kidney disease who would usually go undetected as part of standard care could have a greater benefit than populations who are usually more likely to have serum creatinine or albuminuria measured. By contrast, the anticipated benefit of screening is lower in populations with a lower prevalence of chronic kidney disease. Several factors influence which high-risk populations should be selected for screening including the benefits and problems after treatment, the consequences of non-treatment, false-positive tests, and the feasibility of detecting the target population and treating identified patients. All these factors have been incompletely studied, and it is difficult to recommend groups for whom screening should be indicated or contraindicated. However, older age, diabetes, or hypertension are potentially attractive criteria—and selective testing for kidney disease in these populations seems likely to be beneficial.[29–31]

The ideal frequency for screening measurements is unknown, but given the persistent nature of chronic kidney disease, covering a high proportion of people at risk should take precedence over frequent screening. Whether serum creatinine or albuminuria analysis, and which assays should be used is also unknown.[22,32] Use of both strategies might be most sensitive (particularly as albuminuria does not universally accompany reduced glomerular filtration rate[33]) but would lower specificity. Follow-up testing will probably be needed irrespective of which strategy is used initially. Finally, the best way to deal with cases identified through screening is uncertain, and will vary by setting: some countries may favour a public health approach (a generic bundle of effective therapies applied to all patients), whereas more individualised treatment (based on severity, stage or cause of chronic kidney disease) might be more appropriate in others.

Despite this uncertainty, effective treatment for diabetes, hypertension and cardiovascular disease will also have beneficial effects on chronic kidney disease, and vice versa. Because detection of chronic kidney disease is unhelpful if lifelong medical therapy is unavailable, establishing and maintaining access to effective treatments is a prerequisite for screening programmes for chronic kidney disease in low-income countries but also in high-income nations that lack universal health-care systems. The highest priority for controlling chronic kidney disease should be to ensure secure, sustainable access to low cost antihypertensive drugs (particularly angiotensin-converting-enzyme inhibitors and angiotensin-receptor blockers), which will prevent kidney failure and also reduce cardiovascular morbidity and mortality.[34] Improved access to treatments that control blood glucose and blood cholesterol, and those that tackle smoking will also improve renal and cardiovascular outcomes. Furthermore, serum creatinine and albuminuria measurements are commonly made in usual clinical practice; chronic kidney disease is usually identified in the absence of organised screening programmes. Management of chronic kidney disease is often suboptimal, and therefore improving the care of all chronic kidney disease is important. Finally, in view of the common causes and consequences of chronic kidney disease with other non-communicable diseases, integration of screening into national or regional disease management programmes will be important.

New health service models for controlling chronic kidney disease

Treatment of chronic kidney disease is an important economic burden within health systems and is grossly inadequate in low-income countries.[35,36] WHO has estimated that in two-thirds of low-income countries there is no access to renal replacement therapy for end-stage renal disease.[37] Therefore, addressing the burden of chronic kidney disease requires preventive measures that include control of generic risk factors (eg, smoke, high salt intake, or hyperlipidaemia) and, in some regions of the world, focusing on specific causes.[38]

Multidisciplinary care and control of general risk factors

The increased awareness that death caused by cardiovascular disease is a more common outcome than progression to end-stage renal disease in patients with chronic kidney disease has led nephrologists to focus on the prevention of cardiovascular disease.[39] However, management of cardiovascular disease is fragmented (and sometimes divergent) among nephrologists, cardiologists, and diabetologists; these issues can be further complicated with the involvement of primary-care physicians, geriatricians, dietitians, pharmacists, and nurses. Two non-exclusive approaches might bring cohesion. The first is education of patients and support of self-management; the second is a multidisciplinary team approach. Interactions between informed patients and proactive multidisciplinary teams might improve health outcomes for people with chronic medical disorders.

