate: comparison of the CKD-EPI and MDRD equations in a large UK cohort with particular emphasis on the effect of age. QJM 2011; 104 (10): 839-847.

168. Office of National Statistics UK, Experimental Statistics, Table EE1, 2007.

169. Ma, YC, Zuo L, Chen JH, Luo Q, Yu, XQ, Li Y, Xu JS, Juang SM, Wand LN, Huang Wang M, Xu GB, Wang HY. Modified glomerular filtration rate estimating equation for Chinese patients with chronic kidney disease. J Am Soc Nephrol 2006; 17 (10): 2937-2944.

170. Kitiyakara C, Yamwong S, Vathesatogkit P, Chittamma A, Cheepudomwit S, Vananan S, Hengprasith B, Sritara P. The impact of different GFR estimating equations on the prevalenve of CKD and risk groups in a Southeast Asian cohort using the KDIGO guidelines. BMC Nephrol 2012; 13: 1.

171. Horio M, Imai E, Yasuda Y, Watanbe T, Matsuo S. Modification of the CKD epidemiology colboration (CKD-EPI) equation for Japanese: accuracy and use for population estimates. Am J Kidney Dis 2010; 56 (1): 32-38.

172. Hsu CY, Bansal N. Measured GFR as "gold standard"-all that glitters is not gold? Clin J Am Soc Nephrol 2011; 6 (8): 1813-1814.

173. Seegmiller JC, Burns BE, Fauq AH, Mukhtar N, Lieske JC, Larson TS. Iothalamate quantification by tandem mass spectrometry to measure glomerular filtration rate. Clin Chem 2010; 56 (4): 568-574.

174. Perrone RD, Steinman TI, Beck GJ et al. Utility of radioisotopic filtration marokers in chronic renal insufficiency: simultaneous comparison of ^{125}Iothalamate, ^{169}Yb-DTPA, 99mTc-DPTA, and inulin. The Modification of Diet in Renal Disease Study. Am J Kidney Dis 1990; 16 (3): 224-235.

175. Sterner G, Frennby B, Hultberg B, et al. Iohexol clearance for GFR-determination in renal failure – single or multiple plasma sampling? Nephrol Dial Transplant 1996; 11: 521.

176. Stolz A, Hoizey G, Toupance O, et al. Evaluation of sample bias for measuring plasma iohexol clearance in kidney transplantation. Transplantation 2010; 89 (4): 440-445.

177. Isaka Y, Yamamoto S, Fujiwara Y, et al. A new analytical method for simultaneous measurement of iothalamate and iohexol. Rinsho Byori 1992; 40 (7): 703-707.

178. Erley CM, Bader, BD, Berger ED, et al. Plasma clearance of iodine contrast media as a measure of glomerular filtration rate in critically ill patients. Crit Care Med 2001; 29 (8): 1544-1550.

179. Schaeffer ES, Van der Giet M, Gaedeke J, Tolle M, Ebert N, Kuhlmann MK, Martus P. The Berlin Initiative Study (BIS): the methodology of exploring kidney function in the elderly by combining a longitudinal and cross-sectional approach. Eur J Epidemiol 2010; 25 (3): 203-210.

180. Alshaer I, Kilbride HS, Stevens P, Eaglestone G, Knight S, Carter J, Delaney M, Farmer C, Irving J, O'Riordan S, Dalton N, Lamb E. External validation of the Berlin equations for estimation of GFR in the elderly. Am J Kidney Dis 2014; 63 (5): 859-867.

181. Stevens L, Coresh J, Schmid C, Feldman H, Froissart M, Kusek J, Rossert J, van Lente F, Bruce R, Zhang Y, Greene T, Levey A. Estimating GFR using serum cystatin C alone and in combination with serum creatinine: A pooled analysis of 3134 individuals with CKD. Am J Kidney Dis 2008; 51 (3): 396-406.

182. Kilbride HS, Stevens P, Eaglestone G, Knight S, Carter J, Delaney M, Farmer C, Irving J, O'Riordan S, Dalton N, Lamb E. Accuracy of the MDRD (Modification of Diet in Renal Disease) study and CKD-EPI (CKD Epidemiology Collaboration) equations for estimation of GFR in the elderly. Am J Kidney Dis 2013; 61 (1): 55-66.

