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INSIDE

Review of

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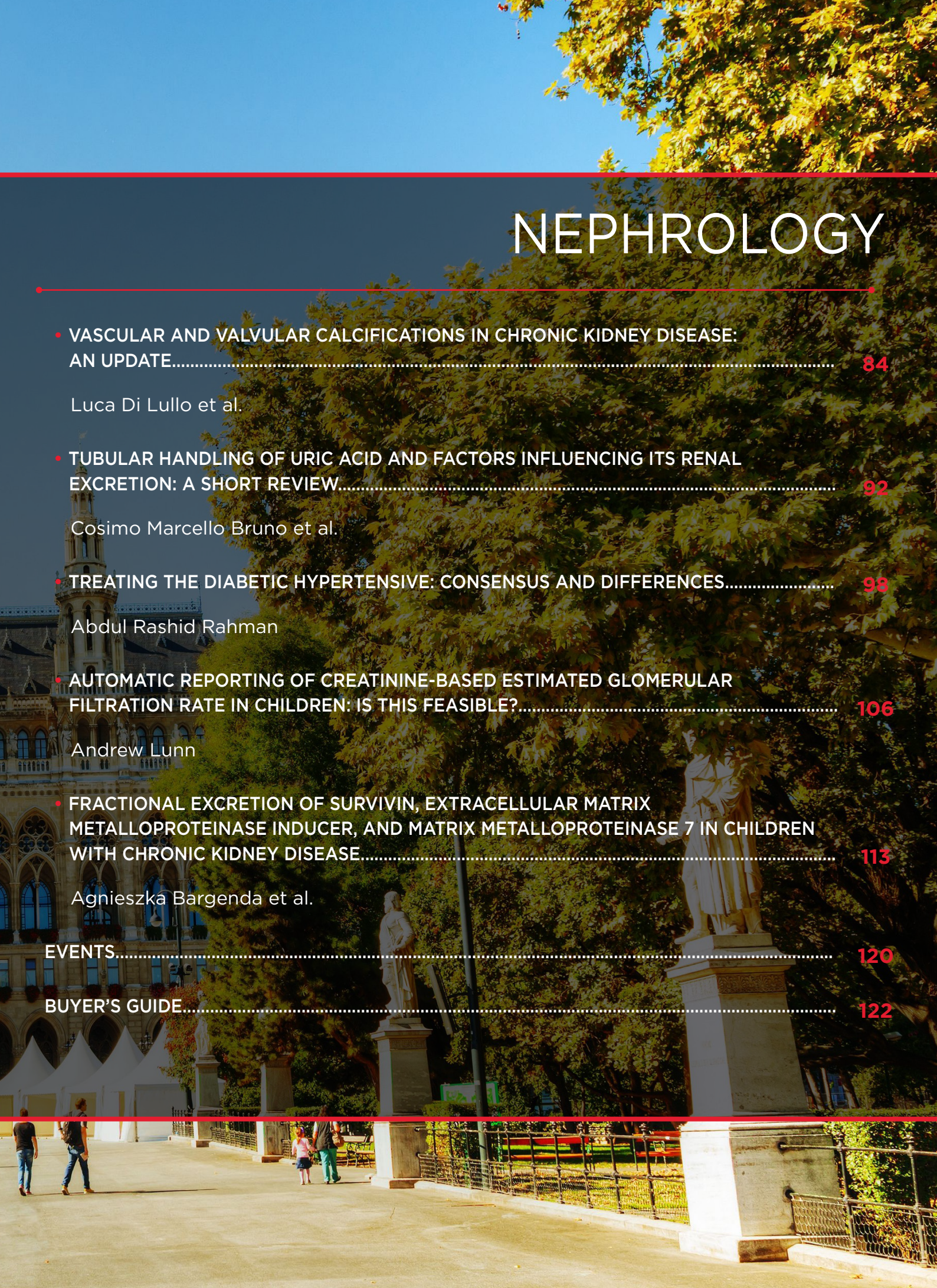
CONTENTS



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Welcome

Welcome to the *European Medical Journal Nephrology*, bringing you all the latest from the world of nephrology as well as highlights from the recent European Renal Association - European Dialysis and Transplantation Association (ERA-EDTA) Congress. We hope you will enjoy catching up on our summary of the congress alongside a selection of brand new peer-reviewed articles, and interviews with our esteemed Editorial Board.

This May, the ERA-EDTA Congress 2016 provided a fascinating 4-day programme of research, discussion, and debate in the historic city of Vienna, Austria. Comprising 56 scientific symposia and 35 free communication sessions, the congress was bursting with information and exciting developments, all focussing on different aspects of this year's main theme, which was "From big data to personalised therapy, biostatistics meets molecular medicine." In our congress review section, you can read up on all of the most exciting news presented throughout the event.

We also bring you abstract reviews of the fascinating research presented at the congress, including Szummer's cardiology perspective on treatments for chronic kidney disease, a discussion of the risks of kidney donation based on long-term follow-up data from Mjøen, and the use of intracellular signals as druggable targets in renal inflammation by Egido and Gómez-Guerrero. Members of our Editorial Board have kindly offered some words of wisdom as well as their take on some of the research presented at this year's congress and beyond. Their combined knowledge and experience of the field make our interviews section a fascinating read for nephrologists from all walks of life.

In this edition we also bring you a selection of the very best peer-reviewed articles from across the field. Di Lullo et al. have put forward an important update on vascular and valvular calcifications in chronic kidney disease, and Bargenda et al. bring us the results of a study into potential biomarkers of paediatric chronic kidney disease. In addition, you will find articles on the topics of uric acid in chronic kidney disease, metastatic renal cell carcinoma patients on dialysis, and much more.

We hope you will find this latest edition of *European Medical Journal Nephrology* an interesting and useful read, looking back on the past year's nephrology advancements and of course the highlights of the ERA-EDTA Congress 2016. We are already looking forward to next year's congress in Madrid, Spain, and all of the new results and developments which will be presented. We hope to see you there!

Spencer Gore

Director, European Medical Journal

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Front cover and contents photograph: Vienna, Austria, home of ERA-EDTA 2016.

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Foreword

Prof Norbert Lameire

Emeritus Professor of Medicine and Nephrology, Medical Faculty of Ghent University, Ghent, Belgium.

Dear Colleagues,

In continuation of a good tradition, this issue of the *European Medical Journal Nephrology* is published following the ERA-EDTA 53rd Congress in Vienna, Austria. The congress took place from 21st–24th May 2016. In keeping with previous years, a superb scientific and clinical programme was presented by an increasingly international group of invited speakers. In addition, the ERA-EDTA congress also offered the opportunity to young, basic, and clinical researchers from Europe and beyond to present their latest research in symposia, free communications, and poster sessions. As in previous years, the whole spectrum of kidney diseases was covered, including kidney physiology, immunology and molecular biology, fluid and electrolyte disturbances, hereditary diseases, pregnancy and the kidney, and paediatric nephrology. Acute kidney injury and intensive care nephrology were discussed along with all aspects of chronic kidney disease and renal replacement therapy. The congress started with a special symposium on 'disaster nephrology', a field to which European nephrology has greatly contributed.

> "The ERA-EDTA congress also offered the opportunity to young, basic, and clinical researchers from Europe and beyond to present their latest research in symposia, free communications, and poster sessions."

The papers included in this issue of the journal reflect the wide range of topics seen at this year's ERA-EDTA congress, with a broad selection of articles. Included is a consideration of creatinine-based reporting of estimated glomerular filtration rate in children from Lunn. We have a number of articles on chronic kidney disease: firstly, an appraisal of the role of uric acid from Galassi et al., and secondly, an up-to-date review on valvular and vascular calcifications from Di Lullo et al. Alongside these we have many other articles and as usual, all papers were externally reviewed, ensuring a high standard of content.

It is an exciting and enlightening period in the field of nephrology, and this is evidenced in this edition of *EMJ Nephrology*. The editorial board of the journal wishes you pleasant reading and hopes to meet some of the readers personally at the next ERA-EDTA congress.

Norbert Lameire

Emeritus Professor of Medicine and Nephrology, Medical Faculty of Ghent University, Ghent, Belgium; Past Chairman of the European Kidney Health Alliance.

ERA-EDTA ANNUAL CONGRESS 2016

THE AUSTRIA CENTER VIENNA,
VIENNA, AUSTRIA
21ST-24TH MAY 2016

Welcome to the *European Medical Journal* review of the 53rd Annual Meeting of the ERA-EDTA Congress

This year's congress stood on the shoulders of the giants of medical history; from Semmelweis and his famous improvements to hygiene in hospitals, to Landsteiner and the discovery of the ABO blood typing system, there is no denying that Vienna, Austria is the perfect location for celebrating and creating medical progress. ERA-EDTA 2016 lived up to this history with many major developments being presented and many future successes discussed.

The congress had almost 8,000 delegates in attendance this year, which reflects the continued success that the event has enjoyed over the last 53 years. The theme this year was indicative of the innovative and futuristic nature of nephrology practice: 'From big data to personalised therapy: Biostatistics meets molecular medicine'. In an interview, Prof Gert Mayer, President of the congress, described these new techniques as complementary to current methods, stating that the two can go "hand-in-hand". This year also saw a video message from Karen Kadenbach, Member of European Parliament, on health policy-making from the perspective of the European Community. This, along with many cross-society discussions, formed a great deal of collaboration between clinicians, academics, policy makers, and industry to help develop European nephrology care.

Prof Andrzej Więcek, President of ERA-EDTA, spoke about the future of both the congress and the society with great promise. Firstly, he discussed the growing role of young nephrologists and encouraged their involvement, saying: "The youngsters are our future, they keep our society young, alive, and continually evolving!" Following this he discussed the current challenges in nephrology and the importance of increasing opportunities for women in the field.

An abundance of awards were received during the congress for some exemplary achievements from across the spectrum of nephrology. The award for the category 'Outstanding Contributions to Nephrology' went to Prof Roseanna Coppo, Italy, for her long-standing positions in educational programmes, scientific committees, and journals. Prof Pierre Ronco, France, was the recipient of the 'Outstanding Basic Science Contributions to Nephrology' award, specifically for his work in the pathophysiology and immunology of renal disease. Prof Christoph Wanner,

Germany, is the pioneer of a number of clinical trials which have developed the field of cardiovascular and renal health in Type II diabetes and revolutionised treatment options, making him a deserving winner of the award for 'Outstanding Clinical Science Contributions to Nephrology'. The Stanley Shaldon award for young investigators went to Dr Emilie Cornec-Le Gall, France, who has consistently presented outstanding work over the last 3 years and is now completing a post-doctoral fellowship at the Mayo Clinic.

In addition to these awards, a number of smaller awards were given for the best abstracts presented during the congress. These covered many topics and represented institutions from across the world. With regards to the presentations that were on display at the event, delegates were treated to fascinating insights and studies that reflected every aspect of the field of nephrology. With topics including chronic kidney disease, renal replacement therapy, and dialysis, this year's ERA-EDTA really had something for everyone to take away from the event.

> "The youngsters are our future, they keep our society young, alive, and continually evolving!"

The team at EMJ has been hugely impressed by the high quality of research and the number of developments that have been made in the world of nephrology this year. We have distilled down the most impactful news and the most interesting presentations and discussions from ERA-EDTA 2016 in this review. We hope that it provides you with some thought-provoking ideas that will help to develop and refine your practice. The 54th edition of ERA-EDTA will take place in Madrid, Spain next year, and we hope to see many of you there!

Update on Miracle Baby Dialysis Machine

MAY 2016 marks the 2-year anniversary of the creation of the innovative baby dialysis machine, CARPEDIEM (cardo-renal paediatric dialysis emergency machine), by Prof Claudio Ronco, Director of Department of Nephrology, Dialysis and Transplantation, International Renal Research Institute, St Bortolo Hospital, Vicenza, Italy.

Prior to the creation of this machine, babies born with acute kidney injury (AKI) had a high risk of mortality due to the lack of adequate dialysis equipment. Adult dialysis machines would be used, only with smaller filters and other applications not specifically suitable for such small patients with significantly smaller blood vessels. These adaptations tended to harm the patient.

AKI in neonates is not a rare phenomenon. In a recent study, 30.3% of the 357 enrolled neonates had AKI, 72.2% of whom were at AKI Stage 1. The number of paediatric AKI patients receiving surgery for congenital heart disease was even higher at 45%. Another study showed the outcome of using adult dialysis machines on neonates with both AKI and sepsis to have a particularly high mortality rate, at 70.2%.

So far, more than 40 babies have been treated and many have been saved. According to Prof Ronco, the mortality rate for babies has been significantly reduced to 30–35%, halving the previous rate.

CARPEDIEM marked a huge turning point for the treatment of AKI in neonates, as symbolised by its first success story, a baby girl named Lisa, and continues to be celebrated as a fantastic innovation which is saving the lives of many babies. So far, more than 40 babies have been treated and many have been saved. According to Prof Ronco, the mortality rate for babies has been significantly reduced to 30–35%, halving the previous rate.

The future of CARPEDIEM is looking bright. According to an ERA-EDTA press release dated 22nd May 2016, a publication is being drafted to detail the success so far of the machine, as well as extensive documentation being recorded in order to properly assess the outcomes of all the babies treated with CARPEDIEM. Additionally, long-term data will be gathered, which will hopefully provide further details of its development.

Peritoneal Dialysis: Providing Options for the Elderly

PRE-EMPTIVE placement of a marsupialised catheter can increase peritoneal dialysis (PD) prevalence. PD has been declining across Europe, with <10% of patients opting for its use due to various socioeconomic factors. However, this form of dialysis can have outcomes comparable to haemodialysis as well as inflicting less stress on the circulatory system, lending itself to use in geriatric patients.

The research, presented in an ERA-EDTA press release dated 22nd May 2016, incorporated 140 patients who had a marsupialised catheter inserted during a pre-dialysis education programme. These patients had an estimated glomerular filtration rate of 10–15 mL/min; mean age of the patients was 65.9±14.5 years and the cohort was 65.7% male.

The process was conducted using a simplified Moncrief–Popovich technique and an incremental dialysis dose (IDD) programme was available. Over a 3-year follow-up data regarding the catheter, patient training time, and PD prescriptions were recorded. The catheter had a short break-in and long survival time (75±291 days and 676±508 days, respectively); there were no severe complications and all catheters were patent at removal. The number of patients opting for PD increased from 22 to 63, representing a prevalence increase from 8% to 19%.

The training in the use of the catheter was simple with 88% completed as a short, outpatient session. Those in the IDD group (n=86) were split into three groups by age: <65 years, 65–75 years, and >75 years old. Those >75 years old maintained the one-night exchange for longer than the other groups (164 days and 285 days versus 480 days) and the same pattern was observed for two exchanges/day (394 days and 440 days versus 711 days). This group saw an increased medium clearance whereas a decline was seen in the other groups. This research thus provides more options for healthy elderly patients who may wish to avoid haemodialysis.

Almost 8,000 delegates

Blood Pressure a Factor in Chronic Kidney Disease Cardiovascular Risk

LOWER blood pressure has been revealed to be beneficial for people with increased cardiovascular risk, including patients with chronic kidney disease (CKD), a recent study has revealed.

> CKD patients have an excessively high cardiovascular mortality and therefore every measure should be taken to reduce it.

The SPRINT study, a randomised trial of >9,000 patients with CKD, showed that better outcomes for a composite primary endpoint of myocardial infarction, acute coronary syndrome, stroke, heart failure, or death were achieved in the patients who received intensive therapy to lower their blood pressure, although no benefit was seen in composite renal outcome. In an ERA-EDTA press release dated 21st May 2016, Prof Carmine Zoccali, Nephrology, Hypertension, and Renal Transplantation Unit, Ospedali Riuniti, Reggio Calabria, Italy, commented that "Because no renal benefits were seen, many doctors believed that there is no advantage of a more intensive blood pressure control in CKD patients. But renal benefits are only one side of the coin, the other side are the cardiovascular benefits."

A meta-analysis of 16 studies showed that lower blood pressure equated to lower cardiovascular risk in those patients considered high-risk, and the reduction was the same for patients with and without CKD. CKD patients with the same high level of cardiovascular risk as non-CKD patients had a cardiovascular benefit if their blood pressure was reduced to <120 mmHg.

Although CKD patients were included in the present study, patients with advanced CKD were excluded (i.e. those with glomerular filtration rate <20 /mL/min/1.73 m^2 or proteinuria 1 g/day). Since lowering blood pressure must be undertaken carefully to avoid side effects, a new randomised trial would need to take place to test if these patients, who are also likely to benefit from the treatment, have similar outcomes. Prof Zoccali counselled that "CKD patients have an excessively high cardiovascular mortality and therefore every measure should be taken to reduce it. A stricter surveillance of the more intensively treated patients is needed."

Elbasvir/Grazoprevir for Heptatitis C Virus-Infected Chronic Kidney Disease Patients

HOPE for treatment of hepatitis C virus (HCV)-infected patients with chronic kidney disease (CKD) may be just around the corner, according to a recent study presented in an ERA-EDTA press release dated 22nd May 2016. Until now there have been few therapeutic options available for HCV infection with advanced kidney disease; both sofosbuvir and ribavirin for example are only used in patients with a good glomerular filtration rate. This Phase III, randomised, controlled C-SURFER study was the first to evaluate an all-oral, ribavirin-free regimen in patients with CKD Stages 4 and 5.

> SVR12 was achieved in 94.6% of all patients...

The study randomised 224 HCV genotype 1a and 1b (52% and 48%, respectively) and CKD Stage 4 and 5 patients, some of whom had received haemodialysis to elbasvir/grazoprevir (EBR/GZR) (50/100 mg) or placebo for 12 weeks. Pharmacokinetic sampling was performed on 11 patients. The primary efficacy endpoint of the trial was sustained virological response at 12 weeks (SVR12). In the genotype 1a patients, the impact of resistance-associated variants (RAVs) of particular amino acids on SVR12 was assessed. Health-related quality of life was evaluated using the SF-36v2® health survey. The placebo patients received EBR/GZR after the placebo therapy was complete.

SVR12 was achieved in 94.6% of all patients (12 failed: relapsed n=3, adverse event discontinuation n=1, administrative reason n=8) and once adjusted for those who discontinued for reasons not associated to the drug this increased to 98.6%. RAVs were present in 11.7% of the genotype 1a patients; SVR12 was achieved in 100% without RAVs and in 84.6% with. During the placebo-controlled phase of the trial adverse events occurred in 16 EBR/GZR patients and 17 placebo patients, discontinuation was 0% and 4%, respectively. In the pharmacokinetic sampling data, dose-adjustment in haemodialysis patients was not indicated.

Low rates of adverse events and high rates of SVR12 suggest a positive future for CKD and HCV-infected patients using once daily EBR/GZR, even in genotype 1a patients.

Vienna

Treating Kidney Disease in Older Patients Needs Specialised Research

CALLS for developments in specialised treatment for older incident dialysis patients (≥65 years old) were proposed at this year's 53rd ERA-EDTA Congress in Vienna, Austria. A statement paper, presented on the 22nd May 2016, discusses the importance of looking into the nutritional derangements in older patients on dialysis, pointing out the need for interdisciplinary collaborations between renal and geriatric clinic studies to assess the impact that diet, as well as lifestyle, have on older dialysis patients.

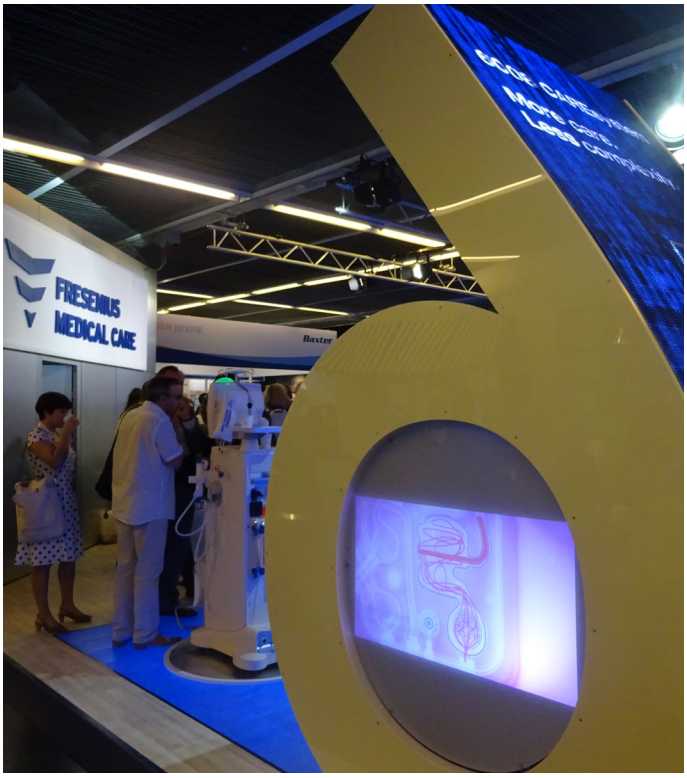
Generally, as people age they are likely to experience muscle performance loss as well as having an increased risk of poor health, disabilities, hospitalisation, and mortality. A high protein diet with enough calories and no amino acid deficiencies, however, can help promote muscle growth and therefore make people less susceptible to frailty and sarcopenia in later years, and therefore should be promoted, according to the paper.

Dr Lina Johansson, Clinical Academic Dietitian, Imperial College Healthcare NHS Trust, London, UK, stated the importance of the need for further research in an ERA-EDTA press release dated 22nd May 2016. "What seems to happen in cases of kidney disease is that frailty and sarcopenia actually occur a lot earlier in life, so people on dialysis are likely to become frail and develop sarcopenia at an earlier age than others, and their rate of muscle mass loss seems to be faster and their nutritional status seems to be worse."

Although this may be the case, Dr Johansson pointed out that no research has been conducted on elderly patients on dialysis specifically. With a dialysis incidence rate of 55% in patients ≥65 years old, nephrologists need to develop specialised healthcare to manage the specific needs of this population of patients, it is argued. Dr Johansson also emphasised the importance of looking at social aspects such as cooking, shopping, and other routine tasks when seeking to address poor nutrition in older people, which are not usually taken into account.

Caution Advised to People Considering High-Protein Diets

MEDICAL examination should be sought before starting a high-protein diet, in recognition of the risks of kidney failure, advise the ERA-EDTA.



This advice is a reflection of international guidelines which suggest that patients with kidney disease who do not yet require dialysis should not exceed 0.80 g of dietary protein intake per kilogram body weight per day.

A high dietary increase of protein for a prolonged period of time, such as during a protein shake diet, could advance the stage of kidney disease and the need for particular medical treatment, according to an ERA-EDTA press release dated 19th May 2016. The organisation warned that people with impaired renal function may cause further damage to their kidneys, leading to a more serious stage of renal failure if they were to sustain high levels of protein in their diet. As a result, they advise people to have a general practitioner examine their renal function before doing so.

This advice is a reflection of international guidelines which suggest that patients with kidney disease who do not yet require dialysis should not exceed 0.80 g dietary protein intake/kg body weight/day. For a person weighing 70 kg, this means that 56 g would be the upper limit. The high level of protein in dietary protein shakes concerns the ERA-EDTA due to it exceeding these guidelines if consumed multiple times a day. For example, some protein shakes contain >30 g of protein per serving and can have further protein added when mixed with milk, which would exceed the guidelines with frequent daily consumption.

Kidney disease often goes undetected and does not produce symptoms for a long period of time. This means that people may be unaware they have the disease while also increasing their dietary intake of protein and potentially increasing their exposure to harm. Furthermore, decreased amounts of protein can help prevent the advance of renal impairment and so an early diagnosis of the disease could assist in slowing its progression.

One Step Closer to Optimised Timing of Renal Replacement Therapy

EARLY commencement of renal replacement therapy (RRT) for patients with acute kidney injury (AKI) has been demonstrated to both improve survival and reduce inflammatory biomarkers, according to an ERA-EDTA press release dated 22nd May 2016.

AKI is a prevalent complication of cardiac surgery, with incidence figures varying between 7% and 40%. Other factors are known to influence the onset of AKI, however the effect of timing in the initiation of renal replacement therapy is unknown, and thus, its optimisation is a challenge. This challenge is further augmented by the knowledge that some patients do recover from AKI, and that cost-efficiency considerations need to be taken into account.

> “Additional evidence suggest that RRT before the onset of severe AKI may attenuate kidney-specific and non-kidney organ injury...”

A recent study examined the outcomes of early and late RRT initiation in 231 patients presenting with plasma-associated lipocalin >150 ng/mL, randomised to either early (Kidney Disease: Improving Global Outcomes [KDIGO] Stage 2) or late (KDIGO Stage 3) RRT. The primary endpoint, survival beyond 90-day follow-up, was achieved in 60.7% of the early group (n=112) compared with 45.3% of the late group (n=119), demonstrating a significant improvement in mortality (hazard ratio: 0.66; 95% confidence interval: 0.45–0.97; p=0.03). Secondary endpoints such as length of hospital stay and recovery of renal function were also significantly improved with early RRT. The concentrations of interleukin 6 and 8, both plasma pro-inflammatory mediators, were also diminished at a statistically significant level in the early group 1 day after initiation (p<0.001).

Although noting the strict enrolment parameters and single-centre nature, Prof Alexander Zarbock, Department of Anaesthesiology and Intensive Care Medicine, University of Münster, Münster, Germany, discussed the positive interpretations; "Additional evidence suggest that RRT before the onset of severe AKI may attenuate kidney-specific and non-kidney organ injury from acidaemia, uraemia, fluid overload, and systemic inflammation, and could potentially translate into improved survival and earlier recovery of kidney function. This might be the mechanism behind the reduced treatment duration and hospitalisation."

Risk of Renal Failure Varies Across Europe

IMPORTANT research has been presented at this year's ERA-EDTA congress concerning potential kidney donors. According to an ERA-EDTA press release dated 22nd May 2016, the study has determined the lifetime risk of renal failure in Europe to be between 0.5% and 1.5%. This is between 2 and 3-times lower than that of the USA, however the team noted that this is not directly comparable under the current research and that a specific risk prediction model should be used for patients in Europe.

Combining data from the ERA-EDTA Registry and population census data from Eurostat, the researchers gauged the population requiring renal replacement therapy (RRT) in 10 European countries to produce an estimate of the average lifetime risk of renal failure in each country. Levels of risk differ across Europe, with Finland having the lowest (0.44% in women and 0.88% in men), and Belgium the highest (1.14% in women and 2.05% in men). It is thought that the disparity in results across Europe may be the result of varying healthcare systems.

It is thought that the disparity in results across Europe may be the result of varying healthcare systems.

The data demonstrate that women have a significantly lower lifetime risk of renal failure than men. It should be noted that the data assessed in this study only observe patients receiving treatment for renal failure, as opposed to all individuals diagnosed with the disease. The researchers also suggested that older women in particular were less likely to receive RRT, which may have affected the results. Another interesting outcome of the analysis established that older populations have a lower lifetime risk of renal failure compared with younger populations. There is some debate over the efficacy of kidneys provided by older living kidney donors, yet the research in question may contribute to future decisions to allow older living kidney donors. The results could prove advantageous in subsequent studies and point toward several other areas that would benefit from further research.

Safety of Steroid Therapy Guidelines Requires Reassessment

BALANCING risk and benefit is a crucial aspect of developing therapeutic strategies, and clinical guidelines are typically built to consider this equipoise. The safety of clinical guidelines recommending steroid therapy for immunoglobulin A nephropathy (IgAN) has however been questioned, an ERA-EDTA press release dated 22nd May 2016 reports.

As the third most common specific cause of kidney failure across Europe, IgAN warrants a robust treatment strategy. Patients exhibiting IgAN and persistent proteinuria are currently recommended steroid therapy, although until recently this guideline had not been tested for safety and efficacy. The multicentre TESTING study therefore set out to evaluate these factors in 262 patients with an estimated glomerular filtration rate of 20–120 mL/min per 1.73 m^2 and persistent proteinuria >1 g/day. Two cohorts were randomised to receive either oral methylprednisolone 0.6–0.8 mg/kg/day weaning over 6–8 months or matching placebo, following 3 months of

blood pressure control and renin-angiotensin system blockade.

At a median follow-up of 1.5 years, serious adverse events (SAE) had occurred in 20 versus 4 patients in the steroid and placebo groups, respectively (p=0.001); the majority of these were due to infection, two of which were fatal. Along with time-averaged proteinuria, the rate of primary efficacy outcome (composite end-stage kidney disease) was significantly lower in the steroid-treated cohort, compared with the placebo arm (1.31 versus 2.19 g/day, p<0.001, and 5.1% versus 14.3%, p=0.019, respectively). Nonetheless, as a result of the excessive frequency of SAE, treatment in both groups was discontinued and the trial recruited no further participants.

Nonetheless, as a result of the excessive frequency of SAE, treatment in both groups was discontinued and the trial recruited no further participants.

Participant follow-up continues to be assessed, however at this junction it can be said that the results of this study necessitate reconsideration of the safety of current clinical recommendations for high doses of steroid therapy for IgAN patients at high-risk of renal progression.

High Sodium Intake Linked to Cardiovascular Disease in Chronic Kidney Disease

CARDIOVASCULAR disease (CVD) risk is increased in chronic kidney disease (CKD) patients who have a high intake of sodium, a recent prospective cohort study has demonstrated. The findings were presented at the ERA-EDTA congress in Vienna, Austria, according to an ERA-EDTA press release dated 24th May 2016.

Researchers from the Chronic Renal Insufficiency Cohort Study were able to determine dietary intake of sodium by measuring urinary sodium excretion in participants diagnosed with CKD. Whilst a positive correlation has long been established between sodium intake and blood pressure, previous research has provided little conclusive evidence of the connection between dietary sodium intake and risk of CVD, with no prior investigations concerning CKD patients.

Almost 2,400 abstracts

The unprecedented results demonstrated that higher levels of urinary sodium excretion were found to have a positive correlation with increased risk of CVD.

The prospective cohort study examined 3,757 participants with CKD by calculating the cumulative mean urinary sodium excretion from three 24-hour urinary measurements. The mean age of the study group was 58 years, and 45% of patients were women. Congestive heart failure, stroke, and myocardial infarction were defined as the composite CVD events, and during the follow-up period (median 6.8 years), 804 CVD events were reported: 575, 148, and 305 of each composite CVD event, respectively. From the lowest (≤2,894 mg/24 hours) to the highest (>4,548 mg/24 hours) quartile of calibrated sodium excretion, 174, 159, 198, and 273 composite CVD events occurred. For heart failure, the cumulative incidence in the highest quartile of calibrated sodium excretion compared to the lowest was 23.2% versus 13.3%, 10.9% versus 7.8% for myocardial infarction, and 6.4% versus 2.7% for stroke, respectively. The unprecedented results demonstrated that higher levels of urinary sodium excretion were found to have a positive correlation with increased risk of CVD.

It is hoped that the study will prove extremely valuable in future research and current practice for nephrologists. Reduction of sodium in the diet may be constructive in preventing CVD in patients diagnosed with CKD, and it is recommended that future research focus on sodium reduction and its influence on CVD risk.

Challenging Clinical Practice for Lipoprotein(a) Lowering Therapies

REDUCTION of a risk factor for coronary artery disease (CAD) may be available for individuals predisposed to high lipoprotein(a), (Lp[a]). Epidemiological studies in the past have shown an association between Lp(a) levels and cardiovascular risk, which has resulted in lipid apheresis being a common practice for lowering Lp(a) in patients with CAD. However, new evidence presented in an ERA-EDTA press release dated 22nd May 2016 suggests that this practice may need to be reconsidered.

The research evaluated the association between Lp(a) and CAD severity in 3,313 patients undergoing coronary angiography in the Ludwigshafen Risk and Cardiovascular Health (LURIC) study. Each patient had serum Lp(a) level measured as well as the status of two single nucleotide polymorphisms (SNPs) (rs10455872, rs3798220), which are associated with elevated Lp(a) throughout life. These tests were validated against 514 patients from the Homburg Cream and Sugar (HCS) study.

Presence of a minor allele at either of the SNPs was associated with a median 250.2% increase in Lp(a), which was strongly associated with angiographically defined CAD (hazard ratio [HR]: 1.89, p<0.001) and time to first myocardial infarction (HR: 1.2, p=0.05). At long-term follow-up, an association between these alleles and all-cause or cardiovascular mortality was not found; the evidence suggested that in patients with prevalent CAD, outcomes and Lp(a) are not associated. Estimated marginal means of Lp(a) decreased following myocardial infarction (-49.4%) compared with patients with stable CAD. Other markers such as CRP, fibrinogen, and interleukin 6 increased. This points to the function of Lp(a) as a negative acute phase protein.

These findings contest the current practice of Lp(a) lowering for CAD patients, suggesting that such therapies would be more effective as a preventative measure for individuals who have a genetic predisposition to either of the two alleles.

These findings contest the current practice of Lp(a) lowering for CAD patients, suggesting that such therapies would be more effective as a preventative measure for individuals who have a genetic predisposition to either of the two alleles. The findings have implications for clinical practice and have

already been validated in both the HCS and KAROLA cohorts.

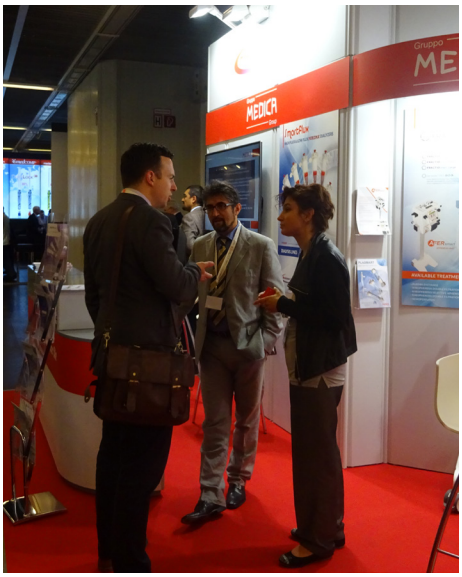
Austria Leading the Way in Chronic Kidney Disease Prevention

THIS YEAR'S ERA-EDTA congress was the second biggest in the world of nephrology. It covered a range of topics including current research and understanding of the treatment of kidney disease and related illnesses, renal replacement therapy, as well as an in-depth discussion on the use of 'big data' and its role in the development of individualised therapy.

> "As far as prevention is concerned, nephrology in Austria is a model to be emulated Europe-wide. Nephrologists and general practitioners work closely together here, and our CKD Prevention Program 60/20 sets an example that many other countries can follow. We can also refer to the specific successes we have achieved: in Austria, the incidence of new CKD cases is declining!"

The main focus of this year's congress was on the prevention of chronic kidney disease (CKD), a frequent consequence of diabetes mellitus or high blood pressure in later life. As the number of patients diagnosed with these disorders is soaring, prevention is becoming a priority for nephrologists worldwide. Prof Gert Mayer, Department of Internal Medicine, Medical University Innsbruck, Innsbruck, Austria, commented in an ERA-EDTA press release dated 19th May 2016: "Demographic change is confronting us with a challenge here, especially since the number of those with diabetes and/or high blood pressure has risen. Present day nephrology sees its mission in protecting these high-risk patients against terminal renal failure."

The Austrian model of prevention is a novel strategy for calculating which patients should be referred to a nephrologist for treatment, named the '60/20' concept. This includes any patients whose kidney function has reduced to ≤60% and a risk constellation exists, as well as those whose kidney function is reduced to 20% for discussion of the best renal replacement therapy. There are also guidelines for the establishment of kidney care centres to accommodate the number of patients being referred and improve the availability of post-mortem donations. "As far as prevention is concerned, nephrology in Austria is a model to be emulated Europe-wide. Nephrologists and general practitioners work closely together here, and our CKD Prevention Program 60/20 sets an example that many other countries can follow. We can also refer to the specific successes we have achieved: in Austria, the incidence of new CKD cases is declining!" stated Prof Dr Karl Lhotta, Department of Nephrology and Dialysis, Landeskrankenhaus Feldkirch, Feldkirch, Austria.



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Thomas Ryzlewicz

Senior Consultant Nephrologist, Dialysis Centre, ViaMedis Riesa, Riesa, Germany; WBR Holiday Kidney Centre, Lana, Italy

Q: Why did you choose to pursue a career in nephrology?

A: At first, engineering seemed to be the target for my profession. I had done one semester in mathematics, but with a blue overall, you will only make small decisions in comparison to those in white overalls. Physiology became very interesting to me whilst I was studying medicine, for its similarities to mechanical engineering; consider measurement, mechanics, the circulation, acid-base-status. I also had interests outside of work, including motor car tuning and sailing together with my brother. This made the handling of machines easier for me. My target was to improve my patient's condition by perfect handling of a machine, which due to my experience in engineering came very easily to me. There was no big difference between adjusting the 4-carburettor system of a car and adjusting the respirator of a breathing system for anaesthesiology. I began working in dialysis 42 years ago. For a normal nephrology doctor it is not necessary to be involved with machines; for me this was dependent on my personal interests and the facilities available.

Q: You have been involved in a variety of technical innovations, such as the development of dialysis machine prototypes. How important do you think it is for doctors to involve themselves with the design and development of new technologies?

A: For me, the development of technology is very important, as I definitely will use it when the improvement is implemented. I myself will notice even the smallest improvements. For example, the current theme guiding our design of the Oxyless Bloodline is the challenge of sheer-stress to red blood cells in the single fibre capillary; reduction of sheer-stress leads to prolonged survival of red blood cells and a much lower dosage of erythropoietin. The experience of the doctors also contributes to the quality of the outcome, but this is never reached with continuous medical education.

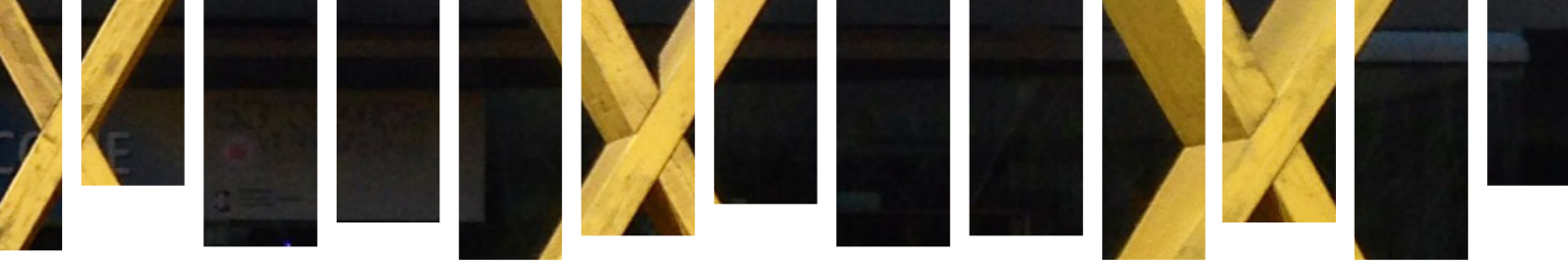
Q: What technological development do you think has been the most impactful in nephrology?

A: The most impactful technology was the invention of the Shaldon's concept of Acetate Dialysis for haemodialysis. This invention was unique! This is the general design of the dialysis monitor of today. With his skill and knowledge, a much larger number of patients with end-stage kidney disease have been treated. An experimental animal trial was not needed, as the patient was best to test this on!

Q: What kind of innovations do you hope we will see in the coming years?

A: For me, the prohibition of acetate as a dialysis fluid is necessary because of calcification, with acidification 3 mmol/L acetate. This is licensed by the medical authorities, as dialysis concentrate is a medical product, giving freedom to the manufacturer. These days, all kinds of medical products are allowed as long as there is a CE mark. This needs to be addressed, and better vigilance for medical products is required. There is also a big problem with vascular access for Stage 5 chronic kidney disease (CKD) patients. In the USA, up to 60% of patients have tunnelled dialysis catheters with a big risk of infection (or in a worst case scenario, sepsis). Unfortunately, the recommendations of KDOQI and DOPPS reports neglected to discuss the arteriovenous fistula as an alternative treatment.

> "My target was to improve my patient's condition by perfect handling of a machine..."



Q: You regularly work in both Italy and Germany; how does nephrology and healthcare in general vary between these countries?

A: The WBR Dialysis Centre in Alto Adige, Italy, is a small centre for holiday patients which is well suited to my work. Generally, the region of Northern Italy severely limits access to the dialysis treatment because of the costs, and when a senior Italian consultant exaggerates the number of Stage 5 CKD patients, his contract will not be renewed. The centre in Riesa (145 patients, ViaMedis, Germany) is a big centre with well-equipped nurses; it was also used for the PEMA Audit for the Oxyless Bloodline.

Q: What do you think is the biggest challenge facing nephrologists today? What could be done to resolve this?

A: I believe the biggest challenge is the need for prohibition of the classical dialysis concentrate, as this prescription contributes to the calcification of coronary vessels and heart valves. As this is a problem of chemical solubility, it is not completely understood by medical doctors. Efforts to involve a material specialist alongside medical doctors in the approval of a medical product should be made by the governing bodies. The second challenge is to reach the switching to the arteriovenous fistula instead of the abuse of tunnelled dialysis catheters. To reach this target, the providers should be involved. An arteriovenous fistula strongly reduces the danger of infection and sepsis. Survival, morbidity, and mortality are joint with this theme. A patient receiving better treatment does live longer!

Q: What can governing bodies and other healthcare providers do to raise awareness of risks to renal health and promote better lifestyles?

A: I would say that the reduction of body weight is very important. Another problem is surgery particularly in relation to Stage 5 CKD patients. There is the problem of the diabetic foot, which involves arterial perfusion and necrosis. Today, 'small surgery' and antibiotics are often used as a supplementation to good nutrition with the result of a selection of bacterial germs, resulting in many days of in-clinic-treatment and increased costs.

Q: How do you think nephrology events like ERA-EDTA contribute to advancements in nephrology and its associated technologies? Were there any specific pieces that caught your eye at this year's conference?

A: EDTA was founded by Dr Shaldon at Lake Geneva. For many decades, EDTA was very important. Today there are many autodidacts with no respect for proven results (see the problem with the tunnelled dialysis catheters). Our group presented the results of our Bloodline at WCN South-Africa, EDTA London, and at ASN in 2015 (by Ian MacDougal, King's College, London, UK). So ERA-EDTA is clearly still important, but in a reduced capacity. The importance of meetings such as ERA-EDTA cannot be denied, and the results of our own research have indeed been presented at an ERA-EDTA meeting in London. I personally would also like to see a rejuvenation of scholarly debate within the literature, the likes of which could be seen to emerge from an international scientific meeting such as ERA-EDTA. The transatlantic acid-base debate in the New England Journal between the Boston School and Siggard-Andersen and his colleagues is a classic example of this kind of stimulating discussion.

Q: What particular research are you currently working on?

A: I designed and am involved with the development of the Oxyless Bloodline technology. I am also working on lectures and editorials in a continuous and growing capacity concerning the calcifying dialysis fluid (acidification with 3 mmol/L acetate). To reach the target of a prohibition of this prescription, I need the assessment of an independent chemist. A prescription with bicarbonate dialysis fluid with acidification with 1 mmol/L citrate does exist on the market as a regular medical product. When using citrate for the acidification of bicarbonate dialysis fluid, there is a second principle of working inside (additional to the first principle of the CO^2 production), the chelate binding of citrate concerning the problem

ions for calcification (Ca^{2+} and Mg^{2+}), so these have been disguised! This context requires qualified chemical knowledge. This is the problem. You may ask me why this is my target. In 1978 I tried to realise a bicarbonate dialysis machine, using an old Milton Roy B II supply system. Coming from motor-cars, I had installed a second pump for the bicarbonate at 8.4%. This equipment had never been used with patients, but I learned a lot about calcification, as I handled the acid base set-up in the Physiologic Institute. The target was to adjust the amount of acid addition, but there always remained a baseline calcification. Nowadays every monitor must be descaled following bicarbonate dialysis. Dr Shaldon's old prescription (acetate dialysis) used acetate as a buffer precursor. There was no bicarbonate inside, the liver had to metabolise this acetate into bicarbonate and CO^2. With acetate dialysis, there was no calcification. This was the real 'Secret of Tassin' (Laurent mentioned in NDT the 8-hour dialysis and the nutrition with only 2.0 g salt per day). So the famous results of Tassin can never be repeated with bicarbonate dialysis, as even in Tassin the new technology (with bicarbonate and calcification) was shown to be far more successful.

Q: What, in your opinion, is the greatest accomplishment of your career thus far?

A: My biggest accomplishment is the facility to continuously improve mechanical and hydraulic systems (i.e. the dialysis set-up) when working. The calcification problem I had seen in the very first days was mechanically occlusive.

> "The importance of meetings such as ERA-EDTA cannot be denied, and the results of our own research have indeed been presented at an ERA-EDTA meeting in London."

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PROTECTING HEART, VESSELS, AND BONE: NEW WAYS TO CONTROL PHOSPHORUS

This symposium took place on 22th May 2016 as part of the European Renal Association - European Dialysis and Transplant Association (ERA-EDTA) Congress 2016 in Vienna, Austria

Chairperson
Markus Ketteler[1]
Co-chairperson
Alexander Rosenkranz[2]
Speakers
Laurent Juillard,[3] Philip Kalra[4]

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Disclosure: Prof Ketteler has served as a consultant for Pfizer, Amgen, Vifor Fresenius Medical Care Renal Pharma, Abbvie, and Sanifit; has received honoraria for lectures on behalf of Amgen, Vifor Fresenius Medical Care Renal Pharma, Sanofi, Shire, Pfizer, and Amgen; and served as a Principle Investigator for Medice. Prof Rosenkrantz has received honoraria for lectures on behalf of Amgen, Mitsubishi, Novartis, Boehringer, Astropharma, Baxter, Fresenius, Takeda, and Abbott/Abbvie. Prof Juillard has received honoraria for lectures from B. Braun, Genzyme, GE Heathcare, and Vifor Fresenius Medical Care Renal Pharma; he has also sat on advisory boards for Baxter, Fresenius Pharma, GE Healthcare, Hemotech, Roche, and Vifor Pharma; and received clinical research/research support from Alexion, Bayer, GlaxoSmithKline, Otsuka, Baxter, Fresenius Pharma, and Genzyme. Prof Kalra has received honoraria for lectures on behalf of Vifor Fresenius Medical Care Renal Pharma, Sanofi, Shire, and received an educational grant from Shire supporting biomarker research. Prof Kalra has also contributed to the design of the VERIFIE:PASS study, sponsored by Vifor Fresenius Medical Care Renal Pharma.
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MEETING SUMMARY

Prof Alexander Rosenkranz and Prof Markus Ketteler welcomed the audience and the expert panel of the symposium, and briefly described the programme of the meeting. Prof Laurent Juillard discussed the challenges faced in achieving phosphate control in patients on haemodialysis, as well as aspects for optimising the management of hyperphosphataemia. Prof Philip Kalra described recent advances in hyperphosphataemia treatment, concentrating on an iron-based, calcium-free phosphate binder that may offer a lower pill burden compared with previous treatments, and thereby address the challenge of patient non-adherence.