Comprehensive, team-based, multidisciplinary interventions for chronic kidney disease are associated with improved blood pressure and metabolic control, preservation of glomerular filtration rate, a smaller percentage of patients needing dialysis, and reduced mortality.[40–44] Multifactorial interventions including lifestyle changes[45–48] and pharmacological interventions to reduce proteinuria and control blood pressure with angiotensin-converting-enzyme inhibitors or angiotensin-receptor blockers, tight diabetic control, and treatment of dyslipidaemia are not only cost-effective measures to reduce the burden of cardiovascular disease but have also a beneficial effect on chronic kidney disease.[49]

The main expectation for the near future is building capacity to establish individualised multidisciplinary care programmes, adapted to the highly diverse medical needs of patients. For patients with chronic kidney disease, high comorbidity or specific frailty (the threshold beyond which the functional reserve of a person is critically reduced and

the tolerance of stress negligible), optimum care will probably be delivered with methods specific to the disorder, including multidisciplinary specialty clinics, aimed at reducing cardiovascular events and reducing or allowing transition to renal replacement therapy.

However, a programme of timely nephrology referral and specialised multidisciplinary follow-up for all patients with chronic kidney disease would quickly overwhelm available resources, and would not be realistic in many settings. Thus, in most parts of the world, primary-care physicians will have an essential role to play in the care of chronic kidney disease. Patients with stable renal disease, low comorbidity, those living in remote areas or in low-income countries could undergo comanagement, with regular monitoring by a primary-care physician, and future nephrology follow-up.[50] Similar considerations apply to the delivery of care to patients with acute kidney injury or with multiple organ failure syndromes (eg, combined hepatic and renal insufficiency; cardiorenal syndrome).

Care models that incorporate nurse practitioners are being increasingly used for the management of chronic diseases.[51] In view of the success in other fields, the large population at risk for chronic kidney disease, and the low availability of trained nephrologists for the number of patients, it is important to test care models in which physicians partner with nurse practitioners to deliver care. The interface of nephrologists with primary-care physicians or other specialists could be implemented even in low-resource setting using new telecommunication technologies. Telehealth initiatives, in which nephrology specialists provide their expertise remotely over the internet, are a model that can be adopted in low-resource settings. This approach has been successfully applied in Bolivia where telehealth care is delivered using online applications to provide expertise at very low cost (Raul Plata Cornejo, Instituto de Nefrología, La Paz, Bolivia, personal communication). The programme not only permits individualised multidisciplinary care and the chance of follow-up by experienced off-site nephrologists, but minimises the burden of travel and its impact on family and employment (panel 3).

***Panel 3:* Telemedicine for renal care**

Low availability of specialist care in developing countries is one of the limiting factors for the prevention and cure of chronic kidney diseases or for proper management of acute events. Geographical barriers, lack of transport or infrastructure, and low availability of trained personnel are among the leading causes for varied management of kidney disease in developing countries. Rather than, or in addition to, providing resources or equipment, health-care systems' primary problem is effectively diffusing knowledge and information. Technology currently offers a chance for this to change.

In many developing countries the rate of growth of digital infrastructure has surpassed that of physical infrastructure. The deployment of mobile networks has reached an estimated 45% global population coverage in 2011, the largest rise in developing countries. Affordable connectivity has strong implications for the future of health care, in particular where accessibility to specialist care is limited.

The ability to communicate through rich, easy to use, multiuser applications capable of transmitting audio or video streams, once available in high-end teleconferencing or telemedicine systems, has become an integral part of everyday lives in both high-income and low-income countries. The availability of these technologies at the consumer level has largely contributed to reduce costs for developing these systems and making them available on a large scale, with minimum initial investments on the infrastructure of information technology as result of cloud computing and platform-as-a-service resources.

New models of specialist health-care provision could emerge from the integration of modern information technology and medicine. Renal care, in particular, is characterised by the need of regular follow-ups over long periods of time. Substantial improvements to morbidity and mortality can be achieved by simple and low-cost actions, if appropriately put in place.

In this setting, several small peripheral centres, typically led by primary-care physicians or nurse practitioners, could be pooled into centrally led virtual nephrology departments. Supported by rich web applications providing clinical information management, direct communication, and real-time data analysis, nephrologists located in these central facilities could lead patient care by relying on peripheral practitioners for in-loco operation.

The opportunity for the scientific community is unique. Information technology has the potential of delivering specialist health care in inaccessible areas, but it also represents a potential collector of data on unprecedented large scales. This collection is possible if data are public and if data collection systems are built with a high degree of interoperability (ie, their interfaces are fully disclosed and they are capable to interact and function with other systems without any access or implementation restrictions). The reliance on transparency and a consensus effort in the definition of the structure and the nature of data to be collected are important.