183. Willems D, Wolff F, Mekahli F, Gillet C. Cystatin C for early detection of renal impairment in diabetes. Clinical Biochemistry 2009: 42 (1-2); 108-110.

184. Dharmidharka VR, Kwon C, Stevens G. Serum cystatin C is superior to serum creatinine as a marker of renal function: a meta-analysis. Am J Kidney Dis 2002: 40; 221-226.

185. Ix JH, Shlipak MG, CHertow GM, Whooley MA. Association of cystatin C with mortality, cardiovascular events, and incident heart failure among persons with coronary heart disease: data from the Heart and Soul Study. Circulation 2007; 115 (2): 173-179.

186. Glassock RJ, Winearls C. An epidemic of chronic kidney disease: fact or fiction? Nephrol Dial Transplant 2008; 23: 1118-1121.

187. Douville P, Martel AR, Talbot J, et al. Impact of age on glomerular filtration estimates. Nephrol Dial Transplant 2009; 24: 97-103.

188. Alan S, Go M, Glenn M, Chertow M et al. Chronic kidney disease and the risks of death, cardiovascular events, and hospitalisation. N Engl J Med 2004; 351: 1296-1305.

189. Rowe, Wright. The Delphi technique as a forecasting tool: issues and analysis. International Journal of Forecasting 1999; 15 (4).

190. Karunaratne K, Stevens P, Irving J, Hobbs H, Kilbride H, Kingston R, Farmer C. The impact of par performance on the control of blood pressure in people with chronic kidney disease stage 3-5. Nephrol Dial Transplant 2013; 28 (8): 2107-2116.

191. James MT, Ghali WA, Knudtson ML, Ravanu P, Tonelli M, Faris P, Pannu N, Manns BJ, Klarenbach SW, Hemmelgarn. Associations between acute kidney injury and cardiovascular and renal outcomes after coronary angiography. Circulation 2011; 123: 409-416.

192. Obermayr R, Temml C, Knechtelsdorfer M, et al. Predictors of new-onset decline in kidney function in a general middle-European population. Nephrol Dial Transplant 2008; 23: 1265-1273.

193. Kronborg J, Solbu M, Njolstad I, et al: Predictors of change in estimated GFR: a population-based 7-year follow-up from the Tromso study. Nephrol Dial Transplant 2008; 23: 2818-2826.

194. Liang L, Chang A, Rostand SG, Hebert L, Appel LJ, Astor BC, Lipkowitz MS, Wright JT, Kendrick C, Wang X, Greene T. A within-patient analysis for time-varying risk factors of CKD progression. J Am Soc Nephrol 2014; 25 (3): 606-613.

195. Singh P, Rifkin DE, Blantz RC. Chronic kidney disease: an inherent risk factor for acute kidney injury. Clin J Am Soc Nephrol 2010; 5: 1690-1695.

196. Khosla N, Soroko SB, Chertow GM, Himmelfarb J, Alp Ikizler T, Paganini E, Mehta RL. Preexisting chronic kidney disease: a potential for improved outcomes from acute kidney injury. Clin J Am Soc Nephrol 2009; 4 (12): 1914-1919.

197. James MT, Hemmelgarn BR, WiebeN, Pannu N,Manns BJ, Klarenbach SW, Tonelli M; Alberta Kidney Disease Network: Glomerular filtration rate, proteinuria, and the incidence and consequences of acute kidney injury: A cohort study. Lancet 2010; 376: 2096–2103.

198. Parikh CR, Devarajan P, Zappitelli M, et al. Postoperative biomarkers predict acute kidney injury and poor outcomes after paediatric cardiac surgery. J Am Soc Nephrol 2011; 22: 1737–1747.

199. Finlay S, Bray B, Lewingtin AJ, Hunter-Rowe CT, Banerjee A, Atkinson JM, Jones MC. Idenitification of risk factors associated with acute kidney injury in patients admitted to acute medical units. Clin Med 2013; 13 (3): 233-238.

200. Schoolwerth AC, Sica DA, Ballermann BJ, Wilcox CS. Renal considerations in angiotensin converting enzyme inhibitor therapy. Circulation 2001; 104: 1985-1991.