Hyperphosphataemia: Understanding the Challenges

Professor Laurent Juillard

In healthy individuals, the kidney is the primary regulator of phosphorus homeostasis. When renal function is preserved, there is a balance between the absorption and urinary excretion of phosphate, together with an equilibrium between bone formation and resorption. A net of 800 mg of phosphate from the diet is being absorbed in an exchangeable phosphate pool consisting of three components: 70% comprises the intracellular phosphate involved in phosphorylation reactions (e.g. signalling); 29% comprises the mineralisation of the skeletal system (bones and teeth); and <1% includes serum phosphate that is usually measured for monitoring purposes. The skeletal component is often ignored due to the daily bone formation/resorption, however the skeleton functions as a phosphate reservoir containing >80% of total body phosphate. This reservoir assimilates phosphate when the bone formation balance is positive.[1] In chronic kidney disease (CKD), impaired renal function (i.e. reduced urinary excretion) results in reduced phosphate excretion and phosphorus homeostasis is lost. Despite reduced gastrointestinal (GI) absorption, excess of bone resorption compared to formation and accumulation of phosphorus in soft tissue and vasculature lead to an overall positive balance of phosphorus in the body and serum, resulting in hyperphosphataemia.[1]

As shown by a large epidemiological trial (N=25,529), serum phosphorus is associated with a significant increase in all-cause (5,857 events) and cardiovascular (CV; 1,930 events) mortality risk in dialysis patients.[2] However, haemodialysis patients prescribed phosphate binders show a significant reduction in mortality (25% lower death rate, without taking into account nutritional factors). Without adjusting for nutritional indicators, mortality rate was higher in patients not administering phosphate binders (versus those prescribed a phosphate binder) at all serum concentrations ≥3.5 mg/dL. Similar patterns were observed when nutritional indicators were adjusted for.[3]

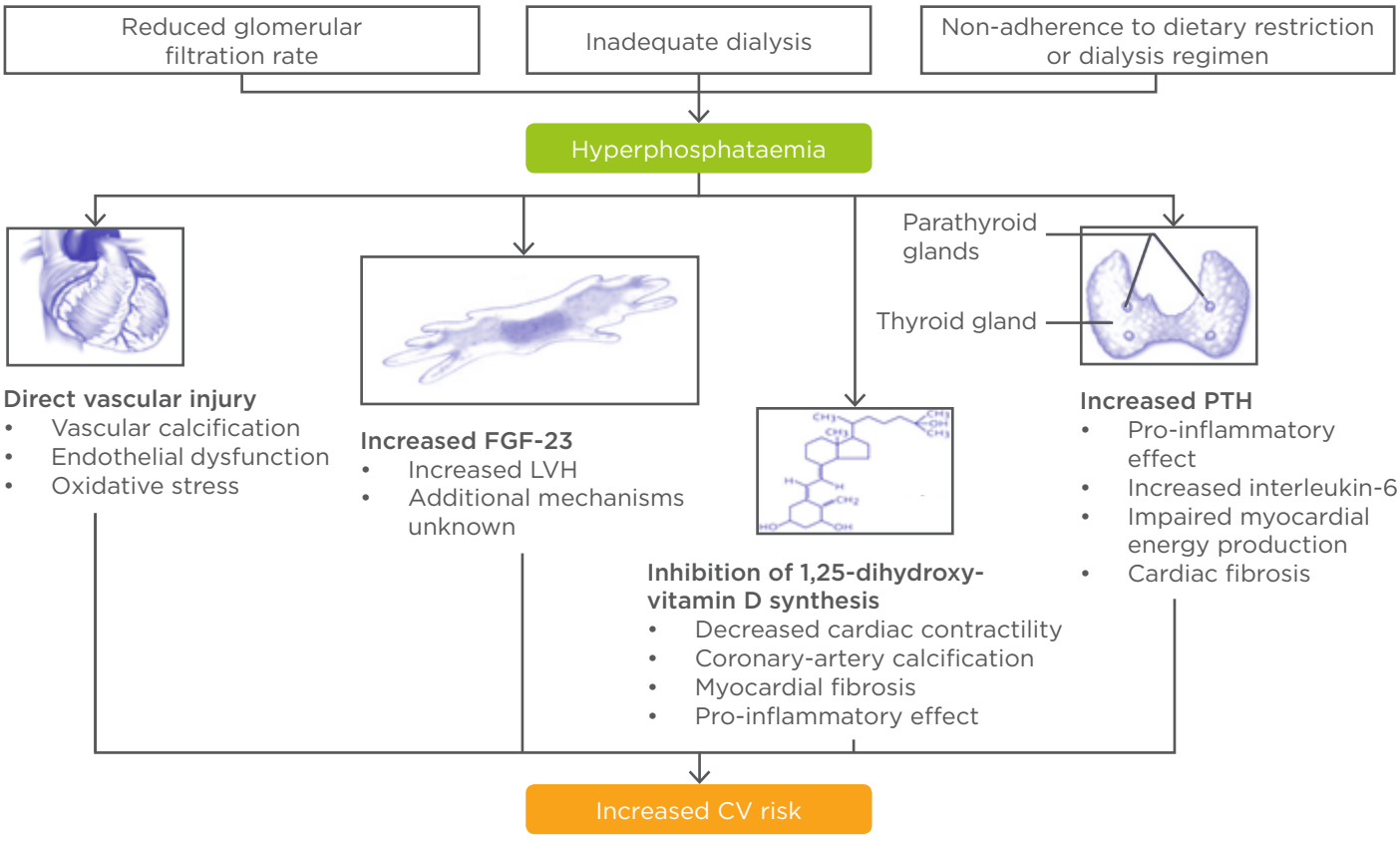

Figure 1: Putative mechanisms linking hyperphosphatemia and cardiovascular disease.
CV: cardiovascular; FGF-23: fibroblast growth factor-23; LVH: left ventricular hypertrophy; PTH: parathyroid hormone.
Adapted from Tonelli M et al.[5] and Faul C et al.[4]

Elevated serum phosphate levels are generally associated with an increased risk of CV disease among patients with and without kidney failure. Hyperphosphataemia occurs in dialysis patients when the glomerular filtration rate is reduced, the dialysis is inadequate, and when the patient is not adhering to dietary restrictions or dialysis regimes. This results in direct effects such as vascular injuries due to, for example, induction of oxidative stress and endothelial dysfunction, and indirect effects. This includes modifications in blood parameters such as increased fibroblast growth factor-23 (FGF-23) leading to increased left ventricular hypertrophy;[4] inhibition of 1,25-dihydroxyvitamin D synthesis leading to decreased cardiac contractility; arterial calcification; myocardial fibrosis; and increases in parathyroid hormone. The result of this is: cardiac fibrosis, inflammation, and impaired myocardial energy production (Figure 1). All these effects increase CV risk in haemodialysis patients.[5]

Abnormal phosphate metabolism is also a major mechanism of vascular calcification. The factors that increase vascular calcification include: failed anti-calcific processes (loss of inhibitors); matrix degradation via elastolysis with metalloproteinases; apoptosis (phosphate inflammation); osteochondrogenic differentiation of vascular cells; and remodelling of bone, leading to circulating nucleation complexes, and paracrine factors contributing to initiation or progression of vascular calcification.[6]

Renal osteodystrophy is a disorder of bone remodelling and is associated with hyperphosphataemia. Insufficient levels of parathyroid hormone characterise low turnover osteodystrophy, where movement of calcium and phosphate from the exchangeable pool into the skeleton is decreased and reductions in bone formation exceed the reductions in bone resorption. As a result, the excess bone resorption contributes to hyperphosphataemia and is illustrated through patients with low turnover osteodystrophy who develop osteoporosis in CKD. In high turnover osteodystrophy, because of the impact of excess parathyroid hormone, high levels of receptor activator of nuclear factor kappa B ligand (involved in osteoclast differentiation) develop, resulting in excess osteoclast activity. Even though bone formation rates are increased, bone resorption rates are excessive, which also contributes to vascular calcification and stiffness.[1] Hyperphosphataemia further affects bone health, which in turn affects mortality. Fractures are 17-times more frequent and occur up to 15 years earlier in patients with Stage 5 CKD than in an age-matched general population.[7] In addition, there is a >2-fold increase in mortality risk associated with hyperphosphataemia compared with patients on dialysis who do not have hip fractures.[8]

As mentioned, increased FGF-23 levels as a result of hyperphosphataemia may lead to heart failure. Additional modifications in levels of vitamin D and parathyroid hormone are also contributors to such consequences, particularly in patients on dialysis. The effects however, can be reversed after kidney transplantation.[9] Significantly increased levels of FGF-23 in patients with CKD increase the left ventricular mass, leading to a considerable remodelling of the heart, reduction in ejection fraction,[4] and inducing mortality in end-stage renal disease.[10]

The management of hyperphosphataemia faces several challenges, the first of which is nutrition. Caution should be exercised when attempting to restrict the amount of phosphorus in the diet of patients so as to not lead to malnutrition; however, more attention should be paid to the quality of food as many processed foods have a much higher phosphorus content compared with their fresh form. Phosphorus additives are particularly common in frozen foods (72%), dry food mixes (70%), packaged meat (65%), bread and baked goods (57%), soup (54%), and yoghurts (51%).[11]

The second challenge in the management of hyperphosphataemia includes the effect of dialysis modalities on phosphate removal from the body: conventional haemodialysis, peritoneal, nocturnal, and daily dialyses all remove significant quantities of phosphorus from the blood.[12] Phosphorus magnetic resonance spectroscopy during haemodialysis in anephric pigs was used to measure the concentration of intracellular phosphate and concentration of adenosine triphosphate, since inorganic phosphate has an impact on cellular energy.[13] The results showed a linear decrease of urea during the dialysis, corrected for acidosis, and calcium in the effluent was almost identical to the levels contained in the dialysis solution. No significant modification in calcium balance and the fact that bone resorption involves more calcium than phosphorus transfer, suggested that the bone was not involved as a

source of phosphorus in dialysis. During dialysis, sustained removal of phosphate was observed over 180 minutes. Extracellular content decreased quickly, then slowly plateaued, and increased before the end of dialysis. Intracellular phosphate was shown to increase at the same time as extracellular phosphate plateaued (the inorganic phosphate to phosphocreatine ratio increased by a very significant 6.9% [p<0.00001]).[13] At the same time, adenosine triphosphate content inside the cell was shown to decrease significantly during the dialysis and was accompanied by a constant increase in intracellular pH.[13] These findings pose the question of whether there is a detrimental cellular effect due to the removal of phosphate during dialysis. This will be assessed in patients with CKD and the outcome is anticipated to influence how dialysis will be performed in the future.

An additional challenge in managing hyperphosphataemia lies with phosphate binders. Their efficacy at lowering serum phosphate levels has been shown to be significant in controlled trials,[14-16] however, data from observational studies as opposed to that taken from a clinical setting show 52% of patients achieved phosphorus concentrations within the Kidney Disease Outcomes Quality Initiative (KDOQI) target range and only 27% of patients had a phosphorus concentration within the Kidney Disease: Improving Global Outcomes (KDIGO) target range.[17]

Haemodialysis patients endure one of the highest pill burdens reported for any chronic disease, with phosphate binders comprising half of a total which can come to 19 pills per day.[18] A systematic review of online databases that analysed the prevalence and determinants of non-adherence to phosphate binding medication in patients with end-stage renal disease showed that adherence varied from 22–74% in a given population; the wide range is attributed to different methods of non-adherence assessment (serum phosphate levels [58% adherence] versus self-report measures [31%]).[19]

In conclusion, Prof Juillard reaffirmed that phosphorus homeostasis is lost in CKD, where impaired renal function leads to a loss of phosphorus excretion resulting in inevitable hyperphosphataemia in end-stage disease. He repeated that hyperphosphataemia is a risk factor for CV disease, vascular and other soft-tissue calcification, bone disease, and increased mortality, and that the management of hyperphosphataemia through dialysis, diet, and phosphate binders is still far from optimal and better treatment options are needed.

Advances in the Management of Hyperphosphataemia

Professor Philip Kalra

As previously described, the mortality rate is very high in dialysis patients. At 25 years of age, men or women on dialysis have around 100-times the risk of CV death versus those in the general population. The gap narrows over time but evens out by the age of 60 or 70 years, when the risk is 10 to 20-fold that of the general population for the same age.[20] There are several aetiological factors for structural CV disease, some of which include: hypertension, renin-angiotensin-aldosterone upregulation, inflammation, intra-dialytic ischaemia, oxidative stress, and anaemia. However, CKD and mineral bone disease play a dominant role incausation of vascular disease and uraemic cardiomyopathy. The prevalence of vascular calcification is very high: vascular calcification is found in around 40% patients with CKD new to dialysis[21] and in up to 83% of patients on established dialysis.[14] The greater the degree of calcification in a patient, the higher the mortality risk: the patients with the greatest calcification have only ~20% all-cause survival at 6 years, compared with 95% survival rate of those with no calcification.[22]

A study in a USA population by Block et al.[23] showed that there is a U-shaped observational association between serum phosphate concentration and mortality in patients with hyperphosphataemia. As serum phosphate concentration increases above 4.0 mg/dL to 5.0 mg/dL, the risk of mortality increases in a stepwise pattern, until at >9.0 mg/dL, the risk of death is twice as high as at 3.1–4.0 mg/dL. The U-shape is attributed to a group within the dialysis population that are often malnourished and have an increased risk of mortality. At the same time, also in the USA population, haemodialysis patients (25%) within the highest range of FGF-23 levels had a nearly 6-times greater risk of mortality compared with those in the lowest quartile.[10] Faul et al.[4] have shown that injecting FGF-23 directly into the murine myocardium causes gross left ventricular hypertrophy in just 1 week; this important animal work ties in with the impact that hyperphosphataemia has on the human body.

Randomised controlled trials investigating phosphate binder intervention in CKD mineral bone disease range from those looking at vascular calcification (Renagel in New Dialysis [RIND] and Treat-to-Goal), or mortality (Dialysis Clinical Outcomes Revisited [DCOR] and RIND follow-up) as endpoints. The RIND study (N=110) included patients who were incident to dialysis and randomised to sevelamer or calcium-based phosphate binders (CCPB) over 18 months.[24] The Treat-to-Goal study involved twice as many patients as RIND, but patients were again randomised to sevelamer or CCPB, and looked at the median percentage change in calcification in the coronary artery and in the aorta.[14] Both studies showed a marked attenuation of calcification in patients treated with calcium-free phosphate binders compared to CCPB.

Including over 2,000 patients on dialysis, the DCOR study is the largest phosphate binder study focussing on mortality and compared patients treated with CCPB to those on sevelamer. The primary outcome measure was all-cause mortality and this was not found to be significantly different between the treatment groups. However, in an apparently pre-specified sub-analysis involving patients >65 years of age, sevelamer therapy resulted in a statistically significant reduction of 22% (p=0.03) in the relative risk for all-cause mortality.[25]

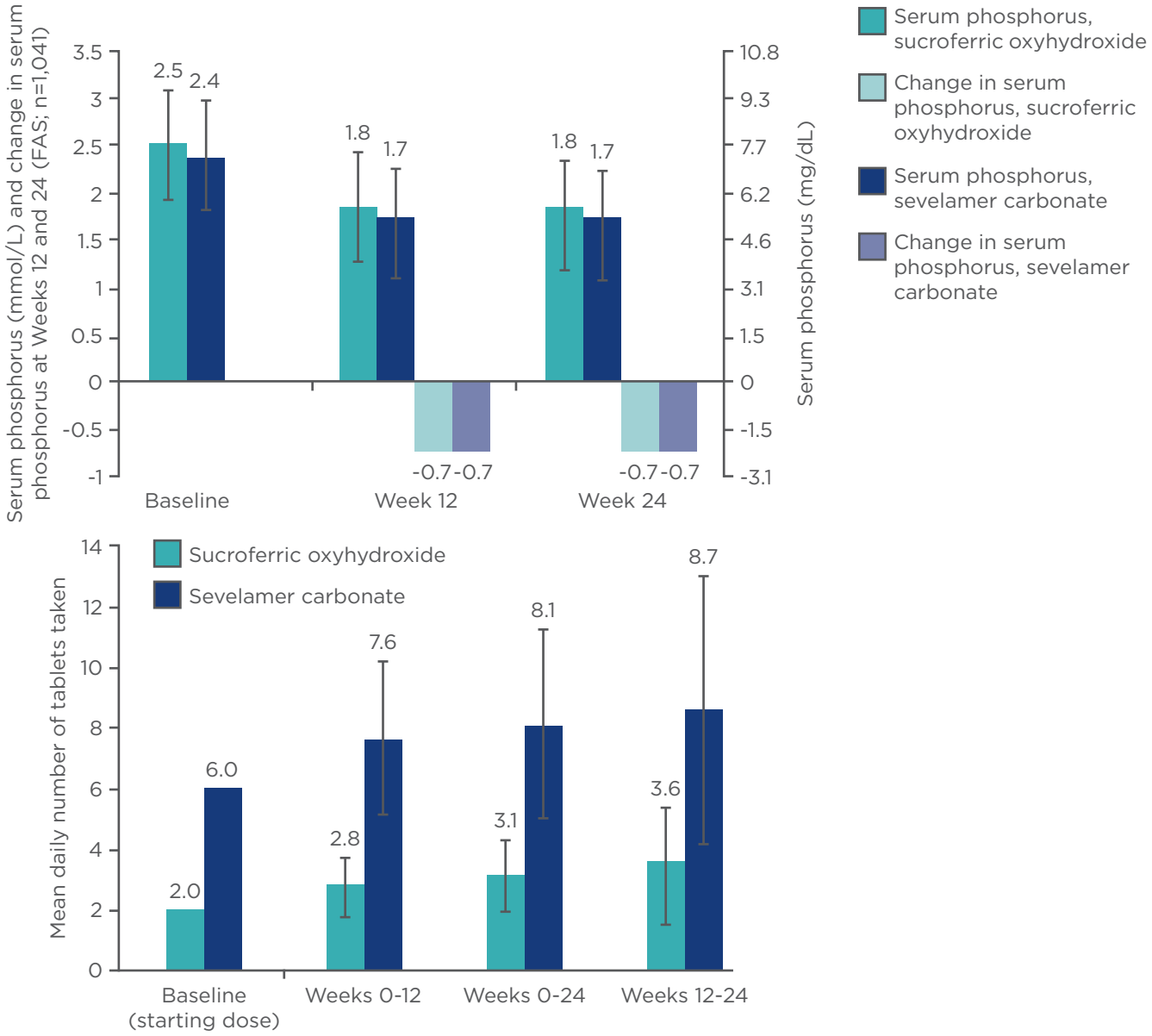

Figure 2: Non-inferiority and lower daily pill burden of sucroferric oxyhydroxide (SFOH) versus sevelamer carbonate.

FAS: full analysis set.

The RIND trial follow-up arm extended to 66 months, during which a significant reduction in mortality was observed amongst patients treated with sevelamer. Mortality rates were 10.6/100 patient years (95% confidence interval [CI]: 6.3–14.9) in subjects treated with calcium containing phosphate binders and 5.3/100 patient years (95% CI: 2.2–8.5, p=0.05) in subjects treated with sevelamer.[26]

In addition, a meta-analysis of 18 studies investigated the effects of CCPB versus non-calcium phosphate binders (NCCPB) on all-cause mortality. Eleven of those studies were randomised and reported an outcome of mortality (4,622 patients; 936 deaths). A 22% reduction in all-cause mortality was observed amongst patients on NCCPB compared with those on CCPB. Over all the 18 studies, NCCPB were associated with a 13% reduction in all-cause mortality versus CCPB (risk ratio: 0.87, 95% CI: 0.77–0.97), which was irrespective of patient dialysis status.[27]

As described previously, the pill burden of phosphate binders in CKD patients is very high, leading to a reduction in treatment adherence. The DOPPS study showed that that the more frequently patients skipped their pills, the more likely they were to have serum phosphate levels higher than the target range of 5.5 mg/dL.[28]

Sucroferric oxyhydroxide (SFOH, [Velphoro®]), is a stabilised polynuclear iron oxyhydroxide-based phosphate binder consisting of iron, sucrose, starch, and water. The iron(III)-oxyhydroxide strongly binds the phosphate by replacing hydroxide groups, giving it its phosphate-binding property, while adding sucrose to iron-oxyhydroxide prevents it from ageing and maintains its phosphate-binding capacity. In a preclinical adenine rat model, SFOH, lanthanum, and sevelamer NCCPB were studied. All three attenuated carotid and aortic calcification (including abdominal, inferior, and superior thoracic aorta calcification) to similar degrees compared with control.[29] A Phase III trial investigating the efficacy and safety of SFOH involved over 1,000 patients. After a washout period of 2–4 weeks in patients who were on phosphate binders, patients were enrolled if their serum phosphate levels were in excess of 5.5 mg/dL. Patients were then randomised to either SFOH or sevelamer, in a ratio of 2:1. The starting dose for SFOH was 1.0 g per day, which is two tablets, while for sevelamer it was 4.8 g per day, which is six tablets. The patients were then followed for 24 weeks, followed by a longer phase extending to 52 weeks.[30,31]

During the Phase III study, non-inferiority of SFOH versus sevelamer carbonate was established; SFOH and sevelamer carbonate showed a comparable phosphate-lowering effect from baseline to Week 24, with a mean daily tablet number of 3.1 and 8.1, respectively: a significant difference in pill burden (Figure 2).[30]

GI disorders were the most frequent treatment-emergent adverse events in both treatment groups, observed in 45.1% of SFOH-treated patients and 33.6% of sevelamer carbonate-treated patients. The incidence of GI-related treatment-emergent adverse events, excluding stool discolouration (due to the iron component), was similar between SFOH (39.0%) and sevelamer carbonate (33.3%) groups. Diarrhoea was reported in 17% of SFOH treated patients during the titration phase of the study but in the maintenance phase diarrhoea had fallen to 5.5% in the SFOH-treated patients compared with 2% in the sevelamer-treated group. Diarrhoea generally presented early in treatment and resolved without treatment change.[30] Looking at iron parameters from baseline to Week 24, median serum ferritin concentrations increased in both treatment groups and a statistically significant but not clinically important increase in median transferrin saturation was observed in the SFOH group. These increases occurred early and plateaued on continuing treatment with SFOH, indicating no accumulation of iron. There were also no significant changes in haemoglobin parameters.[30] Over the extension period of the study (to 52 weeks), reduction in serum phosphate concentrations was maintained for both treatment groups, while incidence of the most frequent GI disorders fell.[32] The extension study has also shown a generally higher adherence with SFOH compared with sevelamer at the pre-defined 70–120% compliance level. Adherence increased over the course of the entire 52 weeks in the SFOH group, possibly due to the lower pill burden in that treatment group over the whole treatment period.[31] There was also a 50% lowering in the FGF-23 levels in 1,055 patients in both treatment groups from baseline over 1 year.[33]

A retrospective database analysis of 693 haemodialysis patients with poorly controlled hyperphosphataemia prescribed SFOH in routine clinical practice was conducted at US Fresenius Medical Care facilities. Observation periods

included a baseline period of 3 months before SFOH prescription, and follow-up of 6 months of SFOH prescription. Serum phosphorus at baseline in these patients was 6.9 mg/dL. An increase in the number of patients achieving targets for serum phosphorus was observed after switching from other phosphorus binders to SFOH, this included sevelamer, lanthanum, and calcium acetate. In addition, reduction in the pill burden was observed (8.6 versus 3.4 pills per day).[34] The analysis confirmed the efficacy of SFOH as well as the reduced pill burden that may translate into improved adherence in a real-world population.[34]

In summary, phosphorus homeostasis is lost in CKD. Impaired renal function results in loss of phosphorus excretion, leading to inevitable hyperphosphataemia, a risk factor for CV disease, vascular and other soft-tissue calcification, bone disease, and increased mortality. Management of hyperphosphataemia through dialysis, diet, and phosphate binders is still far from optimal and better treatment options are needed. Several studies have shown that NCCPB appear to attenuate calcification. Adherence to high pill load is a problem in managing phosphate in dialysis patients but SFOH (Velphoro) now presents a new alternative NCCPB therapy as an efficacious option that has a low pill requirement.

REFERENCES

1. Hruska KA et al. Hyperphosphatemia of chronic kidney disease. Kidney Int. 2008; 74(2):148-57.

2. Tentori F et al. Mortality risk for dialysis patients with different levels of serum calcium, phosphorus, and PTH: the Dialysis Outcomes and Practice Patterns Study (DOPPS). Am J Kidney Dis. 2008; 52(3):519-30.

3. Lopes AA et al. Phosphate binder use and mortality among hemodialysis patients in the Dialysis Outcomes and Practice Patterns Study (DOPPS): evaluation of possible confounding by nutritional status. Am J Kidney Dis. 2012; 60(1):90-101.

4. Faul C et al. FGF-23 induces left ventricular hypertrophy. J Clin Invest. 2011; 121(11):4393-408.

5. Tonelli M et al. Oral phosphate binders in patients with kidney failure. N Engl J Med. 2010;362(14):1312-24.

6. Giachelli CM. The emerging role of phosphate in vascular calcification. Kidney Int. 2009;75(9):890-7.

7. Coco M, Rush H. Increased incidence of hip fractures in dialysis patients with low serum parathyroid hormone. Am J Kidney Dis. 2000;36(6):1115-21.

8. Mittalhenkle A et al. Increased risk of mortality associated with hip fracture in the dialysis population. Am J Kidney Dis. 2004;44(4):672-9.

9. Wolf M. Forging forward with 10 burning questions on FGF-23 in kidney disease. J Am Soc Nephrol. 2010;21(9):1427-35.

10. Gutiérrez O et al. Fibroblast growth factor 23 and mortality among patients undergoing hemodialysis. N Engl J Med. 2008;359(6):584-92.

11. Léon JB et al. The prevalence of phosphorus-containing food additives in top-selling foods in grocery stores. J Ren Nutr. 2013;23(4):265-70.

12. Kooienga L. Phosphorus balance with daily dialysis. Sem Dialysis. 2007; 20(4):342-5.

13. Lemoine S et al. Intracellular Phosphate Dynamics in Muscle Measured by Magnetic Resonance Spectroscopy during Hemodialysis. J Am Soc Nephrol. 2015. [Epub ahead of print].

14. Chertow GM et al. Sevelamer attenuates the progression of coronary and aortic calcification in hemodialysis patients. Kidney Int. 2002;62(1):245-52.

15. Hutchison A et al. Safety, tolerability and efficacy of lanthanum carbonate in haemodialysis patients: A 12-month study. Poster 301. 40th ERA-EDTA World Congress of Nephrology, Berlin, Germany, 8–12 June 2003.

16. Qunibi WY et al. Treatment of hyperphosphatemia in hemodialysis patients: The Calcium Acetate Renagel Evaluation (CARE Study). Kidney Int. 2004;65(5):1914-26.

17. Fernández-Martin JL et al. COSMOS: the dialysis scenario of CKD-MBD in Europe. Nephrol Dial Transplant. 2013; 28(7):1922-35.

18. Chiu YW et al. Pill burden, adherence, hyperphosphatemia, and quality of life in maintenance dialysis patients. Clin J Am Soc Nephrol. 2009;4(6):1089-96.

19. Karamanidou C et al. A systematic review of the prevalence and determinants of nonadherence to phosphate binding medication in patients with end-stage renal disease. BMC Nephrol. 2008;9:2.

20. Levey AS et al. Controlling the epidemic of cardiovascular disease in chronic renal disease: what do we know? What do we need to learn? Where do we go from here? National Kidney Foundation Task Force on Cardiovascular Disease. Am J Kidney Dis. 1998;32(5):853-906.

21. Russo D et al. Coronary artery calcification in patients with CRF not undergoing dialysis. Am J Kid Dis. 2004;44(6):1024-30.

22. Blacher A et al. Arterial calcifications, arterial stiffness, and cardiovascular risk in end-stage renal disease. Hypertension. 2001;38(4);938-42.

23. Block GA et al. Mineral metabolism, mortality, and morbidity in maintenance hemodialysis. J Am Soc Nephrol. 2004; 15(8):2208-18.

24. Block GA et al. Effects of sevelamer and calcium on coronary artery calcification in patients new to hemodialysis. Kidney Int. 2005;68(4):1815-24.

25. Suki WN et al. Effects of sevelamer and calcium-based phosphate binders on mortality in hemodialysis patients. Kidney Int. 2007;72(9):1130-7.

26. Block GA et al. Mortality effect of coronary calcification and phosphate binder choice in incident hemodialysis patients. Kidney Int. 2007;71(5):438-41.

27. Jamal SA et al. Effect of calcium-based versus non-calcium-based phosphate binders on mortality in patients with chronic kidney disease: an updated systematic review and meta-analysis. Lancet. 2013;382(9900):1268-77.

28. Tentori F et al. Self-reported non-adherence with Phosphate binder prescription is associated with high serum phosphorus and PTH: Results from the DOPPS. SA-PO665. American Society of Nephrology Kidney Week, 30 October–4 November 2012.

29. Phan O et al. Effects of Sucroferric Oxyhydroxide Compared to Lanthanum Carbonate and Sevelamer Carbonate on Phosphate Homeostasis and Vascular Calcifications in a Rat Model of

Chronic Kidney Failure. Biomed Res Int. 2015;2015:515606.

30. Floege J et al. A phase III study of the efficacy and safety of a novel iron-based phosphate binder in dialysis patients. Kidney Int. 2014;86(3):638-47.

31. Floege J et al. Long-term effects of the iron-based phosphate binder, sucroferric oxyhydroxide, in dialysis patients. Nephrol Dial Transplant. 2015;30(6):1037-46.

32. Sprague SM et al. Efficacy of PA21, a novel iron-based phosphate binder, maintained to 52 weeks in dialysis patients with hyperphosphatemia. TH-OR027. American Society of Nephrology Kidney Week, 5–10 November 2013.

33. Ketteler M et al. CKD-MBD markers in a phase 3 study of sucroferric oxyhydroxide in dialysis patients with hyperphosphatemia. MON-465. ISN World Congress of Nephrology, 13–17 March 2016.

34. Ficociello L et al. Serum phosphorus and pill burden in hemodialysis patients switched to sucroferric oxyhydroxide (so) as part of routine care. Poster 194. National Kidney Foundation, Spring Clinical Meetings. 2016.

NEW INSIGHTS INTO IRON METABOLISM AND DEFICIENCY

This symposium took place on 23rd May 2016, as part of the European Renal Association - European Dialysis and Transplant Association (ERA-EDTA) Congress 2016 in Vienna, Austria

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MEETING SUMMARY

The symposium provided an overview of the prevalence of iron deficiency and the associated disease burden in patients with chronic kidney disease (CKD). Prof Kai-Uwe Eckardt gave an overview of the prevalence of iron deficiency in patients with CKD not undergoing dialysis and addressed the challenge of diagnosing iron deficiency in this patient population based on the definitions currently used. Prof Tomas Ganz then reviewed the pathophysiology of iron metabolism, and explained the complex interplay of hepcidin in making iron available for erythropoiesis. The symposium concluded with a presentation from Prof Jolanta Małyszko who reviewed the methods of determining iron status among patients with CKD and compared data on the benefits and risks of intravenous (IV) and oral iron therapy.

Prevalence of Iron Deficiency in Patients with Chronic Kidney Disease: A Matter of Definition?

Professor Kai-Uwe Eckardt

The three main causes of renal anaemia are erythropoietin (EPO) deficiency, iron deficiency, and inflammation. Although iron deficiency is a cause of renal anaemia, it can also manifest in other ways. Diagnostic methods of measuring tissue iron content include bone marrow biopsy (an invasive procedure) and liver magnetic resonance imaging, which is not routinely available. The use of surrogate markers such as ferritin and the transferrin saturation (TSAT) as diagnostic tools is routine clinical practice, although they come with several limitations, including being influenced by

the presence of inflammation. In the general population, the threshold for the normal levels of these surrogate markers is lower than if measured in a population of patients with CKD on dialysis, but the appropriate cut-offs for patients with CKD not on dialysis (ND-CKD) is less clear. A recent systematic review concluded that guidelines[1] recommend the use of higher thresholds of ferritin and TSAT in patients with ND-CKD. When looking at a range of interventional iron studies in patients with ND-CKD, the inclusion criteria for iron parameters also stipulate higher threshold values for ferritin and TSAT. However, when using such high threshold values in patients with ND-CKD, the frequency of 'iron deficiency' in this patient population is high. An analysis of data from the National Health and Nutrition Examination Survey (NHANES) found that the majority of patients with CKD had levels of serum ferritin <100 ng/mL or TSAT <20%.[2]

The German Chronic Kidney Disease (GCKD) study is an observational prospective cohort study that aims to increase the understanding of the natural course of CKD, by identifying and validating the risk factors and markers for the manifestation, progression, and complications of CKD. This study recruited over 5,000 patients[3] and an analysis of iron parameters revealed that the majority of patients do not meet the target levels established in CKD haemodialysis patients. Whether this indicates a high prevalence of iron deficiency or impropriety of such cut-off values in patients with less advanced CKD, is difficult to define. In conclusion, diagnosis of iron deficiency in patients with ND-CKD remains a challenge. The risk-benefit relationship for treatment of these patients rather than specific laboratory values should guide therapy.

Iron Pathophysiology: Its Complexity and Our Knowledge Gaps

Professor Tomas Ganz

There are two kinds of iron regulation in the body. The first is systemic regulation, whereby the organism regulates its dietary iron absorption, the concentration of iron in extracellular fluid, and iron storage. The second is cellular regulation, whereby iron uptake and subcellular distribution are controlled at the level of each individual cell.

Erythrocytes are made in the bone marrow and contain iron; each millilitre of packed erythrocytes represents a milligram of iron. The lifecycle of an erythrocyte is 110–120 days, after which it is taken up by macrophages in the spleen and liver, and the iron is transferred to the plasma, where it binds to transferrin and circulates until it is taken up by the bone marrow again to make more erythrocytes (Figure 1).

Due to the lack of excretory mechanisms, very little iron is normally lost from the body; however, in patients with CKD and those on dialysis this loss is increased. The usual homeostatic processes ensure that increased losses in iron are compensated, either by increased absorption of iron in the small intestine and duodenum, or the use of iron from the liver where surplus iron is stored. These compensatory mechanisms are greatly affected during infection and inflammation, leading to a reduction in plasma iron concentrations known as 'hypoferraemia of inflammation', eventually leading to the development of anaemia due to a reduced production of erythrocytes.

Erythropoietic stimulation, a process that results in the production of more erythroid precursors involved in the generation of erythrocytes, requires additional iron to be absorbed into the duodenum, or taken up from hepatocytes into the plasma.

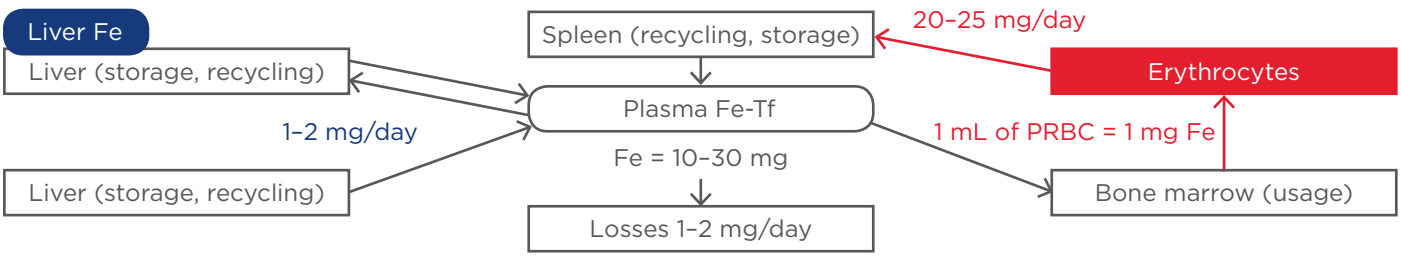


Figure 1: Overview of systemic iron metabolism.
Fe: iron; PRBC: packed red blood cells; Tf: transferrin.

This is a normal physiological process and often occurs during bleeding or if EPO is administered. The chief regulator of iron homeostasis is hepcidin, which is secreted by hepatocytes as the 84-amino-acid preprohepcidin and then cleaved to a 25-amino-acid bioactive hepcidin by the prohormone convertase furin.[4] Hepcidin regulates intestinal iron absorption and iron distribution in tissues by binding to the ferroportin receptor, a 12-transmembrane-segment protein that is present in macrophages, in the duodenum, on hepatocytes, and in the placenta.[5-7] Binding of hepcidin to the ferroportin receptor results in its degradation[8] and decreased cellular iron export. When hepcidin is low, duodenal enterocytes absorb dietary iron and export it into the blood, but high hepcidin inhibits these processes. Thus hepcidin regulates dietary iron absorption and the influx of iron to the plasma at the level of iron absorption, and also similarly at the level of iron recycling and at the level of release from stores.

Hepcidin levels are regulated by levels of iron in the plasma, iron stores in the liver, and erythropoietic signals from the bone marrow. Administration of iron and subsequent measurement of hepcidin levels have shown that, in response to iron, there is a spike in serum iron with an increase in serum and urinary hepcidin.[9] Detection of plasma iron takes place via a complex of transferrin receptors (transferrins 1 and 2) and a human haemochromatosis molecule on the external membrane of hepatocytes that senses the concentration of holo-transferrin and conveys this message intracellularly, resulting in increased hepcidin messenger ribonucleic acid (mRNA) and consequently increased hepcidin production.

Intestinal iron absorption is greatly increased after the administration of EPO and in forms of anaemia in which erythropoiesis is active, such as non-transfused β-thalassaemia.[10] These observations suggest that there is a circulating factor that connects erythropoiesis to iron regulation, and that this factor is likely produced in the bone marrow. One study of five male volunteers who were given EPO has shown that serum hepcidin levels drop 9–24 hours after administration and this effect lasts for at least 5 days, with minor reductions in transferrin, ferritin, and levels of the transferrin receptor.[11] Searches for the factor connecting erythropoiesis to iron regulation has led to the identification of erythroferrone, which is highly expressed in EPO-stimulated erythroblasts and acts as an erythroid regulator of iron metabolism. During anaemia or hypoxia, the kidneys produce EPO which stimulates erythroferrone production and in turn suppresses the production of hepcidin in the liver, increasing iron absorption and making more iron available for erythropoiesis.

During infection or inflammation, hepcidin levels increase and serum iron levels decrease, as demonstrated in an *in vivo* human endotoxaemia model.[12] By contrast, in hepcidin knockout mice that were given an inflammatory stimulus, increased levels of iron were evident,[13] demonstrating that hypoferraemia of inflammation is dependent on hepcidin. Patients with CKD have hepcidin-dependent anaemia, hepcidin-independent anaemia, and a relative lack of EPO. Hepcidin-dependent effects in these patients are mediated by an increase in inflammatory cytokines (e.g. interleukin [IL]-6) that increase hepcidin and cause iron trapping in macrophages, resulting in a reduction in available iron and the restriction of haem and haemoglobin synthesis and erythropoiesis. Hepcidin-independent effects in patients with CKD include shortened erythrocyte lifespan and the direct suppression of erythropoiesis by cytokines. In patients with progressive CKD, levels of hepcidin were higher and directly proportional to the severity of kidney disease, compared with paediatric or adult controls.[14,15] Increased circulating hepcidin, resulting from inflammatory stimulation of hepcidin production and decreased hepcidin clearance, restricts the release of iron into the plasma, causing hypoferraemia. IV iron administration loads macrophages with iron, thereby stimulating ferroportin synthesis in macrophages. Increased ferroportin facilitates the export of iron trapped in macrophages out of the cell for erythropoiesis, overcoming the effect of high levels of hepcidin in the plasma and making more iron available for stimulated erythropoiesis.

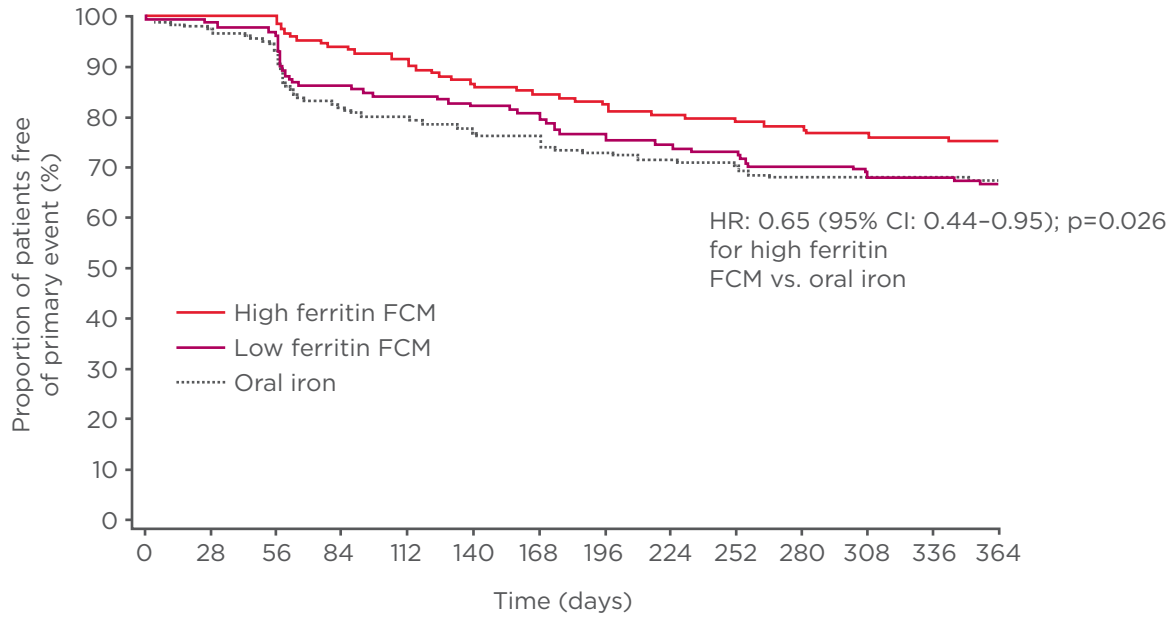
Diagnosing and Treating Iron Deficiency/Iron-Deficiency Anaemia: Meeting Your Patient's Needs

Professor Jolanta Małyszko

The typical patient undergoing haemodialysis has impaired EPO production and EPO receptor function, impaired iron absorption, iron loss during their haemodialysis sessions, inflammation,

and increased iron utilisation (following the administration of EPO-stimulating agents), all of which can lead to iron deficiency and anaemia. Although patients with ND-CKD appear to be less anaemic, they are still iron deficient as a result of impaired iron absorption and repeated venepuncture, with approximately 60% of patients with ND-CKD who start on dialysis being deficient in iron.[16] Assessment of iron deficiency prior to iron therapy is important; this is usually done by measuring the levels of serum ferritin, serum iron, TSAT, and total iron binding capacity; and assessing reticulocyte haemoglobin content, measuring occult blood in stools, determining red blood cell indices, and measuring levels of haemoglobin.[17] Iron stores should be evaluated and non-renal causes of anaemia should be excluded from this assessment.[17] Often, defining iron deficiency using serum ferritin levels and TSAT is difficult in the CKD population, as these biochemical markers can often be affected by acute-phase reactions, particularly those seen in inflammatory disease states such as diabetes and cardiovascular disease, diseases that commonly occur in this population. The advantages of assessment of iron stores using serum ferritin levels as a measure include: high specificity of low levels of this haematological parameter being indicative of iron deficiency;[18] its correlation with body iron stores in healthy individuals;[19] and its ease-of-use, moderate cost, and wide availability. However, normal or high serum ferritin does not exclude functional iron deficiency[20] and there are also observed gender differences in this measurement.[21] In contrast, TSAT is a more reliable measure of iron deficiency than serum ferritin as it is more sensitive[18] and the absence (or near-absence) of sustainable iron in the bone marrow correlates with TSAT <20%.[21] It must be noted that, in patients undergoing dialysis, there is 17–70% diurnal variation in TSAT levels[20,21] and levels can be affected by inflammation, malnutrition, and chronic disease, interfering with its reliability as a measurement of iron deficiency.[21]

Clinical guidelines recommend treating iron-deficiency anaemia with oral or IV iron before initiating other anaemia management,[22,23] as optimal red blood cell production requires iron for haemoglobin synthesis.[24,25] Iron losses in patients with CKD undergoing haemodialysis can be attributed to repeated laboratory tests, accidental losses during haemodialysis and other bleeding events, blood retention in the artificial kidney and tubing, and normal iron losses; these incremental losses can result in the loss of up to 3,000 mg of iron per year.[18]



High serum ferritin FCM	153	153	150	139	134	126	123	118	114	112	110	107	106	81
Low serum ferritin FCM	152	150	145	129	124	121	117	109	106	103	96	93	93	75
Oral iron	308	299	278	241	229	219	212	203	198	194	185	185	184	151

Figure 2: Primary endpoint results of the FIND-CKD study.[30]
FCM: ferric carboxymaltose; HR: hazard ratio; CI: confidence interval.

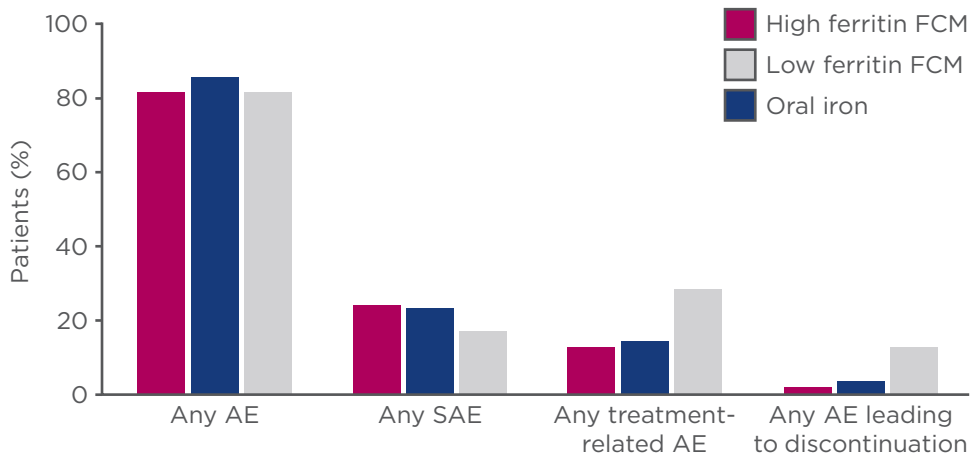


Figure 3: Safety results across treatment groups in the FIND-CKD study.[30]
FCM: ferric carboxymaltose; AE: adverse events; SAE: serious adverse events.

In patients with CKD on haemodialysis, the administration of parenteral iron is routinely employed due to the loss of blood associated with haemodialysis, the need for adequate levels of iron in response to EPO administration, and because patients are often unable to respond to oral iron. In ND-CKD patients, oral or IV iron therapy is initiated depending on the severity of anaemia. ND-CKD patients with severe anaemia may have gastrointestinal intolerance for oral iron therapy and their iron deficiency is unlikely to be corrected within 3 months of receiving oral iron administration. As in patients on haemodialysis, those receiving EPO-stimulating agents are also recommended for IV iron therapy.[22]

Oral iron treatment offers several advantages: it is widely used, inexpensive, and easily administered,[20,26] with no requirement for outpatient visits.[20] However, adherence can be a problem, the underlying blood loss pathology is often not resolved,[27] and iron absorption can be inhibited due to other medications or diet.[28] Oral iron can also lead to frequent gastrointestinal side effects, such as nausea, constipation, and diarrhoea.[28]

IV iron treatment has shown benefits in patients with ND-CKD. The FIND-CKD study was the largest and one of the longest (56-week) randomised studies comparing IV and oral iron in patients with ND-CKD.[29] The study recruited >600 patients with a haemoglobin level of 9–11 g/dL, serum ferritin <100 µg/L, or serum ferritin <200 µg/L + TSAT <20%. The three treatment groups were IV ferric carboxymaltose (FCM) (200 and 1,000 mg) and oral ferrous sulphate (200 mg iron/day), with a primary endpoint of time to initiation of an alternative treatment for anaemia or occurrence of a haemoglobin trigger (specified as two consecutive haemoglobin values <10 g/dL on or after Week 8, without an increase of ≥0.5 g/dL between consecutive values). Secondary endpoints included the percentage of patients with an increase of haemoglobin ≥1 g/dL, and a change in haematological and iron indices. Results showed that 76% of patients with ND-CKD maintained a haemoglobin level ≥10 g/dL or did not require further anaemia treatment when treated with FCM targeting high serum ferritin levels (Figure 2).

FCM targeting of a higher ferritin level also achieved a faster and greater increase in haemoglobin levels versus oral iron. High ferritin FCM also resulted in the desired serum ferritin targets being achieved, and TSAT levels were maintained within guideline recommendations versus oral iron for all time points ($p<0.001$).[29] There were no changes in adverse events between the FCM groups targeting high and low serum ferritin levels, but there were higher rates of adverse events leading to treatment discontinuation in the oral iron group (Figure 3). Importantly, there was no sign of renal toxicity in the FCM group targeting ferritin levels of 400–600 µg/L.

IV iron therapy is well established in patients on haemodialysis, however the benefits of IV iron therapy beyond red blood cell management is still a point of discussion in ND-CKD patients, although the FIND-CKD, a 1-year study with FCM, suggests a faster and greater haemoglobin response with IV iron compared with oral iron in ND-CKD patients.[29] Further research to establish benefits and risks of IV iron therapy is desired.[31]

REFERENCES

1. Peyrin-Biroulet L et al. Guidelines on the diagnosis and treatment of iron deficiency across indications: A systematic review. Am J Clin Nutr. 2015;102(6):1585-94.

2. Fishbane S et al. Iron indices in chronic kidney disease in the National Health and Nutritional Examination Survey 1988-2004. Clin J Am Soc Nephrol CJASN. 2009;4(1):57-61.

3. Eckardt KU et al. The German Chronic Kidney Disease (GCKD) study: Design and methods. Nephrol Dial Transplant. 2012;27(4):1454-60.

4. Jordan JB et al. Hepcidin revisited, disulfide connectivity, dynamics, and structure. J Biol Chem. 2009;284(36): 24155-67.

5. Donovan A et al. The iron exporter ferroportin/Slc40a1 is essential for iron homeostasis. Cell Metab. 2005;1(3): 191-200.

6. McKie AT, Barlow DJ. The SLC40 basolateral iron transporter family (IREG1/ferroportin/MTP1). Pflüg Arch Eur J Physiol. 2004;447(5):801-6.

7. Abboud S, Haile DJ. A novel mammalian iron-regulated protein involved in intracellular iron metabolism. J Biol Chem. 2000;275(26):19906-12.

8. Nemeth E et al. Hepcidin regulates cellular iron efflux by binding to ferroportin and inducing its internalization. Science. 2004;306(5704):2090-3.

9. Ganz T et al. Immunoassay for human serum hepcidin. Blood. 2008;112(10): 4292-7.

10. Finch C. Regulators of iron balance in humans. Blood. 1994;84(6):1697-702.

11. Ashby DR et al. Erythropoietin administration in humans causes a marked and prolonged reduction in circulating hepcidin. Haematologica. 2010;95(3): 505-8.

12. Kemna E et al. Time-course analysis of hepcidin, serum iron, and plasma cytokine levels in humans injected with LPS. Blood. 2005;106(5):1864-6.

13. Kim A et al. A mouse model of anemia of inflammation: Complex pathogenesis with partial dependence on hepcidin. Blood. 2014;123(8):1129-36.

14. Zaritsky J et al. Hepcidin--a potential novel biomarker for iron status in chronic kidney disease. Clin J Am Soc Nephrol. 2009;4(6):1051-6.

15. Zaritsky J et al. Reduction of serum hepcidin by hemodialysis in pediatric and adult patients. Clin J Am Soc Nephrol. 2010;5(6):1010-4.

16. Valderrábano F et al. PRE-dialysis survey on anaemia management. Nephrol Dial Transplant. 2003;18(1):89-100.

17. Małyszko J et al. Iron metabolism in solid-organ transplantation: how far are we from solving the mystery? Pol Arch Med Wewn. 2012;122(10):504-11.

18. Kalantar-Zadeh K et al. The fascinating but deceptive ferritin: to measure it or not to measure it in chronic kidney disease? Clin J Am Soc Nephrol. 2006;1 Suppl 1: S9-18.

19. Crichton RR et al. Iron Therapy with special emphasis on intravenous administration. (2005) Bremen: UNI-MED Verlag AG.

20. Macdougall IC. Monitoring of iron status and iron supplementation in patients treated with erythropoietin. Curr Opin Nephrol Hypertens. 1994;3(6): 620-5.

21. Wish JB. Assessing iron status: beyond serum ferritin and transferrin saturation. Clin J Am Soc Nephrol. 2006;1 Suppl 1: S4-8.

22. KDIGO Clinical Practice Guideline for Anemia in Chronic Kidney Disease. Kidney Int Suppl. 2012;2(4):283-7.

23. National Institute for Health and Care - excellence Clinical Guideline. Chronic kidney disease: Managing anaemia. 2015. Available at: https://www.nice.org.uk/guidance/ng8/chapter/introduction. Last accessed: 3 June 2016.

24. Besarab A et al. Iron metabolism, iron deficiency, thrombocytosis, and the cardiorenal anemia syndrome. Oncologist. 2009;14 Suppl 1:22-33.

25. Beard JL. Iron biology in immune function, muscle metabolism and neuronal functioning. J Nutr. 2001;131(2S–2): 568S-79S.