Telemedicine has had a rich history in the past two decades. For the first time, this model is becoming cost effective for developing and high-income countries, marking the path towards global health-care provision and new opportunities for scientific advancement.

Control of region-specific risk factors for chronic kidney disease

In addition to global risk factors, the burden of chronic kidney disease has specific characteristics that should be recognised and addressed in certain regions of the world. Although most patients with chronic kidney disease in high-income countries have diabetes or hypertension, as many as 40–50% of patients have a different cause in low-income countries.[52] Chronic glomerulonephritis and interstitial nephritis resulting from bacterial, viral, parasite or toxic causes represent a substantial proportion of chronic kidney disease in some areas of the world.[52] An example is HIV infection.[53] Compared with controls without HIV, patients with HIV have an increased prevalence of impaired kidney function (six times), albuminuria (five times), and end-stage renal disease (ten times).[54,55] Between 770 000 and 2·6 million individuals in sub-Saharan Africa have HIV nephropathy. The existing guidelines for management and referral of patients with HIV for nephrology care in high-income countries are not applicable in sub-Saharan Africa and a recently proposed screening algorithm that starts with the determination of microalbuminuria deserves careful assessment.[53]

Registries should be a key priority for control of chronic kidney disease, and could document the total burden of

kidney disease in each country or region and progress made over time—as well as quantifying the comparative contribution of common and specific regional risk factors. The feasibility of large, cross-sectional studies to assess the prevalence of chronic kidney disease was confirmed in a study[56,57] done in several countries (Bangladesh, Bolivia, Georgia, and Nepal), with the support of the International Society of Nephrology in both the general population and in patients at high-risk of the disease. Advances in diagnostic testing will help the expansion of such programmes—including availability of cheap point-of-care testing for kidney function and albuminuria (with appropriate attention to assay standardisation and calibration), and validation of new creatinine-based or cystatin-based prediction equations for estimation of glomerular filtration rate in low-income countries.

New approaches for drug and clinical development

Drug and clinical development have become lengthy and expensive as a result of the number of newly marketed drugs by the pharmaceutical industry.[58] Few drugs to treat kidney disease have been developed in the past 15 years, despite a large number of potential beneficiaries.

Reduction in the time and costs of pharmacology research requires not only an understanding of the pathophysiology of the targeted disease but also an early test of the drug's effects on human physiology and pathology. Early clinical trials in patients are now more practical since there is new guidance from the international conference on harmonisation of technical requirements for registration of pharmaceuticals for human use that introduces exploratory clinical trials as first-in-human studies that assess a drug's distribution in vivo as well as its physiological and pharmacological effects in a few patients.[59] These studies notably reduce the duration of preclinical assessment. These latest regulations for pharmaceutical practice as well as newer, state-of-the-art, efficient strategies for preclinical and clinical development require a thorough understanding by the investigator.[60]

Exploratory clinical trials are important for drug development, especially in kidney diseases for which experimental animals mimicking human disease are difficult to obtain, and where clinical endpoints such as renal death are elusive.

A renewed partnership between pharmaceutical industry and academia is needed. Large clinical trials required for marketing authorisation are the responsibility of the pharmaceutical industry. By contrast, academia selects the compounds to be tested by industry, by assessing the physiological and pharmacological relevance of candidate molecules. Of importance, neither academia nor the pharmaceutical industry pay sufficient attention to orphan diseases[61] or less common kidney diseases.[62,63]

In the next few years, this new vision for the development of new drugs in clinical trials should be extended from high-income to low-income countries. It will be important to ensure that the capacity for clinical trials is developed locally in low-resource regions. Thus, dissemination of clinical trials will require opportunities for training in clinical trial methodology, design, and statistics. Nevertheless, the important task will be to protect emerging countries from pharmaceutical companies taking advantage of patients in these countries who are rarely treated according to standard guidelines.