201. Rifkin DE, Coca SG, Kalantar-Zadeh K. Does AKI truly lead to CKD? J Am Soc Nephrol 2012; 23 (6): 979-84.

202. Triverio PA, Martin PY, Romand J, Pugin J, Perneger T, Saudan P. Long-term prognosis after acute kidney injury requiring renal replacement therapy. Nephrol Dial Transplant 2009; 24 (7): 2186-2189.

203. Askenazi DJ, Feig DI, Graham NM, Hui-Stickle S, Goldstein SL. 3–5 year longitudinal follow-up of paediatric patients after acute renal failure. Kidney Int 2006; 69: 184–189.

204. Bucaloiu ID, Kirchner HL, Norfolk ER, Hartle JE, Perkins RM. Increased risk of death and de novo chronic kdieny disease following reversible acute kidney injury. Kidney Int 2012; 81 (5): 477-485.

205. Robertson S, Newbigging K, Isles CG, Brammah A, Allan A, Norrie J. High incidence of renal failure requiring short-term dialysis: a prospective observational study. QJM 2002; 95: 585-590.

206. United States Renal Disease Survey, 2006. United States Renal Data System. Annual data report, 2006 (USRDS)

207. Thakar CV, Christianson A, Himmelfarb J, et al. Acute kidney injury episodes and chronic kidney disease in diabetes mellitus. Clin J Am Soc Nephrol 2011; 6: 2567-2572.

208. Wald R, Quinn RR, Luo J, Li P, Scales DC, Mamdani MM, Ray JG. Chronic dialysis and death among survivors of acute kidney injury requiring dialysis. JAMA 2009; 302 (11): 1179-1185.

209. Chawla LS; Amdur RL, Amodeo S, Kimmel PL, Palant CE. The severity of acute kidney injury predicts progression to chronic kidney disease. Kidney Int 2011; 79: 1361-1369.

210. Chawla LS, Kimmel P. Acute kidney injury and chronic kidney disease: an integrated syndrome. Kidney Int 2012; 82 (5): 516-524.

211. Alebioscu CO. An update on 'progression promoters' in renal diseases. J Nat Med Assoc 2003. 95 (1): p 30-42.

212. Lafrance, J.P. and D.R. Miller. Defining acute kidney injury in database studies: the effects of varying the baseline kidney function assessment period and considering CKD status. Am J Kidney Dis 2010; 56 (4): 651-660.

213. Davies, A, Green, C, Hutton J. Severe Sepsis: a European estimate of the burden of disease in ICU. Int Care Med 2001; 27: S284.

214. Ftouh S, Thomas M. Acute kidney injury: summary of NICE guidance. BMJ 2013; 347: f4930.

215. Shrier RW, Wang W. Acute renal failure and sepsis. N Eng J Med 2004; 351: 159-169.

216. Vitorio D, Maciel AT. Acute kidney injury induced by Systemic Inflammatory Response Syndrome is an avid and persistent sodium-retaining state. Case Rep Crit Care 2014; 2014; 471658.

217. Daniels R, Nutbeam T, McNamara G, Galvin C. The sepsis six and the severe sepsis resuscitation bundle: a prospective observational cohort study. Emerg Med J 2011; 28: 507-512.

218. Fortescue EB, Bates DW, Chertow. Predicting acute renal failure after coronary bypass surgery: cross-validation of two-risk stratification algorithms. Kidney Int 2000; 57: 2594-2602.

219. Mehran R, Aymong ED, Nikolsky E, Lasic Z, Iakovou, Fahy M, Mintz G, Lansky AJ, Moses JW, Stone G, Leon MB, Dangas G. A simple risk score for prediction of contrast-induced percutaneous coronary intervention. J Am Coll Cardiol 2004; 44 (7) 1393-1399.

220. Tangri N, Stevens LA, Griffith J, Tihiouart H, Djurdjev O, Naimark D, Levin A, Levey. A predictive model for progression of chronic kidney disease to kidney failure. JAMA 2011; 305 (15): 1553-1559.

221. Kellum JA, Mehta RL, Levin A, Molitoris BA, Warnock DG, Shah SV, Joannidis M, Ronco C, for the Acute Kidney Injury Network (AKIN): Development of a clinical research agenda for acute kidney injury using an international, interdisciplinary, three-step modified Delphi process. Clin J Am Soc Nephrol 2008; 3: 887–894.

Appendix