26. Del Vecchio L, Locatelli F. Anemia in chronic kidney disease patients: Treatment recommendations and emerging therapies. Expert Rev Hematol. 2014;7(4):495-506.

27. Brugnara C, Beris P, "Iron therapy," Beaumont C et al. (eds.), Disorders of erythropoiesis, erythrocytes and iron metabolism (2009), Paris: European School of Haematology, pp.512-28.

28. Macdougall IC. Iron supplementation in the non-dialysis chronic kidney disease (ND-CKD) patient: Oral or intravenous? Curr Med Res Opin. 2010;26(2):473-82.

29. Macdougall IC et al. The FIND-CKD study--a randomized controlled trial of intravenous iron versus oral iron in non-dialysis chronic kidney disease patients: background and rationale. Nephrol Dial Transplant. 2014;29(4):843-50.

30. Macdougall IC et al. FIND-CKD: A randomized trial of intravenous ferric carboxymaltose versus oral iron in patients with chronic kidney disease and iron deficiency anaemia. Nephrol Dial Transplant. 2014;29(11):2075-84.

31. Macdougall IC et al. Iron management in chronic kidney disease: Conclusions from a "Kidney Disease: Improving Global Outcomes" (KDIGO) Controversies Conference. Kidney Int. 2016;89(1):28-39.

SECONDARY CARNITINE DEFICIENCY IN DIALYSIS PATIENTS: SHALL WE SUPPLEMENT IT?

Summary of presentations from the Sigma-Tau Pharma International Sponsored Symposium, held at the 53rd Congress of the European Renal Association and European Dialysis and Transplant Association (ERA-EDTA) in Vienna, Austria, on 24th May 2016

Chairs

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MEETING SUMMARY

Carnitine, essential for fatty acid β-oxidation, is obtained from diet and through *de novo* biosynthesis. The organic cation/carnitine transporter 2 (OCTN2) facilitates carnitine cellular transport and kidney resorption. Carnitine depletion occurs in OCTN2-deficient patients, with serious clinical complications including cardiomyopathy, myopathy, and hypoketotic hypoglycaemia. Neonatal screening can detect OCTN2 deficiency. OCTN2-deficiency is also known as primary carnitine deficiency. Carnitine deficiency may result from fatty acid β-oxidation disorders, which are diagnosed via plasma acylcarnitine profiling, but also under other conditions including haemodialysis.

Given the importance of the kidney in maintaining carnitine homeostasis, it is not unexpected that long-term haemodialysis treatment is associated with the development of secondary carnitine deficiency, characterised by low endogenous L-carnitine levels and accumulation of deleterious medium and long-chain acylcarnitines. These alterations in carnitine pool composition have been implicated in a number of dialysis-related disorders, including erythropoietin-resistant renal anaemia. The association between erythropoietin resistance and carnitine levels has been demonstrated, with the proportion of medium and long-chain acylcarnitines within the total plasma carnitine pool positively correlated with erythropoietin resistance. Recent research has demonstrated that carnitine supplementation results in a significant reduction in erythropoietin dose requirements in patients with erythropoietin-resistant anaemia.

Few studies have been conducted assessing the treatment of carnitine deficiency and haemodialysis-related cardiac complications, particularly in children. Thus, a study was recently conducted which showed that intravenous carnitine in children receiving haemodialysis significantly increased plasma carnitine

levels and improved the acylcarnitine to free carnitine ratio. Cardiac function was also significantly improved (determined by longitudinal strain rate using speckle-tracking echocardiography). A study in children receiving continuous renal replacement therapy (CRRT) showed that prevalence of carnitine deficiency increased with the time on CRRT. The impact of carnitine deficiency resulting from CRRT is not well known, although it has been associated with increased mortality in critically ill children. An ongoing, randomised controlled clinical trial is assessing the impact of carnitine supplementation on myocardial function in children receiving CRRT.

Thus, carnitine deficiency is a disorder with significant clinical impact, particularly in patients undergoing renal replacement therapy, which can be simply diagnosed. Carnitine supplementation can be used to effectively treat carnitine deficiency.

Carnitine Metabolism in Human Health and Disease Notably in Genetic Metabolic Diseases

Professor Doctor Ronald J.A. Wanders

Carnitine Biosynthesis and Homeostasis

Carnitine was first discovered in muscle extracts in 1905.[1] Subsequently, Dr G. S. Fraenkel discovered a key role of carnitine through his study of insects and found that carnitine is essential for the beetle *Tenebrio molitor*. Experimentally, excluding carnitine from culture medium resulted in the death of beetle larvae and accumulation of fat was noted. This observation was the first evidence that carnitine may have a role in fatty acid oxidation. In 1955, Dr I. B. Fritz demonstrated that carnitine stimulates fatty acid oxidation, and it has since been established that fatty acid β-oxidation is fully dependent on carnitine.[1]

Humans obtain carnitine from several major dietary sources (meat, fish, and dairy products), and can also biosynthesise carnitine from the amino acid lysine.[1] For omnivores, 75% of body carnitine originates from the diet. In contrast, vegetarians and vegans need to biosynthesise >75% of their body carnitine and may have low carnitine levels as a result. The *de novo* synthesis of carnitine involves four enzymatic steps and is shown in Figure 1.[1]

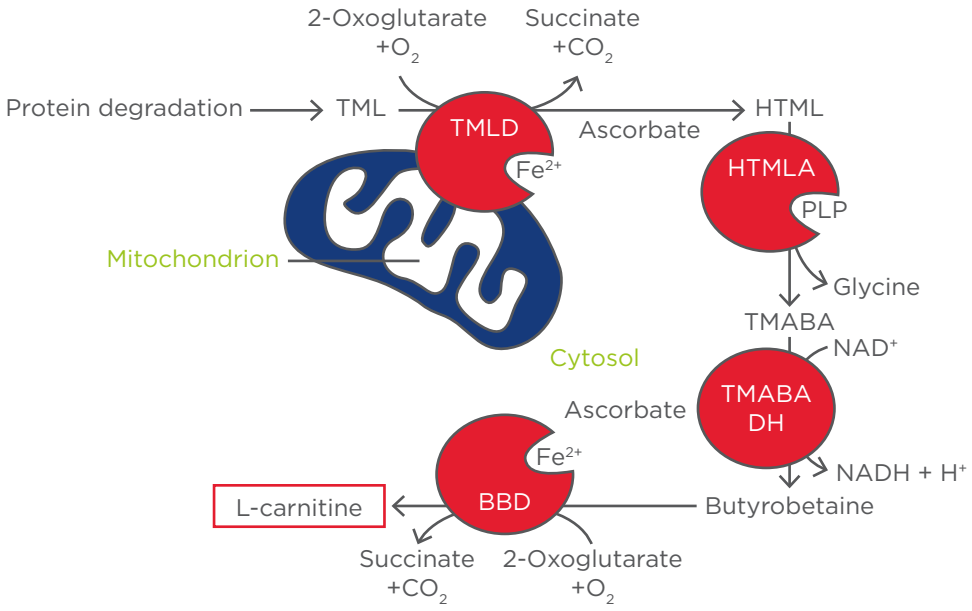

Figure 1: Carnitine biosynthesis.[1]
BBD: γ-butyrobetaine dioxygenase; HTML: β-hydroxy-ε-N-trimethyllysine; HTMLA: β-hydroxy-ε-N-trimethyllysine aldolase; TMABADH: γ-trimethylaminobutyraldehyde dehydrogenase; TML: ε-N-trimethyllysine; TMLD: ε-N-trimethyllysine dioxygenase; PLP: pyridoxal 5'-phosphate; NAD: nicotinamide adenine dinucleotide.
Adapted from Vaz FM and Wanders RJ.[1]

Importantly, humans have a low capacity for carnitine synthesis. Moreover, this biosynthetic process proceeds at a fixed rate and cannot be induced to produce more carnitine.

Carnitine homeostasis in humans reflects the balance between *de novo* synthesis and dietary uptake, with loss via urine and faeces. The protein OCTN2 plays a crucial role in carnitine homeostasis. This plasma membrane-based protein actively facilitates the sodium-dependent transport of carnitine from the plasma (typical carnitine concentrations 20–40 µmol/L) to the cytosol of cells (typical carnitine concentrations >2,000 µmol/L) in all tissues. In the kidney, OCTN2 activity also results in carnitine resorption.

Functions of Carnitine

Carnitine has several important physiological functions in humans. An essential role for carnitine is in fatty acid β-oxidation in the mitochondria, known as the carnitine cycle (Figure 2). In carnitine deficiency, this carnitine cycle is unable to complete effectively.

Carnitine is also involved in the transfer of peroxisomal fatty acid β-oxidation end-products to the mitochondria for full oxidation to carbon dioxide and water.[1] The third role of carnitine is to remove acyl-coenzyme (CoA) species from the mitochondria and the cell. This function is very important as it is the only way to remove acyl-CoA, which is deleterious to cells. Within mitochondria, acyl-CoA is converted to acyl-carnitine which is transported out of the cells by carnitine-acyl-carnitine translocase (mitochondrion membrane) and OCTN2 probably via one of the OCTNs in the plasma membrane. Plasma acyl-carnitine is then excreted via urine and faeces.

Carnitine-Related Disorders

To date, only a few disorders of carnitine biosynthesis have been identified. The most frequently occurring condition is OCTN2 deficiency, a genetic disorder involving the gene *SLC22A5*, which codes for OCTN2.[1] Deficiency of OCTN2 has several manifestations, in particular, defective carnitine uptake into cells and resorption by the kidneys. Consequently, plasma carnitine concentrations are low (<1 µmol/L) and urinary carnitine levels are high. Critically, fatty acid β-oxidation is impaired in virtually all tissues.

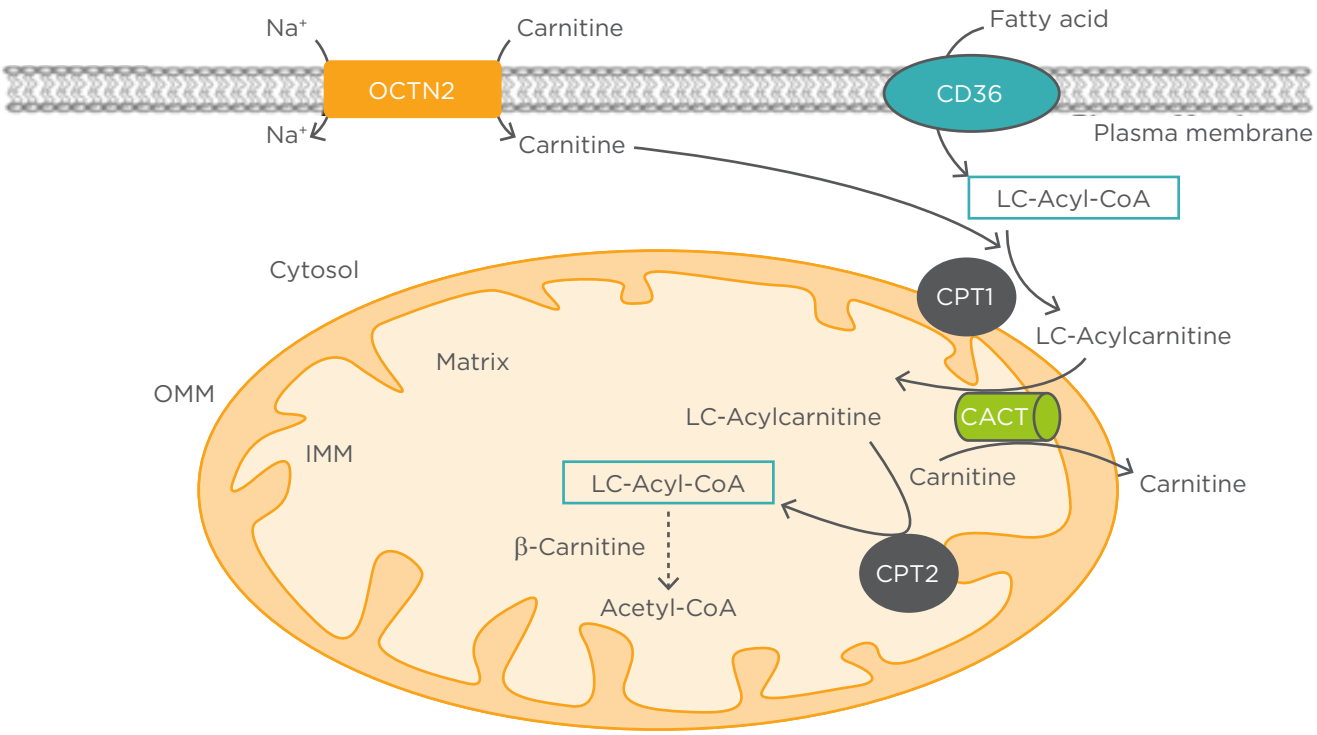

Figure 2: The carnitine cycle.
CACT: carnitine-acyl-carnitine translocase; CoA: coenzyme A; CPT: carnitine palmitoyl transferase; LC: long chain; OCTN2: carnitine/organic cation transporter 2; OMM: outer mitochondrial membrane; IMM: inner mitochondrial membrane.

Clinical signs and symptoms of OCTN2 deficiency are variable and wide ranging. In early-onset presentation of this condition, the following can occur: acute metabolic decompensation, hypoketotic hypoglycaemia, Reye's syndrome, and sudden infant death.[1] Presentation of OCTN2 deficiency later in life is more insidious with chronic myopathy, with or without muscle weakness, which may result in sudden (cardiac) death.

A key challenge facing clinicians is to identify patients with OCTN2 deficiency as soon as possible, ideally through neonatal screening.[2] Diagnosis is important as carnitine therapy is lifesaving, and can correct clinical signs and symptoms of carnitine deficiency. The first neonatal screen for an inborn error of metabolism was pioneered by Dr Guthrie, who developed a test for phenylketonuria in a dried blood spot. Currently, the newborn heel prick, utilising a Guthrie card, is routinely used for screening numerous metabolic diseases, including a range of fatty acid oxidation disorders. Testing for OCTN2 deficiency using this method is available in the USA and in some European countries.

Many inborn errors of fatty acid β-oxidation are known.[2] One of the most frequent deficiencies is a defect in medium chain acyl-CoA dehydrogenase (MCAD). This enzyme catalyses the first step in the degradation of octanoyl-CoA (C8:0-CoA) and decenoyl-CoA (C10:1-CoA) to carbon dioxide and water, and ketone bodies. When MCAD is deficient, C8:0-CoA and C10:1-CoA build up in the mitochondria. Subsequently, C8:0-carnitine and C10:1-carnitine levels are increased in the plasma and urine. Profiling plasma acylcarnitines provides valuable insight into which enzymes and/or transporters are defective in fatty acid β-oxidation.[2] The diagnosis is then confirmed by enzymatic and molecular analyses. Carnitine deficiency can also occur secondary to other conditions or illnesses, the best described of which is dialysis-related carnitine deficiency, which is detailed in the coming section.

Characteristics of Dialysis-Related Carnitine Deficiency: Effectiveness of L-Carnitine for the Treatment of Erythropoietin-Resistant Renal Anaemia

Doctor Stephanie E. Reuter

The kidney has a critical role in carnitine homeostasis through the maintenance of normal endogenous carnitine levels.[3] Given its low molecular weight and polarity, carnitine is extensively filtered at the glomerulus, but then undergoes extensive saturable resorption in the proximal convoluted tubule to avoid extensive loss into the urine. Whilst haemodialysis provides a valuable replacement for kidney function in end-stage renal disease (ESRD), it is unable to compensate for all homeostatic mechanisms and, in the case of carnitine, results in the development of dialysis-related carnitine deficiency.

Dialysis-Related Carnitine Deficiency

The impact of haemodialysis on endogenous carnitine levels has been established, with a single 3-hour dialysis session found to result in a substantial (74%) reduction in plasma L-carnitine concentrations during the intra-dialytic period.[4] However, in the 2-day inter-dialytic period, plasma L-carnitine levels were restored as body carnitine levels re-equilibrated and L-carnitine moved out of the tissue stores and replenished the plasma carnitine pool. Whilst a 74% loss of a plasma L-carnitine pool that only comprises <1% of total body carnitine is unlikely to significantly impact on carnitine pool composition following a single dialysis session, ongoing 3-times per week haemodialysis over an extended period would result in substantial changes in the endogenous carnitine pool, particularly in a setting with decreased dietary intake and a protein-restricted diet, along with a reduction in carnitine biosynthesis by the damaged kidney.

The relationship between dialysis age and carnitine concentrations was subsequently examined in patients with ESRD during the first year of haemodialysis treatment and in the longer term (>12 months).[5] During the first year of dialysis, plasma carnitine levels declined significantly from baseline levels, with the majority of change occurring within the first few months. Concentrations continued to decline with increasing dialysis age, such that all long-term haemodialysis patients had plasma L-carnitine levels below that previously established for healthy controls (Figure 3).[5,6] Examination of muscle L-carnitine concentrations also indicated a significant decline with increasing time on dialysis treatment.

Previous research has demonstrated that administration of intravenous L-carnitine at the end of each dialysis session results in the

replenishment of total body carnitine stores.[4] As expected, haemodialysis results in a substantial clearance of exogenous carnitine. However, carnitine administration at the end of dialysis resulted in a pharmacokinetic profile consistent with the distribution of exogenous carnitine into the peripheral compartments and incorporation into the tissue stores. This has subsequently been illustrated using pharmacokinetic modelling.[7]

Interestingly, in these previous studies it was noted that whilst plasma L-carnitine levels were substantially affected by haemodialysis treatment, total plasma carnitine concentrations (i.e. L-carnitine and the sum of all acylcarnitines) were relatively unaffected. Examination of the relative compositions of the endogenous plasma carnitine pool in haemodialysis patients indicated that haemodialysis treatment is associated with not only loss of L-carnitine, but also accumulation of medium and long-chain acylcarnitines.[5] Strikingly, in long-term haemodialysis patients, these carnitine esters comprise approximately 25% of the total plasma carnitine pool, compared with negligible levels in healthy controls (Figure 4).

Examination of the pattern of acylcarnitine accumulation with haemodialysis treatment indicated that 29 out of 31 individual acylcarnitines quantified were significantly higher in long-term haemodialysis patients compared with healthy controls.[8] Furthermore, the dialytic removal of acylcarnitines was inversely related to carbon-chain length of the carnitine ester, such that the longest acylcarnitines were not removed at all by dialysis. This is likely a result of increasing molecular size and increased protein binding associated with increasing carbon chain length.

Clinical Relevance of Dialysis-Related Carnitine Deficiency

In patients receiving long-term haemodialysis treatment, perturbation of carnitine homeostasis has been linked to several common dialysis-related conditions such as erythropoietin-resistant renal anaemia, cardiac dysfunction, dialytic symptoms (muscle fatigue, asthenia, cramps), and poor quality of life.[9] In December 1999, based on the above data establishing the development of dialysis-related carnitine deficiency, levocarnitine (Carnitor®, Sigma-Tau Pharmaceuticals, Gaithersburg, MD, USA) was approved by the US Food and Drug Administration (FDA) for the prevention and treatment of carnitine deficiency in patients with ESRD receiving dialysis treatment. Following this FDA approval, consensus guidelines, developed in September 2002 by the National Kidney Foundation (NKF) Kidney Disease Outcomes Quality Initiative (KDOQI), recommended intravenous L-carnitine supplementation for the treatment of a number of dialysis-related conditions.[10] Additionally, intravenous L-carnitine received coverage by the US Centers for Medicare and Medicaid Services for the treatment of erythropoietin-resistant anaemia and/or intradialytic hypotension in carnitine-deficient patients receiving long-term haemodialysis treatment.[11] However, the benefit of L-carnitine supplementation for these conditions remains a matter for debate.

Extensive research has been conducted examining the potential benefit of L-carnitine as an adjunct for the treatment of renal anaemia.[9] Whilst most studies have demonstrated improvements with respect to haematocrit or erythropoietin dose, many were poorly designed with short treatment periods, small sample sizes, and/or uncontrolled/unblinded study designs. Furthermore, a number of studies employed the use of oral L-carnitine, a treatment option that is not recommended due to poor bioavailability, potential accumulation of trimethylamine-N-oxide from the gastrointestinal biodegradation of carnitine to trimethylamine, and possible acylation of L-carnitine during oral absorption. Interestingly, from these studies some authors noted that some patients 'responded' better to carnitine supplementation whilst others did not. Amongst a number of possible explanations, it was proposed that the patients who exhibited a greater response to carnitine treatment were those who displayed more disturbed carnitine profiles.

To explore this further, the relationship between endogenous plasma carnitine pool composition and erythropoietin requirements were assessed in long-term haemodialysis patients.[12] In order to consider both erythropoietin dose and effectiveness of that dose, the erythropoietin resistance index (ERI) was determined for each patient, calculated as dose/kg/week/g haemoglobin (Hb); based on NKF practice recommendations erythropoietin resistance was defined as >0.02 μg/kg/week/g Hb. A significant negative correlation between ERI and plasma L-carnitine levels was demonstrated, such that all patients classified as erythropoietin resistant exhibited subnormal L-carnitine levels (<30 μM).

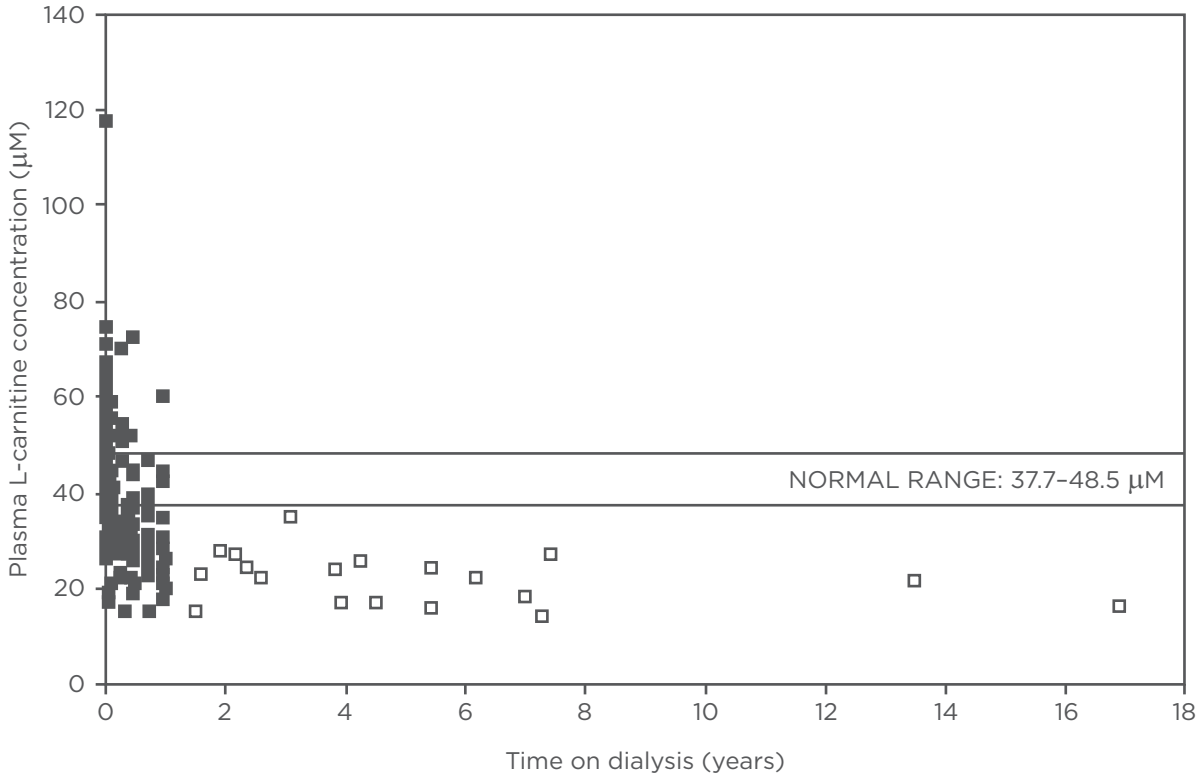

Figure 3: Long-term effect of haemodialysis on plasma carnitine concentrations.[5]
Endogenous plasma L-carnitine concentrations as a function of dialysis age in longitudinal patients during the first 12 months of haemodialysis treatment (solid squares) and in single-session patients having received dialysis for >12 months (open squares).

A stronger positive correlation between ERI and the proportion of medium and long-chain acylcarnitines within the total plasma carnitine pool was found, thereby indicating that a more disturbed carnitine profile is associated with erythropoietin resistance. Whilst the exact mechanism needs to be fully elucidated, it is proposed that this is mediated through effects on carnitine palmitoyltransferase (CPT) activity, an important enzyme involved in the incorporation of fatty acids into the erythrocyte membrane. Previous studies have illustrated that CPT can be regulated by carnitine and acylcarnitine levels, such that high levels of L-carnitine increase CPT activity whereas acylcarnitine inhibits CPT. Feasibly, the pattern of secondary carnitine deficiency seen in long-term haemodialysis treatment would result in inhibition of CPT by high acylcarnitine levels, which is not counteracted by the low levels of L-carnitine. This would conceivably lead to decreased acyl trafficking and reduced erythrocyte membrane repair and stabilisation. It is hypothesised that administration of L-carnitine in haemodialysis would result in a better balance of CPT activity thereby increasing the lifespan of the erythrocyte, providing an adjunct to erythropoietin in the treatment of anaemia.

Interestingly, only one published study has assessed the benefit of L-carnitine supplementation for renal anaemia in a specific group of patients who exhibit erythropoietin-resistance. However, this study employed a non-controlled, open-label assessment of oral L-carnitine. Recently, a randomised, double-blind, placebo-controlled study (ACCORD [Assessment of Carnitine for Clinical Outcomes in Renal Disease]) examined the effect of intravenous L-carnitine administration (20 mg/kg after each dialysis session for 6 months, or a matched placebo) in a group of patients classified as erythropoietin-resistant (>0.02 µg/kg/week/g Hb). L-carnitine treatment was shown to result in a significant improvement in erythropoietin requirements over time (~40% reduction), an effect that was significantly greater than that seen in placebo-treated patients (publication submitted for consideration). These results provide compelling evidence for the use of L-carnitine for the treatment of erythropoietin-resistant anaemia. Further studies examining the potential benefit of

L-carnitine for other dialysis-related conditions is currently being conducted.

Carnitine Levels and Myocardial Function in Children Receiving Chronic Haemodialysis

Professor Doctor Asha Moudgil

Children with ESRD receiving chronic haemodialysis are at risk for carnitine deficiency and cardiac complications. Since carnitine is essential for cardiac muscle function, carnitine deficiency in children may contribute to cardiac complications and supplementation may help improve cardiac function.

Improved Cardiac Function with Carnitine

In adults receiving chronic haemodialysis, oral[13,14] and intravenous[15,16] carnitine treatment improved ejection fraction (a marker of systolic left ventricular [LV] function), and intravenous carnitine supplementation improved myocardial fatty acid imaging.[17] Few studies have investigated the effect of carnitine supplementation on cardiac function in children receiving haemodialysis, with mixed results. Oral carnitine was shown to improve some measures of LV function in two studies,[18,19] intravenous carnitine supplementation did not improve cardiac function as measured by standard echocardiography in another study.[20]

In a prospective, longitudinal, pilot study, we recruited patients (n=9) aged 2–21 years with ESRD and receiving haemodialysis for ≥3 months, on a stable erythropoietin dose and no underlying heart disease.[21] The study included a 3-month observation phase (no carnitine) followed by a 6-month intravenous carnitine (20 mg/kg/dialysis treatment) intervention phase. The retrospective control group (n=8) consisted of children receiving chronic haemodialysis (no carnitine supplementation) with data from two echocardiograms (≥6 months apart).

Patient demographics and baseline clinical characteristics were similar between groups. In the study group, pretreatment total carnitine and free carnitine plasma levels were low; mean (standard error of the mean [SEM]) values were 49 µmol/L (1.7) and 29 µmol/L (1.2), respectively.

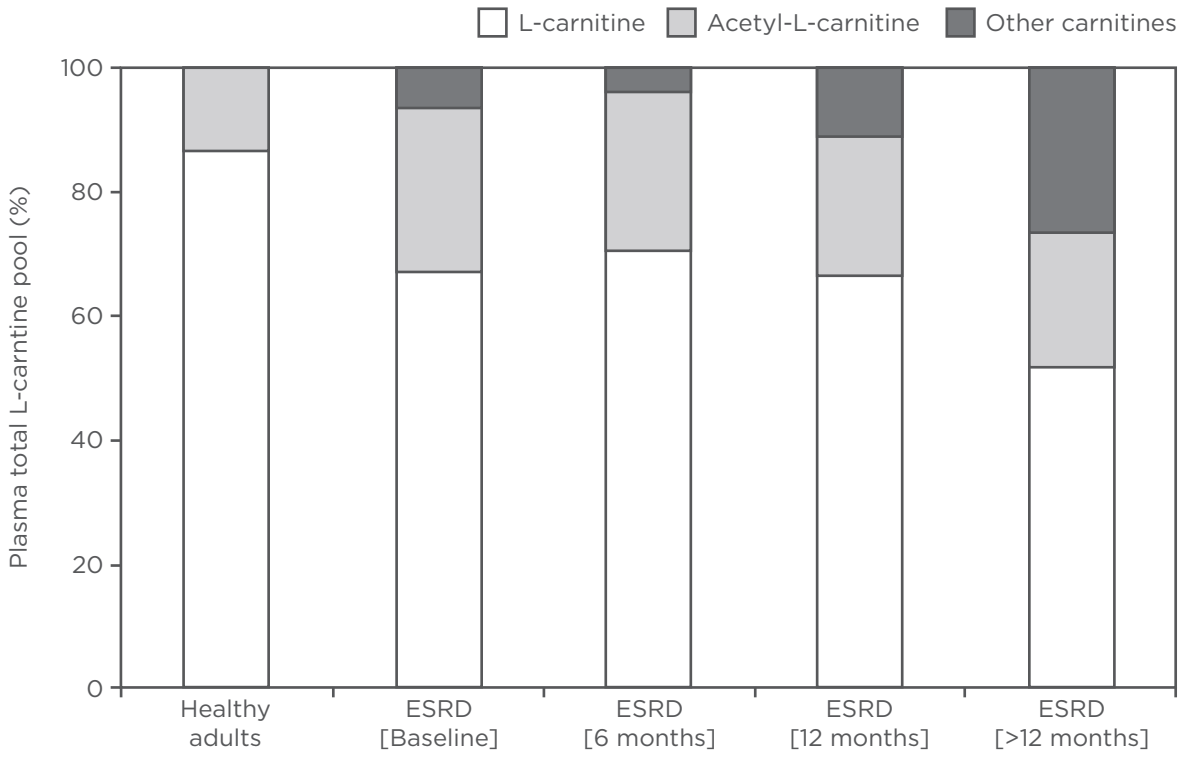

Figure 4: Impact of haemodialysis on plasma carnitine pool composition.[5]
Relative composition of the endogenous plasma carnitine pool in healthy controls and end-stage renal disease (ESRD) patients prior to commencing haemodialysis treatment (baseline) and after undergoing haemodialysis treatment for 6, 12, and >12 months.

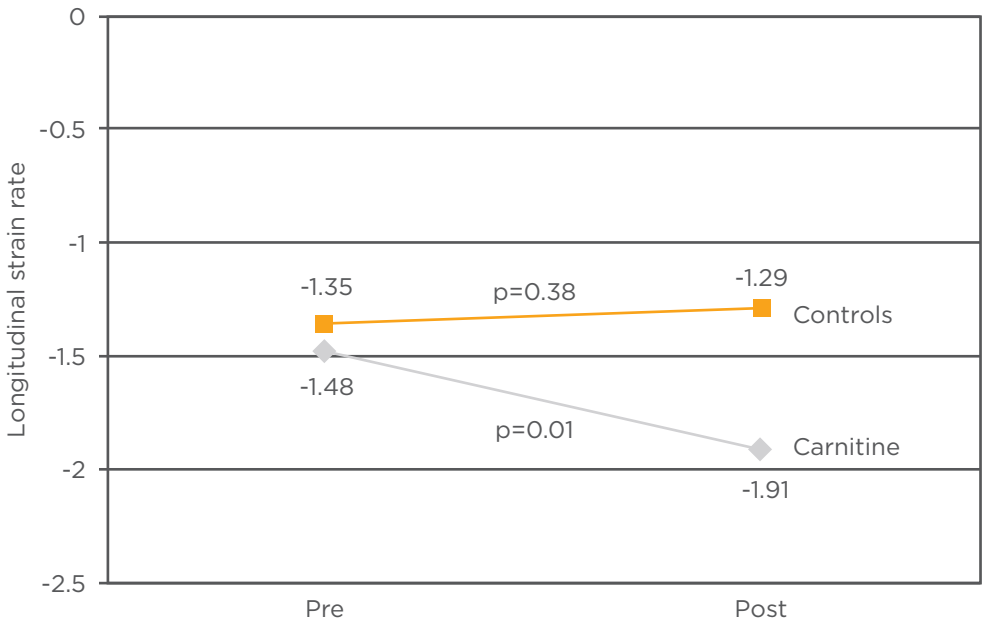

Figure 5: Longitudinal strain rate pre and post-carnitine treatment in children receiving haemodialysis.

After carnitine supplementation, total and free carnitine levels were markedly higher versus pretreatment; mean (SEM) values were 298 µmol/L (31.8) and 180 µmol/L (19.2), respectively, both p<0.0001.[21] Moreover, the acylcarnitine to free carnitine ratio was significantly reduced post-treatment versus pretreatment (mean ± SEM: 0.73±0.04 versus 0.65±0.05; p=0.02), although this ratio was not reduced to normal values.

No differences in LV function were seen in standard echocardiograms between the pre and post-carnitine treatment phases in the study group. However, with speckle-tracking echocardiography (a novel and sensitive technique that evaluates myocardial motion in three planes), the longitudinal strain rate was significantly improved by 33% with carnitine supplementation versus pretreatment values (mean ± SEM: -1.91±0.12 versus -1.48±0.11, respectively; p=0.01) (Figure 5). For this parameter, a more negative value equates to improved heart contractibility. In contrast, longitudinal strain rate in the control group was not significantly different between assessments (mean ± SEM: -1.35±0.13 versus -1.29±0.09, respectively; p=0.38). Overall, this study showed that in children receiving chronic haemodialysis, intravenous carnitine supplementation for 6 months improved plasma carnitine levels and improved LV function as measured by speckled tracking.[21]

Carnitine Deficiency in Continuous Renal Replacement Therapy

Carnitine homeostasis has not been well-studied in CRRT. Children receiving CRRT are highly likely to be carnitine deficient due to constant carnitine removal by CRRT; lack of production by the kidney and/or the liver; no dietary carnitine intake; and comorbidities associated with critical illness known to deplete carnitine (e.g. sepsis, muscle wastage, systemic inflammatory response syndrome). Preliminary data from a pilot study on the kinetics of carnitine removal in adults receiving continuous veno-venous haemofiltration (CVVH) indicated complete passage and efficient removal of free carnitine through the CVVH membrane, based on calculated sieving coefficients.[22] Carnitine deficiency, dyslipidaemia, and muscle catabolism were reported in a case report of a critically ill adult receiving CRRT for 4 months.[23]

Carnitine deficiency in children and young adults (n=42; 0–26 years old) with acute kidney injury receiving CRRT was assessed in a recent study at Children's National Health System in Washington DC, USA.[24] Mean (SEM) age was 7.9 years (1.1), 52% of recruited patients were male, and the mean (SEM) length of stay in the intensive care unit was 68.9 days (10.4). The range of the Paediatric Logistic Organ Dysfunction score was 2–19 (on a scale of 0–33). At baseline, approximately one-third of patients had carnitine deficiency: mean (SEM) for free carnitine was

25.2 µmol/L (4.4).[24] The proportion of carnitine-deficient patients (based on free carnitine plasma levels) was 70%, 90%, and 100% at Weeks 1, 2, and 3 of CRRT, respectively. Seventy-two percent of patients died. The odds ratio for death, adjusting for age, sex, and race, was 4.9 (p=0.03) based on free carnitine deficiency versus patients with normal carnitine levels. Thus, carnitine deficiency occurs rapidly in children receiving CRRT, and is associated with increased mortality.[24]

Currently, a clinical trial is in progress (NCT01941823) to compare the effects of carnitine supplementation (Carnitor at 20 mg/kg/day, intravenously) in children (1–17 years old) receiving CRRT with those not receiving carnitine (control). The key outcome measures are cardiac function and prevalence of carnitine deficiency.

Summary and Conclusions

Overall, the evidence demonstrates that carnitine is essential to human health. Although carnitine can be synthesised *de novo*, this capability is limited and dietary sources are crucial in maintaining carnitine body stores in healthy humans. Carnitine has several key roles, particularly in fatty acid β-oxidation and detoxification of resulting end products within the cell, as well as the removal of deleterious acyl-CoA species.

Primary carnitine deficiency arises from certain genetic defects, such as in OCTN2 and fatty acid, and amino acid oxidation pathways. Secondary carnitine deficiency results from other conditions/illness, the most well-studied of which is kidney failure and dialysis. Patients with ESRD receiving long-term haemodialysis have significant disruption of carnitine homeostasis with reduced plasma and tissue carnitine, and increased proportions of deleterious medium and long-chain acyl carnitines in the plasma. Similar disruptions occur in patients receiving CRRT. Carnitine deficiency, regardless of cause, has many clinical manifestations including erythropoietin-resistant renal anaemia, and cardiac dysfunction, particularly in children.

Carnitine deficiency is easily diagnosed either by early screening (in the case of primary deficiency) or by monitoring plasma carnitine levels in patients known to be at high risk, such as those receiving renal replacement therapy. Based on the clear evidence to date, carnitine deficiency, regardless of cause, should be treated with carnitine supplementation.

REFERENCES

1. Vaz FM, Wanders RJ. Carnitine biosynthesis in mammals. Biochem J. 2002;361(Pt 3):417-29.

2. Wanders RJ et al. The enzymology of mitochondrial fatty acid beta-oxidation and its application to follow-up analysis of positive neonatal screening results. J Inherit Metab Dis. 2010;33(5):479-94.

3. Reuter SE, Evans AM. Carnitine and acylcarnitines: pharmacokinetic, pharmacological and clinical aspects. Clin Pharmacokinet. 2012;51(9):553-72.

4. Evans AM et al. Pharmacokinetics of L-carnitine in patients with end-stage renal disease undergoing long-term hemodialysis. Clin Pharmacol Ther. 2000; 68(3):238-49.

5. Evans AM et al. Impact of hemodialysis on endogenous plasma and muscle carnitine levels in patients with end-stage renal disease. Kidney Int. 2004;66(4): 1527-34.

6. Reuter SE et al. Determination of the reference range of endogenous plasma carnitines in healthy adults. Ann Clin Biochem. 2008;45(Pt 6):585-92.

7. Fornasini G et al. A pharmacokinetic model for L-carnitine in patients receiving haemodialysis. Br J Clin Pharmacol. 2007; 64(3):335-45.

8. Reuter SE et al. Impact of haemodialysis on individual endogenous plasma acylcarnitine concentrations in end-stage renal disease. Ann Clin Biochem. 2005; 42(Pt 5):387-93.

9. Reuter SE et al. L-carnitine supplementation in the dialysis population: are Australian patients missing out? Nephrology. 2008;13(1):3-16.

10. National Kidney Foundation. K/DOQI clinical practice guidelines for chronic kidney disease: Evaluation, classification, and stratification. Am J Kidney Dis. 2002; 39(2 Suppl 1):S1-266.

11. Centers for Medicare and Medicaid Services (eds). Levocarnitine for End-Stage Renal Disease. Report CAG-00077n (2002). Baltimore, MD: Centers for Medicare and Medicaid Services.

12. Reuter SE et al. Endogenous plasma carnitine pool composition and response to erythropoietin treatment in chronic haemodialysis patients. Nephrol Dial Transplant. 2009;24(3):990-6.

13. Matsumoto Y et al. Effects of L-carnitine supplementation on cardiac morbidity in hemodialyzed patients. Am J Nephrol. 2000;20(3):201-7.

14. Khosroshahi HT et al. Effects of oral L-carnitine on cardiac abnormalities of maintenance hemodialysis patients. Biomed Int. 2010;1:30-3.

15. van Es A et al. Amelioration of cardiac function by L-carnitine administration in patients on haemodialysis. Contrib Nephrol. 1992;98:28-35.

16. Romagnoli GF et al. Beneficial effects of L-carnitine in dialysis patients with impaired left ventricular function: an observational study. Curr Med Res Opin. 2002;18:172-5.

17. Nishimura M et al. Effects of intravenous l-carnitine on myocardial fatty acid imaging in hemodialysis patients: responders or non-responders to l-carnitine. Springerplus. 2015;4:353.

18. Khoss AE et al. L-carnitine therapy and myocardial function in children treated with chronic hemodialysis. Wien Klin Wochenschr. 1989;101(1):17-20.

19. El-Metwally TH et al. Dyslipidemia,

oxidative stress and cardiac dysfunction in children with chronic renal failure: effects of L-carnitine supplementation. Ann Saudi Med. 2003;23(5):270-7.

20. Topaloğlu R et al. Effect of carnitine supplementation on cardiac function in hemodialyzed children. Acta Paediatr Jpn. 1998;40(1):26-9.

21. Sgambat K et al. Carnitine supplementation improves cardiac strain rate in children on chronic hemodialysis. Pediatr Nephrol. 2012;27(8):1381-7.

22. Pacitti A et al. A pilot study aimed to evaluate the loss of carnitine during intermittent and continuous veno-venous hemofiltration in acute kidney injury patients. Abstract 74. Poster presented at the 18th International Conference on Continuous Renal Replacement Therapies. San Diego, CA, Feb 12-15, 2013.

23. Bonafé L et al. Carnitine deficiency in chronic critical illness. Curr Opin Clin Nutr Metab Care. 2014;17:200-9.

24. Sgambat K, Moudgil A. Carnitine deficiency in children receiving continuous renal replacement therapy. Hemodial Int. 2016;20(1):63-7.

NOVEL ORAL ANTICOAGULANTS OR WARFARIN IN CHRONIC KIDNEY DISEASE: A CARDIOLOGIST'S PERSPECTIVE

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This presentation focussed on the use of either warfarin or novel oral anticoaglants (NOAC) in the setting of atrial fibrillation in patients with chronic kidney disease (CKD).

The current cardiology guidelines for atrial fibrillation (both the European Society of Cardiology [ESC] guidelines,[1] with a new version expected later in 2016, and the American College of Cardiology [ACC]/American Heart Association [AHA] guidelines[2]) recommend estimating a patient's risk of stroke by calculating the CHA_2DS_2-VASc score and to reduce the risk of bleeding by identifying modifiable factors included in the HAS-BLED score. If the CHA_2DS_2-VASc score is ≥2 an anticoagulant is recommended. The preceding speakers at the same session raised a concern that these risk scores have never been examined in the dialysis population. Regarding treatment, both warfarin and NOAC are considered equivalent in safety and efficacy, whereas antiplatelet agents are no longer recommended. The large-scale clinical trials have indicated a lower risk (almost halved) of intracranial haemorrhage with NOAC.

Renal function should be estimated with the Cockcroft–Gault formula and the dose adjusted when NOAC are used. Dabigatran is eliminated renally in about 80%, apixaban in 27%, and rivaroxaban in 33%. In the clinical trials, only patients with a moderately reduced renal function were included (dabigatran and rivaroxaban 30 mL/min, and apixaban 25 mL/min). Based on pharmacokinetics and dynamics studies, cautious use of both rivaroxaban and apixaban are approved down to a creatinine clearance of 15 mL/min.

The most problematic treatment group are patients with terminal renal function, where no evidence exists, and dialysis patients where only observational reports are available. Based on these reports, the Kidney Disease: Improving Global Outcomes (KDIGO) guidelines[3] advise against the use of warfarin due to the associated increased risk in bleeding. In comparison, also based on observational data, the cardiology guidelines[1,2] still recommend warfarin to prevent stroke as they consider the risk of bleeding acceptable.

There is a lack of evidence for use of NOAC in dialysis patients. Only two pharmacokinetic/dynamic studies exist with apixaban[4] and rivaroxban[5] in a limited number of haemodialysis patients. However, both dabigatran and rivaroxaban has been used off-label in haemodialysis patients[6] with an increased risk of bleeding when compared to warfarin.

The future may yet be promising, if large-scale clinical trials are undertaken and evaluate an appropriately adjusted dose of a NOAC in the dialysis population. Alternatively, novel treatment strategies which obviate the need for long-term anticoagulants, such as left atrial appendage occlusion devices, could be considered. These have never been tested in CKD patients.

CONCLUSION

Cardiology and nephrology guidelines differ in their recommendations for Stage 5 CKD dialysis patients. Cardiologists generally recommend warfarin whereas nephrologists recommend no therapy due to the increased risk of bleeding.

Limited dose-finding data exist for apixaban and rivaroxaban in haemodialysis patients, but there is a lack of clinical data and no clinical trial has evaluted their safety and effectiveness.

REFERENCES

1. Camm AJ et al. 2012 focused update of the ESC Guidelines for the management of atrial fibrillation: an update of the 2010 ESC Guidelines for the management of atrial fibrillation. Developed with the special contribution of the European Heart Rhythm Association. Eur Heart J. 2012;33(21):2719-47.

2. January CT et al. 2014 AHA/ACC/HRS guideline for the management of patients With atrial fibrillation: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines and the Heart Rhythm Society. J Am Coll Cardiol. 2014;64(21):e1-76.

3. KDIGO. KDIGO 2012 Clinical practice guideline for the evaluation and management of chronic kidney disease. Kidney Int Supplements. 2013;3(1).

4. Wang X et al. Pharmacokinetics, pharmacodynamics, and safety of apixaban in subjects with end-stage renal disease on hemodialysis. J Clin Pharmacol. 2016;56(5):628-36.

5. De Vriese AS et al. Dose-finding study of rivaroxaban in hemodialysis patients. Am J Kidney Dis. 2015;66(1):91-8.

6. Chan KE et al. Dabigatran and rivaroxaban use in atrial fibrillation patients on hemodialysis. Circulation. 2015;131(11): 972-9.

PSYCHOLOGICAL OUTCOMES AFTER LIVING KIDNEY DONATION

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Psychological outcomes after living kidney donation (LKD) have been an important consideration from the very beginning of living donor transplantation. This is principally due to the morally questionable act of performing a surgical procedure on one person for the primary physical benefit of another. They are only important within the context of living donation if one believes that the physical harms caused by donor surgery should be compensated for with some degree of gain.

There is currently no consensus on how donors should be assessed pre-operatively and how much weight likely psychological outcomes should be given within the donor assessment process.[1] Psychological outcomes after LKD have been summarised in two systematic reviews,[2,3] which show that psychological outcomes were mostly positive for the majority of donors, as demonstrated by low levels of depression and anxiety, and higher levels of self-esteem, self-worth, and self-confidence. Donors also reported a new appreciation of life and a positive outlook for the future. On the other hand, negative outcomes included feelings of fear, vulnerability, heightened health anxiety, and a sense of loss. Social issues included marital problems, familial conflicts, and financial hardship. Symptoms of depression, anxiety, and distress were also noted, being more prevalent in those whose recipients had experienced poor outcomes or where donor recovery had been suboptimal. Recipient death or graft loss was associated with feelings of devastation, guilt, despair, and inadequacy. Regret was commonly low but increased with recipient graft loss or complications.

By far the most commonly measured outcome in LKD is health-related quality of life (HRQoL). A systematic review of prospective studies[4] demonstrated that donors typically had a lower HRQoL shortly after donation. However, this either returned to pre-operative baseline within 3–12 months after donation, or was slightly reduced (but still comparable to the general population). One must be cautious when interpreting HRQoL data within the context of LKD simply because these measures will only inform you of quality of life outcomes related to health. Whilst this data is useful in comparing surgical techniques or to help monitor recovery from surgery, they do not provide sufficient data to understand how else the donors' quality of life may have been affected by donation. Using these measures in isolation should therefore be avoided.

When interpreting the LKD literature one must always be cautious of the methodological issues raised by Clemens and colleagues.[3] This review highlighted a scarcity of prospective studies, many of which had minimal or no demographic information as well as considerably variable follow-up times. Response rates were not always calculable and non-responders were not always

investigated. On occasions where questions related to LKD were specifically designed, very few were validated or piloted. With this in mind, one must pay more attention to those studies performed in more recent years which have sought to address many of these methodological issues and which provide a more comprehensive assessment of the donor as a whole.

In the modern era of living donor transplantation, we are faced with increasing physical risks to both the donor and the recipient. Put simply, we are transplanting kidneys from donors who previously would not have been allowed to donate (due to their age and comorbidities) into recipients who, in years gone by, would never have been considered transplantable due to their medical, immunological, and anatomical complexity. With this in mind, it is therefore more important than ever to appreciate and understand psychological outcomes after living donation in order to best assess and inform the living kidney donor prior to donation.

REFERENCES

1. Duerinckx N et al. Predonation psychosocial evaluation of living kidney and liver donor candidates: A systematic literature review. Transpl Int. 2014;27(1):2-18.

2. Tong A et al. The motivations and experiences of living kidney donors: A thematic synthesis. Am J Kidney Dis. 2012;60(1):15-26.

3. Clemens KK et al. Psychosocial health of living kidney donors: A systematic review. Am J Transplant. 2006;6(12):2965-77.

4. Wirken L et al. The course and predictors of health-related quality of life in living kidney donors: A systematic review and meta-analysis. Am J Transplant. 2015;15(12):3041-54.

LONG-TERM RISKS IN KIDNEY DONORS

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A kidney transplant from a living donor is the best treatment for end-stage renal disease (ESRD). We owe it to all donors to perform studies on short and long-term risks, both to ensure the safety of donors and to be able to communicate possible risks. It is important that studies on kidney donors include controls who are healthy enough to donate a kidney themselves. Donors are relatively young, with a mean age of around 40 years in most studies, and potential adverse effects of donation are likely to be small. Consequently, studies evaluating potential increased risks of death and ESRD in donors should have a long follow-up.