Focus on research for rare and genetic kidney diseases

The specialty of rare and genetic kidney diseases is expected to change fundamentally during the next decade. The International Rare Disease Research Consortium has formulated two key objectives to be reached by 2020: to establish diagnostic tests for most rare diseases, and to find medical treatments for 200 rare disorders.[64] These targets suggest the most important needs, and foreseeable accomplishments, in rare kidney diseases.

The most imminent progress is expected in the identification of causes for genetic disease. The advent of next-generation sequencing allows screening of the exome quickly and cost effectively.[65] This technique will accelerate the identification of new disease genes but also pose challenges in bioinformatic processing and define a need for new methods for high-efficiency functional assessment of gene variants by cell, tissue, and animal models. Cell and tissue modelling may be boosted by current advances in inducible stem cell and transdifferentiation technology. Conventional transgenic rodent disease models will probably be complemented by refined lower vertebrate models (such as zebrafish and xenopus frogs) suitable for rapid phenotypic and functional screening of candidate proteins and their mutants.[66]

The proportion of patients with an unambiguous genetic diagnosis will increase notably. The development of targeted sequencing arrays covering all genes associated with a particular disease or disease group will substantially improve diagnostic time and cost efficacy. Targeted sequencing will also avoid the ethical dilemmas associated with incidental discovery of mutations in genes unlinked to the disease of interest that can occur with whole-exome sequencing.[67]

In patients with rare kidney disorders in whom a genetic diagnosis cannot be made, new technologies with systems-biology approaches integrating DNA variants, gene transcript patterns, urine proteomic, and metabolomic profiles will soon be available to complement clinical trials by molecular phenotyping. These strategies will identify molecular markers that individually (as disease biomarkers) or in combination (as molecular signatures) will lead to a mechanistically based molecular spectrum of rare kidney diseases.

The availability of reliable genetic and molecular diagnostics will affect clinical disease management—eg, by replacing histopathology, accurately defining the need

for and susceptibility to pharmacological interventions, and by predicting the risk of post-transplant disease recurrence. In families with congenital kidney diseases, accurate prenatal diagnosis and risk assignment will allow individualised genetic counselling and help development of early intervention and secondary prevention strategies.

To date, the development of therapeutics in rare diseases has been lagging behind the advances in genetic and pathophysiological mechanisms, since the altered protein products of the disease-causing genes are commonly not treatable or there is no obvious molecular approach to bypass gene deficiencies. Notwithstanding these challenges, the widespread application of exome sequencing is expected to increase the number of molecular drug targets in rare kidney diseases.[68] Rare diseases with a renal phenotype are usually systemic disorders with multi-organ involvement. Hence, understanding the molecular pathophysiology of these diseases might contribute to knowledge about other organ-specific diseases. For example, research in rare complement kidney diseases will advance the global understanding of complement-mediated tissue and organ damage.[69] Furthermore, podocyte-specific proteins deficient in inherited glomerulopathies are also involved in acquired glomerulopathies, such as diabetic nephropathy, and study of these proteins could help understand the mechanism of glomerular disease.[70] Studying the pathophysiology of tubulopathies might have immediate relevance in understanding regulation of blood pressure, formation of kidney stones, and kidney disease progression. Finally, systems biology approaches that integrate molecular characteristics of different kidney disorders and phenotypes of disease progression may help identify common pathways that lead to kidney disease progression.[71,72] If new targets for pharmacological nephroprotection can be identified, progress in research on rare kidney disease could help with understanding several progressive kidney disorders.

Promotion of research in developing countries

Local health problems in low-income countries indicate the importance of economic and social development: relevant research must focus on the biological causes of such illnesses, but also on how to break the vicious cycle of economic development and new emerging disease. For example, growing urbanisation and pollution have led to rising rates of environmental illness, including sick-building syndrome[73] and sick-house syndrome.[74] Several kidney diseases that are associated with environmental causes (such as glomerular nephropathies associated with organic solvents, common in high-income countries in the last century[75]) have begun to emerge in low-income countries.

The number of researchers moving out of their home countries that are low income increases the gap between north and south, and reduces capacity to address local issues in these regions.[76–79] Taiwan has implemented several policies to keep researchers, including financial incentives reimbursement for the costs of repatriation and grants for business development, which reversed previous trends in migration of Taiwanese scientists.[80] Scientists, political leaders, and decision makers in low-income and high-income countries must collaborate to produce policies and education systems that promote and enable research and development. Easy communication, quick travel, and greater collaboration between high-income and low-income countries are increasingly common and should help expatriate professionals to contribute to their countries of origin.

A capacity gap remains between low-income or middle-income countries and high-income nations in health science, including nephrology. According to a WHO report[81] public health-care systems receive only 4·3–6·3% of the GDP in low-income or middle-income countries compared with 11% in high-income countries. Physician density in low-income or middle-income countries was 10·1 per 10 000 population, as opposed to 28·6 per 10 000 population in high-income countries. High-income countries and the global kidney research community should help low-income countries to increase their funding for primary health systems but also to increase their local capacity for research on local problems. The fellowship, sister renal centres and educational ambassadors programmes from the International Society of Nephrology are important mechanisms to strengthen kidney research capacity in low-income countries.[82] The global kidney research community should also focus on developing global clinical practice guidelines, which are suitable for patients in low-income countries.

At the same time, a new gap in capacity has appeared between scientifically proficient emerging countries (Argentina, Brazil, Chile, China, India, Malaysia, Mexico, and South Africa) and other emerging countries, the so called South–South gap. However, there are examples of increasing South–South cooperation that are helping to close this gap:[83] these initiatives must be promoted in renal medicine as well.

Today, even important ideas and studies from low-income or middle-income countries have little chance to reach international journals and are ignored.[84] Access to health information in low-income and middle-income countries should be improved. The gap between evidence and practice can have profound health effects when highly effective interventions exist.[85] Encouraging original research in low-income or middle-income countries should also increase visibility in these ideas to a wider audience—perhaps providing specific space in international journals for papers focusing on local problems needing specific solutions.

Renal replacement therapies

Despite the success of strategies for preventing progression of chronic nephropathies,[86] kidney failure

remains an important clinical problem. The outcomes associated with chronic dialysis have not substantially improved over the past two decades and further work is needed in this area to improve renal replacement therapy, either for acute kidney injury and chronic kidney injury.

New dialysis research includes cheaper treatments, home-based therapies, and simpler methods of blood purification, objectives that can be achieved with new disciplines such as miniaturisation and nanotechnology. In the field of renal replacement therapy, technical innovation can be the result of a joint effort of not-for-profit organisations, rather than industrial investment, when considering the needs of small populations. For example, the development of equipment for miniaturised renal replacement therapy for newborn babies and very young infants.[87]

Although kidney transplantation is the best available treatment for kidney failure, the supply of renal allografts is insufficient to meet the demands. New and more effective strategies are needed, including the use of self-repair of human tissues and organs.

The human kidney has an intrinsic capacity to repair after injury.[88] The repair process is accomplished by migration of stem or progenitor cells into the damaged region, with eventual reconstitution of a functional epithelium. Such progenitors have been identified in resident epithelial cells[89] and glomerular parietal epithelial cells,[90,91] but stem cells with broader regenerative properties are also found in the proximal tubuli, glomeruli, papilla, and peritubular capillaries, and in urine.[92]

Understanding how these unspecialised precursors are maintained and regulated has practical implications, as the regenerative potential of tissue-specific progenitor cells can be therapeutically used to boost the repair activity of cells in models of chronic kidney disease.[91,93] Efforts are also directed to replenish the renal stem-cell pool and potentiate the regenerative repairing process by transplantation of mesenchymal stromal cells from bone marrow or other tissue sources. This regenerative cell-based approach has been applied in rodent models with damaged renal tissue and is more effective in acute kidney injury than in chronic kidney injury.[94–96] However the main barrier to effective implementation of therapies based on mesenchymal stromal cells is the absence of specific homing of exogenously infused cells and the inability to direct these cells to the diseased tissue. Genetic modification of mesenchymal stromal cells with retroviral vectors that encode homing receptors[97] or preconditioning of mesenchymal stromal cells before infusion with compounds possessing promigratory properties (and possibly without side-effects)[98] are now being explored to direct therapy to diseased cells. Stem-cell therapies, however, might not be acceptable since some studies have suggested an increase in interstitial fibrosis.[99]

Investigators are trying to bioengineer kidneys[100] but this work is in its infancy and ex-vivo kidney regeneration with extracellular matrix scaffolds will probably not be clinically viable for at least a decade, despite a recent clinical attempt.[101]

Recently in-vivo experiments in athymic rats showed the development of renal organoids from embryonic murine cells, indicating that generation of vascularised glomeruli attached to nephrons with filtration and active re-uptake, from simple cell suspension is possible.[102] As engineering and nanotechnology advance, implantable artificial devices that could provide both glomerular and tubular function may be developed.