We published a study in 2014 that found increased risks for all-cause mortality, cardiovascular mortality, and ESRD in kidney donors.[1] The risk of all-cause mortality was 1.30 (95% confidence interval: 1.11–1.52) for donors compared with controls, with a corresponding increase in cardiovascular death. The relative risk of ESRD was greatly increased at 11.38 (4.37–29.6). A paper by Muzaale et al.[2] published in 2014 found around an 8 to 10-times increased risk of ESRD in those who had donated a kidney. This paper included 96,217 kidney donors who donated a kidney in the time period 1994–2011. Median follow-up was 7.6 years.

Since recent studies have found increased risks associated with kidney donation, the interpretation of these risks in relation to clinical practice is important. There are several aspects to consider when evaluating long-term risk. Many of these aspects have been described elsewhere by Robert Steiner.[3] Firstly, the potential donor's lifetime risk at baseline must be considered. Secondly, the incremental risk incurred by donor nephrectomy should be taken into consideration. Diseases that may be contracted later in life, such as diabetes, hypertension, or primary kidney disease, may worsen remaining renal function and lead to symptomatic renal disease at an earlier age than in a similar individual with two kidneys. Reduced renal function in the donor may be a risk factor for other diseases, most importantly cardiovascular disease. This association is known from studies in chronic kidney disease populations.

A normal donor evaluation is more reassuring in an older donor than in a younger donor. Since older

people in general tend to have more diseases than younger individuals, an older donor with a normal evaluation is relatively healthier than a similar younger donor. Most importantly, a younger donor will spend more remaining years with only one kidney. When evaluating long-term risks based on these facts, one may infer that remaining cumulative lifetime risk is higher in a healthy 25-year-old male than in a 60-year-old, otherwise healthy male with mild hypertension. Likewise, basic demographic factors of age and sex may have more impact on baseline risk than the occurrence of isolated medical abnormalities, for example, mild hypertension.

In light of results from recent studies on long-term risks in kidney donors, we have changed the written information to potential donors to include that donation may be associated with increased risks. Hopefully, in the future we will have more long-term data available to guide information to, and selection of, donors.

REFERENCES

1. Mjøen G et al. Long-term risks for kidney donors. Kidney Int. 2014;86(1):162-7.

2. Muzaale AD et al. Risk of end-stage renal disease following live kidney donation. JAMA. 2014;311(6):579-86.

3. Steiner RW. 'Normal for now' or 'at future risk': A double standard for selecting young and older living kidney donors. Am J Transplant. 2010;10(4):737-41.

LOOKING THROUGH RIFLE, AKIN, AND KDIGO GLASSES: DO WE SEE THE SAME?

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Acute kidney injury (AKI) is a syndrome that affects 13–18% of patients admitted to hospital and is particularly common in patients in the intensive care unit.[1] The impact and prognosis vary considerably depending on severity, acute and chronic comorbidities, and geographical location.[1-3]

The definition of AKI has evolved from the Risk, Injury, Failure, Loss, End-stage (RIFLE) criteria in 2004 to the AKI Network (AKIN) classification in 2007 (Table 1).[4,5] In 2012, both were merged, resulting in the Kidney Disease: Improving Global Outcomes (KDIGO) classification.[6]

The key differences are:

- The AKIN and KDIGO classification use a smaller change in serum creatinine to define AKI, compared to the RIFLE definition
- The RIFLE criteria include a 7-day window whereas the AKIN and KDIGO classification have incorporated a 48-hour window
- The AKI classification stipulates that adequate fluid resuscitation should have been undertaken and urinary obstruction excluded before the criteria are applied. This was not specified in the RIFLE and KDIGO classification
- The RIFLE definition includes glomerular filtration rate criteria and allows the use of the Modification of Diet in Renal Disease formula to back-calculate baseline renal function
- The AKIN and KDIGO classification include renal replacement therapy, as a separate criterion to define AKI Stage 3, irrespective of serum creatinine

Several studies have shown that all three classifications demonstrate an association between AKI and clinical outcomes. However, the incidence and stages of AKI vary when two or three classifications are applied to the same patient population.

Importantly, all three classifications are based on changes in serum creatinine and/or urine output. However, creatinine and urine output are markers of excretory function only and do not provide any information about any other roles of the kidney, i.e. metabolic, endocrine, or immunological functions. They are also not kidney specific and may change irrespective of renal function.

Table 1: RIFLE, AKIN, and KDIGO classifications for acute kidney injury.

	Serum creatinine criteria	Urine output criteria
RIFLE criteria		
Risk	Creatinine rise ≥1.5 to 2-fold from baseline or GFR decrease >25%	<0.5 mL/kg/h for >6 hours
Injury	Creatinine rise >2 to 3-fold from baseline or GFR decrease >50%	<0.5 mL/kg/h for >12 hours
Failure	Creatinine rise >3-fold from baseline or creatinine rise ≥354 µmol/L with an acute rise of ≥44 µmol/L or GFR decrease >75%	<0.3 mL/kg/h for 24 hours or anuria for 12 hours
Loss	Complete loss of kidney function for >4 weeks	
End-stage kidney disease	End-stage kidney disease >3 months	
AKIN classification		
Definition	An abrupt (within 48 hours) reduction in kidney function defined as an absolute increase in serum creatinine of either ≥0.3 mg/dL (≥26.4 µmol/L) or an increase ≥50% (1.5-fold) from baseline or a reduction in urine output (after exclusion of hypovolaemia and obstruction).	
Stage 1	Creatinine rise by ≥26 µmol/L (>0.3 mg/dL) or creatinine rise 1.5 to 2-fold from baseline	<0.5 mL/kg/h for >6 hours
Stage 2	Creatinine rise 2 to 3-fold from baseline	<0.5 mL/kg/h for >12 hours
Stage 3	Creatinine rise 3-fold or more from baseline or creatinine rise to ≥354 µmol/L with an acute rise of ≥44 µmol/L or RRT irrespective of serum creatinine	<0.3 mL/kg/h for 24 hours or anuria for 12 hours
KDIGO classification		
Definition	AKI is diagnosed if serum creatinine ≥26 µmol/L over ≤48 hours, or rises to ≥1.5-fold from baseline which is known or presumed to have occurred in the preceding 7 days.	
Stage 1	Creatinine rise ≥26.5 µmol/L in 48 hours or creatinine rise 1.5 to 1.9-times from baseline	<0.5 mL/kg/h for 6–12 hours
Stage 2	Creatinine rise 2.0 to 2.9-times from baseline	<0.5 mL/kg/h for ≥12 hours
Stage 3	Creatinine rise 3-times from baseline, or creatinine rise to ≥353.6 µmol/L or initiation of RRT irrespective of serum creatinine	<0.3 mL/kg/h for ≥24 hours or anuria for ≥12 hours

RIFLE: risk, injury, failure, loss, end-stage; AKIN: acute kidney injury network; KDIGO: kidney disease improving global outcomes; GFR: glomerular filtration rate; RRT: renal replacement therapy.

As a result, there are patients who have AKI but do not meet the RIFLE, AKIN, or KDIGO criteria, and there are also patients who fulfil the criteria for AKI but have not had a significant change in their renal function (Table 2). Until more sensitive and specific biomarkers are routinely used in clinical practice, it is essential to interpret changes in serum creatinine and urine output within the

clinical context. Finally, AKI is a syndrome and may have numerous different aetiologies. The RIFLE, AKIN, and KDIGO classification only serve to diagnose and stage AKI but do not provide any information about the underlying aetiology.

In conclusion, the RIFLE, AKIN, and KDIGO classifications are important tools to diagnose, stage, and prognosticate AKI, but need to be interpreted within the clinical context.

REFERENCES

1. Hoste EA et al. Epidemiology of acute kidney injury in critically ill patients: The multinational AKI-EPI study. Intensive Care Med. 2015;41(8):1411-23.

2. Lewington AJ et al. Raising awareness of acute kidney injury: A global perspective of a silent killer. Kidney Int. 2013;84(3): 457-67.

3. Mehta RL et al. International Society of Nephrology's 0by25 initiative for acute kidney injury (zero preventable deaths by 2025): A human rights case for nephrology. Lancet. 2015;385(9987):2616-43.

4. Bellomo R et al. 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(4):R204-12.

5. Mehta RL et al. Acute Kidney Injury Network: Report of an initiative to improve outcomes in acute kidney injury. Crit Care. 2007;11(2):R31.

6. ISN. KDIGO Clinical Practice Guideline for Acute Kidney Injury. Kidney Int Suppl. 2012;2(1):1-138.

Table 2: Potential pitfalls of AKI definition based on creatinine or urine output.

Clinical scenario	Consequence
Administration of drugs which interfere with tubular secretion of creatinine (i.e. cimetidine, trimethoprim)	Misdiagnosis of AKI (rise in serum creatinine without change in renal function)
Reduced production of creatinine (i.e. muscle wasting, liver disease, sepsis)	Delayed or misdiagnosis of AKI
Ingestion of substances which lead to increased generation of creatinine independent of renal function (i.e. creatine, red meat)	Misdiagnosis of AKI
Obesity	Over diagnosis of AKI if urine output criteria are applied to actual weight
Conditions associated with physiologically increased GFR (i.e. pregnancy)	Delayed diagnosis of AKI
Interference with analytical measurement of creatinine (i.e. 5-fluorocytosine, cefoxitin, bilirubin)	Misdiagnosis and delayed diagnosis of AKI (depending on the substance)
Fluid resuscitation and overload	Delayed diagnosis of AKI (dilution of serum creatinine concentration)
Hypovolaemia and physiological oliguria	Misdiagnosis of AKI

AKI: acute kidney injury; GFR: glomerular filtration rate.

SENSING ACID

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The daily intake and metabolism of an average Western diet as well as changes in physical activity or disease processes can cause an acid load to the body that requires buffering and eventually elimination. The lungs and kidneys participate in this critical task by first increasing ventilation then with some delay, enhancing renal acid excretion and bicarbonate regeneration. However, the mechanism by which these organs sense pH or its changes and then communicate to co-ordinate the compensatory response is only recently emerging.

We had hypothesised that a small group of three proton-activated G protein-coupled receptors (GPCRs) may contribute to proton sensing.[1,2] These three GPCRs: TDAG8, OGR1, and GPR4 are expressed in almost all organs and cell types and are activated in the physiological pH range with low activity at pH 7.6 and maximal stimulation at pH 6.8. These receptors have now been linked to a variety of physiological and disease processes such as bone resorption (TDAG8), asthma, activation of T cells during encephalitis in a murine model of multiple sclerosis (TDAG8), inflammatory bowel disease (all three receptors), pH and calcium regulation in brain granule cells (OGR1), fear-conditioning (OGR1), VEGF-dependent neovascularisation of solid tumours (GPR4), insulin secretion and sensitivity (OGR1, GPR4), and regulation of renal acid-base transporters (OGR1 and GPR4).

The kidney expresses both OGR1 and GPR4. Mice lacking OGR1 show a normal adaption to an acid load but do not hyperexcrete calcium during acidosis due to enhanced expression of the TRPV5 calcium channel as well as the intracellular calbindin-D28k calcium buffer. In contrast, GPR4 deficient mice have been reported to have a reduced renal capacity to excrete acid.[3] While renal acid excretion is only slightly reduced, respiratory adaption to metabolic acidosis as well as elevated CO_2 is severely blunted in the absence of GPR4 suggesting a role of GPR4 in the control of breathing.[4] GPR4 plays a critical role in a major subset of neurons in the retrotrapezoid nucleus in the medulla oblongata. However, GPR4 has no impact on the chemosensitivity for oxygen.

Another interesting feature of the proton-activated GPCRs is their emerging role in inflammatory processes. Small clinical trials and animal studies both support a role of acidosis in the progression of chronic kidney disease. Several effector pathways including the complement system and the endothelin-aldosterone axis have been implicated but the upstream pH-sensors have remained elusive. Preliminary data suggest increased expression of the proton-activated GPCRs in kidney tissue. Whether the receptors play a role in disease progression remains to be tested but would provide interesting therapeutic targets.

REFERENCES

1. Brown D, Wagner CA. Molecular mechanisms of acid-base sensing by the kidney. J Am Soc Nephrol. 2012;23(5):774-80.

2. Ludwig MG et al. Proton-sensing G-protein-coupled receptors. Nature. 2003; 425(6953):93-8.

3. Sun X et al. Deletion of the pH sensor GPR4 decreases renal acid excretion. J Am Soc Nephrol. 2010;21(10):1745-55.

4. Kumar NN et al. Regulation of breathing by CO_2 requires the proton-activated receptor GPR4 in retrotrapezoid nucleus neurons. Science. 2015;348(6240):1255-60.

FIRST DIALYSIS MODALITY CHOICE IN DIABETIC PATIENTS WITH END-STAGE KIDNEY DISEASE

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Diabetes is the most frequent cause worldwide of end-stage kidney disease (ESKD) requiring chronic renal replacement therapy, yet the question as to what could be the optimal dialysis technique for treating diabetic patients, be it peritoneal dialysis (PD) or haemodialysis (HD), still remains unanswered. Constant exposure to glucose in the dialysate may worsen glycaemic control in diabetic patients when on PD. On the other hand, PD therapy may be better tolerated than HD because of a more stable blood pressure, particularly in subjects with overt autonomic neuropathy. No less important, the creation of good vascular access in the presence of advanced diabetic vasculopathy may be challenging, and fistula failure episodes may become frequent.

In the recent past, randomised controlled trials comparing PD to HD have been demonstrated to be very problematic, due to recruitment and equipoise problems. Proper clinical guidelines

are therefore limited and there is considerable heterogeneity in practice across countries with regard to the information given to patients on the dialysis modality recommended as first option. A recent survey conducted in the USA among nephrologists has shown that people with diabetes had half the odds of being recommended for PD. Such an observation was in complete opposition to another similar survey among Canadian, British, and American nephrologists, showing that diabetics tends to favour PD slightly. The issue of whether first dialysis choice may impact on hard clinical outcomes for diabetic patients has been specifically addressed by the European Renal Best Practice (ERBP) Group as part of their recent diabetes guidelines.[1] The evidence systematically retrieved was mostly confined to observational studies assessing the mid to short-term risk of death in PD versus HD in incident cohorts.[2] Results were highly inconsistent, potentially influenced by selection and lead-time bias along with other methodological pitfalls, and varied across study designs, follow-up period, and subgroups.

Although no evidence-based arguments were found in favour of or against a particular dialysis modality as first choice treatment in patients with diabetes and ESKD, some concerns seem to arise about choosing PD in elderly and frail patients, since this technique was associated with a higher risk of death, particularly within the first 3 years. Sparse data were obtained on the risk of infectious complications. Conversely, no information was available on the impact of dialysis modality choice on quality of life, patient satisfaction, major and minor morbid events, hospital admissions, deterioration of residual renal function, functional status, glycaemic control, access to transplantation, or survival of the technique. In the absence of targeted studies, specifically designed to clarify such an issue, modality selection in diabetic patients should still be driven by subjective preferences and individual conditions, after unbiased patient information about the various available treatment options. Making sure that all the different renal replacement therapy modalities can be made equally available for all patients is indispensable to allow free modality choice.

REFERENCES

1. Guideline Development Group. Clinical Practice Guideline on management of patients with diabetes and chronic kidney disease Stage 3b or higher (eGFR<45 mL/min). Nephrol Dial Transplant. 2015;30(Suppl 2):ii1-142.

2. Couchoud C et al. Dialysis modality choice in diabetic patients with end-stage kidney disease: A systematic review of the available evidence. European Renal Best Practice (ERBP) Diabetes Guideline Development Group. Nephrol Dial Transplant. 2015;30(2):310-20.

MANAGEMENT OF POLYCYSTIC KIDNEY DISEASE FROM CHILDHOOD TO ADULTHOOD

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Autosomal recessive polycystic kidney disease (ARPKD) is a rare disorder (1/20,000 live births); however, it is the most common cystic disease in childhood. It is generally diagnosed *in utero* or at birth, and within the first year of life, approximately 23–30% of affected infants die. Although ARPKD is considered a paediatric disorder, overall survival has significantly improved (15-year survival is 67–79%), thanks to improved neonatal care and renal replacement therapy.[1] Furthermore, some patients are diagnosed only in adolescence or adulthood with renal function ranging from normal to end-stage renal disease. Unfortunately, long-term data on outcome and natural history of this population are scarce. Recently, an international registry has been initiated to collect longitudinal data on ARPKD patients

(www.ARegPKD.org).[2] This will help to understand the complete phenotype of this rare disorder.[3]

Autosomal dominant polycystic kidney disease (ADPKD) is the most common monogenic cause of end-stage renal disease in humans. Although it is considered an adult disease, it is supposed that renal injury begins with the formation of the first cyst, frequently *in utero*,[4] and hypertension, urinary concentrating defect, proteinuria, and nephromegaly have been described in paediatric populations.[5,6] Testing of asymptomatic offspring for APDKD remains controversial due to the possible psychological stress and financial implications such as employment or insurance issues. We would therefore like to highlight the recent Kidney Disease: Improving Global Outcomes (KDIGO) guidelines on controversies in ADPKD, which recommend to discuss the advantages and disadvantages of testing asymptomatic offspring with the families, and to respect their decision.[7]

REFERENCES

1. Büscher R et al. Clinical manifestations of autosomal recessive polycystic kidney disease (ARPKD): Kidney-related and non-kidney-related phenotypes. Pediatr Nephrol. 2014;29(10): 1915-25.

2. ARegPKD - an international registry study for autosomal recessive polycystic kidney disease. Available at: https://www.aregpkd.org/index.php?id=about_aregpkd. Last accessed: 22 June 2016.

3. Ebner K et al. Rationale, design and objectives of ARegPKD, a European ARPKD registry study. BMC Nephrol. 2015;16:22.

4. Grantham JJ. Rationale for early treatment of polycystic kidney disease. Pediatric Nephrol. 2015:30(7);1053-62.

5. Reddy BV, Chapman AB. The spectrum of autosomal dominant polycystic kidney disease in children and adolescents. Pediatric Nephrol. 2016. [Epub ahead of print].

6. Mekahli D et al. Similar renal outcomes in children with ADPKD diagnosed by screening or presenting with symptoms. Pediatric Nephrol. 2010;25(11):2275-82.

7. Chapman AB et al. Autosomal-dominant polycystic kidney disease (ADPKD): Executive summary from a Kidney Disease: Improving Global Outcomes (KDIGO) Controversies Conference. Kidney Int. 2015:88(1):17-27.

OPTIMISING THE MANAGEMENT OF CYSTIC KIDNEY DISEASE AND CILIOPATHIES

Genetic Testing Holds the Key to Accurate Diagnosis and Effective Treatment

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Cystic nephropathies and ciliopathies encompass a clinically and genetically heterogeneous group of diseases. Differentiation of the various entities can be very difficult. Important contributions to a classification are provided by genetics, a field which has improved considerably in recent years with the advent of high-throughput, next-generation sequencing (NGS). The analysis of an increasing number of genes is largely benefitted by NGS-based approaches, which allow the parallel analysis of all disease genes (e.g. through the use of multi-gene panels).

A close interdisciplinary co-operation and dialogue between treating physicians and geneticists is very helpful and beneficial. Opportunities and shortcomings of the various genetic approaches in different clinical settings need to be balanced. Interpretation of data is the most challenging part of the analysis and requires expert knowledge in genetics and of the respective medical field. Currently, a one-size-fits-all approach is not reasonable and there are many arguments in favour of a more differentiated approach, dependent on the patient's phenotype.

Genetics might prove useful in the context and discussion of different clinical settings. Accurate genetic counselling with discussion of the expected clinical course and spectrum of symptoms is only possible if the underlying genotype is known.

The following conclusions can be drawn:

- NGS often provides significant advantages in terms of diagnostic cost, efficiency, and accuracy
- NGS is increasingly replacing the classical stepwise analysis ('gene-by-gene' analysis) for heterogeneous diseases
- Genetic diagnosis often provides a clear-cut assessment of the disorder and improved clinical care (especially important for young patients)
- Determination of recurrence risk for patient's own offspring (50% in the case of dominant inheritance versus practically no risk in the case of recessive disease)
- Assessment of recurrence risk for other family members (e.g. no risk for the rest of the family with a *de novo* mutation)
- Genetic diagnosis allows assessment of comorbidities and possible complications
- Mutation may have consequences for therapeutic approach, for prognosis, and recurrence risk in the context of transplants

INTRACELLULAR SIGNALS AS DRUGGABLE TARGETS IN RENAL INFLAMMATION

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Inflammation is a prominent feature of almost any renal disease. Leukocytes mediate initiation and progression of tissue damage by direct cytotoxicity, the secretion of soluble factors, or regulation of the immune response. A large array of chemokines and cytokines are involved in the recruitment of mononuclear cells into the kidney where effector functions are activated. Contrasting with other inflammatory and immune diseases such as rheumatoid arthritis, the number of anti-inflammatory drugs available in clinical practice for renal diseases is much more limited. In fact, not a single drug exists on the market to treat diabetic nephropathy, a paradigm of renal disease in which inflammation plays a pivotal role.

During recent years, tremendous efforts and advances have been made in order to develop more specific anti-inflammatory compounds as the general anti-inflammatory therapies can cause adverse effects associated with global immunosuppression, which render the patient more prone to infections. At present, anti-inflammatory drugs on the market span from general immune-suppressing therapies to highly specific monoclonal antibodies targeting distinct adhesion molecules, cytokines, or downstream intracellular signalling proteins (Table 1). In recent years, the treatment of inflammatory and autoimmune diseases such as colitis and arthritis has been revolutionised by the introduction of tumour necrosis factor-specific therapies. Other approaches to reducing the induction of inflammation currently under clinical evaluation include the inhibition of the master regulator nuclear factor kappa B (NF-κB) by small molecule inhibitors of upstream activators such as the IκB kinases (IKKs), c-Jun N-terminal kinases, Janus kinases (JAKs), or mitogen-activated protein kinases. Although in the past few years there has been substantial progress in the use of biologics in the treatment of certain glomerulonephritides that has transformed the outlook for those patients, our presentation mainly focussed on diabetic nephropathy due to the prominent role of inflammation in this common clinical condition.

Our group has been particularly interested in the role and therapeutic modulation of two key intracellular signalling pathways such as the

NF-κB and JAK/signal transducer and activator of transcription (STAT) in the micro and macrovascular diabetic complications. The canonical NF-κB pathway mediated by the inhibitor of IKK regulates the transcription of inflammatory genes involved in the pathogenesis of many renal diseases including diabetes. The NF-κB essential modulator binding domain (NBD) contained in IKKα/β is essential for IKK complex assembly. We have recently investigated the functional consequences of targeting the IKK-dependent NF-κB pathway in the progression of diabetes-associated nephropathy. In apolipoprotein E-deficient mice with diabetes induced by streptozotocin, treatment with a cell-permeable peptide derived from the IKKα/β NBD region resulted in renal protection, as evidenced by dose-dependent decreases in albuminuria, renal lesions (mesangial expansion, leukocyte infiltration, and fibrosis), intranuclear NF-κB activity, and pro-inflammatory and pro-fibrotic gene expression. This nephroprotective effect was accompanied by a decline in systemic T helper 1 cytokines. *In vitro*, NBD peptide prevented IKK assembly/activation, p65 nuclear translocation, NF-κB-regulated gene expression, and cell proliferation induced by either high glucose or inflammatory stimulation. Targeting NF-κB using natural compounds (berberine, celastrol, quercetin, and astragaloside IV) have also improved renal function and inflammation in experimental diabetic kidney disease. The small molecule bindarit, which downregulates the classical NF-κB pathway by acting on a specific subpopulation of NF-κB dimers, reduced albuminuria and urinary CCL2 levels in a 24-week, Phase II, randomised, controlled trial in patients with lupus nephritis. Twelve weeks of bindarit therapy was also reported to reduce albuminuria in another small, Phase II, randomised, controlled trial in 100 patients with diabetic kidney disease, but the full results have not been published and no new trials of this agent are ongoing.

Activation of JAK/STAT signalling is another pathway for hyperglycaemia-induced kidney injury. Dysregulation of the JAK/STAT pathway has been documented in human progressive diabetic kidney disease. Inhibitors of JAK2 (such as AG 490), JAK3 (Janex 1), and STAT1 (fludarabine) have been proposed as anti-diabetic agents. Our group have demonstrated that the over-expression of the intracellular negative regulators of JAK/STAT signalling, suppressors of cytokine signalling (SOCS) 1 and SOCS3, is also protective in experimental diabetic kidney disease. In addition, we have noted that a cell-permeable peptide mimicking SOCS1 protects against nephropathy by suppressing JAK/STAT-mediated renal cell responses to diabetic conditions. Remarkably, administration of SOCS1 peptide at both early and advanced stages of diabetes ameliorated renal STAT1/STAT3 activity and resulted in renal protection, as evidenced by reduced albuminuria, renal histological changes, kidney leukocyte recruitment, and expression levels of pro-inflammatory and pro-fibrotic markers, independently of lipid and glycaemic changes. Preclinical data implicate heat shock protein 90 (HSP90) as a promising anti-inflammatory target in a number of inflammatory conditions such as rheumatoid arthritis, systemic lupus erythematosus, and uveitis, among others. We have recently investigated whether pharmacological HSP90 inhibition ameliorates diabetes-associated renal damage. Treatment of diabetic mice with 17-dimethylaminoethylamino-17-demethoxygeldanamycin (DMAG) for 10 weeks improved renal function, as evidenced by dose-dependent decreases in albuminuria, renal lesions (mesangial expansion, leukocyte infiltration, and fibrosis), and expression of pro-inflammatory and profibrotic genes. Mechanistically, the renoprotective effects of DMAG are mediated by the induction of protective HSP70 along with inactivation of NF-κB, STAT, and target gene expression, both in diabetic mice and in cultured cells under hyperglycaemic and pro-inflammatory conditions. On the whole, our studies have identified that the modulation of two key intracellular signalling pathways such as the NF-κB and JAK/STAT could be of interest in the treatment of renal inflammation. However, further studies are needed in order to introduce drugs modulating those pathways into the clinical practice, as has been done in other inflammatory conditions such as rheumatoid arthritis.

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Table 1: Selected potential anti-inflammatory therapies in renal inflammation.

Therapy type	Potential therapies
TNF-α neutralisation	• Infliximab
CCR2/CCR5 antagonists	• CCX140B, PF04634817, BMS813160 • CCL2/CXCL12 spiegelmers
ICAM1 neutralisation	• Monoclonal antibodies
NF-κB inhibitors	• Bindarit • Natural products, parthenolide
JAK/STAT inhibitors	• Baricitinib • AG-490, Janex-1, SOCS, fludarabine

TNF-α: tumour necrosis factor alpha; JAK/STAT: Janus kinase-signal transducer and activator of transcription; NF-κB: nuclear factor kappa B; SOCS: suppressors of cytokine signalling; ICAM1: intercellular adhesion molecule 1.

THE DIALYSIS OUTCOMES AND PRACTICE PATTERNS STUDY (DOPPS) PROGRAMME: CELEBRATING 20 YEARS AND LOOKING AHEAD

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BRUCE ROBINSON: THE DIALYSIS OUTCOMES AND PRACTICE PATTERNS STUDY PROGRAMME CONTINUES TO GROW AND INVITES COLLABORATORS

The Dialysis Outcomes and Practice Patterns Study (DOPPS) Programme is celebrating its 20th anniversary in 2016. Throughout the history of the DOPPS Programme, a key motivation has been to understand the reasons for international variation in dialysis and chronic kidney disease (CKD) outcomes. The DOPPS is committed to partnerships that maximise the scientific value of the wealth of data made possible by all the participating facilities and patients. To continue the tradition of providing unique opportunities for scientific investigation, the DOPPS invites interested researchers to learn more about possible collaboration. For more information, please visit www.DOPPS.org.

MICHEL JADOUL: KEY LESSONS FROM THE FIRST 20 YEARS OF DIALYSIS OUTCOMES AND PRACTICE PATTERNS STUDY

Research from the DOPPS over the past 20 years has highlighted that arteriovenous fistulae (AVF) are the first choice vascular access in haemodialysis (HD), and are associated with the best outcomes. Vascular access use still varies greatly within and across countries. In countries such as the UK and USA, a culture devoted to raising AVF use has led to commendable improvements. By contrast, fistula use has fallen and/or catheter use has risen in other countries.

The DOPPS has been of paramount importance in monitoring trends in HD practices worldwide. Thus, the impact of policy or guideline changes in some countries or globally and their associations with outcomes have been studied, for example in the anaemia or CKD-mineral bone disorder field. The DOPPS has demonstrated the value of longer dialysis session length, and session length has been extended in recent years in many DOPPS countries, in part due to specific policy incentives.

DOPPS publications have further demonstrated that lower quality of life and symptoms of depression predict higher mortality rates amongst haemodialysis patients. As the renal community becomes more aware of the importance of quality of life as a key outcome, DOPPS data can be leveraged to identify the factors that are of greatest importance to patients and develop novel instruments to assess them.

SOPHIE LIABEUF: GUIDELINE ADHERENCE AND DRUG PRESCRIPTION IN EURODOPPS - FINDING FROM THE FIRST PROPOSAL CALL

EURODOPPS is a joint venture of the DOPPS and the ERA-EDTA to collect and analyse data to address questions that are of specific interest to the European community. Since its start in 2014, EURODOPPS has grown to be a fruitful collaboration among investigators on both sides of the ocean. The enthusiasm and number of European researchers applying to use EURODOPPS data to address specific research projects demonstrates the interest of the community in this endeavour.

ZIAD MASSY: CHRONIC KIDNEY DISEASE OUTCOMES AND PRACTICE PATTERNS STUDY - IMPROVING OUTCOMES IN ADVANCED CHRONIC DISEASE AND THE TRANSITION TO DIALYSIS

The Chronic Kidney Disease Outcomes and Practice Patterns Study (CKDOPPS) has now launched in Brazil, France, Germany, Japan, and the USA. CKDOPPS is part of the CKD-REIN cohort in France. The CKDOPPS was developed to address the difficulty of studying patients as they transition from advanced CKD to kidney failure. The motivation for the study is that much of the variation in patient outcomes for advanced CKD patients is likely attributable to practice differences in real-world settings.

SIMON DAVIES: PERITONEAL DIALYSIS OUTCOMES AND PRACTICE PATTERNS STUDY - THE LARGEST INTERNATIONAL STUDY OF PERITONEAL DIALYSIS PRACTICES

The Peritoneal Dialysis Outcomes and Practice Patterns Study (PDOPPS) was launched in 2012. It is co-ordinated by Arbor Research in collaboration with the International Society for Peritoneal Dialysis (ISPD). Data are being collected in Australia, Canada, Japan, the UK, and the USA.

Even at this early stage, the PDOPPS sample already represents the largest international study of peritoneal dialysis (PD) patients. With data collected, the study serves as an invaluable resource and research platform for the international PD community, and provides a means to understand variation in PD practices and outcomes, to identify optimal practices, and ultimately to improve outcomes for PD patients.

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EDITOR'S PICK

Targeted therapies (vascular endothelial growth factor/vascualar endothelial growth factor receptor targeting agents and/or mammalian target of rapamycin inhibitors) for the treatment of metastatic renal cell carcinoma have changed the treatment landscape of patients suffering from end-stage renal disease requiring dialysis. Data about the pharmacokinetics of these drugs in renal failure are scarce and the timely paper by Guida et al. that follows provides an informative summary of the available literature on this topic. This paper can be highly recommended to nephrologists and oncologists involved in the difficult management of these patients.

Prof Norbert Lameire

TARGETED AGENTS IN PATIENTS WITH METASTATIC RENAL CELL CARCINOMA ON DIALYSIS: MYTHS AND REALITY

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ABSTRACT

Agents targeting the vascular endothelial growth factor (VEGF)/VEGF receptor (VEGFR) pathway, as well as mammalian target of rapamycin (mTOR) inhibitors have revolutionised the therapeutic landscape of metastatic renal cell carcinoma (mRCC) in the past decade, greatly improving the survival rates of these patients. However, translating results of registrative Phase III trials into everyday clinical practice is often troublesome, since real-world patients are completely different from those enrolled in randomised controlled Phase III trials. Prospective data on active oncological treatments in mRCC patients on dialysis are dramatically lacking. This literature review summarises and critically comments on available data relative to mRCC patients on dialysis receiving either VEGF/VEGFR-targeting agents, or mTOR inhibitors. Although prospective studies would definitely be warranted in these specific patient populations, all the available data suggest that mRCC patients on dialysis have the same outcome, both in terms of efficacy and safety, as mRCC patients with normal or marginally impaired kidney function, when treated with VEGF/VEGFR-targeting agents and/or mTOR inhibitors.

Keywords: Vascular endothelial growth factor (VEGF), vascular endothelial growth factor receptor (VEGFR) targeting agents, mammalian target of rapamycin (mTOR) inhibitors, dialysis, end-stage renal disease (ESRD).

INTRODUCTION

A number of molecularly targeted therapies (i.e. agents targeting the vascular endothelial growth factor [VEGF]/VEGF receptors [VEGFR] pathway, as well as mammalian target of rapamycin [mTOR] inhibitors) have revolutionised the therapeutic landscape of metastatic renal cell carcinoma (mRCC) in the past decade, greatly improving the survival of these patients[1] irrespective of their baseline prognostic features.[2,3]

However, translating the results of a Phase III trial into everyday clinical practice is often troublesome since real-world patients are completely different from those enrolled in large, global, randomised, controlled Phase III trials, usually characterised by very strict inclusion and exclusion criteria. Indeed, we now know that patients not suitable for consideration for a clinical trial have a very poor prognosis.[4] It is therefore crucial to confirm the efficacy, as well as the safety profile, of these novel agents in specific patient subpopulations, typically excluded from clinical trials.

A better knowledge of the outcome of treatment in these subpopulations may in fact allow us to improve the way we take care of patients with complicated cases, as well as to understand whether the decision to exclude some of these subpopulations from clinical trials is sensible or not.

In this literature review, we summarise and critically comment on the available data relative to mRCC patients on dialysis receiving either VEGF/VEGFR-targeting agents or mTOR inhibitors. Since the available data are scarce, exclusively retrospective, and mainly coming from small series or even single case reports, the derived evidence is highly biased and is endowed with a low level of supporting data. Nevertheless, we strongly believe that, in the absence of reasonable alternatives, these data could be practically quite useful and scientifically hypotheses-generating.[5]

CANCER AND DIALYSIS

Data from cancer registries show a high incidence of cancer in patients with end-stage renal disease (ESRD).[6] In patients undergoing dialysis, a number of pro-carcinogenic conditions are often present, including immune suppression, the presence of uraemic toxins, chronic oxidative stress, and cytokine-mediated inflammatory responses. The development of kidney neoplasms, ranging from adenoma to metastatic carcinoma, is the most serious complication of acquired cystic kidney disease (ACKD). ACKD-associated renal cell carcinoma is seen predominantly in males, occurs approximately 20 years earlier than in the general population, and is frequently bilateral (9%) and multicentric (50%).[7]

ACKD occurs in patients who are on dialysis for ESRD. It is generally accepted that ACKD develops as a consequence of sustained uraemia and can first manifest in stages of chronic kidney disease, even before dialysis is initiated. The prevalence of ACKD is directly related to the duration of dialysis and the risk of cancer is directly related to the presence of cysts.[8]

In the CANDY (Cancer and Dialysis) study, Janus et al.[9] retrospectively analysed a population of 178 patients who developed cancer after initiation of chronic dialysis. The mean period between the beginning of dialysis and cancer diagnosis was 30.8 months, and the main primary cancer sites were genito-urinary (21%), haematologic (15%), lung (13%), gastrointestinal (13%), prostate (8%), and head and neck cancers (7%), while the remaining were miscellaneous malignancies. Only 28% of these patients received active anti-cancer treatments, including agents for which no recommendations in dialysis were available. Seventy-two of the patients received at least one drug that required a dosage adjustment or for which there were no data in dialysis. This lead to the development of iatrogenic toxicity in 44% of the treated patients; 34% related to drugs requiring dosage adjustment, and 17% related to additional drugs with no existing management recommendations in dialysis patients. Overall, 88% of those who received an active oncological treatment needed specific drug management in terms of dose adjustment and/or time of administration according to the dialysis session of at least one anti-cancer drug. Notably, just 11% of the anti-cancer agents administered to studied patients were represented by target therapies, either monoclonal antibody (7%), or tyrosine kinase inhibitors (TKIs) (4%). Fifty-eight percent of the CANDY patients died during the 2-year follow-up period after cancer diagnosis and about half of those cases were due to cancer, with median survival time being 13.5 months after the diagnosis of malignancy. Furthermore, 38% of the CANDY patients died within a period of 2 years after dialysis onset versus 28% in the French Renal Epidemiology and Information

Network (REIN) registry.[10] Notably, in the European Renal Association and European Dialysis and Transplantation Association (ERA-EDTA) registry, mortality from malignancies was 2.9-times higher in dialysis patients than in the general population.[11]

Renal Cell Carcinoma

Renal cell carcinoma represents 2–3% of all malignancies in adults and generally occurs during the sixth and seventh decades of life.[12] Approximately a third of all patients with newly diagnosed renal cell carcinoma present with metastatic disease, and as many as 50% of those completely resected for a localised disease develop a local or distant relapse. Five percent of patients present with a bilateral renal mass with or without a known hereditary renal cancer syndrome.[13]

In regards to surgery, over the years less invasive techniques (compared with radical nephrectomy) have been developed for smaller tumours; indeed, according to the 2015 guidelines of the European Association of Urology (EAU), partial nephrectomy is recommended in patients with T1a tumours (i.e. a tumour ≤4 cm maximum dimension, limited to the kidney), while it is recommended to favour partial nephrectomy over radical nephrectomy in patients with T1b tumours (i.e. a tumour >4 cm but ≤7 cm maximum dimension, limited to the kidney), whenever feasible.[14] These conservative surgical approaches achieved similar oncological outcomes as compared with radical nephrectomy, but allowed the maintenance of an adequate renal function in a larger number of patients. Despite this, in patients with pre-existing kidney disease, especially the elderly and those with relevant cardiovascular comorbidities, the surgical excision (radical or partial) of a renal malignancy may contribute to the development of *de novo* kidney function impairment, or to the worsening of pre-existing chronic kidney disease.

Our improved knowledge of the molecular mechanisms underlying kidney carcinogenesis have led to the development of active systemic therapies, which ultimately improved the natural history of tumours in these patients, expecially when administered sequentially.[15] These novel agents include four VEGFR-TKIs (sorafenib, sunitinib, pazopanib, and axitinib), the anti-VEGF monoclonal antibody bevacizumab (which is given together with interferon-α [IFN-α]), as well as two mTOR inhibitors (everolimus and temsirolimus).[16] More recently, a further two agents have been registered (at least in many parts of the world): cabozantinib,[17] which is a VEGFR/C-Met inhibitor, and the anti-programmed death 1 checkpoint inhibitor, nivolumab.[18]

This review will focus on just the first seven agents, thus excluding cabozantinib and nivolumab; data for patients on dialysis treated with these agents are presently not available, due to their very recent implementation in clinical practice.

VASCULAR ENDOTHELIAL GROWTH FACTORS AND THEIR RECEPTORS' TYROSINE KINASE INHIBITORS TO TREAT METASTATIC RENAL CELL CARCINOMA

Sorafenib Tosylate

Originally identified as an inhibitor of Raf kinase, sorafenib proved to be endowed with a significant anti-angiogenic activity, characterised by the ability to inhibit, at pharmacological concentrations, all three VEGFRs (VEGFR-1, 2, and 3) along with platelet-derived growth factor receptor (PDGFR)-α and β, in addition to a number of other kinases during its pre-clinical development.[19] Sorafenib is administered orally at the fixed dose of 400 mg twice a day and has a safety profile characterised by a high incidence of fatigue, hypertension, hand–foot skin reaction (HFSR), hypothyroidism, and diarrhoea.[20]

The sorafenib registrative study was a placebo-controlled, Phase III trial comparing this multikinase inhibitor with placebo in patients with treatment-refractory (mainly cytokine-refractory) mRCC. Sorafenib almost doubled progression-free survival (PFS) (the primary endpoint of the study) compared with placebo (5.5 versus 2.8 months), a difference that was not only statistically significant, but also equivalent to a reduction in the risk of progression or death of 56%.[21] Sorafenib is metabolised in the liver by cytochrome CYP3A4; approximately 19% of the administrated dose is recovered in urine as metabolites.[22]

Sunitinib Malate

Sunitinib is an oral multikinase inhibitor selectively directed against all three VEGFRs (VEGFR-1, 2, and 3), against PDGFR-α and β, as well as against a range of other kinases.[23] From Phase I studies, the dose of 50 mg per day within a 4 weeks on,

2 weeks off schedule emerged as the one to be used in later stages of development,[23] although alternative schedules (expecially the 2 weeks on, 1 week off) have been recently proposed to alleviate its toxicity profile,[24] which is characterised mainly by hypertension, diarrhoea, myelotoxicity, skin toxicity (expecially HFSR), hypothyroidism, and fatigue.[25]

The pivotal sunitinib study was a randomised, controlled, Phase III trial, in which 750 treatment-naïve mRCC patients were randomised to receive either sunitinib or IFN-α (given subcutaneously at a loading dose of 9 MU 3-times per week), PFS being the primary endpoint of the study.[26] Median PFS in sunitinib-treated patients was significantly longer than in those treated with IFN-α (11 versus 5 months), corresponding to a reduction in the risk of progression or death of 58%. From a pharmacokinetic (PK) viewpoint, as with all VEGFR-TKIs sunitinib is metabolised by cytochrome CYP3A4, and renal eliminations account for 16% of the administrated dose.[22]

Bevacizumab

Bevacizumab is a recombinant humanised monoclonal antibody directed against VEGF; it is able to selectively bind and neutralise all active isoforms of VEGF (also known as VEGF-A), but not other members of the family of VEGF, i.e. VEGF-B, C, and D.[27] In mRCC, it is administered intravenously at a dose of 10 mg/kg every 2 weeks; its safety profile includes hypertension, proteinuria, wound healing impairment, an increased risk of haemorrhage, intestinal perforation, and thromboembolic events.[28] However, since in mRCC it is administered together with IFN-α, patients treated with this combination usually also experience IFN-α-related adverse events (AEs) such as fever and flu-like syndrome.

In the AVOREN pivotal trial,[29] the combination of bevacizumab plus IFN-α was compared with IFN-α plus placebo, overall survival (OS) being the primary endpoint of the study. The combination was approved based on preliminary results showing a significant benefit in terms of the secondary endpoint PFS (median 10.2 versus 5.4 months; hazard ratio [HR]: 0.63, equivalent to a reduction in the risk of progression or death of 37%). Surprisingly, OS did not differ between the two treatment arms, mainly due to the confounding role of subsequent active treatments.[30] As with all large molecular size monoclonal antibodies, bevacizumab is mainly metabolised by the reticuloendothelial system and has no renal excretion.[22]

Pazopanib

Pazopanib is another oral multikinase inhibitor capable of inhibiting the activation of different tyrosine kinases heavily implicated in the mechanisms of angiogenesis (mainly VEGFR-1, 2, and 3, but also PDGFR-α and β, and others).[31]

The recommended dose resulting from a Phase I study, which showed a correlation between plasma concentrations of pazopanib and development of hypertension, was 800 mg/day.[32] The safety profile of pazopanib is similar to that of sunitinib, but with a less detrimental effect on the quality of life of mRCC patients, as subsequently demonstrated by both the COMPARZ and PISCES studies;[33,34] compared with sunitinib, pazopanib induces more hepatic toxicity, but less myelotoxicity and HFSR.

Pazopanib's pivotal trial was conducted in a population of mRCC patients who were either treatment-naïve, or cytokine pre-treated. The study was placebo-controlled, and its primary endpoint was once again PFS.[35] A significant benefit in terms of PFS in favour of pazopanib was observed in both groups of patients, with a median PFS of 11.1 months in treatment-naïve patients (versus 2.8 months for placebo-treated subjects, hazard ratio [HR]: 0.4), and 7.4 months (versus 4.2, HR: 0.54) in cytokine pre-treated patients.[35] Like all VEGFR-TKIs, pazopanib is metabolised by cytochrome CYP3A4; renal eliminations are particularly low at <4% of the administered dose.[22]

Axitinib

Axitinib is a so-called third-generation VEGFR-TKI,[36] characterised by a particular selectivity of action for all three VEGFRs, and a high power. Commonly observed axitinib-related AEs include hypertension, diarrhoea, and fatigue.[37] The pivotal Phase III axitinib trial[38] was conducted in a second-line setting, in patients pre-treated with a variety of first-line treatment, and was the very first study on renal cell carcinoma which compared two active drugs head-to-head, sorafenib having been chosen as the control arm. In this study (AXIS study), axitinib proved to be superior in terms of PFS (primary endpoint of the study) to sorafenib; indeed, median PFS was 6.7 months with axitinib compared with 4.7 months with sorafenib, equivalent to a 33.5% reduction in the risk of progression.[38] The biggest advantage of axitinib over sorafenib was

evidenced in patients pre-treated with cytokines; however, when just sunitinib pre-treated patients were considered, both drugs performed quite well, axitinib maintaining an advantage over the older agent.[37,39] Again, as a VEGFR-TKI, axitinib is metabolised by cytochrome CYP3A4, with renal eliminations accounting for 23% of the administrated dose.[22]

MAMMALIAN TARGET OF RAPAMYCIN INHIBITORS TO TREAT METASTATIC RENAL CELL CARCINOMA

Temsirolimus

Temsirolimus, a water-soluble derivative of sirolimus, is a highly selective inhibitor of mTOR; binding the FKBP1 domain of mTOR, it inhibits kinase activity, preventing phosphorylation of substrate proteins such as 4E-BP1 and S6K1, and consequently blocking the cell cycle in the G1 phase.[40] Furthermore, inhibition of mTOR by temsirolimus leads to a suppression of various other proteins involved in the processes of angiogenesis, such as the hypoxia-inducible factor-1α, and ultimately also VEGF.[41] Temsirolimus-induced AEs include fatigue, stomatitis, anaemia, dyslipidaemia, hyperglycaemia, drug-induced pneumonitis, as well as an increased risk of infections.[42]

Its registrative study was a randomised, Phase III trial, aimed at investigating the efficacy of temsirolimus alone or in combination with IFN-α, compared with IFN-α alone in patients with poor prognosis features, according to the Memorial Sloan-Kettering Cancer Center (MSKCC) classification,[2] OS being the primary endpoint of the study. Treatment with temsirolimus was associated with a reduction in the risk of death by 27%, with an OS of 7.3 months in the group treated with IFN-α alone, 8.4 months in the group treated with the combination of the two drugs, and 10.9 months in the group treated with temsirolimus alone.[43] Temsirolimus is metabolised by cytochrome CYP3A4, with renal eliminations accounting for 4.6% of the administrated dose.[22]

Everolimus

Everolimus is another derivative of rapamycin, endowed with inhibitory activity on the mTOR, developed, unlike temsirolimus, as an oral medication.[44] Its safety profile is similar to that of temsirolimus, more common AEs being anaemia, hyperglycaemia, dyslipidaemia, stomatitis, drug-induced pneumonitis, and an increased risk of infection.[45] RECORD-1, everolimus' registrative Phase III trial was a randomised (2:1), placebo-controlled, Phase III study, in which mRCC patients who had failed treatment with sunitinib, sorafenib, or both, were enrolled; the majority of patients treated within this study had also failed other previous treatments, mainly (but not exclusively) cytokines.[46]

The RECORD-1 study showed, at an interim analysis, a statistically significant improvement in median PFS (primary endpoint of the study) in favour of everolimus. Indeed, median PFS was 4 months in the everolimus arm, and just 1.9 months in the placebo arm, with a percentage of patients free of progression at 6 months of 26% (compared with 2%), again in favour of everolimus.[46] Regarding OS, the high percentage of patients who crossed-over from the placebo to the active drug precluded any chance to observe a significant difference between the two arms, even though a subsequent statistical analysis, used to correct the estimate of the effect of treatment taking into account the bias generated by cross-over, showed an OS 1.9-times longer in favour of everolimus-treated patients.[47] Everolimus renal excretion accounts for just 2% of the administered dose.[22]

TARGETED THERAPIES FOR METASTATIC RENAL CELL CARCINOMA AND DIALYSIS

When treated for their cancer, patients with mRCC on dialysis are usually treated as patients without renal impairment, but data about the PKs of these drugs in this context are extremely scarce, at best. Our treatment decisions in this setting thus rely solely on small retrospective series, or even on single case reports. The following is a summary of current literature regarding VEGF/VGFR targeting agents and mTOR inhibitors.

Vascular Endothelial Growth Factor/Vascular Endothelial Growth Factor Receptor Inhibitors in Dialysis

Among VEGF/VEGFR-targeting agents, sorafenib and sunitinib are more frequently used in dialysis and more thoroughly described in the literature, being the very first agents to be registered for the treatment of mRCC, back in 2005/2006.

Kennoki et al.[48] examined PK parameters in patients on dialysis treated with sorafenib for mRCC. In this study, 10 patients received 200 mg of sorafenib once daily (i.e. one-quarter of the standard dose) as the initial dose. Regarding treatment activity, the authors observed one complete response, two partial responses, and disease stabilisation in four more patients. The median PFS and OS were 6.3 and 14.9 months, respectively. AEs were also collected and were generally serious; when the incidence of the AEs observed in patients on dialysis were compared with that of patients with normal renal function from the same institution, the authors found that the incidence of serious AEs was higher in patients on dialysis, the most common being hypertension, thrombocytopenia, and haemorrhagic events. They also reported a Grade 5 subarachnoid haemorrhage, and a Grade 4 cerebellar haemorrhage (in patients without brain metastasis). The PK study was performed in just six of these patients. The geometric mean of C_{max} (maximum level concentration of the day), C_{min} (minimum level concentration of the day), and AUC_{0-10} (area under the curve from 0–10 hours after taking 200 mg of sorafenib) on haemodialysis as compared with non-haemodialysis days was related to the objective responses observed and with the number of AEs of Grade 3 or higher; no significant relationship between the PK parameters and the occurrence of serious AEs, and between PK parameter and clinical efficacy was observed.[48] The authors therefore suggested that the higher incidence of sorafenib-related serious AEs in patients on dialysis is likely associated with the compromised general conditions of these patients, and not with the high plasma exposure of sorafenib. They concluded that sorafenib treatment is also effective in patients on dialysis, but suggested the use of lower sorafenib doses due to the particularly high incidence of AEs, especially cardiovascular ones.[48] This study was performed on Japanese mRCC patients, who, like all Asian populations, are known to poorly tolerate VEGFR-targeting agents.

A retrospective analysis conducted in several centres in the UK and USA in 2010, Josephs et al.[49] reported the outcome of sunitinib treatment in terms of both efficacy and safety. Nineteen patients were included, 10 of whom were undergoing haemodialysis. Of the nine nondialysis-dependent patients at drug initiation, the median estimated glomerular filtration rate was 27 mL/min/1.73 m^2 (range 23–29). The estimated median PFS of the whole cohort was 43 weeks (range 7 to >158), progression having not yet been reached in six patients at the time of publication. Partial response or stable disease was observed as best response in 15 patients, while the most common treatment-related AEs included fatigue, diarrhoea, HFSR, nausea/vomiting, and rash. Grade 3 treatment-related AEs including fatigue (seven patients), HFSR (two patients), diarrhoea (one patient), rash (one patient), and stomatitis (one patient) occurred in a total of 12 patients. Only one patient experienced a Grade 4 AE (HFSR). Diarrhoea, HFSR, and neutropenia were more common in patients undergoing haemodialysis compared with nondialysis-dependent patients. Three haemodialysis, and four nondialysis-dependent patients started at a dose of 50 mg; whereas five and two of the patients undergoing haemodialysis started at doses of 37.5 mg and 25 mg daily, respectively, compared with four and one of the nondialysis-dependent patients. Dose reductions during treatment were performed in eight patients but only one patient required complete discontinuation.[49]

Khosravan et al.[50] published data from a Phase I, open-label study aimed at evaluating PK data relative to sunitinib and its metabolite SU12662. Twenty-four patients were enrolled, eight for each of the following groups: normal renal function (creatinine clearance >80 mL/min/1.73 m^2), severe impairment renal function (creatinine clearance <30 mL/min/1.73 m^2), and ESRD requiring dialysis. PKs in subjects with severe renal impairment appeared similar to those with normal renal function. Indeed, plasma exposure to sunitinib and SU12662 was lower in subjects with ESRD compared with subjects with normal renal function or severe renal impairment. Notably, in haemodialysis patients, the incidence of AEs was quite low, with no drug discontinuations due to AEs. Considering the above data, the authors concluded that sunitinib, given at standard dose and schedule, seems to be an effective and safe option for patients with mRCC undergoing dialysis, yielding results in line with those observed in patients without renal impairment.[50]

In 2012 Masini et al.[51] reported a multicentre, retrospective study on 24 patients with mRCC and ESRD requiring haemodialysis treated with sorafenib (n=8) and sunitinib (n=16), aimed at retrospectively describing targeted agents' administered doses, treatment-related AEs, and clinical response. Sunitinib was administered at the dose of 50 mg daily, 4 weeks on, 2 weeks off, in six

patients; at 37.5 mg daily, 4 weeks on, 2 weeks off, in seven patients (one of them subsequently increased the dose to 50 mg daily); at 25 mg daily, 4 weeks on, 2 weeks off, in two patients; and finally at 12.5 mg daily, 4 weeks on, 2 weeks off, in one patient. Among the eight patients treated with sorafenib, four patients received 800 mg daily (400 mg twice daily), three patients 400 mg daily, and one patient 200 mg daily, all with a continuous schedule. The estimated median PFS and OS in this cohort of patients was 10.3 months and 22.6 months, respectively. With regard to tolerability and safety, no unexpected AEs were registered and no Grade 4 haematological or nonhaematological toxicities were reported. In this series, sunitinib and sorafenib proved to be not contraindicated in patients with mRCC undergoing dialysis, the outcome of this patient population being similar to that observed in patients with normal renal function treated with VEGFR-TKIs.[51]

In another retrospective study, Shetty et al.[52] reported the outcome of 14 mRCC patients on dialysis treated sequentially with different targeted therapies in the USA. The median number of targeted agents received per patient was three (range: 1–4), resulting in a median time on treatment (for all the agents used) of 28 months. Eighty-eight percent of all toxicities were Grade 1–2, without cases of Grade 4 AEs; treatment discontinuations included three patients treated with sorafenib (due to HFSR, intolerable fatigue, and squamous cell skin cancer development, respectively), two patients treated with pazopanib (due to intolerable fatigue and increased transaminase levels), and one patient treated with everolimus (due to non-bacterial pneumonitis).

Table 1: Summary of literature reports on the safety and efficacy of vascular endothelial growth factor/vascular endothelial growth factor receptor-targeting agents in metastatic renal cell cancer.

Author (year)	No. of patients	VEGFR/VEGFR-targeting agents	Dose reductions required	Toxicities observed (Grades ≥3)
Garnier-Viougeat et al.[54] (2006)	1	Bevacizumab*	No	None
Maroto Rey, Villavicencio[55] (2008)	2	Sunitinib (1 patient) Sorafenib (1 patient)	No Yes	None
Ruppin et al.[56] (2008)	1	Sorafenib	No	None
Zastrow et al.[57] (2009)	2	Sunitinib	No (1 patient), Yes (1 patient)	Increase in amylase and lipase (1 patient)
Ferraris et al.[58] (2009)	2	Sorafenib	No (1 patient), Yes (1 patient)	Asthenia, gastritis, dyspnoea (1 patient)
Hilger et al.[59] (2009)	2	Sorafenib	Yes (2 patients)	Not reported
Vickers et al.[60] (2009)	2	Sunitinib	Yes (1 patient), No (1 patient)	Hypothyroidism and fatigue (1 patient)
Park[61] (2009)	1	Sunitinib	No	None
Reckova et al.[62] (2009)	1	Sunitinib	Yes	Thrombocytopenia, hypertension, decreased LVEF
Izzedine et al.[63] (2009)	1	Sunitinib	No	None
Castagneto et al.[64] (2010)	1	Sorafenib	Yes	None
Shinsako et al.[65] (2010)	1	Sorafenib	No	None
Yoon et al.[66] (2010)	2	Sunitinib	No†	None
Park et al.[67] (2010)	6	Sunitinib	Yes	Mucositis, anorexia, fatigue
Khosravan et al.[50] (2010)	8	Sunitinib	No	None
Josephs et al.[49] (2011)	10	Sunitinib	Yes	Fatigue, stomatitis, HFSR, diarrhoea
Kennoki et al.[48] (2011)	10	Sorafenib	Yes	Subarachnoid and cerebellar haemorrhage
Casper et al.[68] (2011)	21	Sunitinib	Yes	Asthaenia, nausea, vomiting, diarrhoea, thrombocytopenia, hypertension, hypotension, decreased LVEF

Table 1 continued.

Author (year)	No. of patients	VEGFR/VEGFR-targeting agents	Dose reductions required	Toxicities observed (Grades ≥3)
Thiery-Vuillemin et al.[69] (2011)	1	Sunitinib	No	None
Masini et al.[51] (2012)	24	Sunitinib (16 patients), sorafenib (8 patients)	No	Nausea, diarrhoea, symptomatic cardiac ischaemia
Yildiz et al.[70] (2014)	2	Sunitinib	No	Acute pulmonary oedema, hypertension
Shetty et al.[52] (2014)	14	Sunitinib, sorafenib, pazopanib (plus everolimus and temsirolimus)‡	Yes (9 patients)	HFSR, fatigue, and squamous cell skin cancer leading to treatment discontinuation in 3 patients; intolerable fatigue and increased transaminases in 2 patients leading to pazopanib discontinuation
Czarnecka et al.[53] (2015)	9	Sunitinib (3 patients), sorafenib (4 patients), pazopanib (1 patient), (plus everolimus in 1 patient)§	Yes (1 patient)	Hypertension, anaemia, fatigue
Bersanelli et al.[71] (2016)	1	Pazopanib	Yes	Diarrhoea, hypertension

*Bevacizumab was administered as a single-agent at the dose of 5 mg/kg, every 14 days.
†An intermittent schedule was used from the beginning.
‡Authors analysed and reported different sequences of targeted agents.
§Two patients received also a second-line targeted agent.
VEGF: vascular endothelial growth factor; VEGFR: vascular endothelial growth factor receptor; LVEF: left ventricular ejection fraction; HFSR: hand–foot skin reaction.

Median OS from initiation of targeted therapies and from time of diagnosis were 28.5 and 35 months, respectively. Once again, the authors concluded that targeted agents were safe, well tolerated, and able to produce an anti-tumour response in patients with mRCC and ESRD receiving dialysis, at the expense of mild-to-moderate AEs consistent with those reported in previous studies conducted in patients not on dialysis.[52]

More recently, Czarnecka et al.[53] retrospectively analysed a large number of consecutive mRCC patients treated with VEGFR-TKIs. Out of a total of 679 patients, 464 (i.e. 68%) were treated with VEGFR-TKIs, and among those just 9 (1.3 and 1.9%, respectively) were treated while on dialysis due to ESRD; 5 of these 9 patients were treated with sunitinib, 3 with sorafenib, and 1 with pazopanib. After first-line treatment, two of them received second-line therapy. PFS of this cohort was within the range reported in the literature for a typical mRCC patient population not on dialysis, i.e. 8–8.5 months. A partial response or a disease stabilisation was observed in one and five patients, respectively; with regard to safety, most AEs were Grade 1 or 2, with no Grade 5 AEs observed. For one patient the dose was decreased, and for another treatment was discontinued; this patient was already hypertensive at the start of treatment. As a whole, the results supported the authors' statement that VEGFR-TKIs treatment in dialysis is safe and effective.[53]

All the above series, together with other case reports and small series published in the literature[54-71] are summarised in Table 1, ultimately supporting the concept that dialysis patients do not differ dramatically from a typical population of mRCC in terms of activity and safety of VEGF or VEGFR-targeting agents.

MAMMALIAN TARGET OF RAPAMYCIN INHIBITORS IN DIALYSIS

In 2008/2009, Lunardi[72,73] demonstrated that temsirolimus concentration assessed immediately before haemodialysis was similar to that assayed 1 hour after the treatment, without unexpected or severe AEs.

Table 2: Summary of literature reports on the safety and efficacy of mammalian target of rapamycin inhibitors in metastatic renal cell carcinoma.

Author (year)	No. of patients	VEGFR/VEGFR-targeting agents	Dose reductions required	Toxicities observed (Grade ≥3)
Lunardi et al.[72,73] (2008 and 2009)	2	Temsirolimus	No	None
Thiery-Vuillemin et al.[74] (2012)	2	Everolimus	Yes*	Asthaenia, hyperglycaemia
Guida et al.[75] (2015)	11	Everolimus	Yes	Thrombocytopenia, anaemia, skin rash, dyspnoea, and non-bacterial pneumonitis
Syrios et al.[76] (2013)	2	Everolimus	No	None
Miyake et al.[77] (2013)	10	Temsirolimus	No	Asthaenia, anaemia (1 patient each), thrombocytopenia (2 patients)
Shetty et al.[52] (2014)	9	Everolimus temsirolimus (plus sunitinib, sorafenib, and pazopanib)†	Yes (1 patient)‡	1 patient discontinued everolimus due to pneumonitis
Omae et al.[78] (2016)	4	Everolimus (4 patients) Temsirolimus (2 patients)	No (3 patients), yes (1 patient) No (1 patient), yes (1 patient)	None

*Everolimus starting dose was 5 mg/day with the possibility of escalation according to the tolerance after the first PK assessment; patient n=1 experienced escalation to 10 mg/day, but required dose reduction to 5 mg/day due to Grade 3 AEs (asthaenia).
†Authors analysed and reported different sequences of targeted agents.
‡Treated with everolimus.
VEGF: vascular endothelial growth factor; VEGFR: vascular endothelial growth factor receptor; AEs: adverse events.

In 2012, Thiery-Vuillemin et al.[74] for the first time reported PK data relative to everolimus treatment during haemodialysis in two mRCC patients pre-treated with sunitinib; in these two patients everolimus was administered at the reduced dose of 5 mg daily. From a PK viewpoint, dialysis did not modify everolimus blood concentrations, as they were close to the predialysis level; moreover, no everolimus was detected in the dialysate, confirming its lack of adhesion to the dialysis membrane.[74]

In 2012, Guida et al.[75] retrospectively collected data on 11 mRCC patients treated with everolimus. Everolimus was administered at the dose of 10 mg daily in 10 patients, and at the reduced dose of 5 mg daily in 1 patient only. Only five Grade 3 AEs were reported: thrombocytopenia, anaemia, cutaneous rash, dyspnoea, and non-bacterial pneumonitis (in the same patient). In this cohort of patients, the estimated PFS was 9 months, while estimated median OS was 15.7 months.[75]

In the already mentioned study from Shetty et al.[52] six patients were treated with everolimus, all at the dose of 10 mg daily. Four everolimus-related AEs were reported, only one severe (Grade 3). One patient discontinued everolimus due to pneumonitis, but overall the median duration of treatment was just 1.9 months.[52] Taking into account a few other cases reported in the literature[76-78] and summarised in Table 2, mTOR inhibitors' efficacy and safety does not significantly differ in mRCC patients on dialysis from those observed in non-dialytic patients.

CONCLUSION

One of the more challenging areas of onco-nephrology is the appropriate management of cancer patients requiring dialysis. Beyond the ethical aspect related to the choice of whether or not to initiate an active oncological treatment in a patient on dialysis (or vice versa),[79]

to date decisions about anti-cancer drug choices and dosing are too often not supported by PK or pharmacodynamic data, making therapeutic decisions extremely difficult.[80] An accurate understanding of the effects of dialysis on general drug clearance (e.g. volume of distribution, protein binding, and molecular size) is mandatory to reasonably estimate the safety of anti-cancer drugs in patients on dialysis.[80]

The above review of the scarce literature available could be useful to drive our everyday clinical decisions in a very complicated patient population such as that of patients undergoing dialysis. Although prospective studies would definitely be warranted in specific patient populations, such as those with chronic kidney disease and on dialysis, all the available data suggest that mRCC patients on dialysis have similar outcomes, both in terms of efficacy and safety, as mRCC patients with normal or marginally impaired kidney function, when treated with VEGF/VEGFR-targeting agents and/or mTOR inhibitors.

REFERENCES

1. Iacovelli R et al. Inhibition of the VEGF/VEGFR pathway improves survival in advanced kidney cancer: a systematic review and meta-analysis. Curr Drug Targets. 2015;16:164-70.

2. Motzer RJ et al. Survival and prognostic stratification of 670 patients with advanced renal cell carcinoma. J Clin Oncol. 1999;17(8):2530-40.

3. Heng DY et al. External validation and comparison with other models of the International Metastatic Renal-Cell Carcinoma Database Consortium prognostic model: a population-based study. Lancet Oncol. 2013;14(2):141-8.

4. Heng DY et al. Outcomes of patients with metastatic renal cell carcinoma that do not meet eligibility criteria for clinical trials. Ann Oncol. 2014;25(1):149-54.

5. Rawlins MD. The Harveian oration of 2008: De testimonio. On the evidence for decisions about the use of therapeutic interventions. Royal College of Physicians. Lancet. 2008;372:2152-61.

6. Holley JL. Screening, diagnosis, and treatment of cancer in long-term dialysis patients. Clin J Am Soc Nephrol. 2007;2(3):604-10.

7. Truong LD et al. Renal neoplasm in acquired cystic kidney disease. Am J Kidney Dis. 1995;26(1):1-12.

8. Choyke PL. Acquired cystic kidney disease. Eur Radiol. 2000;10(11):1716-21.

9. Janus N et al. Management of anticancer treatment in patients under chronic dialysis: results of the multicentric CANDY (CANcer and DialYsis) study. Ann Oncol. 2013;24(2):501-7.

10. Lassalle M et al. The essential of 2012 results from the French Renal Epidemiology and Information Network (REIN) ESRD registry. Nephrol Ther. 2015; 11:78-87.

11. Vogelzang JL et al. Mortality from infections and malignancies in patients treated with renal replacement therapy: data from the ERA-EDTA registry. Nephrol Dial Transplant. 2015;30(6):1028-37.

12. Rini BI et al. Renal cell carcinoma. Lancet. 2009;373(9669):1119-32.

13. Bratslavsky G, Linehan WM. Long-term management of bilateral, multifocal, recurrent renal carcinoma. Nat Rev Urol. 2010;7(5):267-7.

14. Ljungberg B et al. EAU guidelines on renal cell carcinoma: 2014 update. Eur Urol. 2015;67(5):913-24.

15. Albiges L et al. A systematic review of sequencing and combinations of systemic therapy in metastatic renal cancer. Eur Urol. 2015;67:100-10.

16. Bex A et al. Challenging the treatment paradigm for advanced renal cell carcinoma: a review of systemic and localized therapies. Am Soc Clin Oncol Educ Book. 2015:e239-47.

17. Choueiri TK et al. Cabozantinib versus Everolimus in advanced renal-cell carcinoma. N Engl J Med. 2015;373(19): 1814-23.

18. Motzer RJ et al. Nivolumab versus Everolimus in advanced renal-cell carcinoma. N Engl J Med. 2015;373(19): 1803-13.

19. Wilhelm SM et al. BAY 43-9006 exhibits broad spectrum oral antitumor activity and targets the RAF/MEK/ERK pathway and receptor tyrosine kinases involved in tumor progression and angiogenesis. Cancer Res. 2004;64(19):7099-109.

20. Porta C et al. Sorafenib tosylate in advanced kidney cancer: past, present and future. Anticancer Drugs. 2009;20(6):409-15.

21. Escudier B et al. Sorafenib in advanced clear-cell renal-cell carcinoma. N Engl J Med. 2007;356(2):125-34.

22. Porta C et al. Renal effects of targeted anticancer therapies. Nat Rev Nephrol. 2015;11(6):354-70.

23. Chow LQ, Eckhardt SG. Sunitinib: from rational design to clinical efficacy. J Clin Oncol. 2007;25(7):884-96.

24. Bracarda S et al. Sunitinib administered on 2/1 schedule in patients with metastatic renal cell carcinoma: the RAINBOW analysis. Ann Oncol. 2016;27(2):366.

25. Porta C et al. Long-term safety of Sunitinib in metastatic Renal Cell Carcinoma. Eur Urol. 2016;69(2):345-51.

26. Motzer RJ et al. Sunitinib versus interferon alfa in metastatic renal-cell carcinoma. N Engl J Med. 2007;356(2): 115-24.

27. Ferrara N et al. Discovery and development of bevacizumab, an anti-VEGF antibody for treating cancer. Nat Rev Drug Discov. 2004;3(5):391-400.

28. Sanborn RE, Sandler AB. The safety of bevacizumab. Expert Opin Drug Saf. 2006;5(2):289-301.

29. Escudier B et al. Bevacizumab plus interferon alfa-2a for treatment of metastatic renal cell carcinoma: a randomised, double-blind phase III trial. Lancet. 2007;370(9605):2103-11.

30. Bracarda S et al. Overall survival in patients with metastatic renal cell carcinoma initially treated with bevacizumab plus interferon-α2a and subsequent therapy with tyrosine kinase inhibitors: a retrospective analysis of the phase III AVOREN trial. BJU Int. 2011;107(2):214-9.

31. Kumar R et al. Pharmacokinetic-pharmacodynamic correlation from mouse to human with pazopanib, a multikinase angiogenesis inhibitor with potent antitumor and antiangiogenic activity. Mol Cancer Ther. 2007;6(7): 2012-21.

32. Hurwitz HI et al. Phase I trial of pazopanib in patients with advanced cancer. Clin Cancer Res. 2009;15(12): 4220-7.

33. Motzer RJ et al. Pazopanib versus sunitinib in metastatic renal-cell carcinoma. N Engl J Med. 2013;369(8): 722-31.

34. Escudier B et al. Randomized, controlled, double-blind, cross-over trial assessing treatment preference for pazopanib versus sunitinib in patients with

metastatic renal cell carcinoma: PISCES Study. J Clin Oncol. 2014;32(14):1412-8.

35. Sternberg CN et al. Pazopanib in locally advanced or metastatic renal cell carcinoma: results of a randomized phase III trial. J Clin Oncol. 2010;28(6):1061-8.

36. Bukowski RM. Third generation tyrosine kinase inhibitors and their development in advanced renal cell carcinoma. Front Oncol. 2012;2:13.

37. Gunnarsson O et al. Evaluating the safety and efficacy of axitinib in the treatment of advanced renal cell carcinoma. Cancer Manag Res. 2015;7: 65-73.

38. Rini BI et al. Comparative effectiveness of axitinib versus sorafenib in advanced renal cell carcinoma (AXIS): a randomised phase 3 trial. Lancet. 2011;378(9807): 1931-9.

39. Escudier B et al. Axitinib versus sorafenib in advanced renal cell carcinoma: subanalyses by prior therapy from a randomised phase III trial. Br J Cancer. 2014;110(12):2821-8.

40. Dudkin L et al. Biochemical correlates of mTOR inhibition by the rapamycin ester CCI-779 and tumor growth inhibition. Clin Cancer Res. 2001;7(6):1758-64.

41. Del Bufalo D et al. Antiangiogenic potential of the Mammalian target of rapamycin inhibitor temsirolimus. Cancer Res. 2006;66(11):5549-54.

42. Bhojani N et al. Toxicities associated with the administration of sorafenib, sunitinib, and temsirolimus and their management in patients with metastatic renal cell carcinoma. Eur Urol. 2008;53(5):917-30.

43. Hudes G et al. Temsirolimus, interferon alfa, or both for advanced renal-cell carcinoma. N Engl J Med. 2007;356(22):2271-81.

44. Tanaka C et al. Identifying optimal biologic doses of everolimus (RAD001) in patients with cancer based on the modeling of preclinical and clinical pharmacokinetic and pharmacodynamic data. J Clin Oncol. 2008;26:1596-602.

45. Porta C et al. Management of adverse events associated with the use of everolimus in patients with advanced renal cell carcinoma. Eur J Cancer. 2011; 47(9):1287-98.

46. Motzer RJ et al. Efficacy of everolimus in advanced renal cell carcinoma: a double-blind, randomised, placebo-controlled phase III trial. Lancet. 2008; 372(9637):449-56.

47. Motzer RJ et al. Phase 3 trial of everolimus for metastatic renal cell carcinoma: final results and analysis of prognostic factors. Cancer. 2010; 116(18):4256-65.

48. Kennoki T et al. Clinical results and pharmacokinetics of sorafenib in chronic hemodialysis patients with metastatic renal cell carcinoma in a single center. Jpn J Clin Oncol. 2011;41(5):647-55.

49. Josephs D et al. Efficacy and toxicity of sunitinib in patients with metastatic renal cell carcinoma with severe renal impairment or on haemodialysis. BJU Int. 2011;108(8):1279-83.

50. Khosravan R et al. Pharmacokinetics and safety of sunitinib malate in subjects with impaired renal function. J Clin Pharmacol. 2010;50(4):472-81.

51. Masini C et al. Use of tyrosine kinase inhibitors in patients with metastatic kidney cancer receiving haemodialysis: a retrospective Italian survey. BJU Int. 2012;110(5):692-8.

52. Shetty AV et al. Outcomes of patients with metastatic renal cell carcinoma and end-stage renal disease receiving dialysis and targeted therapies: a single institution experience. Clin Genitourin Cancer. 2014;12(5):348-53.

53. Czarnecka AM et al. Feasibility, efficacy and safety of tyrosine kinase inhibitor treatment in hemodialyzed patients with renal cell cancer: 10 years of experience. Future Oncol. 2015;11(16):2267-82.

54. Garnier-Viougeat N et al. Pharmacokinetics of bevacizumab in haemodialysis. Nephrol Dial Transplant. 2007;22(3):975.

55. Maroto Rey P, Villavicencio H. Sorafenib: tolerance in patients on chronic hemodialysis: a single-center experience. Oncology. 2008;74(3-4):245-6.

56. Ruppin S et al. Successful sorafenib treatment for metastatic renal cell carcinoma in a case with chronic renal failure. Eur Urol. 2009;55(4):986-8.

57. Zastrow S et al. Treatment of metastatic renal cell cancer with sunitinib during chronic hemodialysis. Urology. 2009; 73(4):868-70.

58. Ferraris E et al. Use of sorafenib in two metastatic renal cell cancer patients with end-stage renal impairment undergoing replacement hemodialysis. Tumori. 2009; 95(4):542-4.

59. Hilger RA et al. Pharmacokinetics of sorafenib in patients with renal impairment undergoing hemodialysis. Int J Clin Pharmacol Ther. 2009;47:61-4.

60. Vickers MM et al. Tolerance of sunitinib in dialyzed patients with metastatic renal cell carcinoma. Clin Genitourin Cancer. 2009;7(3):E104-6.

61. Park CY. Successful sunitinib treatment of metastatic renal cell carcinoma in a patient with end stage renal disease on hemodialysis. Anticancer Drugs. 2009;20(9):848-9.

62. Reckova M et al. Treatment of hemodialyzed patient with sunitinib. Ann Oncol. 2009;20(2):392-3.

63. Izzedine H et al. Pharmacokinetics of sunitinib in hemodialysis. Ann Oncol. 2009;20(1):190-2.

64. Castagneto B et al. Sustained response following sorafenib therapy in an older adult patient with advanced renal cancer on hemodialysis: a case report. Med Oncol. 2011;28(4):1384-8.

65. Shinsako K et al. Tolerable sorafenib therapy for a renal cell carcinoma patient with hemodialysis: a case study. Int J Clin Oncol. 2010;15(5):512-4.

66. Yoon SH et al. Novel sunitinib strategy in metastatic renal cell carcinoma on hemodialysis: intermittent dose of sunitinib after hemodialysis. Cancer Res Treat. 2010;42(3):180-4.

67. Park S et al. Treatment of hemodialyzed patients with sunitinib in renal cell carcinoma. Chemotherapy. 2010;56: 485-91.

68. Casper J et al. Efficacy and safety of sunitinib in patients with metastatic renal cell carcinoma on hemodialysis. Abstract 4646. ASCO Annual Meeting 2011, Chicago, Illinois, USA. June 3-7.

69. Thiery-Vuillemin A et al. Impact of sunitinib pharmacokinetic monitoring in a patient with metastatic renal cell carcinoma undergoing hemodialysis. Ann Oncol. 2011;22(9):2152-4.

70. Yildiz I et al. Intolerance to sunitinib treatment in hemodialysis patients with metastatic renal cell carcinoma. Korean J Urol. 2014;55(1):74-6.

71. Bersanelli M et al. Pazopanib in Renal Cell Carcinoma dialysis patients: a mini-review and a case report. Curr Drug Targets. 2016. [Epub ahead of print].

72. Lunardi G et al. Temsirolimus in patients with renal cancer on hemodialysis. J Clin Oncol. 2008;26(34):5652-3.

73. Lunardi G et al. Comparison of temsirolimus pharmacokinetics in patients with renal cell carcinoma not receiving dialysis and those receiving hemodialysis: a case series. Clin Ther 2009;31(8):1812-9.

74. Thiery-Vuillemin A et al. Hemodialysis does not affect everolimus pharmacokinetics: two cases of patients with metastatic renal cell cancer. Ann Oncol. 2012;23(11):2992-3.

75. Guida A et al. Retrospective analysis on safety and efficacy of everolimus in treatment of metastatic renal cancer patients receiving dialysis. Future Oncol. 2015;11(23):3159-66.

76. Syrios J et al. Treatment of patients with metastatic renal cell carcinoma undergoing hemodialysis: case report of two patients and short literature review. BMC Nephrol. 2013;14:84.

77. Miyake H et al. Efficacy and safety of temsirolimus in Japanese patients with metastatic renal cell carcinoma on hemodialysis. Int J Clin Oncol. 2013;18(6):1054-9.

78. Omae K et al. Use of mammalian target of rapamycin inhibitors after failure of tyrosine kinase inhibitors in patients with metastatic renal cell carcinoma undergoing hemodialysis: A single-center experience with four cases. Hemodial Int. 2016. [Epub ahead of print].

79. Berman N. End-of-life matters in chronic renal failure. Curr Opin Support Palliat Care. 2014;8(4):371-7.

80. Cosmai L et al. Onco-nephrology: a decalogue. Nephrol Dial Transplant. 2015. [Epub ahead of print].

URIC ACID IN CHRONIC KIDNEY DISEASE: A CLINICAL APPRAISAL

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ABSTRACT

A consistent body of evidence supports an independent association between uric acid (UA) level and the risk of chronic kidney disease (CKD) in humans. It has been observed in experimental data that UA is capable of inducing renal damage through several pathways, including activation of the renin-angiotensin-aldosterone system (RAAS), oxidative stress, and inflammation. Treatment with urate lowering agents and RAAS inhibitors prevented renal insult mediated by UA in animal models. Both of the xanthine oxidase inhibitors available in clinical practice, allopurinol and febuxostat, were efficient in controlling gout flares. However, data from randomised controlled trials are still inconsistent in relation to their benefit for slowing CKD progression. This review discusses the metabolism of urates in humans as well as the experimental and clinical evidence linking UA to CKD. Current evidence about the effect of allopurinol and febuxostat on CKD progression is also considered.

Keywords: Uric acid (UA), chronic kidney disease (CKD), allopurinol, febuxostat, humans.

INTRODUCTION

Stepwise mutations of the uricase promoter gene occurred in lesser and great apes between 9 and 15 million years ago. Uricase silencing led to an inability in hominoids to convert uric acid (UA) into allantoin, which is disposable via renal excretion; this led to higher circulating levels of UA compared with their ancestors.[1] Increased UA concentration, induced by poor nutritional salt intake, may have been adjuvant in the tertiary development of bipedal locomotion. The maintenance of high blood pressure through activation of the renin-angiotensin-aldosterone system (RAAS), afferent arteriosclerosis, and increased salt sensitivity are suggested as the main favourable adaptive responses offered by uricase silencing during the Miocene period.[2] The contemporary Western diet, which is richer in proteins, alcohol, and fructose, compared with the presumed nutritional profile of the Quaternary period, may have led to an excessive load of UA with potential disadvantageous effects thereafter (a maladaptive response).[3]

Gout represents the traditional consequence of UA excess in contemporary subjects as they are predisposed to UA deposition in joints and soft tissues. Several trials have demonstrated how lowering UA synthesis by the administration of xanthine oxidase inhibitors was effective in reducing the incidence of gout flares.[4-8] More recently, observational studies have linked UA to a variety of unfavourable outcomes, such as cardiovascular disease,[9,10] hypertension,[11,12] metabolic syndrome,[13] diabetes,[14] mortality,[10,15] and chronic kidney disease (CKD).[16] This review will consider the metabolism of UA in humans and the experimental and clinical evidences linking UA to CKD. The discussion will also cover the potential benefits offered by xanthine oxidase inhibitors on CKD progression.

URIC ACID METABOLISM IN HUMANS

UA is the final enzymatic end product of purine metabolism in humans. UA is derived from the endogenous degradation (500–600 mg/day) of adenosine triphosphate, nucleic acids, and amino acids, as well as from nutrients (100–200 mg/day) rich in proteins, alcohol, and fructose.[3,17] Transformation of xanthine into UA by xanthine oxidase represents the last reaction of the purine catabolic pathway. The synthesis of UA mainly occurs in the liver and intestine, with less production observed in peripheral tissues such as the muscle, the kidney, and the endothelium.[18] UA is thereafter excreted in urine (70%), and in biliary fluid to a lesser extent (30%).[17,18] Almost all of the circulating UA is filtered by glomeruli. Although the handling of UA in the proximal convoluted tubule (PCT) is far more complex, including pre-secretory reabsorption, secretion, and post-secretory reabsorption in the first, second, and third segment of PCT, respectively,[17] it is accepted that the PCT reabsorbs almost 90% of the filtered UA,[19] principally through the action of the urate anion transporter (URAT) 1, glucose transporter (GLUT) 9, and organic anion transporters 1 and 3.[17,20] Notably, GLUT and URAT are inhibited by probenecid and benzbromarone,[17] while losartan and angiotensin II receptor blockers can inhibit URAT1.[17,21]

Circulating UA exists as a urate anion at the physiological pH due to its pK_a of 5.75, while it is present in urine at pH of 5–6 in its acidic form.[20] The solubility limit of circulating UA corresponds to 6.8 mg/dL at 37°C and decreases at colder temperatures, reaching 4.5 mg/dL at 30°C. Hyperuricaemia is variably defined as serum UA levels >6.8 mg/dL or 7.0 mg/dL in adults,[22,23] while guidelines for gout management recommend to lower serum UA below 6 mg/dL.[24]

URIC ACID AND CHRONIC KIDNEY DISEASE PROGRESSION: MECHANISMS

The solubility of UA in urine is highly dependent on several factors beyond uricosuria. This also exposes individuals with normal renal excretion of urates to the risk of renal stones, irrespective of concomitant predisposing conditions such as persistently low urinary pH or reduced urine volume. Although UA nephrolithiasis represents the most known cause of renal disease, a recent body of evidence was reported on how UA overload may induce asymptomatic kidney damage through crystal independent pathways including oxidative stress, activation of RAAS, reduced synthesis of nitric oxide, and inflammation.[25]

Hyperuricaemic rats developed hypertension and ischaemic renal injury independent of urate crystal deposition.[26] Notably, hyperuricaemia was associated with increased renal vascular resistance and reduced tubular excretion of sodium, mediated by increased synthesis of renin and a reduced synthesis of nitric oxide.[26] Hypertension was consequently prevented by the administration of allopurinol, enalapril, and L-arginine.[26] Arteriolopathy of the afferent glomerular vessels improved after administration of enalapril but not after administration of hydrochlorothiazide despite a similar blood pressure control, revealing that UA induced renal vascular damage independently from the hypertensive mechanism.[27] Furthermore, correcting high UA levels through the administration of febuxostat improved afferent arteriolar morphology and tubulointerstitial fibrosis, reduced proteinuria, and preserved renal function in rats with CKD.[28]

Apoptosis of proximal tubular cells occurred under exposition to high UA levels *in vitro,* due to its pro-oxidant effects mediated by the upregulation of nicotinamide adenine dinucleotide phosphate oxidase and *NOX-4*.[29] Apoptotic events were prevented by the addition of URAT1 blockers (probenecid and losartan), showing that UA may elicit oxidative effects at an intracellular level.[29] Furthermore, repeated experimental data has demonstrated how UA may induce renal damage through the activation of proinflammatory cascades, mediated by tumour necrosis factor alpha, cyclo-oxygenase, and inflammatory transcription factor nuclear factor kappa B.[30] This hypothesis was confirmed by the renal protective effect elicited by allopurinol in diabetic rats, resulting from the inhibition of an inflammatory cascade triggered by intracellular adhesion molecule 1 in tubular epithelial cells.[31]

UA may also contribute to CKD progression through indirect processes. UA has been proposed as a promoter of the cardiorenal metabolic syndrome, which is currently defined as an interactive milieu of risk factors for CKD and cardiovascular disease, including insulin resistance, obesity, dyslipidaemia, hypertension, and diastolic dysfunction.[25] Repeated experimental data raised evidence on how UA may induce the synthesis

of lipids and insulin resistance, both of which are involved in the onset of metabolic syndrome, resulting in a higher risk of CKD.[32] UA-dependent intracellular and mitochondrial oxidative stress resulted in a relevant induction of hepatic lipogenesis.[33] UA-induced oxidative and inflammatory reactions in the adipocytes of fructose-fed rats, leading to insulin resistance and an unbalanced leptin–adiponectin ratio, were improved by allopurinol administration.[34,35] Oxidative stress was reported in insulin-secreting islet cells exposed to a high concentration of UA, raising the risk of islet dysfunction.[36] Furthermore, normalisation of circulating UA levels with febuxostat reduced renal vasoconstriction and afferent arteriolar area, and improved insulin sensitivity as well as blood and glomerular pressure in rats affected by metabolic syndrome.[32,37]

Xanthine oxidase inhibitors have also been suggested as the cause of organ damage attributed to high UA levels.[18,32] The pro-oxidant effects of xanthine oxidase are well known; however, no studies have investigated the effect of xanthine oxidase on CKD progression independent of the action of UA in humans.

URIC ACID AND CHRONIC KIDNEY DISEASE: OBSERVATIONAL DATA

The association between UA and the risk of CKD has been repeatedly investigated in observational studies.[16,18] A recent meta-analysis by Li et al.[18] reported a significant association between hyperuricaemia and new onset CKD in five studies (odds ratio [OR]: 2.11; 95% confidence interval [CI]: 1.70–2.619). A 6% higher risk of incident CKD was observed for each 1 mg/dL increase of UA (OR: 1.06; 95% CI: 1.04–1.08).[18] Subgroup analysis revealed a slightly stronger association between UA levels and CKD in Western populations compared with Asian populations, potentially accounted for by the higher purine intake in the Occidental compared with the Eastern diet.[18] The risk of CKD progression was also addressed in a large cohort of volunteers in California over a median follow-up of 25.7 years.[38] Subjects in the fourth quartile of UA levels (6.00–14.90 mg/dL) had a 2-fold increased risk of requiring dialysis or renal transplantation compared with those in the lowest quartile (0.10–4.17 mg/dL; hazard ratio [HR]: 2.15; 95% CI: 1.65–2.77).[38] More recently, UA levels >6.5 mg/dL were retrospectively associated with the risk of progression to end-stage renal disease (ESRD) among 803 CKD patients (HR: 3.39; 95% CI: 1.55–7.42).[39] Similar results, but with a significant gender interaction, were reported among 48,177 adults in Okinawa.[40] Hyperuricaemic women (UA levels >6 mg/dL) presented an independent higher risk of progression toward ESRD compared with normouricaemic women (HR: 5.77; 95% CI: 2.31–14.21).[40] In the multivariate Cox analysis in men, hyperuricaemia was not a significant risk factor for ESRD (UA levels >7.0 mg/dL),[40] however data concerning differences in UA toxicity between males and females remains conflicted. The prevalence of hyperuricaemia and gout were higher in males[41,42] and several authors reported a greater association between UA and CKD in men.[43,44] Furthermore, the meta-analysis by Li et al.[18] showed a descriptively higher risk of CKD for each 1 mg/dL increase of UA in males (OR: 1.43; 95% CI: 1.05–1.94) compared with females (OR: 1.21; 95% CI: 1.04–1.41).

The aforementioned link between high UA levels and CKD is not universally confirmed. In the Mild to Moderate Kidney Disease Study (MMKD), the association between UA levels and progression of CKD, defined as the doubling of serum creatinine or dialysis need, lost significance after adjustment for basal kidney function.[45] A *post hoc* analysis of the Modification of Diet in Renal Disease (MDRD) study did not confirm any association between UA levels and the risk of kidney failure in Stage 3–4 CKD patients.[46] Several factors accounting for this negative result were proposed by the authors, such as the enrolment of patients with considerable risk factors for CKD progression, the presence of patients randomised to the low blood pressure group with consequent delayed kidney failure potentially mediated by strict blood pressure control and, thirdly, the adjustment for measured glomerular filtration rate (GFR).[46] Thus, several authors have raised the hypothesis that UA should be taken as a marker of reduced GFR rather than a potential effector of renal damage, accounting for the inconsistent association between UA and CKD progression after adequate adjustments for GFR.[18] More recently, a polymorphism of GLUT9 was strongly associated with serum UA levels and with a significant risk of CKD progression.[47]

A negative association between UA and CKD has also been reported. In a retrospective cohort of 94,422 patients in Taiwan, a minority of 50 patients with UA levels <2.0 mg/dL presented a higher risk of CKD progression.[48] More recently,

Kanda et al.[49] showed a U-shaped association between UA level and loss of kidney function in healthy subjects.[49] Oxidative stress induced by renal hypouricaemia was suggested as a potential mechanism underlying the loss of GFR due to UA levels <2.0 mg/dL. Although the real impact of hypouricaemia on renal function remains unknown, these preliminary data raise concerns about the adequacy of a 'the lower the better' approach to UA targets in CKD.

XANTHINE OXIDASE INHIBITORS: OVERVIEW AND LESSON FROM GOUTY PATIENTS

Two classes of drugs are currently available to reduce UA levels: xanthine oxidase inhibitors (allopurinol and febuxostat) and uricosuric agents (probenecid, benzbromarone, and sulfinpyrazone).

Allopurinol works as a purine substrate for xanthine oxidase, blocking the synthesis of xanthine and UA.[50] Allopurinol is thereafter hydroxylated by xanthine oxidase to its active catabolite oxypurinol, which is considered more active than allopurinol as it is more stable, more easily available, and has a longer half-life.[51] Allopurinol is also transformed into other nucleotide derivatives by hypoxanthine-guanine phosphoribosyltransferase and orotate phosphoribosyltransferase.[50] Allopurinol and its metabolites are then capable of inhibiting T cell function, which may result in allopurinol hypersensitivity syndrome.[52] Furthermore, allopurinol and oxypurinol accumulation may lead to tissue injury with secondary immune reaction, induced by antigen release.[53] Metabolism of allopurinol is based on hepatic conversion to oxypurinol, which is then excreted in urine. Renal patients are thus exposed to a higher risk of allopurinol toxicity, requiring proper dose adjustments.[50] Although the approved dose of allopurinol ranges from 50–800 mg, the mean delivered dose corresponds to 300 mg,[54] with even lower dosages in renal patients leading to a risk of inadequate efficacy.[55]

Febuxostat is differentiated from allopurinol by several characteristics. The non-purine based structure of febuxostat delivers selective inhibition of both the reduced and the oxidative form of xanthine oxidase.[50] Febuxostat is metabolised by liver enzymes to acyl-glucuronide, generating only a minimal amount of oxidative metabolites. Compared with allopurinol, febuxostat provides a stronger and quicker reduction of UA levels without exposing renal patients to drug accumulation.[50]

Trials that have compared allopurinol and febuxostat in patients with gout have provided significant outcomes.[4-8] Although febuxostat lowered UA levels in a greater proportion of patients and to a greater extent than allopurinol, none of the trials demonstrated a significant difference in the incidence of gout flare and tophi reduction between the treatment arms.[4-8] Notably, the absolute risk of gout flares was highly sensitive to the length of follow-up, reaching a rate of flares close to zero after 40 months of treatment.[6,7] Urate lowering therapy was followed by a transitory increase of gout flares, especially in patients receiving febuxostat at high dosage;[4-8] this inspired an experimental extension of prophylaxis with low dose colchicine ≤6 months in the more recent CONFIRMS trial.[8] Liver function test abnormalities were a common cause of drug discontinuation; the few cardiovascular events were not attributed to the study drug.[4-8] Notably, the APEX trial included a minority of 35 patients with serum creatinine 1.5–1.9 mg/dL.[5] None of the 10 patients randomised to allopurinol 100 mg/day achieved UA levels <6 mg/dL, compared with 44%, 45%, and 60% of those receiving febuxostat 80 mg, 120 mg, and 240 mg, respectively.[5] The CONFIRMS trial included 35% of patients with mild or moderate renal impairment (defined as an estimated creatinine clearance of 60–89 mL/min and 30–59 mL/min, respectively).[8] Patients were randomised in a 1:1:1 fashion to febuxostat 40 mg, febuxostat 80 mg, and allopurinol (300 mg/day for subjects with normal renal function and mild renal impairment or 200 mg in the presence of moderate renal impairment). The proportion of subjects with mild-to-moderate renal impairment achieving UA levels <6 mg/dL was higher in the treatment arm with febuxostat 80 mg (71.6%) compared with those with febuxostat 40 mg (49.7%) and allopurinol (42.3%) ($p<0.001$ for each comparison). *Post hoc* analysis of the FOCUS trial estimated an improvement of 1 mL/min of GFR for every 1 mg/dL reduction of UA level.[56]

XANTHINE OXIDASE INHIBITORS AND CHRONIC KIDNEY DISEASE PROGRESSION: CLINICAL TRIALS

In 2012, the Kidney Disease: Improving Global Outcomes (KDIGO) guidelines declared that there was a lack of evidence to support or refute the use

of urate lowering agents in renal patients in order to delay the progression of CKD.[57] Although the evidence remains inconclusive, recent investigations added encouraging results on the topic. The meta-analysis by Bose et al.[58] included eight randomised controlled trials comparing the effect of allopurinol with placebo or no treatment on renal outcomes. There was a slight but significant reduction of creatinine in favour of allopurinol in three trials conducted among CKD patients (-0.4 mg/dL; 95% CI: -0.8–0.0, p=0.03), though treatment with allopurinol was not associated with any benefit in terms of GFR change, proteinuria, and progression to ESRD.[58] More recently Goiocoechea et al.[59] randomised 113 CKD patients to allopurinol 100 mg versus standard therapy. After 84 months of follow-up, patients receiving allopurinol experienced a mild but significant reduction in circulating UA level (from 7.8±2.1 mg/dL to 6.6±1.5 mg/dL, p=0.04) and a lower GFR decline (-6.5±1.6 mL/min/1.73 m^2) compared with standard therapy (-13.3±5.0 mL/min/1.73 m^2). Allopurinol was also associated with a lower risk of cardiovascular events (HR 0.43; 95% CI: 0.21–0.88).[59] Sircar et al.[60] randomised 108 CKD patients to febuxostat 40 mg versus placebo. Six months of treatment with febuxostat was associated with a stronger reduction in UA levels (from 9.0±2.0 to 5.2±1.5 mg/dL) and with improvement of GFR (+3.2 ml/min/1.73 m^2)[60] compared with that observed with allopurinol by Goiocoechea et al.[59]

CONCLUSIONS

The majority of observational data support an independent association between high UA levels and the risk of CKD. However, conflicting results have also been published in regards to this. Experimental evidence has been reported on how UA may directly induce renal damage through the activation of RAAS, inflammation, and intracellular oxidative cascades. RAAS inhibitors or urate lowering agents were shown to be effective in correcting the renal damage induced by UA in animal models. Several randomised controlled trials thereafter investigated the effect of xanthine oxidase inhibitors on renal outcomes, leading to encouraging, but still inconsistent results. Febuxostat may represent a safe and effective medication for lowering UA levels, avoiding the risk of toxicity, or underpowered efficacy of allopurinol in renal patients. Although UA can be considered as a risk factor as well as an effector of renal damage, currently the strength of evidence remains insufficient to recommend a widespread adoption of xanthine oxidase inhibitors for slowing the progression of CKD.

REFERENCES

1. Johnson RJ et al. The planetary biology of ascorbate and uric acid and their relationship with the epidemic of obesity and cardiovascular disease. Med Hypotheses. 2008;71(1):22-31.

2. Watanabe S et al. Uric acid, hominoid, and the pathogenesis of salt-sensitivity. Hypertension. 2002;40:355-60.

3. Johnson RJ et al. Lessons from comparative physiology: could uric acid represent a physiologic alarm signal gone awry in western society? J Comp Physiol B. 2009;179(1):67-76.

4. Becker MA et al. Febuxostat compared with allopurinol in patients with hyperuricemia and gout. N Engl J Med. 2005;353(23):2450-61.

5. Schumacher HR et al. Effects of febuxostat versus allopurinol and placebo in reducing serum urate in subjects with hyperuricemia and gout: a 28-week, phase III, randomized, double-blind, parallel-group trial. Arthritis Rheum. 2008;59: 1540-8.

6. Becker MA et al. Clinical efficacy and safety of successful longterm urate lowering with febuxostat or allopurinol in subjects with gout. J Rheumatol. 2009; 36:1273-82.

7. Schumacher HR et al. Febuxostat in the treatment of gout: 5-yr findings of the FOCUS efficacy and safety study. Rheumatology (Oxford). 2009;48(2): 188-94.

8. Becker MA et al. The urate-lowering efficacy and safety of febuxostat in the treatment of hyperuricemia of gout: the CONFIRMS trial. Arthritis Res Ther. 2010;12(2):R63.

9. Huang H et al. Uric acid and risk of heart failure: a systematic review and meta-analysis. Eur J Heart Fail. 2014;16(1):16-24.

10. Xu Y et al. Hyperuricemia as an independent predictor of vascular complications and mortality in type 2 diabetic patients:a meta-analysis. PLoS One. 2013;8(10):e78206.

11. Grayson PC et al. Hyperuricemia and incident hypertension: a systematic review and meta-analysis. Arhritis Care Res (Hoboken). 2011;63(1):102-10.

12. Wang J et al. Hyperuricemia and risk of incident hypertension: a systematic review and meta-analysis of observational studies. PLoS One. 2014;9(12):e114259.

13. Yuan H et al. Serum uric acid levels and risk of metabolic syndrome: a dose-response meta-analysis of prospective studies. J Clin Endocrinol Metab. 2015; 100(11):4198-207.

14. Lv Q et al. High serum uric acid and increased risk of type 2 diabetes: a systematic review and meta-analysis of prospective cohort studies. PLoS One. 2013;8(2):e56864.

15. Zhao G et al. Baseline serum uric acid level as a predictor of cardiovascular disease related mortality and all cause mortality: a meta-analysis of prospective studies. Atherosclerosis. 2013;231(1):61-8.

16. Li L et al. Is hyperuricemia an independent risk factor for new-onset chronic kidney disease?: A systematic review and meta-analysis based on observational cohort studies. BMC Nephrol. 2014;15:122.

17. Fathallah-Shaykh SA, Cramer MT. Uric acid and the kidney. Pediatr Nephrol.

2014;29:999-1008.

18. Jalal DI et al. Uric acid as a target of therapy in CKD. Am J Kidney Dis. 2012; 61(1):134-46.

19. Edwards NL. The role of hyperuricemia and gout in kidney and cardiovascular disease. Cleve Clin J Med. 2008;75(suppl5):S13-6.

20. Wright AF et al. A 'complexity' of urate transporters. Kidney Int. 2010;78(5): 446-52.

21. Enomoto A et al. Molecular identification of a renal urate anion exchanger that regulates blood urate levels. Nature. 2002;417(6887):447-52.

22. Neogi T. Clinical practice. Gout. N Engl J Med. 2011;364(5):443-52.

23. Terkeltaub R. Update on gout: new therapeutic strategies and options. Nat Rev Rheumatol. 2010;6(1):30-8.

24. Khanna D et al. 2012 American College of Rheumatology guidelines for management of gout part I: systematic non pharmacolic and pharmacologic therapeutic approaches to hyperuricemia. Arthritis Case Res (Hoboken). 2012; 64(10):1431-46.

25. Chaudhary K et al. Uric acid- key ingredient in the recipe for cardiorenal metabolic syndrome. Cardiorenal Med. 2013;3(3):208-20.

26. Mazzali M et al. Elevated uric acid increases blood pressure in the rat by a novel crystal-independent mechanism. Hypertension. 2001;38(5):1101-6.

27. Mazzali M et al. Hyperuricemia induces a primary renal arteriolopathy in rats by a blood pressure-independent mechanism. Am J Physiol Renal Physiol. 2002;282(6):F991-7.

28. Sanchez-Lozada LG et al. Effect of febuxostat on the progression of renal disease in 5/6 nephrectomy rats with and without hyperuricemia. Nephron Physiol. 2008;108(4):69-78.

29. Verzola D et al. Uric acid promotes apoptosis in human proximal tubular cells by oxidative stress and the activation of the NADPH oxidase NOX 4. PLoS One. 2014;9(12):e115210.

30. Han HJ et al. Uric acid inhibits renal proximal tubule cell proliferation via at least two signaling pathways involving PKC, MAPK, cPLA2, and NF-kappaB. Am J Physiol Renal Physiol. 2007;292(1): F373-81.

31. Kosugi T et al. Effect of lowering uric acid on renal disease in the type 2 diabetic db(db mice. Am J Physiol Renal Phsyiol. 2009;297(2):F481-8.

32. Johnson RJ et al. Sugar, uric acid, and the etiology of diabetes and obesity. Diabetes. 2013;62(10):3307-15.

33. Lanaspa MA et al. Uric acid induces hepatic steatosis by generation of mitochondrial oxidative stress: potential role in fructose-dependent and -independent fatty liver. J Biol Chem. 2012;287(48):40732-44.

34. Baldwin W et al. Hyperuricemia as a mediator of the proinflammatory endocrine imbalance in the adipose tissue in a murine model of the metabolic syndrome. Diabetes. 2011;60(4):1258-69.

35. Lanaspa M et al. Uric acid and metabolic syndrome: what is the relationship? Curr Rheum Rev. 2011;7(2):162-9.

36. Roncal-Jimenez CA et al. Sucrose induces fatty liver and pancreatic inflammation in male breeder rats independent of excess energy intake. Metabolism. 2011;60(9):1259-70.

37. Sanchez-Lozada LG et al. Effects of febuxostat on metabolic and renal alterations in rats with fructose-induced metabolic syndrome. Am J Physiol Renal Physiol. 2008;294(4):F710-8.

38. Hsu C et al. Risk factors for end-stage renal disease: 25-year follow-up. Arch Intern Med. 2009:169(4):342-50.

39. Uchida S et al. Targeting uric acid and the inhibition of progression to end-stage renal disease - a propensity score analysis. PLoS One. 2015;10(12):e0145506.

40. Iseki K et al. Significance of hypeuricemia as a risk factor for developing ESRD in screened cohort. Am J Kidney Dis. 2004;44(4):642-50.

41. Wallace KL et al. Increasing prevalence of gout and hyperuricemia over 10 years among older adults in a managed care population. J Rheumatol. 2004;31(8): 1582-8.

42. Trifirò G et al. Epidemiology of gout and hyperuricemia in Italy during the years 2005-2009: a nationwide population-based study. Ann Rheum Dis. 2013;72(5):694-700.

43. Chen LK et al. Uric acid: a surrogate of insulin resistance in older women. Maturitas. 2008;59(1):55-61.

44. Xue C et al. Prevalence of chronic kidney disease in Jing adults in China: a village-based study. Clin Nephrol. 2013;79(1):50-6.

45. Sturm G et al. Uric acid as a risk factor for progression of non-diabetic chronic kidney disease? The Mild to Moderate Kidney Disease (MMKD) study. Exp Gerontol. 2008;43(4):347-52.

46. Madero M et al. Uric acid and long-term outcomes in CKD. Am J Kidney Dis. 2009;53(5):796-803.

47. Testa A et al. Association of a polymorphism in a gene encoding a urate transporter with CKD progression. Clin J Am Soc Nephrol. 2014;9(6):1059-65.

48. Ogino K et al. Clinical significance of hypouricemia in hospitalized patients. J Med. 1991;22(2):76-82.

49. Kanda E et al. Uric acid level has a U-shaped association with loss of kidney function in healthy people: a prospective cohort study. PLoS One. 2015;10(2):e0118031.

50. Stockert AL, Stechschulte M. Allopurinol or fenuxostat: how far have we come? Clin Med Insight: therap. 2012;2:927-45.

51. Spector T. Inhibition of urate production by allopurinol. Biochem Pharmacol. 1977;26(5):355-8.

52. Takano Y et al. Selectivity of febuxostat, a novel non-purine inhibitor of xanthine oxidase/xanthine dehydrogenase. Life Sciences. 2005;76(16):1835-47.

53. Arellano F, Sacristan JA. Allopurinol Hypersensitivity Syndrome: a Review. Ann Pharmacother. 1993;27(3):337-43.

54. Hoskison TK, Wortmann RL. Advances in the management of gout and hyperuricaemia. Scand J Rheumatol. 2006;35(4):251-60.

55. Dalbeth N, Stamp L. Allopurinol dosing in renal impairment: walking the tightrope between adequate urate lowering and adverse events. Semin Dial. 2007;20(5):391-5.

56. Whelton A et al. Renal function in gout: long-term treatment effects of febuxostat. J Clin Rheumatol. 2011;17(1): 7-13.

57. 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):1-150.

58. Bose B et al. Effects of uric acid-lowering therapy on renal outcomes: a systematic review and meta-analysis. Nephrol Dial Transplant. 2014;29(29): 406-13.

59. Goiocoechea M et al. Allopurinol and progression of CKD and cardiovascular events: long-term follow-up of a randomized clinical trial. Am J Kidney Dis. 2015;65(4):543-9.

60. Sircar D et al. Efficacy of febuxostat for slowing the GFR declive in patients with CKD and asymptomatic hyperuricemia: a 6 month, double-blind, randomized, placebo-controlled trial. Am J Kidney Dis. 2015;66(6):945-50.

VASCULAR AND VALVULAR CALCIFICATIONS IN CHRONIC KIDNEY DISEASE: AN UPDATE

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ABSTRACT

In chronic kidney disease (CKD) and end-stage renal disease patients cardiovascular disease is the main cause of morbidity and mortality, with incidence of cardiac related mortality increasing as renal function declines. Even after controlling for traditional cardiovascular risk factors such as smoking, age, gender, dyslipidaemia, and arterial hypertension, patients with CKD have a higher incidence of major cardiovascular events. CKD is characterised by the presence of many other non-traditional cardiovascular risk factors, such as chronic inflammation and accelerated atherosclerosis, oxidative stress, and especially, secondary hyperparathyroidism. This review will summarise the current evidence on vascular calcifications and valvular heart disease in CKD patients, from pathophysiology to therapeutic strategies.

Keywords: Chronic kidney disease (CKD), vascular calcifications, valvular calcifications, secondary hyperparathyroidism.

BACKGROUND

In chronic kidney disease (CKD) and end-stage renal disease patients cardiovascular disease is the main cause of morbidity and mortality, with incidence of cardiac related mortality increasing as renal function declines. Even after controlling for traditional cardiovascular risk factors such as smoking, age, gender, dyslipidaemia, and arterial hypertension, patients with CKD have a higher incidence of major cardiovascular events. CKD is characterised by the presence of many other non-traditional cardiovascular risk factors, such as chronic inflammation and accelerated atherosclerosis, oxidative stress, and especially, secondary hyperparathyroidism.[1]

It has been postulated that the high incidence of valvular and vascular calcifications could be mainly due to systemic chronic inflammation;[2] however, new findings have indicated that chronic inflammation alone cannot explain the extent of degenerative calcifications (involving the arterial media layer) that are observed in CKD patients.[3,4] This article will review the latest insights into cardiovascular calcifications, which are potent predictors of cardiovascular morbidity and mortality in CKD patients.

CLINICAL FEATURES AND DIAGNOSIS

Two types of vascular calcification affect the majority of patients with long-standing CKD and ESRD: arterial media calcification (calcific arteriosclerosis or Mönckeberg's sclerosis), and accelerated calcification of the intimal plaque (calcific atherosclerosis).[3,4] The latter probably represents the last step in classical atherosclerosis,

whereas medial calcification is non-inflammation based and associated with the duration of haemodialysis, calcium phosphate disorders, diabetes, and ageing.[3,4] Cardiac calcifications (i.e. myocardial or valvular calcifications) mainly involve valve leaflets, discussed forthwith, although pathophysiological pathways seem to be similar to those of vascular calcifications. Another rare form of vascular calcification is calcific uraemic arteriolopathy (also known as calciphylaxis),[5,6] but discussion of this topic lies beyond the scope of this review.

The presence of vascular and valvular calcifications can be associated with other cardiovascular findings related to arterial stiffness and myocardial fibrosis. Vascular calcifications are frequently related to reduced arterial elasticity, higher pulse pressures, and pulse-wave velocities resulting in cardiomyocyte hypertrophy and left ventricular hypertrophy (LVH).[7-9] Uraemia-associated valvular disease (i.e. stenotic mitral and aortic valvular disease) is therefore accountable for increased afterload and contributes to worsening of LVH. CKD-associated LVH is related to cardiomyocyte and interstitial fibrosis, which can induce systolic and/or diastolic heart failure as well as arrhythmias, and is responsible for higher rates of sudden cardiac death compared with the general population.[10] Diagnosis of vascular and valvular calcifications can be provided by ultrasound or plain X-ray radiograms. Echocardiography is crucial in detecting and staging both mitral and aortic calcifications and it is fundamental to the evaluation of younger CKD patients and those awaiting a kidney transplant.

PATHOPHYSIOLOGY AND CLINICAL CORRELATIONS

Vascular and valvular calcifications usually result from an imbalance between promoting and inhibiting factors (Table 1). The main constituents of calcifications are calcium and phosphate, especially as hydroxyapatite aggregates.[3] It has been well established that higher mortality rates in CKD patients can be explained by high calcium and phosphate levels, which lead to osteoblastic modifications of vascular smooth muscle cells (VSMCs) and fibrosis development.[3] CKD patients also show higher concentrations of bone morphogenic protein (BMP) 2, which is involved in the calcification process.[3] Unbalanced calcium and phosphate levels are also associated with high parathyroid hormone and fibroblast growth factor 23 (FGF23) levels.[11,12]

Osteocytes produce FGF23 in response to dietary phosphate overload.[11] FGF23 increases renal phosphate excretion and reduces the levels of 1,25-dihydroxyvitamin D and parathyroid hormone. In CKD patients, increased levels of FGF23 therefore act as a compensatory mechanism to normalise phosphate concentrations as renal function worsens. The FGF23-mediated compensation of phosphate balance may have deleterious trade-offs. High levels of FGF23 are associated with mortality, CKD progression, and calcification in CKD patients.[11] Indeed, high FGF23 leads to an increased risk of secondary hyperparathyroidism due to the inhibition of 1-alpha-hydroxylase.

Other extracellular matrix molecules may play a role in vascular mineralisation and arterial remodelling, such as Type 1 collagen, bone sialoprotein, fibronectin, and decorin.[13,14] Arterial remodelling results in VSMCs switching to osteoblastic cells, leading to vascular calcification. Matrix metalloproteinases and cathepsin can also modulate and promote vascular calcifications and their levels are elevated in the blood even in patients with early CKD.[15,16] Among calcification-promoting agents, palmitic acid can promote mineralisation through activation of acyl-coenzyme A synthetases;[17] therefore oxidised low-density lipoprotein can also induce osteoblastic switching in VSMCs.[18] Finally, chronic systemic diseases (diabetes and hypertensive diseases) have been associated with cardiovascular calcifications. Drugs such as warfarin could promote calcifications by inhibiting the vitamin K cycle and reducing fetuin-A levels.[19-22] Calcification inhibitors that provide tight control of calcium metabolism and precipitation will now be discussed.

Fetuin-A is a systemic inhibitor of ectopic calcification as several experimental studies have shown.[20-24] In haemodialysis patients, low blood fetuin-A levels are associated with chronic inflammation and vascular/valvular calcifications. It has also been demonstrated that small vesicular structures derived from VMSCs accumulate fetuin-A to prevent calcification;[23] fetuin-A acts by binding BMP2, BMP4, and BMP6, blocking osteochondrogenic activity.[24] Matrix Gla protein is another calcification inhibitor as demonstrated in studies where matrix Gla protein-deficient mice have died as a result of massive bleeding caused

by calcified vessel rupture.[25,26] Osteoprotegerin is mainly expressed on VMSCs and endothelial cells and it inhibits osteoclastic function and bone reabsorption; its deficiency can lead to accelerated osteoporosis and vascular calcification, while high blood levels are associated with atherosclerotic disease.[27,28] Osteopontin (OPN) is an extracellular phosphoprotein with a high affinity for hydroxyapatite and it can be found in mineralised tissues; its deficiency is associated with accelerated vascular calcification.[29] OPN usually inhibits calcium crystal growth and promotes osteoclastic function; it is not present in normal arteries.[30]

Calcification inhibitor/promoter imbalances lead to deep modifications in VSMC phenotype and function. Before the deposition of calcium in the vessel wall, VSMCs undergo differentiation into osteoblastic-like cells and downregulate production of smooth muscle specific genes such as *smooth muscle actin* and *SM22*. Simultaneously, as previously discussed, these cells upregulate markers of osteochondrogenesis such as OPN, osteocalcin, and alkaline phosphatase. Osteoblast/chondrocyte-like VSMCs are able to produce a collagen matrix and form calcium and phosphorus-enriched matrix vesicles (MV) promoting vascular wall mineralisation and beginning the calcification pathway.[30-32] Endothelial and vascular calcification also seems to be increased by α-elastin, an elastin-derived peptide; however elastin degradation secondary to ESRD in mice has demonstrated a lack of medial calcification development.[30-32] As previously described, calcium and phosphorus-enriched MV seem to promote vascular calcification. MV have been found in atherosclerotic plaques associated with intimal calcification and in non-atherosclerotic patients with arterial medial calcification, such as those with CKD.[33] In CKD patients, elevated calcium and phosphate levels have been shown to induce release of 30–300 nm large MV from cultured VSMCs, and elevated calcium levels also enhance mineral formation from MV.[34] New evidence demonstrates that macrophages also play a role in the histogenesis of procalcific vesicles similar to MV at atherosclerotic plaque sites.[35]

Other extracellular vesicles are represented by apoptotic bodies that can contribute to CKD vascular calcifications; it has been shown that elevated calcium and phosphate levels can be accountable for VMSC apoptosis.[36] Released apoptotic bodies can accumulate calcium and lead to widespread calcifications along the vessels walls.[36] Elevated levels of MV are also associated with cardiovascular mortality in ESRD patients and they are often correlated with aortic pulse wave velocity and carotid intimal media thickness.[37] Circulating MV are detectable in menopausal women with coronary calcifications and coronary heart disease and they have been directly correlated with coronary artery calcium score.[38]

Heart valvular calcifications mainly occur on mitral and aortic valves. Prevalence of mitral annulus calcifications (MAC) is between 8% and 15% in the general population and there is a median prevalence of 14–18% in CKD patients dependent on CKD stage, with a higher incidence in those presenting other cardiovascular risk factors.[39-43]

Table 1: Promoters and inhibitors of cardiovascular calcifications.

Promoters	Inhibitors
BMP2, BMP4	Matrix Gla protein
Osteocalcin	Osteopontin
Bone sialoprotein	Osteoprotegerin
Alkaline phosphatase	Fetuin-A
Calcium and phosphate ions	Klotho
Oxidative stress	Pyrophosphate
Inflammatory cytokines (IL-6, IL-1, TNF)	Carbonic anhydrase
Diabetes	Vitamin K
Coumadin derivatives	Magnesium
Matrix vesicles and apoptotic bodies	Sodium thiosulfate

BMP: bone morphogenic protein; IL: interleukin; TNF: tumour necrosis factor.

MAC are thought to be an age-related process but, especially in CKD patients, they represent the culmination of an active process quite similar to medial and atherosclerotic calcification.[44] Presence of MAC is also related to incidence of arrhythmias and sudden cardiac death.[45,46] MAC can be defined as a chronic degenerative process of the fibrous component of the mitral valve[45,47] and can be easily detected by standard two dimensional echocardiography, both in M and B-mode. Echocardiography usually detects MAC as a hyperechoic band beneath the posterior mitral leaflet with the M-mode; B-mode (Figure 1) ultrasound allows physicians to localise MAC in the angle between the left ventricular posterior wall and the posterior mitral leaflet.[45,48] Unfortunately, echocardiography does not distinguish between calcification and collagen; electron beam computed tomography (CT) and multislice (spiral) CT are more effective, non-invasive techniques for assessing cardiac and coronary calcifications.[40,49] New three dimensional ultrasound devices provide a cheaper alternative to CT and they provide complete information regarding localisation and extension of calcifications, allowing evaluation of the mitral valve area.[50]

MAC was initially considered a degenerative, age-related process only;[51] however, accumulating evidence points toward a regulated process with features similar to both medial and atherosclerotic cardiovascular calcification on the basis of pathological findings and strong correlations with other pre-existing cardiovascular risk factors.[41,52] A strong correlation has also been demonstrated between MAC and carotid atherosclerotic disease, peripheral artery disease, and coronary artery disease.[53] Degenerative calcification of the mitral annular area is accelerated by conditions that increase mitral valve stress such as hypertension, aortic stenosis, and hypertrophic cardiomyopathy,[45,54] with a strong relationship between LVH and severity of MAC.[55] MAC is common in patients with CKD because of an increased prevalence and severity of cardiovascular risk factors and atherosclerotic disease.[56]

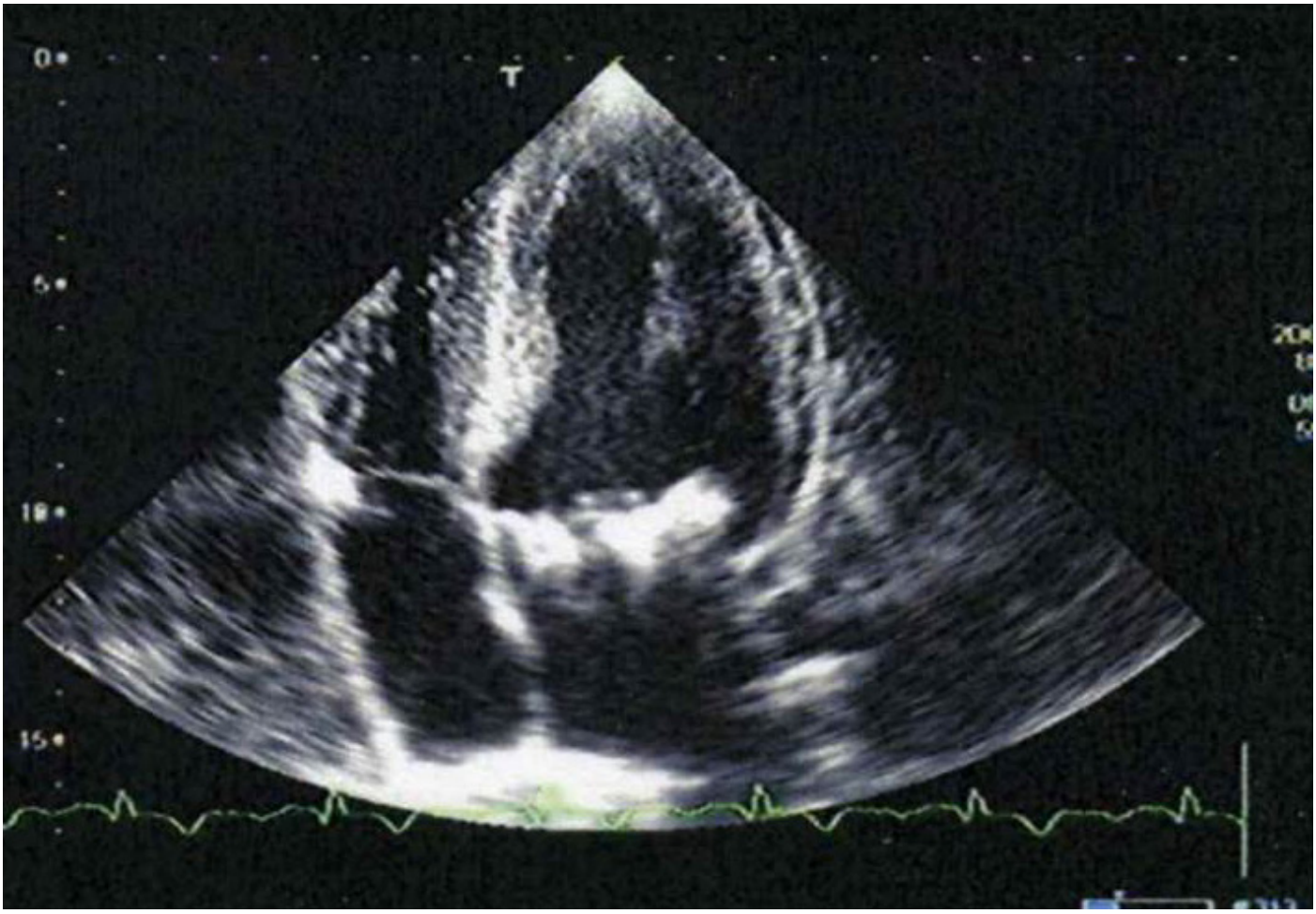

Figure 1: Large mitral annulus calcification in a Stage 4 chronic kidney disease patient.

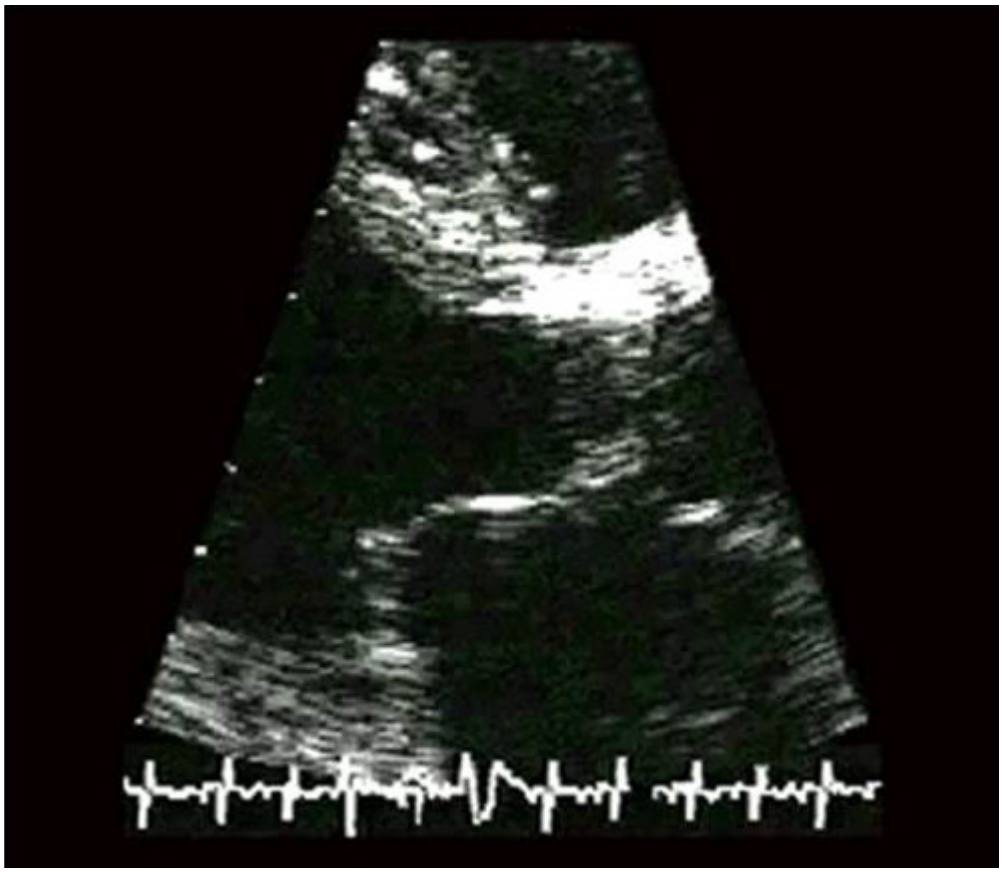

Figure 2: Aortic valvular calcification in a haemodialysis patient.

Furthermore, there is growing evidence that the abnormal calcium-phosphorus metabolism observed in patients with chronic renal failure has a direct role in the pathogenesis of MAC.[45,57]

MAC is usually an incidental finding in patients evaluated by ultrasound for cardiovascular disease (CVD). Calcified mitral annulus is not always associated with symptoms, precluding true evaluation of MAC prevalence in the general population and in CKD patients. In large observational studies, the prevalence of MAC was 42% in elderly patients with known CVD and 28% in CKD patients at any stage of disease.[31] MAC generally has little or no effect on left ventricular inflow or mitral valve function because leaflets are usually spared;[45] severe mitral annulus involvement may occasionally lead to mitral regurgitation or stenosis.[45,58,59] Patients with MAC also have a higher incidence of arrhythmias such as atrioventricular block, bundle branch block, intraventricular conduction delay, and especially in CKD patients, sudden cardiac death probably due to direct extension of calcific deposits in the atrioventricular node and bundle.[46,60] Several community-based studies have evidenced a strong association between the presence of mitral valve calcifications and atrial fibrillation (AF) that seems to be independent of other risk factors for AF and development of acute coronary syndrome.[61] In patients with MAC, AF is probably a result of involvement of the inter and intra-atrial conduction systems and only partially mediated by left atrial dilation.[61,62]

As previously mentioned, >50% of deaths among ESRD patients are caused by CVD. Valvular heart disease is common in ESRD patients, with an incidence rate 5-times greater than that reported in the general population; its prevalence ranges from 9.5–36%.[63-66] Aortic valve calcification (AVC; Figure 2) is the most common valvular abnormality in the general population as well as in patients on haemodialysis.[52] In the general population, incidence of AVC increases with age, occurring mainly in those >65 years old,[67,68] while in CKD patients AVC is seen at a younger age and with associated secondary hyperparathyroidism. FGF23 levels could be considered as strong predictors of aortic valve disease in CKD patients.[69]

TREATMENT STRATEGIES

Moderate-to-severe CKD (with estimated glomerular filtration rate [eGFR] <30 mL/min) is characterised by an impairment of calcium-phosphate metabolism with low calcium and high phosphate levels due to secondary hyperparathyroidism. Haemodialysis patients worldwide are treated with bicarbonate dialysis using sodium bicarbonate as the base. Post-dialysis alkalosis may result in precipitation of calcium phosphate in soft tissues, including vessel walls, especially in the presence of high serum calcium levels (e.g. due to high dialysate calcium or treatment with 1,25-dihydroxyvitamin D) and may contribute to the pathogenesis of CVD.[70] The appropriate dialysate calcium concentration should be chosen on the basis of the clinical characteristics of each patient for personalised dialysis therapy.[71] For instance, in order to avoid positive calcium balance and increased risk of vascular calcification, 1.25 mmol/L dialysate calcium concentration should be chosen for patients taking supplements of calcium salts, active vitamin D metabolites, and calcium-containing phosphate binders. In contrast, for patients prone to cardiac arrhythmias, special caution is warranted when reducing from a 1.5 to 1.25 mmol/L dialysate calcium concentration. Current opinion encourages calcium levels of 1.25 mmol/L or lower; however some authors suggest an individualised approach.[72,73] On the other hand, phosphate blood levels increase in patients with moderate-to-severe renal failure. Advanced CKD stages (with eGFR <30 mL/min) are characterised by hyperphosphataemia, also due to excessive dietary intake. Together with diet, phosphate binder therapy should be started with calcium-based or calcium free phosphate binders; the majority of clinical studies show better outcomes with calcium-free phosphate binders.[74,75]

Some studies have shown the relevance of vascular calcification progression on mortality rate and the critical role of diet and phosphate intake on survival.[76-79] Indeed, slow progression of cardiovascular calcifications, and even delayed progression of kidney disease may be attained in patients who follow a phosphate-restricted diet plus sevelamer in comparison with either unrestricted diet alone or with calcium acetate. Concerning vitamin D levels and the effects of its supplementation on the onset and progression of vascular and valvular calcifications, recently reported data are conflicting.[80] The ADVANCE study has investigated the role of calcimimetics (e.g. cinacalcet) regarding the calcification progress in haemodialysis patients; the trial was conducted in 360 patients randomised to cinacalcet plus low-dose calcitriol or vitamin D analogue. The primary endpoint was to evaluate the percentage change in coronary artery calcification from baseline to Week 52; results did not differ between the two arms, although excessive use of vitamin D analogue in the combination arm could be a confounding factor.[81,82] A small pilot study has investigated the potential role of a magnesium-containing phosphate binder[83] but no data are available to substantiate the hypothesis that magnesium interferes with calcification in CKD. Finally, in 50 patients with CKD Stages 3–4, alendronate was tested but results showed that progression of vascular calcification compared with placebo over 18 months was not affected.[84]

CONCLUSIONS

Different factors and several pathophysiological pathways can contribute to vascular and heart valvular calcification development in CKD patients. These various patterns (atherosclerotic calcification, media sclerosis, and valvular disease) could overlap but VSMCs are the most directly involved in the calcification process using the mechanisms described in this article. Therapy has to be aimed at both correcting the imbalance between calcification promoters and delaying cardiovascular calcification progression, reducing phosphate levels, and monitoring the clinical and biochemical features of secondary hyperparathyroidism.

REFERENCES

1. Schlieper G et al. The vulnerable patient with chronic kidney disease. Nephrol Dial Transplant. 2015;pii: gfv041. [Epub ahead of print].

2. Zoccali C, London G. Con: Vascular calcification is a surrogate marker, but not the cause of ongoing vascular disease and it is not a treatment target in chronic kidney disease. Nephrol Dial Transplant. 2015;30(3):352-7.

3. Ketteler M et al. Calcification and cardiovascular health: new insights into an old phenomenon. Hypertension 2006;47(6):1027-34.

4. Lanzer P et al. Medial vascular calcification revisited: review and perspectives. Eur Heart J. 2014;35(23): 1515-25.

5. Russo D et al. Multimodal treatment of calcific uraemic arteriolopathy (calciphylaxis): A case series. Clin Kidney J. 2016;9(1):108-12.

6. Barbera V et al. [Calciphylaxis: an enigma to the nephrologist]. G Ital Nefrol. 2012;29(6):674-82.

7. London G et al. Arterial aging and arterial disease: interplay between central hemodynamics, cardiac work, and organ flow-implications for CKD and cardiovascular disease. Kidney Int Suppl. 2011;1(1):10-2.

8. Russo D et al. Pulse pressure and presence of coronary artery calcification. Clin J Am Soc Nephrol. 2009;4(2):316-22.

9. Schlieper G et al. Ultrastructural analysis of vascular calcifications in uremia. J Am Soc Nephrol. 2010;21(4):689-96.

10. Di Lullo L et al. Sudden cardiac death and chronic kidney disease: From pathophysiology to treatment strategies. Int J Cardiol. 2016;217:16-27.

11. Jüppner H. Phosphate and FGF-23. Kidney Int Suppl. 2011;79(Suppl 121): S24-27.

12. Henley C et al. The calcimimetic AMG 641 abrogates parathyroid hyperplasia, bone and vascular calcification abnormalities in uremic rats. Eur J Pharmacol. 2009;616(1-3):306-13.

13. Uitto J et al. Pseudoxanthoma elasticum: molecular genetics and putative pathomechanisms. J Invest Dermatol. 2010;130(3):661-70.

14. Pereira L et al. Pathogenetic sequence for aneurysm revealed in mice underexpressing fibrillin-1. Proc Natl Acad Sci USA. 1999;96(7):3819-23.

15. Basalyga DM et al. Elastin degradation and calcification in an abdominal aorta injury model: role of matrix metalloproteinases. Circulation. 2004;110(22):3480-7.

16. Aikawa E et al. Arterial and aortic valve calcification abolished by elastolytic cathepsin S deficiency in chronic renal disease. Circulation. 2009;119(13):1785-94.

17. Kageyama A et al. Palmitic acid induces osteoblastic differentiation in vascular smooth muscle cells through ACSL3 and NF-kappa B, novel targets of eicosapentaenoic acid. PLoS One. 2013;8(6):e68197.

18. Taylor J et al. Oxidized low-density lipoprotein promotes osteoblast differentiation in primary cultures of vascular smooth muscle cells by up-regulating Osterix expression in an Msx2-dependent manner. J Cell Biochem. 2011; 112(2):581-8.

19. Buendía P et al. Endothelial microparticles mediate inflammation-induced vascular calcification. FASEB J. 2015;29(1):73-181.

20. Schäfer C et al. The serum protein alpha 2-Heremans-Schmid glycoprotein/ fetuin-A is a systemically acting inhibitor of ectopic calcification. J Clin Invest. 2003;112(3):357-66.

21. Westenfeld R et al. Fetuin-A protects against atherosclerotic calcification in CKD. J Am Soc Nephrol. 2009;20(6): 1264-74.

22. Ketteler M et al. Association of low fetuin-A (AHSG) concentrations in serum with cardiovascular mortality in patients on dialysis: a cross-sectional study. Lancet. 2003;361(9360):827-33.

23. Reynolds JL et al. Multifunctional roles for serum protein fetuin-a in inhibition of human vascular smooth muscle cell calcification. J Am Soc Nephrol. 2005; 16(10):2920-30.

24. Demetriou M et al. Fetuin/alpha2-HS glycoprotein is a transforming growth factor-beta type II receptor mimic and cytokine antagonist. J Biol Chem. 1996; 271(22):12755-61.

25. Luo G et al. Spontaneous calcification of arteries and cartilage in mice lacking matrix GLA protein. Nature. 1997;386 (6620):78-81.

26. Brancaccio D et al. Matrix GLA protein gene polymorphisms: Clinical correlates and cardiovascular mortality in chronic kidney disease patients. Am J Nephrol. 2005;25(6):548-52.

27. Bucay N et al. osteoprotegerin-deficient mice develop early onset osteoporosis and arterial calcification. Genes Dev. 1998;12(9):1260-8.

28. Kazama JJ et al. Increased circulating levels of osteoclastogenesis inhibitory factor (osteoprotegerin) in patients with chronic renal failure. Am J Kidney Dis. 2002;39(3):525-32.

29. Speer MY et al. Inactivation of the osteopontin gene enhances vascular calcification of matrix Gla protein-deficient mice: evidence for osteopontin as an inducible inhibitor of vascular calcification in vivo. J Exp Med. 2002; 196(8):1047-55.

30. Giachelli CM et al. Regulation of vascular calcification: roles of phosphate and osteopontin. Circ Res. 2005;96(7): 717-22.

31. Barasch E et al. Clinical significance of calcification of the fibrous skeleton of the heart and aortosclerosis in community dwelling elderly. The Cardiovascular Health Study (CHS). Am Heart J. 2006; 151(1):39-47.

32. Pai A et al. Elastin degradation and vascular smooth muscle cell phenotype change precede cell loss and arterial medial calcification in a uremic mouse model of chronic kidney disease. Am J Pathol. 2011;178(2):764-73.

33. Tanimura A et al. Matrix vesicles in atherosclerotic calcification. Proc Soc Exp Biol Med. 1983;172(2):173-7.

34. Kapustin AN et al. Calcium regulates key components of vascular smooth muscle cell-derived matrix vesicles to enhance mineralization. Circ Res. 2011;109(1):e1-12.

35. New SE et al. Macrophage-derived matrix vesicles: an alternative novel mechanism for microcalcification in atherosclerotic plaques. Circ Res. 2013; 113(1):72-7.

36. Reynolds JL et al. Human vascular smooth muscle cells undergo vesicle-mediated calcification in response to changes in extracellular calcium and phosphate concentrations: a potential mechanism for accelerated vascular calcification in ESRD. J Am Soc Nephrol. 2004;15(11):2857-67.

37. Amabile N et al. Predictive value of circulating endothelial microparticles for cardiovascular mortality in end-stage renal failure: a pilot study. Nephrol Dial Transplant. 2012;27(5):1873-80.

38. Jayachandran M et al. Characterization of blood borne microparticles as markers of premature coronary calcification in newly menopausal women. Am J Physiol Heart Circ Physiol. 2008;295(3):H931-8.

39. Fertman MH, Wolff L. Calcification of the mitral valve. Am Heart J. 1946;31: 580-9.

40. Allison MA et al. Mitral and aortic annular calcification are highly associated with systemic calcified atherosclerosis. Circulation. 2006;113(6):861-6.

41. Kanjanauthai S et al. Relationships of mitral annular calcification to cardiovascular risk factors: the Multi-Ethnic Study of Atherosclerosis (MESA). Atherosclerosis. 2010;213(2):558-62.

42. Asselbergs FW et al. Association of renal function with cardiac calcifications in older adults: the cardiovascular health study. Nephrol Dial Transplant. 2009; 24(3):83440.

43. Foley PW et al. Incidental cardiac findings on computed tomography imaging of the thorax. BMC Res Notes. 2010;3:326.

44. Johnson RC et al. Vascular calcification: pathobiological mechanisms and clinical implications. Circ Res. 2006;99(10): 1044-59.

45. Nestico PF et al. Mitral annular calcification: clinical, pathophysiology, and echocardiographic review. Am Heart J. 1984;107(5 Pt 1):989-96.

46. Di Lullo L et al. [Chronic kidney disease and sudden death]. G Ital Nefrol. 2014;31(3).

47. Korn D et al. Massive calcification of the mitral annulus. A clinicopathological study of fourteen cases. N Engl J Med. 1962;267:900-9.

48. D'Cruz I et al. Submitral calcification

or sclerosis in elderly patients: M mode and two dimensional echocardiography in "mitral annulus calcification." Am J Cardiol. 1979;44(1):31-8.

49. Eisen A et al. Calcification of the thoracic aorta as detected by spiral computed tomography among stable angina pectoris patients: association with cardiovascular events and death. Circulation. 2008;118(13):1328-34.

50. Hahn R. Recent advances in echocardiography for valvular heart disease. F1000Res. 2015;4(F1000 Faculty Rev):914.

51. Sell S, Scully RE. Aging changes in the aortic and mitral valves. Histologic and histochemical studies, with observations on the pathogenesis of calcific aortic stenosis and calcification of the mitral annulus. Am J Pathol. 1965;46:345-65.

52. Boon A et al. Cardiac valve calcification: characteristics of patients with calcification of the mitral annulus or aortic valve. Heart. 1997;78(5):472-4.

53. Adler Y et al. Mitral annulus calcification—a window to diffuse atherosclerosis of the vascular system. Atherosclerosis. 2001;155(1):1-8.

54. Silbiger JJ. Anatomy, mechanics, and pathophysiology of the mitral annulus. Am Heart J. 2012;164(2):163-76.

55. Elmariah S et al. Associations of LV hypertrophy with prevalent and incident valve calcification: Multi-Ethnic Study of Atherosclerosis. JACC Cardiovasc Imaging. 2012;5(8):781-8.

56. Fox CS et al. Cross-sectional association of kidney function with valvular and annular calcification: The Framingham heart study. J Am Soc Nephrol. 2006;17(2):521-7.

57. Umana E et al. Valvular and perivalvular abnormalities in end-stage renal disease. Am J Med Sci. 2003;325(4):237-42.

58. Muddassir SM, Pressman GS. Mitral annular calcification as a cause of mitral valve gradients. Int J Cardiol. 2007;123(1): 58-62.

59. Labovitz AJ et al. Frequency of mitral valve dysfunction from mitral anular calcium as detected by Doppler echocardiography. Am J Cardiol. 1985; 55(1):133-7.

60. Takamoto T, Popp RL. Conduction disturbances related to the site and severity of mitral anular calcification: a 2-dimensional echocardiographic and electrocardiographic correlative study. Am J Cardiol. 1983;51(10):1644-9.

61. Fox CS et al. Mitral annular calcification is a predictor for incident atrial fibrillation. Atherosclerosis. 2004;173(2):291-4.

62. Pekdemir H et al. Assessment of atrial conduction time by tissue Doppler echocardiography and P-wave dispersion in patients with mitral annulus calcification. J Electrocardiol. 2010;43(4):339-43.

63. Ikee R et al. Differences in associated factors between aortic and mitral valve calcification in hemodialysis. Hypertens Res. 2010;33(6):622-6.

64. Maher ER et al. Aortic and mitral valve calcification in patients with end-stage renal disease. Lancet. 1987;2(8564):875-7.

65. Schott CR et al. Mitral annular calcification. Clinical and echocardiographic correlations. Arch Intern Med. 1977;137(9):1143-50.

66. Forman MB et al. Mitral anular calcification in chronic renal failure. Chest. 1984;85(3):367-71.

67. Stewart BF et al. Clinical factors associated with calcific aortic valve: Cardiovascular Health Study. J Am Coll Cardiol. 1997;29(3):630-4.

68. Otto CM et al. Characterization of the early lesion of "degenerative" valvular aortic stenosis: histological and immunohistochemical studies. Circulation. 1994;90(2):844-53.

69. Di Lullo L et al. Fibroblast growth factor 23 and parathyroid hormone predict extent of aortic valve calcifications in patients with mild to moderate chronic kidney disease. Clin Kidney J. 2015;8(6):732-6.

70. Tentori F et al. Association of dialysate bicarbonate concentration with mortality in the Dialysis Outcomes and Practice Patterns Study (DOPPS). Am J Kidney Dis. 2013;62(4):738-46.

71. Bosticardo G et al. Optimizing the dialysate calcium concentration in bicarbonate haemodialysis. Nephrol Dial Transplant. 2012;27(6):2489-96.

72. Gotch FA et al. The KDIGO guideline for dialysate calcium will result in an increased incidence of calcium accumulation in hemodialysis patients. Kidney Int. 2010;78(4):343-50.

73. Messa P. The ups and downs of dialysate calcium concentration in haemodialysis patients. Nephrol Dial Transplant. 2013;28(1):3-7.

74. Chertow GM et al. Sevelamer attenuates the progression of coronary and aortic calcification in hemodialysis patients. Kidney Int. 2002;62(1):245-52.

75. Di Iorio D et al. Mortality in kidney disease patients treated with phosphate binders: A randomized study. Clin J Am Soc Nephrol. 2012;7(3):487-93.

76. Russo D et al. Progression of coronary artery calcification and cardiac events in patients with chronic renal disease not receiving dialysis. Kidney Int. 2011; 80(1):112-8.

77. Di Iorio B et al. Acute effects of very-low-protein diet on FGF23 levels: A randomized study. Clin J Am Soc Nephrol. 2012;7(4):581-7.

78. Russo D et al. Effects of phosphorus-restricted diet and phosphate-binding therapy on outcomes in patients with chronic kidney disease. J Nephrol. 2015; 28(1):73-80.

79. Di Lullo L et al. Progression of cardiac valve calcification and decline of renal function in CKD patients. J Nephrol. 2013; 26(4):739-44.

80. Morrone LF et al. Vitamin D in patients with chronic kidney disease: A position statement of the Working Group "Trace Elements and Mineral Metabolism" of the Italian Society of Nephrology. J Nephrol. 2016. [Epub ahead of print].

81. Raggi P et al. The ADVANCE study: a randomized study to evaluate the effects of cinacalcet plus low-dose vitamin D on vascular calcification in patients on hemodialysis. Nephrol Dial Transplant. 2011;26(4):1327-39.

82. Ureña-Torres PA et al. Protocol adherence and the progression of cardiovascular calcification in the ADVANCE study. Nephrol Dial Transplant. 2013;28(1):146-52.

83. Spiegel DM, Farmer B. Long-term effects of magnesium carbonate on coronary artery calcification and bone mineral density in hemodialysis patients: a pilot study. Hemodial Int. 2009;13(4): 453-9.

84. Toussaint ND et al. Effect of alendronate on vascular calcification in CKD stages 3 and 4: a pilot randomized controlled trial. Am J Kidney Dis. 2010; 56(1):57-68.

TUBULAR HANDLING OF URIC ACID AND FACTORS INFLUENCING ITS RENAL EXCRETION: A SHORT REVIEW

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ABSTRACT

In this review, the authors briefly examine the most recent evidence concerning the role of several proteins involved in tubular urate transport. They also analyse the influence of extracellular volume, electrolyte disorders, acid-base imbalance, and insulin-resistance on renal clearance of uric acid.

Keywords: Uric acid, urate transport, tubular handling, urate clearance.

INTRODUCTION

Uric acid is produced from purine nucleotide metabolism. The formation of uric acid involves purine degradation to inosinic acid and hypoxanthine. The latter is converted by the xanthine oxidase enzyme to xanthine and uric acid. In most mammals, uric acid is then converted to a more soluble allantoin by hepatic uricase.[1] In humans it is not further degraded and constitutes a metabolic end-product that is mainly excreted by the kidneys and, to a lesser extent, by the gastrointestinal tract.[2] Uric acid is a weak acid with a dissociation costant of pK 5.75. Thus, at a physiological plasma pH of 7.4, uric acid is in its more soluble deprotonated form. Its serum level in normal conditions is 3.5–6.8 mg/dL and is determined by the balance of synthesis and excretion rates. Higher values are associated with gout, nephrolithiasis, progression of chronic kidney disease, diabetes mellitus, hypertension, and cardiovascular damage.[3-5] Since the majority of uric acid is excreted by the kidney following the filtration, reabsorption, and secretion processes, the knowledge of uric acid tubular handling helps us to understand the main pathophysiological mechanisms underlying the alterations of its metabolism and to identify new therapeutic targets.

TUBULAR HANDLING OF URIC ACID

The kidney plays a pivotal role in uric acid homeostasis and excretion. Traditionally, uric acid was considered to be freely filtered by the glomerulus and subsequently reabsorbed and secreted along the renal tubule with a fractional excretion of ~10%. Nevertheless, the molecules responsible for bidirectional transcellular urate transport remain partially unknown. In recent years, many studies have demonstrated the role of several proteins belonging to the organic anion transporter (OAT) family, which is involved in urate transport.

Initially, urate transporter (URAT) 1, encoded by the *SLC22A12* gene, was identified as a reabsorptive URAT on the apical membrane of the renal proximal tubule.[6] In humans, a mutation of this encoding gene has been associated to Type 1 renal idiopathic hypouricaemia, characterised by low serum level and high renal excretion of uric acid.[7] Several drugs, such as probenecid and losartan, could decrease the serum urate levels by exercising

inhibitory effects on URAT1, therefore confirming the important role of this molecule in the urate transport.

Besides URAT1, other molecules belonging to the OAT family are expressed on the tubular cell membrane. They form a renal tubular secretory pathway for organic anions including drugs and toxins, however, recent evidence suggests that these proteins also play a role in urate transport.[8-10] OAT1 and OAT3, encoded by the *SLC22A6* and *SLC22A8* genes, respectively, are localised to the basolateral membrane of the renal proximal tubules. In experimental murine models, the absence of OAT1 and OAT3 determines a defective renal excretion of urate;[10] OAT4, encoded by the *SLC22A11* gene, is instead expressed in the apical cell membrane.

Glucose transporter (GLUT) 9, encoded by the *SLC2A9* gene, was initially reported as a fructose or GLUT but recent evidence suggests that it is involved in voltage-dependent urate transport.[11-13] Two isoforms have been identified in experimental studies, GLUT9a and GLUT9b, localised on the basal and apical side of the cellular membrane, respectively. Thus, it has been hypothesised that GLUT9b regulates the luminal uptake and GLUT9a regulates the interstitial exit of urate.[14-16] In humans however, exogenous expression of GLUT9 has been confirmed only at the basolateral side of renal proximal tubules.

Breast cancer resistance protein, ATP-binding cassette sub-family G member 2 (ABCG2), is a protein expressed on the epithelial cells of several organs, especially the placenta, liver, and intestine, and mediates the transport of various chemical compounds including anti-cancer drugs. In the kidney, it is expressed at the apical side of proximal tubules. It has recently been reported to excrete urate, however given its higher expression in the liver and intestine, it likely contributes to the regulation of intestinal urate rather than renal excretion.[17,18] Paradoxically, in *ABCG2* knockout mice an increased renal excretion of urate has been reported, likely due to a compensatory effect or decreased intestinal excretion.[19] NPT (sodium-phosphate cotransporter) 1 and NPT4 proteins belong to the SLC17 family and they are expressed in the apical cellular membrane of proximal tubules. They were initially identified as sodium-dependent phosphate transporters[20] but subsequent studies revealed that they can contribute to *in vivo* excretion of several organic anions, including urate.[21]

In summary, URAT1, GLUT9, and OAT4 ensure the reabsorption of uric acid on the apical and basolateral sides of the tubular cell membrane, respectively. ABCG2, NPT1, and NPT4, provide exit from the cells in the tubular lumen (Figure 1). Other molecular mechanisms contributing to the renal uptake of uric acid at proximal tubular cells have been identified but their specific role and importance in human pathophysiology is still under investigation. Table 1 reports the various URATs, their localisation, and their pathogenetic role in human and animal disease states.

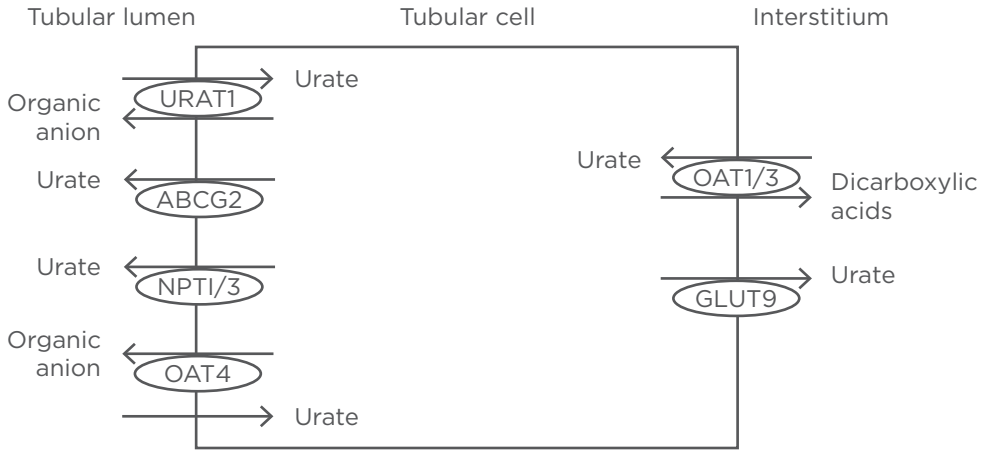

Figure 1: Schematic representation of the location and function of urate transporters on the cell membrane.

URAT1: Uric acid transporter 1; OAT: organic anion transporter; GLUT9: glucose transporter 9; NPT: sodium-phosphate cotransporter; ABCG2: ATP-binding cassette sub-family G member 2.

Table 1: Uric transporters, localisations, and pathogenetic role in human and animal disease states.

Urate transporters	Localisation	Pathogenetic role/related disease in human and animal
URAT1	Apical membrane of proximal tubule	Reabsorption of uric acid from tubular lumen/Type 1 renal idiopathic hypouricaemia in humans
OAT1/3	Basolateral membrane of proximal tubule	Luminal excretion of uric acid/ impaired renal excretion of uric acid in murine models
OAT4	Apical membrane of proximal tubule	Reabsorption of uric acid/ hyperuricaemia and gout in humans
GLUT9	Basolateral membrane of proximal tubule	Reabsorption of uric acid and interstitial exit/hyperuricaemia if upregulated, increased renal excretion if downregulated in humans. Increased renal excretion of uric acid in knockout mice
ABCG2	Apical membrane of proximal tubule, liver, intestine	Excretion of uric acid/ hyperuricaemia and gout with increased renal excretion of urate as compensatory function in humans

URAT1: Uric acid transporter 1; OAT: organic anion transporter; GLUT9: glucose transporter 9; ABCG2: ATP-binding cassette sub-family G member 2.

FACTORS INFLUENCING RENAL EXCRETION OF URIC ACID

The renal clearance of uric acid can be affected by several factors. Genome-wide association studies have demonstrated that single nucleotide polymorphisms of genes encoding for URATs can determine impaired or increased excretion of urate with consequent alteration of serum uric acid values.[22] Electrolyte alterations, especially sodium and chloride, may influence serum urate levels. It has been reported that a higher long-term sodium intake is associated with an increase of serum uric acid.[23] It has been established that each 1 g increase of sodium intake is associated with a 1.2 µmol/L increase in serum uric acid. Elevated serum uric acid could reflect the endothelial dysfunction of these subjects in response to high sodium intake, but it could also be consequent to altered function of NPTs in response to elevated sodium load into the proximal tubule. NPT activity can also be influenced by tubular chloride concentration as a result of its chloride dependence and inhibitor sensitivity.[24] In addition, electrolyte concentration can affect peritubular oncotic pressure and peritubular hydrostatic pressure or transtubular electrochemical gradient, thereby facilitating or slowing uric acid reabsorption. On the other hand, the state of hydration of the extracellular fluid compartment exerts an important influence on the tubular reabsorption of sodium and other filtered ions, including urate.[25] Contraction of extracellular fluid volume determines an activation of the renin-angiotensin-aldosterone system, leading to increased sodium ion (Na^+) reabsorption. This enhanced tubular uptake of sodium is associated with an up to 47% decrease in the clearance of uric acid and hyperuricaemia. Conversely, volume expansion results in a decrease in the net tubular reabsorption of uric acid.

As renal sodium reabsorption is the main method of regulating the extracellular volume, these findings suggest a close relation between reabsorption of sodium and urate in the proximal tubule. Other studies have suggested a similar relation between phosphate and uric acid reabsorption.[26] However, it has been reported that the enhanced urate clearance observed in patients with syndrome of inappropriate antidiuretic hormone secretion is not only due to an increase of effective vascular volume but also by an undetermined mechanism of vasopressin-1 receptor stimulation.[27] The well-known diuretic-induced hyperuricaemia may be attributed to reduced vascular volume but it has also been proposed that URAT1 could mediate urate-furosemide exchanges because furosemide is secreted into the lumen while enhancing urate uptake.[6]

Acid-base status can also influence uric acid clearance. We must first consider that OATs are not urate-specific; organic anions such as sulfate, phosphate, lactate, and ketones; as well as drugs,

metabolites, or other fixed acids excreted by the kidney can therefore interfere with the binding of uric acid to the receptors. Several studies[28,29] have demonstrated that alkalisation of urine increases uric acid excretion and that there is a direct relationship between the amount of excreted uric acid and luminal pH. Urinary pH largely changes in response to an acid or alkali load such as protein intake. Dairy consumption and an alkaline diet can reduce uric acid levels, increasing urate excretion.

In response to an acid load, the kidney increases reabsorption of filtered bicarbonate ions and excretion of hydrogen ions (H^+). H^+ is secreted in the lumen by Na^+/H^+ exchanger 3 (NHE3) and to a lesser extent by a proton pump. The Na^+/H^+ exchange by NHE3 plays a pivotal role for reabsorption of filtered bicarbonate ions and ammoniagenesis.[30] Within the tubular lumen, the secreted H^+ combines with filtered bicarbonate ions leading to carbonic acid formation. The latter is then split into H_2O and CO_2, which diffuses into the cell where it is rehydrated to carbonic acid. Ammonium (NH_3) is formed in the kidney by deamination of glutamine to glutamic acid. Once NH_3 has been protonated to ammonia (NH_4^+), this is secreted into the tubular lumen and subsequently excreted into the urine as ammonium chloride (NH_4^-Cl). Ammoniagenesis is the main renal system for the buffering and removal of H^+ from the body.

In the distal tubule, urine acidification is primarily achieved by a H^+-ATPase proton pump. In this instance, Na^+ reabsorption indirectly influences H^+ or potassium ion (K^+) excretion because the removal of cationic Na^+ from the tubular fluid makes the lumen more electronegative, thereby promoting the secretion of H^+ or K^+ into the lumen.[31] A low K^+ availability secondary to hypokalaemia as well as an acidic state determines an increased H^+ excretion in this site that contributes to urine acidification and maintenance of acid-base balance. For the same reason, anion chloride ions (Cl^-) can modify the negative charge into the tubular lumen, contributing to H^+ excretion and low urinary pH. Interestingly, urinary pH is simply a measure of free H^+ ions in urine but does not reflect renal net acid excretion because NH_4^+ and titratable acidity determine the majority of renal acid excretion. A reduced generation of NH_4^+ can determine an increase of urinary free H^+ and low urinary pH, consequently impairing renal uric acid excretion.[32,33]

Furthermore, as uric acid is a weak acid with a pK value of 5.35 in urine, a low urine pH can determine uric acid crystallisation and subsequent stone formation even if its excretion rate is normal. Observational studies as well as clinical experience suggests that the majority of pure uric acid stone formers exhibit lower urinary pH and fractional urate excretion when compared to healthy controls.[33,34] It has been postulated that the reason for a lower urinary pH in these subjects is a defective urinary ammonia excretion, likely associated to an insulin-resistant state.[33]

Diabetes mellitus and insulin resistance have been associated to low renal clearance of uric acid. The impact of insulin resistance, evaluated by a homeostasis model assessment, showed an inverse correlation between homeostasis model assessment and clearance of uric acid in patients with normal renal function.[35] Unfortunately in this study the relationship between insulin resistance and urinary pH was not investigated. Moreover, in healthy subjects, uric acid excretion was investigated after insulin infusion using the euglycaemic clamp technique. Insulin caused a statistically significant decline in fractional renal urate excretion.[36] Furthermore, uric acid stone formers showed a higher prevalence of metabolic syndrome, which is characterised by insulin resistance.[37]

Sodium-glucose cotransporter 2 (SGLT2) inhibitors are a new class of drug for the treatment of Type 2 diabetes mellitus, that act by blocking the tubular SGLT2-mediated glucose uptake. Patients treated with SGLT2 inhibitors showed that besides increased glycosuria, lower serum uric acid levels and increased urate excretion were present.[38,39] These findings support the hypothesis that hyperinsulinaemia, glucose levels, and tubular glucose uptake influence uric acid excretion. This influence can occur by regulation of GLUT9 activity which is also involved in glucose transport. Alternatively, we can hypothesise that insulin resistance determines low NH_4^+ production and that low urinary pH conditioning creates reduced uric acid excretion. Both these hypotheses could explain the epidemiological link between diabetes mellitus, hyperuricaemia, gout, and stone formation. In Table 2 the main factors influencing renal handling of uric acid in human diseases are summarised.

Table 2: Uric transporters, localisations, and pathogenetic role in human and animal disease states.

Factors influencing renal uric acid excretion	Related diseases
High sodium intake	Hypertension
Electrolyte disorders	Hypokalaemia, diarrhoea
Extracellular fluid volume	Dehydration, furosemide, SIADH
Acid-base balance	Acidosis, alkalosis
Urinary pH	Tubular acidosis, stone formation, impaired ammonia generation
Insulin resistance	Diabetes mellitus, metabolic syndrome

SIADH: syndrome of inappropriate antidiuretic hormone secretion.

CONCLUSIONS

Hyperuricaemia is associated with gout, hypertension, diabetes mellitus, and renal and cardiovascular damage. Serum uric acid levels mainly depend on uric acid synthesis and its renal excretion. Several proteins identified in recent years are involved in tubular urate transport and influence its renal clearance. In addition, clinical and experimental studies have shown that several haemodynamic and metabolic derangements can affect uric acid renal excretion. Therefore, genetic mutations, drugs, electrolyte disorders, acid-base imbalance, variation of effective vascular volume such as renin-angiotensin system alteration, reduced ammonia generation, and insulin resistance can influence urate renal clearance. These are all contributing factors to the occurrence of hyperuricaemia, gout, renal stone formation, and renal failure. The knowledge of the intrinsic homeostatic systems regulating uric acid excretion has a great impact in clinical practice because it provides us with a better understanding of pathophysiological alterations as well as aiding the development of further therapeutic strategies and novel drugs.

REFERENCES

1. Wu XW et al. Two independent mutational events in the loss of urate oxidase during hominoid evolution. J Mol Evol. 1992;34(1):78-84.

2. Sica DA, Schoolwerth AC. "Renal handling of organic anions and cations: excretion of uric acid", Brenner BM (ed.), The Kidney (2000), Philadelphia: WB Saunders, pp.680-700.

3. Feig DI et al. Uric acid and cardiovascular risk. N Engl J Med. 2008;359(17):1811-21.

4. Mazzali M et al. Elevated uric acid increases blood pressure in the rat by a novel crystal-independent mechanism. Hypertension. 2001;38(5):1101-6.

5. Mazzali M et al. Hyperuricemia induces a primary renal arteriolopathy in rats by a blood pressure-independent mechanism. Am J Physiol Ren Physiol. 2002;282(6): F991-7.

6. Enomoto A et al. Molecular identification of a renal urate anion exchanger that regulates blood urate levels. Nature. 2002;417(6887):447-52.

7. Ichida K et al. Clinical and molecular analysis of patients with renal hypouricemia in Japan influence of URAT1 gene on urinary urate excretion. J Am Soc Nephrol. 2004;15(1):164-73.

8. Ahn SY, Nigam SK. Toward a systems level understanding of organic anion and other multispecific drug transporters: a remote sensing and signaling hypothesis. Mol pharmacol. 2009 Sep;76(3):481-90.

9. Motohashi H et al. Gene expression levels and immunolocalization of organic ion transporters in the human kidney. J Am Soc Nephrol. 2002;13(4):866-74.

10. Sekine T et al. Molecular physiology of renal organic anion transporters. Am J Physiol. Renal Physiol 2006;290(2): F251-61.

11. Doblado M, Moley KH. Facilitative glucose transporter 9, a unique hexose and urate transporter. Am J Physiol Endocrinol Metabol. 2009;297(4):E831-5.

12. Vitart V et al. SLC2A9 is a newly identified urate transporter influencing serum urate concentration, urate excretion and gout. Nature genetics 2008;40(4):437-42.

13. Anzai N et al. Plasma urate level is directly regulated by a voltage-driven urate efflux transporter URATv1 (SLC2A9) in humans. J Biol Chem. 2008; 283(40):26834-8.

14. Anzai N et al. Urate transport: Relationship with serum urate disorder. Curr Rheumatol Rev. 2011;7:123-31.

15. Augustin R et al. Identification and characterization of human glucose transporter-like protein-9 (GLUT9):alternative splicing alters trafficking. J Biol Chem. 2004;279(16): 16229-36.

16. Reginato AM et al. The genetics of hyperuricaemia and gout. Nat Rev Rheumatol. 2012;8(10):610-21.

17. Woodward OM et al. Identification of a urate transporter, ABCG2, with a common functional polymorphism causing gout. Proc Natl Acad Sci U S A. 2009;106(25): 10338-42.

18. Noguchi K et al. Human ABC transporter ABCG2/BCRP expression in chemoresistance: Basic and clinical perspectives for molecular cancer therapeutics. Pharmacogenomics Pers Med. 2014;7:53-64.

19. Ichida K et al. Decreased extra-renal urate excretion is a common cause of

hyperuricemia. Nat Commun. 2012;3:764

20. Murer H et al. Proximal tubular phosphate reabsorption: molecular mechanisms. Physiol Rev. 2000;80(4): 1373-409.

21. Jutabha P et al. Human sodium phosphate transporter 4 (hNPT4/ SLC17A3) as a common renal secretory pathway for drugs and urate. J Biol Chem. 2010;285(45):35123-32.

22. Anzai N et al. Recent advances in renal urate transport: Characterization of candidate transporters indicated by genome-wide association studies. Clin Exp Nephrol. 2012;(1):89-95.

23. Forman JP et al. Association between sodium intake and change in uric acid, urine albumin excretion, and the risk of developing hypertension. Circulation. 2012; 125(25):3108-16.

24. Iharada M et al. Type 1 Sodium-dependent Phosphate Transporter (SLC17A1 Protein) is a Cl(-)-dependent Urate Exporter. J Biol Chem. 2010;285(34): 26107-13.

25.Weinman EJ et al. The influence of the extracellular fluid volume on the tubular reabsorption of uric acid. J Clin Invest. 1975; 55(2):283-91.

26. Staum BB et al. Tracer microinjection study of renal tubular phosphate reabsorption in the rat. J Clin Invest. 1972; 51(9):2271-6.

27. Decaux G et al. Evidence in hyponatremia related to inappropriate secretion of ADH that V1 receptor stimulation contributes to the increase in renal uric acid clearance. J Am Soc Nephrol. 1996;7(5):805-10.

28. Kanbara A et al. Urine alkalization facilitates uric acid excretion. Nutr J. 2010; 9:45.

29. Kanbara A et al. Effect of urine pH changed by dietary intervention on uric acid clearance mechanism of pH-dependent excretion of urinary uric acid. Nutr J. 2012;11:39.

30. Bruno CM, Valenti M. Acid-base disorders in patients with chronic obstructive pulmonary disease: A pathophysiological review. J Biomed Biotechnol. 2012;915150.

31. Eaton MP, Stone AM, "Neuromuscular Blockade", Rose BD, Post TW, (e.ds), Clinical physiology of acid-base and electrolyte disorders (2001), New York: McGraw-Hill, pp.406-26.

32. Kamel KS et al. Studies on the pathophysiology of the low urine pH in patients with uric acid stones. Kidney Int. 2002;61(3):988-94.

33. Sakhaee S et al. Pathophysiologic basis for normouricosuric uric acid nephrolithiasis. Kidney Int. 2002;62(3): 971-9.

34. Pak CYC et al. Biochemical profile of idiopathic uric acid nephrolithiasis. Kidney Int. 2001;60(2):757-61.

35. Perez-Ruiz F et al. Renal clearance of uric acid is linked to insulin resistance and lower excretion of sodium in gout patients. Rheumatol Int. 2015;35(9): 1519-24.

36. Ter Maaten JC et al. Renal handling of urate and sodium during acute physiological hyperinsulinaemia in healthy subjects. Clin Sci. 1997;92(1):51-8.

37. Fraile JM et al. Uric acid metabolism in patients with primary gout and the metabolic syndrome. Nucleosides Nucleotides Nucleic Acids. 2010;29(4-6): 330-4.

38. Ferrannini E et al. A Phase IIb, randomized, placebo-controlled study of the SGLT2 inhibitor empagliflozin in patients with Type 2 diabetes. Diabetes Obesity Metab. 2013;15(8):721-8.

39. Lytvyn Y et al. Glycosuria-mediated urinary uric acid excretion in patients with uncomplicated Type 1 diabetes mellitus. Am J Physiol Renal Physiol. 2015;308(2): F77-83.

TREATING THE DIABETIC HYPERTENSIVE: CONSENSUS AND DIFFERENCES

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ABSTRACT

Hypertension and diabetes commonly coexist. Both are major modifiable risk factors for cardiovascular diseases. There has been a substantial shift in the recommendations of several expert committees on the management of hypertension in diabetics. It was once unanimously agreed by almost all major guidelines that the threshold for initiating diabetic patients with antihypertensive therapy is when blood pressure is >130/80 mmHg. The blood pressure target for treatment was also unanimously agreed to be <130/80 mmHg. These recommendations were, however, based on expert opinions and not on findings from major randomised controlled trials.

Since then, there have been several randomised controlled trials looking at blood pressure-lowering in the diabetic population. These include the ADVANCE and ACCORD, and a subanalysis of the INVEST trials. Together with the earlier UKPDS and HOT trials, one would expect there to be more agreement in the most recent recommendations, but in fact the opposite is the case. There are now two different systolic targets (<130 mmHg and <140 mmHg) and three different diastolic targets (<90 mmHg, 85 mmHg, and <80 mmHg). The reason for this involves the choice of trials included in the recommendation, and the interpretation of results from these trials by various guideline committees.

The recommendation for diabetic hypertensives will be more consistent if future trials begin by asking a relevant research question that has not yet been answered: does treating diabetics with different thresholds of blood pressure levels impact on clinical outcomes? The trial must not only determine a primary research question, but it must also be adequately powered to answer it. Only when this question is answered should the next questions be asked. Does it matter how blood pressure is lowered? And are some drugs better than others? In the meantime, guideline committees should try to narrow the gap in recommendations, particularly if the guidelines originate from the same country or region.

Keywords: Diabetes, hypertension, treatment, blood pressure (BP).

INTRODUCTION

Hypertension and diabetes are major contributors to cardiovascular (CV) events and total mortality. The World Health Organization (WHO) has identified both as top causes of total mortality worldwide for more than a decade.[1] These two major risk factors also commonly coexist. In recent mega trials on diabetes, up to 80% of the patients were hypertensive at baseline.[2,3] Diabetes is now regarded as a vascular disease with accompanying dyslipidaemia. Vascular diseases, particularly macrovascular disease, predate the onset of dysglycaemia.[4,5] The importance of blood pressure (BP) control in diabetics was highlighted by the UKPDS analysis which showed that while tighter control of BP improves macrovascular outcome, the same was not seen with tight diabetes control.[6] Surveys have shown that BP control in diabetic hypertensives is required.[7,8] It has been estimated that better control of BP, as in clinical trials in diabetes, could prevent up to 1.5 million deaths worldwide over a 4-year period.[9]

TARGET BLOOD PRESSURE: EVOLUTION OF EVIDENCE

The first insight into what level BP should be lowered to by treatment was provided by the HOT study.[10] Analysis of the diabetic subpopulation in this study showed that, unlike in the main study population, patients who were treated to a diastolic of <80 mmHg had significantly fewer CV events than those treated to a diastolic <90 mmHg. The study's diabetic population (n=1,915), however, constituted only 8% of the total study population. In the same year, the UKPDS 38 showed that, of newly diagnosed diabetics, patients whose BP was tightly controlled (achieved BP was 144/82 mmHg) had significantly fewer strokes than patients whose BP was less tightly controlled (achieved BP was 154/87 mmHg).[6] No significant difference was seen with myocardial infarction or all-cause mortality. It should be noted that the number of patients studied in the UKPDS BP-lowering arm was small (758 in the 'tight group' versus 390 in the 'less tight group'). In other words, both the HOT and UKPDS substudies were, strictly speaking, 'hypothesis-generating' and not definitive. The first study that looked at the effect of different levels of BP was the ABCD 2 trial.[11] In that study, 480 diabetic hypertensives with a mean baseline BP of 136/84 mmHg were randomised to placebo or active treatment (with nisoldipine or enalapril). The achieved BP in the treated group was 128/75 mmHg compared with 137/81 mmHg in the placebo-treated group. There were significantly fewer strokes, development of macroalbuminuria, and progression of retinopathy in the treatment group. Glomerular filtration rate estimated by a 24-hour creatinine clearance performed every 6 months over the 5-year study period, rather surprisingly, did not differ between the active treatment group compared with the placebo-group. These three studies (HOT, UKPDS, and ABCD 2) were the only available evidence at that time and, unsurprisingly, almost all major guidelines (for both hypertension and diabetes) at the turn of the century recommended that BP should be reduced to <130/80 mmHg in diabetic hypertensives. This is despite the fact that all three studies were either subanalyses with small sample sizes or small trials, which made them underpowered and not definitive evidence.

The first mega trial which looked at BP-lowering intervention in a diabetic population was the ADVANCE trial.[12] In this diabetes dedicated study, half of the 11,140 patients with baseline BP of 145/81 mmHg were randomised to either a single pill combination of perindopril 4 mg plus indapamide 1.5 mg (Coversyl Plus®) or placebo. One of the research questions asked in this trial was whether in the diabetic population, lowering systolic BP to <145 mmHg will provide additional benefits. The level of 145 mmHg was chosen because at the time that the study was being designed, the only available evidence for systolic level was from UKPDS 38, which managed to lower BP in the intensive arm to 144/82 mmHg. The ADVANCE trial showed that, in the treated group (achieved BP 135/75 mmHg), there was a significant reduction in CV death and all-cause mortality compared with the placebo group (achieved BP 140/77 mmHg). The ADVANCE trial was a 2-by-2 factorial design which also had a glucose-lowering arm with either standard diabetic care or intensive care, with the addition of gliclazide modified release (Diamicron® MR). In the glucose arm HbA1C dropped from a baseline of 7.1% to 6.5% in the intensive group and to 7.2% in the standard care group. The results from the glucose-lowering arm (10% reduction in combined macro and microvascular events with no impact on mortality) was not as exciting as the BP-lowering arm. A combined analysis of the BP and glucose-lowering intervention showed that the best outcomes were seen in the group that received both intensive BP and glucose lowering, with a significant 18% reduction of total mortality.[13]

The first trial which specifically looked at the effects of different achieved levels of BP on active treatment was the ACCORD trial.[14] In this open-label trial, more than 4,733 diabetics with a baseline BP of 139/75 mmHg were randomised to an intensive arm (systolic BP <120 mmHg) or a standard therapy (systolic BP <140 mmHg). In the intensive arm, the achieved BP was 119/64 mmHg while the BP achieved on the standard arm was 134/71 mmHg. Except for stroke, there were no differences in clinical outcomes between the intensive and standard therapy. There was, however, significantly more serious adverse events with the intensive group. The lack of benefit from intensive BP control was corroborated by a subanalysis of a mega trial, INVEST,[15] which looked at 22,576 hypertensives with underlying ischaemic heart disease. Patients were randomised to receive either atenolol with a thiazide as the second drug, compared with trandolapril with verapamil as a second drug. In a separately published subanalysis of INVEST,[16] 6,321 diabetics were categorised into

those with achieved systolic BP of <130 mmHg (tight BP control), 130–139 mmHg (usual BP control), and >140 mmHg (uncontrolled BP). As expected, the uncontrolled group showed significantly higher events (death, myocardial infarction, and stroke). There was no difference between the control group and the tight group.

One of the largest CV trials to enrol diabetics is the ONTARGET trial.[17] In this study, 25,620 patients were randomised to ramipril, telmisartan, or a combination of both. Of these participants, 9,612 patients (37.5%) were diabetic. In the overall population, there was no difference in primary outcome between those randomised to the two different monotherapies, while those randomised to the combination had worse renal outcomes and more adverse events. In a *post hoc* analysis of the diabetic population, CV events were significantly higher than the non-diabetic population at each level of baseline or achieved on-treatment BP.[18] This *post hoc* analysis did not suggest a BP threshold, which may be potentially harmful to the diabetic population.

SHIFTING PARADIGM: CONSENSUS AND DIFFERENCES

The latest round of hypertension guidelines was published in 2011 by the National Institute of Health Care and Excellence (NICE) UK.[19] No specific recommendation was made for target BP in diabetics. However, reference was made to the NICE Diabetes Guideline which recommended a BP target of <140/80 mmHg.[20] The next published guideline was the European Society of Hypertension/European Society of Cardiology (ESH/ESC) guidelines in 2013.[21] The recommendation made by the ESH/ESC guidelines was a BP of <140/85 mmHg. This was followed by the Canadian Hypertension Education Programme (CHEP)[22] guidelines, the Japanese Society of Hypertension (JSH) guidelines,[23] and the Taiwan Society of Hypertension and Taiwan Society of Cardiology (TSOC) guidelines,[24] all of which recommended a target level of <130/80 mmHg. Three American-based guidelines including the American Heart Association/American College of Cardiology (AHA/ACC),[25] the Eighth Joint National Committee (JNC 8),[26] and the American Society of Hypertension/International Society of Hypertension (ASH/ISH)[27] all recommended a BP target of <140/90 mmHg. On the diabetic guideline front, until very recently, the American Diabetes Association (ADA) guidelines[28] concurred with British Diabetes NICE guidelines by recommending a BP target of <140/80 mmHg. The most recent ADA guideline of 2016, however, has revised the recommended BP for diabetics to a target of <140/90 mmHg.[29] There are therefore four different target BPs recommended to doctors by the different guidelines, as opposed to only one not so long ago (130/80 mmHg). These differing recommendations may leave practitioners confused; what makes it more perplexing is that the same evidence was quoted to justify the new recommendations.

WHY DIFFERENT RECOMMENDATIONS?

There are several reasons why this happened, the first of which was the particular selection of trials to provide the evidence-base. In some guidelines, studies were quoted only if it was primarily designed to test the hypothesis that separation of BP to pre-specified levels produces different outcomes. This was why JNC 8 did not accept the ADVANCE trial as evidence, even though the achieved diastolic BP in both the active and placebo treated groups in ADVANCE was <80 mmHg. JNC 8 argued that ADVANCE was not a hypertension study in the diabetic population because both hypertensives and normotensives were recruited. However, it is worth emphasising that the baseline BP in ADVANCE was 145/81 mmHg, which at that time was considered high for diabetics. Both ESH/ESC and JNC 8 quoted the ADVANCE trial but did not use it to justify their diastolic BP target recommendation (<85 mmHg in ESH/ESC and <90 mmHg in JNC 8). JNC 8's recommendation of BP <140/90 mmHg is based on expert opinion because none of the available studies were considered to be high level evidence, according to their strict criteria for grading of evidence. The AHA/ACC guidelines, meanwhile, do not quote primary data or studies in making its recommendation, but has made the decision to update its recommendation by reviewing all available evidence working together with the National Heart Lung Blood Institute (NHLBI). The update is due to be released in 2016.[30] The CHEP guidelines classified their recommendation for a systolic BP of <130 mmHg as Grade C while that for diastolic BP of <80 mmHg as Grade A evidence, but no reference was quoted. The HOT trial was quoted in the CHEP recommendation but was not used to justify this recommendation and the ACCORD, UKPDS, and

ADVANCE trials were not quoted. CHEP has not revised this recommendation in their subsequent yearly update.[31] The Canadian Diabetes Association (CDA) guidelines[32] meanwhile give the same recommendation for BP targets by quoting UKPDS, ABCD2, and HOT trials. Table 1 summarises the various recommendations thus far.

Another reason for the divergence of recommendations was the interpretation of the trial results. The ESH/ESC guidelines justified the diastolic BP target of <85 mmHg by quoting the UKDPS and HOT trials. These two trials, however, studied a small number of diabetic hypertensives and, strictly speaking, the recommendation was based on a subanalysis and is thus hypothesis-generating. The ASH/ISH meanwhile justified their recommendation by arguing that the previously recommended BP of <130/80 mmHg in diabetics lacks evidence and thus the goal of <140/90 mmHg should generally be used. The JNC 8 as mentioned above did not think any of the available evidence was good enough to be quoted and chose expert opinion for their recommendation.

The Japanese guidelines justified the decision to maintain the recommended BP target of <130/80 mmHg by quoting HOT, UKPDS, and the older recommendations from the ADA 2003, JNC 7, and the ESH/ESC 2007 guidelines. Meanwhile the Taiwanese Guideline justified their recommended target of <130/80 mmHg by highlighting the reality of the status of diabetes control in Taiwan, and quoted the Japanese guidelines target BP recommendation and the latest International Diabetes Federation (IDF) guidelines recommendation as supporting evidences.[33]

It is worth asking some basic questions of the interpretation of existing data from randomised control trials. While there is general agreement that available randomised control trials that specifically address the issue in question are lacking, there is obviously a lack of congruence in the interpretation, as discussed elsewhere by the author.[34] In the ADVANCE trial meanwhile, the achieved diastolic BP in the actively treated group was 75 mmHg compared with the control group which was 77 mmHg.

Table 1: Guidelines for blood pressure targets in diabetic hypertensives.

Guidelines	Year published	BP targets (mmHg)	Studies quoted
CHEP	2013, 2014, 2015	<130/80	Not specified
CDA	2013	<130/80	UKPDS, HOT, ABCD2
Japanese	2014	<130/80	UKPDS, HOT
Taiwan	2014	<130/80	UKPDS, HOT
Malaysian Diabetes	2015	<135/75	ADVANCE, ACCORD
NICE Diabetes and NICE Hypertension	2015 2011	<140/80	UKPDS, HOT
Malaysian Hypertension	2014	<140/80	ADVANCE
ADA	2015	<140/80	HOT
ESH/ESC	2013	<140/85	HOT, UKPDS
AHA/ACC	2014	<140/90	Not specified
ASH/ISH	2014	<140/90	Not specified
JNC 8	2014	<140/90	Expert opinion
ADA	2016	<140/90	HOT

CHEP: Canadian Hypertension Education Program; CDA: Canadian Diabetes Association; NICE: National Institute of Health Care and Excellence; ADA: American Diabetes Association; ESH: European Society of Hypertension; ESC: European Society of Cardiology; AHA: American Heart Association; ACC: American College of Cardiology; ASH: American Society of Hypertension; ISH: International Society of Hypertension; JNC 8: Eighth Joint National Committee; UKPDS: UK Prospective Diabetes Study; HOT: Hypertension Optimal Treatment; ADVANCE: Action in Diabetes and Vascular Disease: Preterax and Diamicron MR Controlled Evaluation; ACCORD: Action to Control Cardiovascular Risk in Diabetes.

However, despite the fact that the ADVANCE trial was the largest diabetic dedicated trial to look at the effects of antihypertensive therapy on clinical outcomes, it was not accepted by many guidelines as a trial to be quoted for targeting BP-lowering in diabetics. This was mainly because as a placebo controlled trial it did not compare active treatment regimens. Although in the HOT trial, the diabetic subpopulation on treatment diastolic BP of <85 mmHg did not have different clinical outcomes compared with those with targeted diastolic BP of <90 mmHg, there was a significant difference between those who were targeted to achieve diastolic BP of <80 compared with <90 mmHg. Why then was the diastolic BP target of <80 mmHg not recommended by the ESH/ESC, which quoted the HOT diabetic subanalysis as their justification for their recommendation? A possible explanation was that even in the HOT trial the actual mean achieved diastolic BP in the intensive treated group was slightly more than 80 mmHg, i.e. 81 mmHg. Meanwhile, the ADA has revised their 2015 guideline, recommending the target diastolic BP of <90 mmHg[29] as opposed to <80 mmHg the previous year. Justification given for this shift was that the earlier recommendation was based on *post hoc* analysis of the HOT trial and for this latest recommendation is consistent with that of JNC 8.

CONCLUSION AND FUTURE RECOMMENDATIONS

Recently recommended BP targets for diabetic hypertensives show significant variation and lead to confusion among readers and practitioners. Many of the recommendations made were based on subanalyses of big studies involving small sample sizes, and are therefore by definition not definitive evidence. With the recent publication of the SPRINT trial[35] and the reopening of the debate on optimum BP to be achieved in hypertensive patients, the time is right for an adequately powered and *a priori* hypothesis-testing study dedicated to the diabetic hypertensive population to be designed and executed. This is especially so because the SPRINT trial excluded patients with diabetes. This also means that the findings from this large study (which showed that clinical outcomes are significantly better with a target systolic BP of <120 compared with <140 mmHg) cannot be extrapolated to the diabetic population. The question as to the best target BP to aim for in the diabetic patient with hypertension will remain unanswered until a SPRINT-type study is carried out in the diabetic population. The SPRINT trial has triggered interesting debates among experts in hypertension with more questions being asked.[36-38] However, on a reassuring note, a recent subanalysis of the SPRINT trials on patients >75 years old reaffirms that lowering systolic BP to <120 mmHg leads to a significant reduction in fatal and non-fatal CV events, also significantly lowering all-cause mortality.[39] While waiting for further studies to provide a definitive answer to the question of target BP for the diabetic hypertensives population, it is important that guideline committees narrow the differing recommendations so as not to create more confusion among practitioners, patients, and policy makers alike. It is also very important that specific countries' hypertension and diabetes associations produce guidelines which concur with each other on their recommendations, as has happened in Canada and more recently in the USA.

In Malaysia the guidelines also differ; the hypertension clinical practice guidelines differ in their recommendations from the Malaysian Diabetes Association guidelines, with the former recommending the target of <140/80 mmHg[40] and the latter <135/85 mmHg.[41] In this author's view, what can be deduced from all the studies done so far is that attaining a BP on treatment as low as 135/75 mmHg (as achieved in the ADVANCE trial) is beneficial for major clinical important outcomes including CV outcomes and even all-cause mortality. Of equal importance, it is safe to lower BP to that level in patients with diabetes. The ADVANCE trial is also the most important and largest study so far looking at diabetic population and BP-lowering treatment. We hope that a critical study to address this issue will one day be conducted and there will be uniformity in future recommendations. In the meantime, it should be noted that BP control rates in diabetics remain poor even based on the latest surveys and systematic reviews. A recent Dutch study showed that rates of hypertension control among Dutch of African-Surinamese origin was only 28.7%, of Ghanaian Origin was 41.7%, and of ethnic Dutch origin was 54.1%.[42] A systematic review involving 25,629 diabetic hypertensives from 19 countries all conducted between 2009 and 2014 revealed a control rate of only 35.7%. The review noted that hypertension control rates were the worst compared with glycaemic control (44.5%) and

cholesterol control (51.4%).[43] There is obviously a lot more work to be done. It will help if future recommendations correspond with one another rather than remaining contradictory.

REFERENCES

1. World Health Organization, "Non Communicable diseases," Health in 2015: From MDG to SDG (2015), Geneva: WHO, pp. 131-50.

2. White WB et al. Alogliptin after acute coronary syndrome in patients with type 2 diabetes. N Engl J Med. 2013;369(14): 1327-35.

3. Scirica BM et al. Saxagliptin and cardiovascular outcomes in patients with type 2 diabetes mellitus. N Engl J Med. 2013;369(14):1317-26.

4. Rahman S et al. Early manifestation of macrovasculopathy in newly diagnosed never treated type II diabetic patients with no traditional CVD risk factors. Diabetes Res Clin Pract. 2008;80(2):253-8.

5. Rahman S et al. Increased arterial stiffness in normoglycaemic offspring of newly diagnosed, never treated type 2 diabetic and impaired glucose tolerance patients. Br J Diabetes Vasc Dis. 2009;9(2):65-8.

6. UK Prospective Diabetes Study Group. Tight blood pressure control and risk of macrovascular and microvascular complications in type 2 diabetes: UKPDS 38. Br Med J. 1998;317(7175):703-13.

7. Patel SA et al. Cardiovascular mortality associated with 5 leading risk factors: National and state preventable fractions estimated from survey data. Ann Intern Med. 2015;163(4):245-53.

8. Flores-Hernandez S et al. Quality of Care: The Challenges of an Increasing Epidemic in Mexico. Results from Two National Health Surveys (2006 and 2012). PLoS One. 2015;10(7):e0133958.

9. Gan D et al., "Diabetes Atlas" (2006) 3rd Edition, Cape Town: International Diabetes Federation.

10. Hansson L et al. Effects of intensive blood pressure lowering and low dose aspirin in patients with hypertension: Principal results of the Hypertension Optimal Treatment (HOT) randomised trial. Lancet. 1998;351(9118):1755-62.

11. Schrier RW et al. Effects of aggressive blood pressure control in normotensive type 2 diabetic patients on albuminuria, retinopathy and strokes. Kidney Int. 2002; 61(3):1086-97.

12. Patel A; ADVANCE Collaborative Group. Effects of a fixed combination of perindopril and indapamide on macrovascular and microvascular outcomes in patients with type 2 diabetes mellitus (the ADVANCE trial): A randomised controlled trial. Lancet. 2007;370(9590):829-40.

13. Zoungas S et al. Combined effects of routine blood pressure lowering and intensive glucose control on macrovascular and microvascular outcomes with type 2 diabetes. New results from the ADVANCE trial. Diabetes Care. 2009;32(11):2068-74.

14. The ACCORD Study Group. Effects of intensive blood-pressure control in Type 2 Diabetes Mellitus. N Engl J Med. 2010;362(17):1575-82.

15. Pepine CJ et al. A calcium antagonist vs a non-calcium antagonist hypertension treatment strategy for patients with coronary artery disease. The International Verapamil-Trandolapril Study (INVEST): A randomized controlled trial. JAMA. 2003;290(21):2805-16.

16. Cooper-DeHoff RM et al. Tight blood pressure control and cardiovascular outcomes among hypertensives with diabetes and coronary artery disease. JAMA. 2010;304(1):61-8.

17. The ONTARGET Investigators. Telmisartan, ramipril, or both in Patients at high risk for vascular events. N Engl J Med. 2008;358(15):1547-59.

18. Redon J et al. Safety and efficacy of low blood pressures among patients with diabetes: Subgroup analysis from the ONTARGET trial. J Am Coll Cardiol. 2012;59(1):74-83.

19. NICE. National Institute for Health and Excellence Clinical Guideline 127: Hypertension in adults: Diagnosis and management NICE guidelines (CG127). August 2011. Available at: http://publications.nice.org.uk/hypertension-cg127/guidance. Last Accessed: 28 February 2016.

20. NICE. National Institute for Health and Excellence Clinical Guideline 28: Type 2 diabetes in adults: management. 2 December 2015. Available at: nice.org.uk/guidance/ng28. Last accessed: 28 February 2016.

21. Mancia G et al. 2013 ESH/ESC Guidelines for the management of arterial hypertension: The task force for the management of arterial hypertension of the European Society of Hypertension (ESH) and the European Society of Cardiology (ESC). Eur Heart J. 2013; 34(28):2159-219.

22. Dasgupta K et al. The 2014 Canadian Hypertension Education Program Recommendation for Blood Pressure Measurement, Diagnosis, Assessment of Risk, Prevention and Treatment of Hypertension. Can J Cardiol. 2014;30(5): 485-501.

23. Lindholm LH, Carlberg B. The new Japanese Society of Hypertension Guidelines for the management of hypertension. Hypertens Res. 2014;37(4): 253-392.

24. Chiang CE et al. 2015 Guideline of the Taiwan Society of Cardiology and the Taiwan Hypertension Society for the management of hypertension. J Chin Med Assoc. 2014;78(1):1-47.

25. Go AS et al. An effective approach to high blood pressure control: A science advisory from the American Heart Association, the American College of Cardiology, and the Centers for Disease Control and Prevention. Hypertension. 2014;63(4):1116-35.

26. James PA et al. 2014 Evidence-Based Guideline for management of high blood pressure in adults. Report from the panel members appointed to the Eighth Joint National Committee (JNC 8). JAMA. 2014;311(5):507-20.

27. Weber MA et al. Clinical Practice Guidelines for the management of hypertension in the community, a statement by the American Society of Hypertension and the International Society of Hypertension. J Clin Hypertens. 2014;16(1):14-26.

28. Standards of Medical Care in Diabetes 2015. Diabetes Care. 2015;38(Suppl 1): 1-94.

29. Standards of Medical Care in Diabetes 2016. Diabetes Care. 2016;39(Suppl 1): 1-119.

30. ACC/AHA: Hypertension Management Guideline Update Underway. Available at: http://www.empr.com/news/acc-aha-hypertension-management. Last accessed: 6 February 2016.

31. The 2015 Canadian Hypertension Education Program. Recommendations for blood pressure, measurement, diagnosis, assessment of risk, prevention, and treatment. Can J Cardiol. 2015;31(5): 549-68.

32. Canadian Diabetes Association Clinical Practice Guidelines Expert Committee. Treatment of Hypertension.

Can J Diabetes. 2013;37(Suppl 1):S117-8.

33. International Diabetes Federation Guideline Development Group. Global guideline for type 2 diabetes. Diabetes Res Clin Pract. 2014;104:1-52.

34. ARA Rashid. Evidence based medicine: Are we on the same page? Bangladesh J Med Sci. 2014;13(2):110-3.

35. Wright JT Jr et al.; The SPRINT Research Group. A randomized trial of intensive versus standard blood-pressure control. N Engl J Med. 2015;373(22): 2103-16.

36. Jones DW et al. SPRINT: What remains unanswered and where do we go from here? Hypertension. 2016:67(2):261-2.

37. Esler M. SPRINT or false start: Toward a lower universal treated blood pressure target in hypertension. Hypertension. 2016;67(2):266-7.

38. Touyz RM, Dominiczak AN. Success of SPRINT; but still some hurdles to cross. Hypertension. 2016;67(2):268-9.

39. Williamson JD et al. Intensive vs standard blood pressure control and cardiovascular disease outcomes in adults aged > 75 years: A randomized clinical trial. JAMA. 2016. [Epub ahead of print].

40. ARA Rashid. Clinical Practice Guidelines. Management of Hypertension (4th Edition). Available at: http://www.moh.gov.my/attachments/CPG_Management_of_Hypertension_4th_Edition.pdf. Last accessed: 22 June 2016.

41. CPG Secretariat. Clinical Practice Guidelines Management of Type 2 diabetes (5th Edition). Available at: https://www.scribd.com/doc/311462750/QR-Management-of-Type-2-Diabetes-Mellitus-5th-Edition. Last accessed: 22 June 2016.

42. Fernald FF et al. Hypertension awareness, treatment and control among diabetes and non-diabetics individuals in a multiethnic population in the Netherlands: the HELIUS Study. J Hypertens. 2016;34(3):539-47.

43. Pinchevsky Y et al. Glycaemic, blood pressure and cholesterol control in 25,629 diabetics. Cardiovasc J Afr. 2015; 26(4):188-92.

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AUTOMATIC REPORTING OF CREATININE-BASED ESTIMATED GLOMERULAR FILTRATION RATE IN CHILDREN: IS THIS FEASIBLE?

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ABSTRACT

Creatinine, although widely used as a biomarker to measure renal function, has long been known as an insensitive marker of renal impairment. Patients with reduced renal function can have a creatinine level within the normal range, with a rapid rise when renal function is significantly reduced. As of 1976, the correlation between height, the reciprocal of creatinine, and measured glomerular filtration rate (GFR) in children has been described. It has been used to derive a simple formula for estimated glomerular filtration rate (eGFR) that could be used at the bedside as a more sensitive method of identifying children with renal impairment. Formulae based on this association, with modifications over time as creatinine assay methods have changed, are still widely used clinically at the bedside and in research studies to assess the degree of renal impairment in children.

Adult practice has moved in many countries to computer-generated results that report eGFR alongside creatinine results using more complex, but potentially more accurate estimates of GFR, which are independent of height. This permits early identification of patients with chronic kidney disease. This review assesses the feasibility of automated reporting of eGFR and the advantages and disadvantages of this in children.

Keywords: Estimated glomerular filtration rate (eGFR), chronic kidney disease (CKD), children, automated reporting of eGFR.

BACKGROUND

Creatinine, although widely used as a biomarker to measure renal function, has long been known as an insensitive marker of renal impairment.[1] Patients with reduced renal function can have a creatinine level within the normal range, with a rapid rise when renal function is significantly reduced. In 1976 Schwartz et al.[2] reported a correlation between height, the reciprocal of creatinine, and measured glomerular filtration rate (GFR) in children. This was used to derive a simple formula for estimated glomerular filtration rate (eGFR) that can be used at the bedside as a more sensitive method of identifying children with renal impairment. At a similar time, this association was also described by Counahan et al.[3] with an adaptation to allow use of SI units to measure creatinine. These formulae, which have been modified as creatinine assay methods have changed over time, are still widely used clinically at the bedside and in research studies to assess the degree of renal impairment in children.

With the advent of computer-generated results, there is the ability to report alongside creatinine results more complex but potentially more accurate estimates of GFR, including those which are independent of height. This would permit early identification of patients with chronic kidney disease (CKD), with evidence in adults demonstrating the benefits of this.[4] For this reason, in some countries, reporting an eGFR is mandated when a creatinine result is reported, with evidence of an increase in early referrals.[5,6]

During this review, I will assess the feasibility of automated reporting of eGFR and the advantages and disadvantages of using this in children. I will review the accuracy of established height-based formulae and more recently published height-independent formulae. After briefly discussing local optimisation of these formula and methods of calculating eGFR, which use other biomarkers (e.g. cystatin C), I will draw conclusions regarding the benefits and feasibility of automated eGFR reporting in children.

BENEFITS OF AUTOMATIC REPORTING OF ESTIMATED GLOMERULAR FILTRATION RATE

In adults, evidence-based strategies have been shown to prevent progression of CKD. In addition, CKD is a marker of increased risk of cardiovascular disease. Early detection of CKD permits early intervention to reduce the risk of cardiovascular mortality and progression to renal replacement therapy (RRT). Even in patients who progress to RRT, early detection (>12 months before onset of RRT) has been shown to reduce mortality rates. This rationale for population-based screening led to automated eGFR reporting in the UK in 2006 following publication of a National Service Framework document.[7] Automated reporting led to increased and earlier referrals,[6] although an evidence-based review suggests there is still a need for more research into the most effective interventions at early stages of CKD.[4]

The rationale for screening in children is less clear. There are multiple risk-factors for progression of CKD in children.[8] Many are amenable to treatment including conditions such as hypertension[9] and proteinuria.[10] The incidence of CKD, however, is much lower than in the adult population and the significance of anxiety caused by false positives is more of a concern. The incidence of CKD-related cardiovascular disease is also lower than in adults. However, studies have detected subclinical cardiovascular changes such as increased left ventricular mass,[11] which suggests that earlier detection and intervention may be of benefit.[12] Locally, in the Nottingham Children's Hospital, we introduced a report accompanying a creatinine result in children <18 years of age including instructions on how to calculate an eGFR based on local optimisation of the Schwartz formula. This was in response to late referrals of patients with CKD. This resulted in a moderate increase in referrals, but we have still received late referrals (unpublished). There are many theoretical benefits of early identification through automated eGFR reporting in children, though there is still a need for more research to determine the effectiveness of early interventions.

LIMITATIONS OF CREATININE-BASED ESTIMATED GLOMERULAR FILTRATION RATE

Both calculated and direct measurements of GFR have limitations that need to be considered when evaluating their use. Height-based eGFR has limitations in relation to accuracy, utility, and feasibility.

Any calculated GFR is an estimate and therefore has a degree of inaccuracy. The standard, used for adult based formulae, requires that 75% of eGFR values are within 30% of the measured GFR value.[13] When assessing the bedside height-independent formula in this way locally, we found that only 55% of measurements met this criterion.[14] This can be attributed to the changes in methods of creatinine measurement since the original studies were performed with a move towards enzymatic measurements or a corrected Jaffe measurement. This variation in creatinine measurement has been less of a concern since standardisation of creatinine methods was developed in order to facilitate automatic eGFR reporting in adults. The creatinine standardisation recommendations[15] by the National Kidney Disease Education (NKDEP) and Laboratory Working Group (LWG) Laboratory Working Group set isotope-dilution mass spectrometry (IDMS) as the reference for creatinine measurements. This significantly reduced variability in creatinine measurements between different methods and manufacturers.[16]

Height and creatinine-based eGFR formulae assume a linear relationship between height and the reciprocal of serum creatinine, although some studies have questioned this relationship.[17] Any factor that may influence this relationship will limit the accuracy of the estimate. This would include extremes of age (e.g. <1 year of age), malnutrition, and neuromuscular disorders.

eGFR formula have not been demonstrated to be reliable when altering medication doses in children with reduced renal function.[18,19] This is likely to be because the formulae are based on children with stable renal function. Children who are unwell and

require chemotherapy or antibiotics are unlikely to be stable and the equations are therefore less accurate. Where possible, drug monitoring should be used.

The derivation of many height and creatinine-based eGFRs in children has been completed in populations of patients with CKD and those post renal transplant. This has important implications for the wider application of the estimate. The calculations assume that creatinine levels are steady and less accurate in patients with acute kidney injury (AKI). Adjustments can be made to the equations to account for the rate of change of creatinine to improve the utility in this situation.[20] However, current AKI detection algorithms remain creatinine-based.[21]

Finally, the limitation of height-based formulae that is probably most significant is the need for an accurate height to be included in the data given to the laboratory that is measuring the creatinine assay. This relies on the clinician recording this data when requesting the blood test, laboratory teams or systems to input this data, and the systems available to perform the calculation. Feedback from local paediatric teams regarding a project to report the height-based formulae with creatinine measurements in children <18 years old included clinician difficulty in having a recent height available when requesting or interpreting the result (unpublished).

Table 1: Formulae discussed in this paper and accuracy expressed as a percentage (%) of values within 30% of reference glomerular filtration rate (P30).

Formulae (publication)	*eGFR (mL/min per 1.73m^2)	P30 (%)	Summary of study conclusions
Height-based formula[2,3]	=40 × height (cm)/SCr(µmol/L)	55	Measured GFR is linearly related to height divided by serum creatinine
Nottingham - optimised height-based formula[14] (Lunn et al. 2015)	=k × height (cm)/SCr(µmol/L) Where k=36 in males ≥13 years of age and k=30 in all other cases	79	Local optimisation improves performance of height-based formula
Belgium–Pottel[27,29] (Pottel et al. 2012)	=107.3/(SCr/Q) with: Q=88.4 × (0.21 + 0.057 × Age − 0.0075 × Age^2 + 0.00064 × Age^3 − 0.000016 × Age^4) for males Q=88.4 × (0.23 + 0.034 × Age − 0.0018 × Age^2 + 0.00017 × Age^3 − 0.0000051 × Age^4) for females	79.6	Equation comparable to the updated Schwartz formula
Lyon–Pottel[33] (De Souza et al. 2015)	107.3/(SCr/Q) with Q=Lyon derived median of PCR of healthy children at a specific age	87	The height-independent equation, with or without an adaptation to the local laboratory, could be used as a screening tool in a general population
BCCH2[24] (Mattman et al. 2006)	=-61.56 + [5886 × [1/SCr(µmol/L)] + [4.83 × Age] + 10.02 (if male)	Not reported	With local derivation of constants, the formula can be used where height is not available.
Modified BCCH2[25] (Zappitelli et al. 2010)	=Inverse ln of: 8.067 + (1.034 × ln[1/SCr(µmol/L)] + (0.305 × ln[age]) + 0.064 if male	74.2–80.9	Height-independent formula with local optimisation performed equally well when compared to the Schwartz formula
Nottingham - optimised Modified BCCH2[14] (Lunn et al. 2015)	=Inverse ln of: 6.064 + (0.554 × ln[1/SCr(µmol/L)]) + (0.254 × ln[age]) + 0.025 if male	85	Height-independent formula with local optimisation performed equally well when compared to the locally-optimised Schwartz formula

*all ages in years
eGFR: estimated glomerular filtration rate; PCR: protein/creatinine ratio; SCr: serum creatinine.

CURRENT HEIGHT-INDEPENDENT CREATININE-BASED ESTIMATED GLOMERULAR FILTRATION RATE FORMULAE IN CHILDREN

The advantage of height-independent creatinine-based eGFR formulae is that no extra data are required in the laboratory to allow reporting whenever creatinine is measured.[22] There is theoretical evidence that this is plausible as uncorrected GFR has a linear relationship with age between the ages of 2 and 16 years.[23] This has led to the development of height-independent formulae, the majority of which use creatinine because of the wide availability within clinical laboratories.

In 2006, Mattman et al.[24] derived two height-independent formulae (Table 1). The first (BCCH2) was a derived formula based on patients undergoing ^{99m}Tc-DTPA (diethylene triamine pentaacetic acid) GFR measurement and creatinine measured by enzymatic method. The aim of the study was to assess the sensitivity and specificity of an estimate to accurately identify patients with a GFR <60 mL/min/1.73 m^2. They also derived a method to determine creatinine cut-offs based on age and gender to serve this purpose. This performed comparably to a locally optimised Schwartz formula with a Pearson correlation coefficient of 0.72 compared with 0.83 for the Schwartz formula. The cut-off based method was also comparable with a sensitivity of 86% and specificity of 93% for the detection of a GFR <60 mL/min/1.73 m^2 (BCCH2 – sensitivity 79%, specificity 99%, locally optimised Schwartz – sensitivity 86%, specificity 97%).

The BCCH2 formula was modified in 2010 by Zappitelli et al.[25] using local data based on iothalamate GFR and enzymatic creatinine measurements in a population of paediatric patients with renal disease pre and post-transplant. Measurements were taken on up to three occasions over a median period of 1.1 years and the ability of the estimates to assess changes in measured GFR over time. The modified formula was comparable to the Schwartz formula with 75.4%, 74.2%, and 80.9% of values within 30% of measured GFR over the three different time periods compared to 76.4%, 82.8%, and 85.1% for the Schwartz formula.

Attempts have been made to apply adult derived formulae to children, the majority without a sufficient degree of accuracy. One exception may be the Lund–Malmö formula. This was originally derived in adults[26] reporting 84% of values within 30% of a measured GFR. This was assessed in a paediatric population of renal and oncology patients (pre-chemotherapy) with an iohexol GFR used as a reference and an enzymatic method used to measure creatinine. Again this was comparable to height-based formulae with 76% of values within 30% of measured GFR compared to 74% in a locally optimised Counahan–Barratt height-based estimate.

Most recently, a height-independent formula was developed based on the concept of a population-normalised serum creatinine by Pottel et al.[27] This used a ^{51}Cr-EDTA GFR as a reference. The serum creatinine measurement was obtained with an enzymatic method in children <5 years of age and a compensated Jaffe assay in children ≥5 years of age in a population of children with renal disease. They reported that 72.8% of values were within 30% of the measured GFR. This was comparable to the 69.4% for the Schwartz formula and better than the previously described height-independent measurements, although this data was not published.[28] A later publication[29] described an age-based formula for derivation of the population-normalised serum creatinine.

There are therefore a number of candidates for estimating GFR in children that are as accurate as the currently used height-based methods. They could be used to report eGFR automatically in children. The majority are tested in a population of children who have renal disease, and a concern would be that they may not be as accurate in a general paediatric population leading to an increase in false positives. The number of false positives is dependent on the definition of the lower limit of normal. Pottel et al.[30] have used large population based data to define a cut-off of 75 mL/min/1.73 m^2. This is at a higher level than recommended in adults, where in the absence of other evidence of renal disease an eGFR of 60 mL/min/1.73 m^2 is used.[31]

Local Optimisation of Results

A method often used is to apply formulae derived externally to internally verify them, and then optimise the constants to improve the accuracy of the formulae. Different methods of creatinine measurement (enzymatic, uncompensated Jaffe assay, or compensated Jaffe assay) have historically

been incomparable. Local optimisation allows adjustment for local methods of creatinine assessment. Table 2 describes different formula derived by local optimisation of the Schwartz formula and compares them with the local creatinine method used. Centres using the same creatinine method derived similar values of k.

We have done this locally in Nottingham Children's Hospital, using the Solver function of Microsoft Excel to optimise the constant k in the formula: k × height (cm)/creatinine (µmol/L) to minimise the sum of the squares of the differences. This produced a k of 36 for males 13 years or older and a k-value of 30 in all other situations.[32] We verified our data following a change in method of creatinine assay and found that the optimised formula was of similar efficacy with 49% of patients within 30% of measured GFR using the Schwartz formula (k=40) and an enzymatic assay, 55% of patients within 30% of measured GFR using the Schwartz formula (k=40) and a compensated Jaffe assay, 72% of patients within 30% of measured GFR using the locally optimised formula and an enzymatic assay, and 79% of patients within 30% of measured GFR using the locally optimised formula and a compensated Jaffe assay.[14,32] This also demonstrates the benefits of the use of the IDMS as a reference for creatinine assays.

This method has not been widely applied to height-independent eGFR formulae in children. Zappitelli et al.[25] modified the BCCH2 formula[24] using a natural logarithmic transformation rather than simple optimisation of the original formula. We applied our original optimisation technique to the Zappitelli formula and altered the constants to minimise the sum of the squares of the differences. In our population, this improved the performance of the equation from 50% of values within 30% of the measured GFR to 85%.[14]

The Pottel formula is unique in that they have introduced the concept of a Q value as a population-normalised serum creatinine.[27] They have defined a method for calculating Q values at different ages and for different genders. This value is based on normal values from their own laboratory and hence could be applied to other laboratories by calculating the local median serum creatinine for different age and gender which will be specific to the local population. This was done in Lyon and demonstrated an improvement in the accuracy of the equation from 79.6% of values within 30% of the measured GFR to 87%.[33]

Table 2: Height-based formulae and local optimisation with the method of serum creatinine assay and reference GFR.

Publication	Patient population	Age range (years)	Gender (% male)	GFR reference method	SCr	K (SCr units: µmol/L)
Schwartz et al.[2] (1976)	CKD	0.5–20	Not reported	Creatinine	Jaffe	48.6
Counahan et al.[3] (1976)	CKD	2–14	Not reported	^{51}Cr-EDTA	Jaffe (compensated)	38.0
Morris et al.[41] (1982)	CKD & normal	2–14	Not reported	^{51}Cr-EDTA	Jaffe (compensated)	40.0
Schwartz et al.[42] (1985)	CKD	3–21	63%	Creatinine	Jaffe	Males >13 years, 61.9 Others, 48.6
Vachvanichsanong et al.[43] (2003)	CKD & normal	0–19	Not reported	Creatinine	Jaffe (compensated)	41.1
Hellerstein et al.[44] (2004)	CKD	4–21	48%	Creatinine (cimetidine)	Jaffe	Males >13 years, 52.2 Others, 44.2
van Rossum et al.[45] (2005)	Tx	4–20	68%	Inulin	Enzymatic	41.2
Schwartz et al.[46] (2009)	CKD	1–16	61%	Iohexol	Enzymatic	36.5

Cr-51 EDTA: chromium-51 ethylene diamine tetracetic acid; CKD: chronic kidney disease; SCr: serum creatinine assay; GFR: glomerular filtration rate; Tx: treatment.

Table 1 summarises the main formulae discussed in this review and the accuracy as assessed by the percentage of values within 30% of the reference GFR.

ALTERNATIVES TO CREATININE-BASED ESTIMATED GLOMERULAR FILTRATION RATE IN CHILDREN

Other height-independent formulae have been derived based on cystatin C either in isolation or in combination with other measurements such as creatinine. They have been shown to be comparable to a local optimised Schwartz formula with 81–91% of values within 30% of the measured GFR[34] when tested using a compensated Jaffe assay and compared to an inulin-measured GFR. Some of these formulae also require height or weight. The use of cystatin C is promising in increasing the accuracy of eGFR measurements and has been recommended in adults,[31] particularly if the creatinine based eGFR is <60 mL/min/1.73 m^2.

NGAL (neutrophil gelatinase-associated lipocalin) has been used as a urine[35] and serum biomarker[36] for AKI in children. Serum NGAL has been shown to correlate with measured GFR, particularly at lower levels of GFR.[37] Beta-trace protein and beta-2 microglobulin have also been suggested as possible candidates for serum biomarkers of GFR but these require further research. They have not been shown to be superior to the Schwartz eGFR.[38,39] A significant limitation of these methods is that they are not yet available in all institutions.

CONCLUSION

In conclusion, automated eGFR reporting in children is now feasible with similar accuracy to currently used adult equations. It would meet accepted criteria[40] for a screening tool for the detection of CKD whenever any child has a serum creatinine measurement. An abnormal eGFR result should therefore prompt a thorough clinical evaluation of the patient and be interpreted in the light of the clinical findings. (The 'e' indicating 'estimated' should never be forgotten).

I suggest that automated eGFR should be introduced for children in conjunction with guidelines for the clinical evaluation of children who are noted to have a reduced eGFR. Either the Pottel formula[27] or the modified BCCH2 formula[25] could be used. Some studies without local optimisation favour the Pottel formula,[22,23] hence local optimisation should be used wherever possible.[14,33] It should be carefully evaluated to note the effect on early detection of CKD, the number of false positives, and the implications for service provision related to increased referrals. This will allow earlier intervention of treatments that have been shown to reduce the rate of progression of renal disease. More research is still required to determine the most effective interventions and their impact on the long-term outcomes in young children with CKD.

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REFERENCES

1. Levey AS et al. Serum Creatinine and Renal Function. Annu Rev Med. 1988;39:465-90.
2. Schwartz GJ et al. A simple estimate of glomerular filtration rate in children derived from body length and plasma creatinine. Pediatrics. 1976;58(2):259-63.
3. Counahan R et al. Estimation of glomerular filtration rate from plasma creatinine concentration in children. Arch Dis Child. 1976;51(11):875-8.
4. Black C et al. Early referral strategies for management of people with markers of renal disease: a systematic review of the evidence of clinical effectiveness, cost-effectiveness and economic analysis. Health Technol Assess. 2010;14(21):1-184.
5. Noble E et al. The impact of automated eGFR reporting and education on nephrology service referrals. Nephrol Dial Transplant. 2008;23(12):3845-50.
6. Richards N et al. The impact of population-based identification of chronic kidney disease using estimated glomerular filtration rate (eGFR) reporting. Nephrol Dial Transplant. 2008;23(2):556-61.
7. Department of Health. National service framework for renal services—part two. Chronic Kidney Disease, Acute Renal Failure and End of Life Care. Available at: http://webarchive.nationalarchives.gov.uk/20130107105354/http://www.dh.gov.uk/prod_consum_dh/groups/

dh_digitalassets/@dh/@en/documents/digitalasset/dh_4102680.pdf. Last accessed: 1 December 2015.

8. Staples AO et al. Association Between Clinical Risk Factors and Progression of Chronic Kidney Disease in Children. Clin J Am Soc Nephrol. 2010;5(2):2172-9.

9. ESCAPE Trial Group. Strict blood-pressure control and progression of renal failure in children. N Engl J Med. 2009;361(17):1639-50.

10. Ardissino G et al. Proteinuria as a predictor of disease progression in children with hypodysplastic nephropathy. Data from the Ital Kid Project. Pediatr Nephrol. 2004;19(2):172-7.

11. Sinha MD et al. Blood pressure control and left ventricular mass in children with chronic kidney disease. Clin J Am Soc Nephrol. 2011;6(3):543-51.

12. Copelovitch L et al. Insights from the Chronic Kidney Disease in Children (CKiD) study. Clin J Am Soc Nephrol. 2011;6(8):2047-53.

13. Swedish Council on Health Technology Assessment. Methods to Estimate and Measure Renal Function (Glomerular Filtration Rate); A Systematic Review. Available at: http://www.sbu.se/upload/Publikationer/Content1/1/Renal_Function.pdf. Last accessed: 1 December 2015.

14. Lunn A et al. A Locally Verified Height Independant Formula For Estimating Glomerular Filtration Rate (eGFR) In Children - Is Automatic eGFR Reporting In Children Feasible? Nephrol Dial Transplant. 2015;30(Supp 3):143.

15. Myers GL et al. Recommendations for improving serum creatinine measurement: a report from the Laboratory Working Group of the National Kidney Disease Education Program. Clin Chem. 2006;52(1):5-18.

16. Jones GRD. Adventures with Creatinine and eGFR - A National, International and Personal Story – AACB Roman Lecture 2014. Clin Biochem Rev. 2015;36(2):75-82.

17. Davies JG et al. Clinical limitations of the estimation of glomerular filtration rate from height/plasma creatinine ratio: a comparison with simultaneous 51Cr edetic acid slope clearance. Arch Dis Child. 1982;57:607-10.

18. Alford EL et al. Glomerular filtration rate equations do not accurately predict vancomycin trough concentrations in pediatric patients. Ann Pharmacother. 2014;48(6):691-6.

19. Bernhardt MB et al. Agreement among measurements and estimations of glomerular filtration in children with cancer. Pediatr Blood Cancer. 2015;62(1):80-4.

20. Chen S. Retooling the creatinine clearance equation to estimate kinetic GFR when the plasma creatinine is changing acutely. J Am Soc Nephrol. 2013;24(6):877-88.

21. National Institute for Health and Care Excellence. Acute kidney injury: prevention, detection and management. Available at: http://nice.org.uk/guidance/cg169. Last accessed: 1 December 2015.

22. Blufpand HN et al. Height-independent estimation of glomerular filtration rate in children: an alternative to the Schwartz equation. J Pediatr. 2013;163(6):1722-7.

23. Pottel H et al. On the relationship between glomerular filtration rate and serum creatinine in children. Pediatr Nephrol. 2010;25(5):927-34.

24. Mattman A et al. Estimating Pediatric Glomerular Filtration Rates in the Era of Chronic Kidney Disease Staging. J Am Soc Nephrol. 2006;17(2):487-96.

25. Zappitelli M et al. Estimating Glomerular Filtration Rate in Children at Serial Follow-up When Height Is Unknown. Clin J Am Soc Nephrol. 2010;5:1763-9.

26. Björk J et al. Prediction of relative glomerular filtration rate in adults: New improved equations based on Swedish Caucasians and standardized plasma-creatinine assays. Scandinavian Journal of Clinical and Laboratory Investigation. 2007;67(7):678-95.

27. Pottel H et al. A simple height-independent equation for estimating glomerular filtration rate in children. Pediatr Nephrol. 2012;27(6):973-9.

28. Nyman U et al. The Lund-Malmö creatinine-based glomerular filtration rate prediction equation for adults also performs well in children. Scand J Clin Lab Invest. 2008;68(7):568-76.

29. Hoste L et al. A new equation to estimate the glomerular filtration rate in children, adolescents and young adults. Nephrol Dial Transplant. 2014;29(5):1082-91.

30. Pottel H et al. Abnormal glomerular filtration rate in children, adolescents and young adults starts below 75 mL/min/1.73 m(2). Pediatr Nephrol. 2015;30(5):821-8.

31. National Institute for Health and Care Excellence. Chronic kidney disease in adults: assessment and management. Available at: https://www.nice.org.uk/guidance/cg182. Last accessed: 10 March 2015.

32. Hay PD, McCrear D. Optimisation of estimated glomerular filtration rate in paediatric patients. Nucl Med Commun. 2012;33:1103-4.

33. De Souza V et al. Can the height-independent Pottel eGFR equation be used as a screening tool for chronic kidney disease in children? Eur J Pediatr. 2015;174:1225-35.

34. Bacchetta J et al. Which Creatinine and Cystatin C Equations Can Be Reliably Used in Children? Clin J Am Soc Nephrol. 2011;6(3):552-60.

35. Zwiers AJ et al. Reference intervals for renal injury biomarkers neutrophil gelatinase-associated lipocalin and kidney injury molecule-1 in young infants. Clin Chem Lab Med. 2015;53(8):1279-89.

36. Dent CL et al. Plasma neutrophil gelatinase-associated lipocalin predicts acute kidney injury, morbidity and mortality after pediatric cardiac surgery: a prospective uncontrolled cohort study. Crit Care. 2007;11(6):R127.

37. Mitsnefes MM et al. Serum neutrophil gelatinase-associated lipocalin as a marker of renal function in children with chronic kidney disease. Pediatr Nephrol. 2007;22(1):101-8.

38. Filler G et al. Beta-trace protein, cystatin C, beta(2)-microglobulin, and creatinine compared for detecting impaired glomerular filtration rates in children. Clin Chem. 2002;48(5):729-36.

39. Benisty K, Zappitelli M. Measures of GFR in Health and Disease. Current Pediatric Reports, 2015;3(1):101-10.

40. Wilson JMG, World Health Organization. Principles and Practice of Screening for Disease. Available at: http://www.who.int/iris/handle/10665/37650. Last accessed: 1 December 2015.

41. Morris MC et al. Evaluation of a height/plasma creatinine formula in the measurement of glomerular filtration rate. Arch Dis Child. 1982;57(8):611-5.

42. Schwartz GJ, Gauthier B. A simple estimate of glomerular filtration rate in adolescent boys. J Pediatr. 1985;106(3):522-6.

43. Vachvanichsanong P et al. Simple estimation of the glomerular filtration rate in sick Thai children. Nephrology (Carlton). 2003;8(5):251-5.

44. Hellerstein S et al. Comparison of two formulae for estimation of glomerular filtration rate in children. Pediatr Nephrol. 2004;19(7):780-4.

45. van Rossum LK et al. Estimation of the glomerular filtration rate in children: which algorithm should be used? Pediatr Nephrol. 2005;20(12):1769-75.

46. Schwartz GJ et al. New equations to estimate GFR in children with CKD. J Am Soc Nephrol. 2009;20(3):629-37.

47. De Souza VC et al. Schwartz formula: is one k-coefficient adequate for all children? PLoS One. 2012;7(12):e53439.

FRACTIONAL EXCRETION OF SURVIVIN, EXTRACELLULAR MATRIX METALLOPROTEINASE INDUCER, AND MATRIX METALLOPROTEINASE 7 IN CHILDREN WITH CHRONIC KIDNEY DISEASE

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ABSTRACT

Background: Epithelial–mesenchymal transition (EMT) is defined as a transformation of tubular epithelial cells into mesenchymal ones. These cells migrate through the extracellular matrix and change into active myofibroblasts, which are responsible for excessive matrix deposition. Such changes may lead to tubular dysfunction and fibrosis of the renal parenchyma, characteristic of chronic kidney disease (CKD). However, there are no data on potential EMT markers in children with CKD. The aim of our study was to assess the usefulness of fractional excretion (FE) of survivin, E-cadherin, extracellular matrix metalloproteinase inducer (EMMPRIN), matrix metalloproteinase (MMP)7, and transforming growth factor beta 1 (TGF-β1) as potential markers of CKD-related complications such as tubular damage and fibrosis.
Methods: Forty-one pre-dialysis children with CKD Stages 3–5 and 23 age-matched controls were enrolled in the study. The serum and urine concentrations of analysed parameters were assessed by an enzyme-linked immunosorbent assay test.
Results: Tubular reabsorption of all analysed parameters was >99% in the control group. All FE values rose significantly in children with CKD, yet they remained <1% in the case of E-cadherin and TGF-β1. The highest FE values in CKD children were those of survivin, EMMPRIN, and MMP7: >1%.
Conclusions: FE of the examined markers may become a useful tool in the assessment of tubular dysfunction during the course of CKD. The FE of survivin, EMMPRIN, and MMP7 warrant further research as potential independent markers of kidney-specific EMT.

Keywords: Epithelial–mesenchymal transition (EMT), fibrosis, tubular damage, E-cadherin, transforming growth factor beta 1 (TGF-β1).

INTRODUCTION

Renal interstitial fibrosis is the final common pathway in chronic kidney disease (CKD), irrespective of its primary cause.[1,2] The origin of myofibroblasts, which play a pivotal role in renal fibrogenesis, is under debate.[3,4] Despite conflicting evidence, the epithelial–mesenchymal transition (EMT) is still considered one of the possible mechanisms responsible for the appearance of fibrotic changes.[5-7] During EMT, epithelial cells lose their epithelial characteristics and gain mesenchymal features.[2,5-7] The newly formed mesenchymal cells migrate through the extracellular matrix and change into the active myofibroblasts, responsible for excessive matrix deposition and the subsequent fibrosis progression.[1,2,6,7] The ongoing discussion suggests that this process may be reversible, and could be a sign of immense kidney cell plasticity, enabling regression of fibrosis.[8,9]

Transforming growth factor beta 1 (TGF-β1) is the main EMT player and the master regulator of fibrosis, triggering early hypertrophy, apoptosis, and the atrophy of tubular epithelial cells and their transdifferentiation in order to gain the phenotype characteristic of matrix-producing myofibroblasts.[10,11] The *in vitro* models and clinical studies have

proven the essential role of TGF-β1 in fibrosis, since the administration of anti-TGF-β antibodies has resulted in the reduction of renal injury and fibrosis.[12] However, recent data show that TGF-β1 alone is responsible for a reversible transition of tubular cells, whereas additional exposure to Type I collagen, the most abundant extracellular matrix protein in renal interstitium, makes this transdifferentiation complete.[13] EMT is thus one of the trigger factors in fibrosis, whereas the irreversible parenchymal damage is a finely tuned process, strengthened by the *in situ* conditions.

The limits of matrix metalloproteinase (MMP) engagement in EMT and renal fibrosis have primarily been drawn as far as the regulation of extracellular matrix content and tissue remodelling. However, recent studies have revealed that the MMP influence may be more significant as they can have pro-fibrotic functions through EMT induction.[14,15] MMP7 occupies a unique position among metalloproteinases. It promotes renal fibrosis through its proteolytical activity in two ways. Firstly, it triggers EMT by shedding E-cadherin, and then aggravates apoptosis through Fas ligand cleavage.[16] Moreover, urinary MMP7 is a biomarker of the pro-fibrotic Wnt/β-catenin activity in the kidney.[17]

The extracellular matrix metalloproteinase inducer (EMMPRIN) also interacts with the fibrotic signalling pathways.[18] Such correlation has been confirmed by our preliminary study concerning children with CKD.[19] E-cadherin is an adhesion molecule released into the circulation as a consequence of the cell-cell detachment.[20-22] Anoikis, a specific form of apoptosis, enables the elimination of these cells thus preventing the reattachment in an inappropriate location and the formation of metastasis.[20,22] The loss of E-cadherin expression, resulting in the molecule accumulation in serum, is a hallmark of EMT, strictly connected with the resistance to anoikis.[20,22] It has scarcely been analysed in the patients with CKD.[23] Survivin is another protein acting in an anti-apoptotic way, enabling the rescue from anoikis through the activation of nuclear factor kappa B.[24] Recent interest has transformed the potential nephrological engagement of survivin, revealing the paramount importance of its expression in mice undergoing the renal proximal tubule recovery after acute kidney damage.[25] However, the role of survivin has not yet been assessed in patients with CKD except for in our preliminary results concerning children.[23]

The idea of analysing the fractional excretion (FE) of various parameters as a substitute of tubular dysfunction is novel in CKD patients and it has been historically used for assessing phosphate metabolism.[26] The aim of this study was therefore to analyse both known and new molecules engaged in EMT, resulting in tubular dysfunction and subsequent fibrosis. We have assessed the FE of survivin, E-cadherin, EMMPRIN, active MMP7, and TGF-β1 in children with CKD Stages 3–5 and in a control group. We have also analysed the potential relations between these parameters and their applicability as markers of CKD-related phenomena such as tubular damage and fibrosis.

METHODS

Patient Characteristics

Sixty-four patients enrolled in the study were divided into two groups. The basic demographic and biochemical data are given in Table 1. The first group consisted of 41 children with CKD Stages 3–5 (17 girls, 24 boys; median age 11 years, interquartile range 4–17 years) treated conservatively (median glomerular filtration rate of 26 mL/min/1.73 m^2, calculated according to the Schwartz formula).[27] The diseases leading to CKD were: reflux nephropathy (n=19), chronic glomerulonephritis (n=10), chronic pyelonephritis (n=6), polycystic kidney disease (n=4), and haemolytic uraemic syndrome (n=2). Twenty-three children (13 girls, 10 boys; median age 10.5 years, range 5–16.5 years) with primary nocturnal enuresis and normal kidney function served as controls.

None of the patients had clinical evidence of infection, diabetes, malignancies, or vasculitis, smoked, or took antibiotics or statins. None of the patients had been treated with corticosteroids or immunosuppressive therapy for at least 12 months. The patients were also free of such comorbidities as cardiovascular disease, peripheral vascular disease, or obesity. In the CKD group, 10 children were normotensive according to the criteria of the European Society of Hypertension (ESH) in children and adolescents.[28] In 31 patients with CKD, blood pressure was well controlled with the use of angiotensin converting enzyme inhibitors (14 children), calcium channel blockers (10 patients), or beta blockers (3 children); 4 patients needed combined therapy. In all CKD patients, phosphate binders and vitamin D metabolites were supplemented.

Table 1: Patient characteristics.

Parameter	Median values (Interquartile range)	
	Control group (n=23)	CKD (n=41)
Age (years)	10.5 (5.0–14.5)	11.0 (4.0–17.0)
Gender	13 female, 10 male	17 female, 24 male
eGFR (mL/min/1.73m^2)	105.0 (97.0–112.3)*	26.0 (16.8–38.0)
Urea (mg/dL)	32.0 (25.5–37.0)*	77.0 (55.0–94.5)
Albumin (g/dL)	NA	4.3 (3.8–4.5)
Haemoglobin (g/dL)	12.8 (11.7–13.9)*	11.2(10.5–12.2)
Parathormone (pg/mL)	NA	125.0 (46.1–223.0)
hsCRP (mg/L)	0.5 (0.24–1.34)	0.6 (0.18–1.37)
Proteinuria (g/L)	0.01 (0.0–0.1)*	0.4 (0.03–0.6)

*Mann–Whitney U test: p<0.001 control group versus CKD.
CKD: chronic kidney disease; eGFR: estimated glomerular filtration rate; hsCRP: high sensitivity C-reactive protein; NA: not assessed.

$$\frac{(\text{Parameter urine concentration})\times(\text{serum creatinine concentration})}{(\text{Parameter serum concentration})\times(\text{urine creatinine concentration})} \times 100$$

Equation 1: Calculation for fractional parameter excretion.

Informed consent was obtained from the subjects, and their parents if necessary. The research project was approved by the university ethics committee, in accordance with the Helsinki Declaration. Blood samples were drawn from peripheral veins after an overnight fast. Samples were clotted for 30 minutes, centrifuged at room temperature for 10 minutes, and then serum was stored at -20°C until assayed. Urine was collected aseptically from the first morning sample, centrifuged at room temperature for 10 minutes, and then stored at -20°C until assayed.

Assay Characteristics

The serum and urine concentrations of survivin (molecular mass 16.5 kDa), E-cadherin (120 kDa), EMMPRIN (35–65 kDa), MMP7 (25 kDa), and TGF-β1 (25 kDa) were evaluated by the commercially available Quantikine® Human Immunoassay ELISA (enzyme-linked immunosorbent assay) kits (survivin: R&D Systems, reagent kit DSV00; E-cadherin: R&D Systems, reagent kit DCADE0; EMMPRIN: R&D Systems, reagent kit DEMP00; MMP7: R&D Systems, reagent kit DMP700; TGF-β1: R&D Systems, reagent kit DB100B). Standards, serum, and urine samples were transferred to 96-well microplates pre-coated with recombinant antibodies to human survivin, E-cadherin, EMMPRIN, MMP7, and TGF-β1. Measurements were performed according to the manufacturer's instructions and results were calculated by reference to standard curves.

The serum and urine creatinine were assessed with the Beckman Coulter® Creatinine (enzymatic) OSR61204 Reagent on the AU2700 Chemistry Analyzer™. High sensitivity C-reactive protein was assessed by immunonephelometry, with Siemens CardioPhase® high sensitivity C-reactive protein reagent, on the BN™ II System analyser. The fractional parameter excretion was calculated according to the formula in Equation 1.

Statistical Analysis

The results are expressed as median values and interquartile ranges. Since the null hypothesis of normality of distribution was rejected by the Shapiro–Wilk test, comparisons in pairs were evaluated by using nonparametric tests (Mann–Whitney U). Relations between parameters were defined by Pearson's correlation coefficient r and additionally pictured by cluster analysis.

Table 2: The serum and urine concentrations, and fractional excretion values of analysed parameters in children with chronic kidney disease and controls.

Analysed Parameters	Median values (Interquartile range)	
	Control group (n=23)	CKD (n=41)
Serum survivin (ng/mL)	44.40 (40.42–47.97)*	98.51 (88.19–107.13)
Serum E-cadherin (ng/mL)	31.45 (30.45–32.68)*	98.50 96.34–103.58)
Serum EMMPRIN (pg/mL)	871.93 (854.86–906.07)*	1175.03 (1150.55–1211.54)
Serum MMP7 (ng/mL)	2.23 (2.17–2.91)*	2.97 (2.27–3.05)
Serum TGF-β1 (ng/mL)	1221.99 (1195.0–1242.9)*	1738.88 (1717.61–1760.18)
Serum creatinine (mg/mL)	0.69 (0.64–0.76)*	2.55 (1.3–3.7)
Urine survivin (ng/mL)	41.49 (38.21–45.42)*	86.50 (80.85–93.84)
Urine E-cadherin (ng/mL)	3.34 (3.17–3.42)*	6.54 (6.30–6.67)
Urine EMMPRIN (pg/mL)	394.39 (375.02–413.56)*	800.42 (767.25–849.63)
Urine MMP7 (ng/mL)	1.05 (1.02–1.11)*	2.36 (2.29–2.38)
Urine TGF-β1 (ng/mL)	48.37 (45.96–49.98)*	195.10 (192.39–199.62)
Urine creatinine (mg/dL)	114.00 (100.00–126.00)*	74.97 (60.0–82.0)
FE survivin (%)	0.73 (0.58–0.75)*	1.99 (1.46–3.30)
FE E-cadherin (%)	0.07 (0.06–0.07)*	0.16 (0.10–0.28)
FE EMMPRIN (%)	0.31 (0.28–0.31)*	1.56 (1.07–2.86)
FE MMP7 (%)	0.25 (0.23–0.29)*	1.89 (1.24–4.16)
FE TG-Fβ1 (%)	0.02 (0.02–0.03)*	0.29 (0.17–0.55)

*Mann-Whitney U test: p<0.0001 control group versus CKD.
FE: fractional excretion; CKD: chronic kidney disease; EMMPRIN: extracellular matrix metalloproteinase inducer; MMP7: matrix metalloproteinase 7; TGF-β1: transforming growth factor beta 1.

Statistical analysis was performed using the package Statistica 10.0 (StatSoft). A p-value <0.05 was considered significant.

RESULTS

Fractional Urinary Excretion of Survivin, E-cadherin, EMMPRIN, MMP7, and TGF-β1

The median serum and urine concentrations of all examined parameters were elevated in CKD patients versus controls (Table 2). The values of urinary FE in all cases were significantly elevated in the CKD children when compared with the control group (Table 2). In particular, the FE values of E-cadherin and TGF-β1 were <1% in both the control and CKD groups, whereas FE for survivin, EMMPRIN, and MMP7 elevated, exceeding 1% in the CKD children (Table 2).

Correlations and Cluster Analysis

All FE values correlated significantly with each other (r>0.96; p<0.000001). Additionally, cluster analysis revealed the similarity of features pictured by EMMPRIN, E-cadherin, and TGF-β1, suggesting that their efficiency as markers is comparable and choosing one out of three would be enough to get the required information (Figure 1). None of the analysed parameters correlated with analysed biochemical markers or proteinuria.

DISCUSSION

We have shown that FE values of survivin, E-cadherin, EMMPRIN, MMP7, and TGF-β1 increase in children with CKD, when compared with controls. In our previous studies, we looked into the urine excretion of the above mentioned parameters and their correlations to the serum concentration.[19,23] The increasing urine concentrations of survivin, E-cadherin, EMMPRIN, MMP7, and TGF-β1 have turned out to be useful markers of EMT and other CKD-related complications such as fibrosis or apoptosis. Moreover, some of the analysed urine parameters were independent of serum

concentrations, suggesting that the increase of urine concentration of survivin, EMMPRIN, MMP7, and TGF-β1 in CKD is more due to the kidney production than to the parameter leakage by the damaged filtration barrier. However, those results were not sufficient to prove the origin of parameters found in urine. Therefore, we decided to verify the credibility of our hypothesis with the usage of FE.

FE has previously found its way to clinical practice in the case of sodium, enabling differentiation between pre-renal acute kidney injury and renal acute kidney injury due to tubular damage. The latter is characterised by increased FE of sodium. The data on the practical use of FE in the population with CKD is restricted to phosphate metabolism.[26] There are also single studies assessing FE of endothelin 1 or heat shock protein 27 in the context of lupus nephritis and CKD.[29,30] However, none of them dealt with a whole group of parameters characterising a particular process. Our focus was on markers picturing the variety of processes leading to tubular damage and fibrosis, which are characteristic features of CKD.

In our study, the FE of all analysed parameters was <1% in the control group, showing that in normal conditions survivin, E-cadherin, EMMPRIN, MMP7, and TGF-β1 are reabsorbed in over 99% by tubular cells. In children with CKD, FE values were significantly higher than in controls, confirming the tubular dysfunction characteristic of CKD. There was however a clear distinction between the examined markers. E-cadherin and TGF-β1 FE values remained <1% in CKD children, whereas survivin, EMMPRIN, and MMP7 FE values exceeded that border. Such an increase is suggestive of renal production of survivin, EMMPRIN, and MMP7, resulting from tubular loss and probably facilitating EMT with subsequent fibrosis and irreversible loss of renal function.

The significance of increased FE values strongly depends on the analysed parameter. Survivin is known for its anti-apoptotic activity and the connections between urine survivin and apoptotic markers have been shown in our previous study.[23]

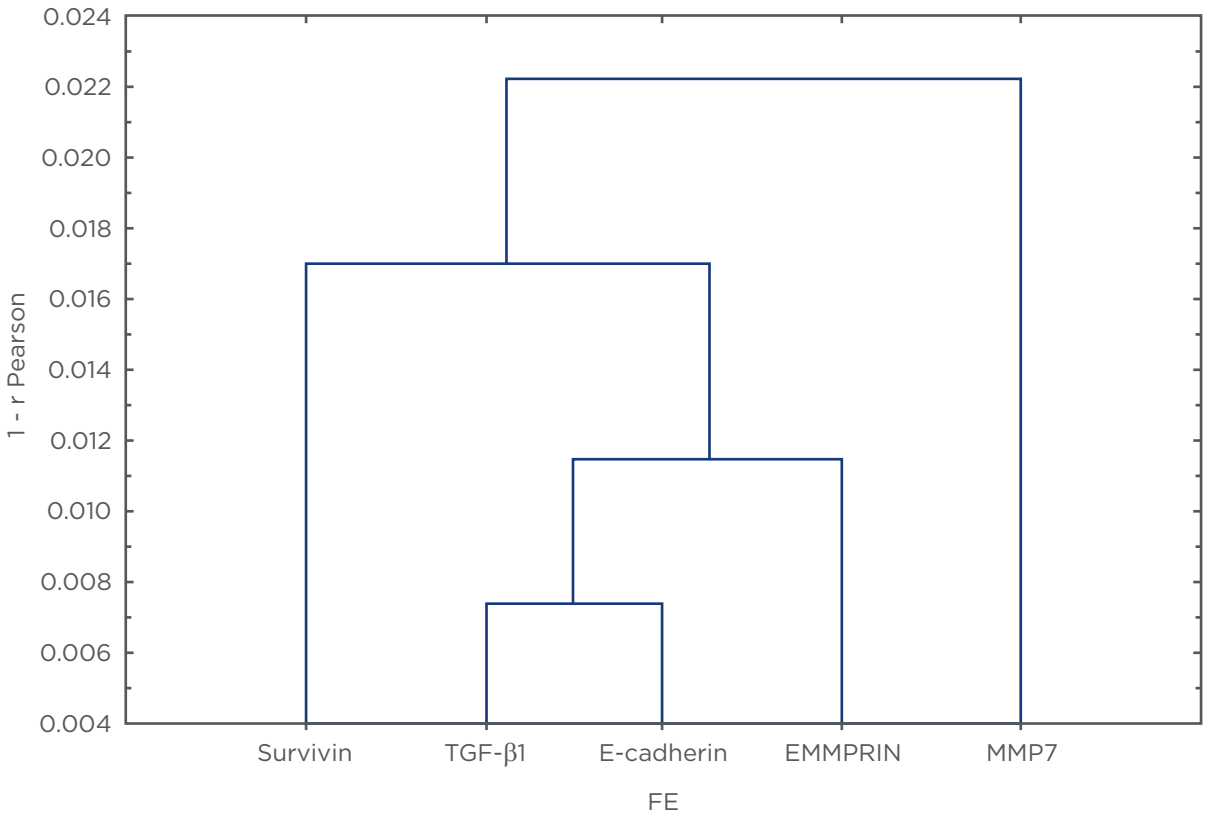

Figure 1: The cluster analysis of fractional excretion values of examined parameters in children with chronic kidney disease.

FE: fractional excretion; TGF-β1: transforming growth factor beta 1; EMMPRIN: extracellular matrix metalloproteinase inducer; MMP7: matrix metalloproteinase 7.

The essential role for survivin in tubular damage recovery after acute kidney injury has also been discovered.[25] Therefore, the increase of FE survivin value in CKD children may mean the protective reaction against aggravated apoptosis in the milieu of renal parenchyma, including damaged tubular cells. The FE survivin values may be treated as an early marker of tubular damage in the course of CKD.

The tight relations in the area of pro-fibrotic activity and EMT triggering, seen between MMPs and their EMMPRIN, have also been known for a while.[15,18] Our previous investigation has confirmed the usefulness of those parameters in assessing CKD-related complications like fibrosis.[19] It has now been shown that both EMMPRIN and MMP7 in urine might originate from the renal parenchyma, giving insight into the actual fibrotic activity of the tissue and, indirectly, the progression of kidney dysfunction. This example is even more convincing in the case of MMP7, because its urine concentration in controls was nearly negligible; the values observed in the urine of CKD children from that perspective seemed overwhelming.[19] Thus, both EMMPRIN and MMP7 FE values >1% in CKD children may indicate the tubular damage characteristic of renal failure progression.

The correlation analysis has shown significant correlations between all analysed FE values, yet the cluster analysis allowed us to eliminate the markers that described similar features. Out of three parameters (FE values of E-cadherin, TGF-β1, and EMMPRIN) we have chosen EMMPRIN FE values >1%, thus forming the homogenous marker group with FE of survivin and FE of MMP7. Our preliminary data have shown the potential usefulness of FE of survivin, EMMPRIN, and MMP7, mainly in assessing the tubular dysfunction characteristic of CKD progression. The use of these parameters as markers of the kidney-specific EMT, however promising, requires further investigation.

There are a number of limitations to this study that have to be acknowledged. The lack of comparative data and reference values for some of the examined parameters, both in adult and paediatric patients, necessitates caution in drawing conclusions. Additionally, there ought to be careful interpretation of the results due to the potential bias of a small study group and transversal design. However, the number of analysed patients and the study design were both conditioned by the overall size of the paediatric population with CKD.

CONCLUSIONS

FE values of the examined markers may become a useful tool in assessment of tubular dysfunction in the course of CKD. FE values of survivin, EMMPRIN, and MMP7 warrant further research as potential independent markers of the kidney-specific EMT.

REFERENCES

1. Eddy AA. Can renal fibrosis be reversed? Pediatr Nephrol. 2005;20(10):1369-75.

2. Prunotto M et al. From acute injury to chronic disease: pathophysiological hypothesis of an epithelial/mesenchymal crosstalk alteration in CKD. Nephrol Dial Transplant. 2012;27 suppl. 3;43-50.

3. Humphreys BD et al. Fate tracing reveals the pericyte and not epithelial origin of myofibroblasts in kidney fibrosis. Am J Pathol. 2010;176(1):85-97.

4. Mack M, Yanagita M. Origin of myofibroblasts and cellular events triggering fibrosis. Kidney Int. 2015;87(2):297-307.

5. Fragiadaki A, Mason RM. Epithelial-mesenchymal transition in renal fibrosis – evidence for and against. Int J Exp Path. 2011;92(3):143-50.

6. Liu Y. New insights into epithelial-mesenchymal transition in kidney fibrosis. J Am Soc Nephrol. 2010;21(2):212-22.

7. Lovisa S et al. Epithelial-to-mesenchymal transition induces cell cycle arrest and parenchymal damage in renal fibrosis. Nat Med. 2015;21(9):998-1009.

8. De Chiara L, Crean J. Emerging Transcriptional Mechanisms in the Regulation of Epithelial to Mesenchymal Transition and Cellular Plasticity in the Kidney. J Clin Med. 2016;5(1);E6.

9. Huang S, Susztak K. Epithelial Plasticity versus EMT in Kidney Fibrosis. Trends Mol Med. 2016;22(1):4-6.

10. Loeffler I, Wolf G. Transforming growth factor-β and the progression of renal disease. Nephrol Dial Transplant. 2014;29 suppl 1:i37-45.

11. Lopez-Hernandez FJ, Lopez-Novoa JM. Role of TGF-β in chronic kidney disease: an integration of tubular, glomerular and vascular effects. Cell Tissue Res. 2012;347(1):141-54.

12. Decleves AE, Sharma K. Novel targets of antifibrotic and anti-inflammatory treatment in CKD. Nat Rev Nephrol. 2014;10(5):257-67.

13. Yen CL et al. Stimulation of transforming growth factor-beta-1 and contact with type I collagen cooperatively facilitate irreversible transdifferentiation in proximal tubular cells. Biomed J. 2016;39(1):39-49.

14. Cheng S, Lovett DH. Gelatinase A (MMP-2) is necessary and sufficient for renal tubular cell epithelial-mesenchymal transformation. Am J Pathol. 2003;162(6): 1937-49.

15. Tan TK et al. Macrophage matrix metalloproteinase-9 mediates epithelial-mesenchymal transition in vitro in murine renal tubular cells. Am J Pathol. 2010;176(3):1256-70.

16. Tan RJ et al. Wnt/β-catenin signaling and kidney fibrosis. Kidney Int Suppl. 2014;4(1):84-90.

17. He W et al. Matrix metalloproteinase-7 as a surrogate marker predicts renal Wnt/ β-catenin activity in CKD. J Am Soc Nephrol. 2012;23(2):294-304.

18. Kato N et al. Basigin/CD147 promotes renal fibrosis after unilateral ureteral obstruction. Am J Pathol. 2011;178(2): 572-9.

19. Musiał K et al. Urine survivin, E-cadherin and matrix metalloproteinases as novel biomarkers in children with chronic kidney disease. Biomarkers. 2015; 20(3):177-82.

20. Onder TT et al. Loss of E-cadherin promotes metastasis via multiple downstream transcriptional pathways. Cancer Res. 2008;68(10):3645-54.

21. Taddei ML et al. Anoikis: an emerging hallmark in health and diseases. J Pathol. 2012;226(2):380-93.

22. Yoo CB et al. γ-secretase-dependent cleavage of E-cadherin by staurosporine in breast cancer cells. Cell Communication and Adhesion. 2012;19(1):11-6.

23. Musiał K et al. Urine matrix metalloproteinases and their extracellular inducer EMMPRIN in children with chronic kidney disease. Ren Fail. 2015;37(6): 980-4.

24. Mehrotra S et al. IAP regulation of metastasis. Cancer Cell. 2010;17(1):53-64.

25. Chen J et al. Survivin mediates renal proximal tubule recovery from AKI. J Am Soc Nephrol. 2013;24(12):2023-33.

26. Dominguez JR et al. Fractional excretion of phosphorus modifies the association between fibroblast growth factor-23 and outcomes. J Am Soc Nephrol. 2013;24(4):647-54.

27. Schwartz GJ et al. New equations to estimate GFR in children with CKD. J Am Soc Nephrol. 2009;20(3):629-37.

28. Lurbe E et al.; European Society of Hypertension. Management of high blood pressure in children and adolescents: recommendations of the European Society of Hypertension. J Hypertens. 2009;27(9):1719-42.

29. Dhaun N et al. Urinary endothelin-1 in chronic kidney disease and as a marker of disease activity in lupus nephritis. Am J Physiol Renal Physiol. 2009;296(6): F1477-83.

30. Lebherz-Eichinger D et al. Hsp27 and Hsp70 serum and urine levels in patients suffering from chronic kidney disease. Clin Chim Acta. 2012;413(1-2):282-6.

UPCOMING EVENTS

The 23rd Budapest Nephrology School

26th–31st August 2016

Budapest, Hungary

The Budapest Nephrology School is a week-long nephrology refresher course organised by the Hungarian Kidney Foundation charity. International speakers will provide updates on nephrology, dialysis, hypertension, and transplantation, with the most recent scientific advances and the current clinical approaches covered. It is an increasingly popular event providing workshops, a series of talks, and visits to quality dialysis and research units.

8th Annual Meeting of the German Society of Nephrology (DGfN)

10th–13th September 2016

Berlin, Germany

The latest research on topics such as experimental nephrology, renal failure, and transplantation will be presented during the 4-day event. Discussions will also be held on important issues in the field that include strategies for organ donation and creating greater gender awareness among professionals. Prof Howard Jacob, Executive Vice President for Genomic Medicine, HudsonAlpha Institute for Biotechnology, will be a guest speaker discussing the future of personalised medicine.

45th European Dialysis and Transplant Nurses Association/ European Renal Care Association (EDTNA/ERCA) International Conference

17th–20th September 2016

Valencia, Spain

This conference will share the latest research, innovations, and technology for practice and research in renal medicine. This year's theme is 'Quality versus Cost: Sustainable Renal Care', which seeks to address the challenge of achieving and maintaining quality renal care despite cost implications. The scientific programme will balance assessments of the latest therapeutic approaches with considerations of the costs faced, while exploring its impact on public health globally.

17th Congress of the International Pediatric Nephrology Association (IPNA)

20th–24th September 2016

Iguaçu, Brazil

Held every 3 years, the congress will meet in September to update its participants on the current knowledge and skills needed for paediatric nephrology, including the areas of critical care, kidney pathology, and rare diseases. It will also provide a series of master classes to support those practising in the areas of glomerular diseases, chronic kidney disease and renal replacement therapy, and inherited and structural diseases, amongst others.

10th International Congress of the International Society for Hemodialysis (ISHD)

22nd–24th September 2016

Marrakech, Morocco

Organised in association with the Moroccan Society of Nephrology (MSN), this will be the first time the ISHD congress is held on the African continent. It will provide a scientific programme on the most recent developments in the field of haemodialysis, but does so with an ambitious aim. It intends to address scientific, practical, and socio-economic challenges that work towards the congress theme: 'For a comprehensive, equitable, and sustainable access to kidney disease care'.

9th UK Annual Dialysis Conference

29th–30th September 2016

Manchester, UK

With an emphasis on care being given in the UK, this 2-day event will discuss the latest progress in dialysis outcomes and the emerging themes and perspectives towards improving dialysis delivery and care. It will also include presentations reporting on the latest efforts of the highest achieving renal units in the UK to inform clinical care in areas such as reducing drop-out in home dialysis and the best practice for eliminating peritoneal dialysis peritonitis.

1st Congress of the French Society of Nephrology, Dialysis and Transplantation (SFNDT)

4th–7th October 2016

Strasbourg, France

The conference will provide high-level scientific sessions that reflect the dynamism of this newly formed society. It also will also offer a Continuous Professional Development programme designed to bring together the latest scientific developments and current clinical practices. The programme will consist of four different sessions with glomerular, dialysis, kidney transplantation, and genetic kidney disease available for participants to choose.

54th European Renal Association - European Dialysis and Transplant Association (ERA-EDTA) Congress

3rd–6th June 2017

Madrid, Spain

This long-established congress will once again provide an attractive programme devoted to reporting on the latest research results and extensive overviews of nephrology with high-quality presentations. The central theme of the event will be: 'Nephrology: much more than kidney disease', which is reflected in the diverse topics to be discussed in presentations by world-renowned scientists, and in the more than 70 mini-lectures and symposia. Topics for discussion include kidney regeneration, bioelectric medicine, and the recreation of life. This is a clear indication that the nephrology field is both an exciting and a growing one, and ERA-EDTA 2017 will be a brilliant opportunity to both witness and contribute to this growth.

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