Whether and when these new technologies will result in significant clinical applications cannot be determined at present. Even more difficult is to predict how much such technologies would cost when applied on a large scale, and whether they would be affordable in low-income countries.

Contributors
All authors contributed to writing and conceptulisation of the report. GR and AS revised and did final editing of the various contributions.

Conflicts of interest
We declare that we have no conflicts of interest.

References

1 Vijayan A, Palevsky PM. Dosing of renal replacement therapy in acute kidney injury. *Am J Kidney Dis* 2012; **59**: 569–76.
2 Cerda J, Lameire N, Eggers P, et al. Epidemiology of acute kidney injury. *Clin J Am Soc Nephrol* 2008; **3**: 881–86.
3 Jha V, Malhotra HS, Sakhuja V, Chugh KS. Spectrum of hospital-acquired acute renal failure in the developing countries—Chandigarh study. *Q J Med* 1992; **83**: 497–505.
4 Callegari JG, Kilonzo KG, Yeates KE, et al. Peritoneal dialysis for acute kidney injury in sub-Saharan Africa: challenges faced and lessons learned at Kilimanjaro Christian Medical Centre. *Kidney Int* 2012; **81**: 331–33.
5 Yeates K, Cruz DN, Finkelstein FO. Re-examination of the role of peritoneal dialysis to treat patients with acute kidney injury. *Perit Dial Int* 2012; **32**: 238–41.
6 Chionh CY, Ronco C, Finkelstein FO, Soni SS, Cruz DN. Acute peritoneal dialysis: what is the 'adequate' dose for acute kidney injury? *Nephrol Dial Transplant* 2010; **25**: 3155–60.
7 George J, Varma S, Kumar S, Thomas J, Gopi S, Pisharody R. Comparing continuous venovenous hemodiafiltration and peritoneal dialysis in critically ill patients with acute kidney injury: a pilot study. *Perit Dial Int* 2011; **31**: 422–29.
8 Kilonzo KG, Ghosh S, Temu SA, et al. Outcome of acute peritoneal dialysis in northern Tanzania. *Perit Dial Int* 2012; **32**: 261–66.
9 Sustainable Kidney Care Foundation. Press release: new dialysis centers in Africa and Asia for acute kidney injury in young adults and children made possible by international medical partnership. http://www.skcf.net/?v=blog&post=9 (accessed Oct 31, 2012).
10 Hossain MP, Goyder EC, Rigby JE, El Nahas M. CKD and poverty: a growing global challenge. *Am J Kidney Dis* 2009; **53**: 166–74.
11 Najar MS, Shah AR, Wani IA, et al. Pregnancy related acute kidney injury: a single center experience from the Kashmir Valley. *Indian J Nephrol* 2008; **18**: 159–61.
12 Prakash J, Niwas SS, Parekh A, et al. Acute kidney injury in late pregnancy in developing countries. *Ren Fail* 2010; **32**: 309–13.
13 Luyckx VA, Bertram JF, Brenner BM, et al. Effect of fetal and child health on kidney development and long-term risk of hypertension and kidney disease. *Lancet* 2013; published online May 31. http://dx.doi.org/10.1016/S0140-6736(13)60311-6.
14 Luyckx VA, Brenner BM. The clinical importance of nephron mass. *J Am Soc Nephrol* 2010; **21**: 898–910.

15 Halim N, Bohara AK, Ruan X. Healthy mothers, healthy children: does maternal demand for antenatal care matter for child health in Nepal? *Health Policy Plan* 2011; **26**: 242–56.

16 Beaglehole R, Yach D. Globalisation and the prevention and control of non-communicable disease: the neglected chronic diseases of adults. *Lancet* 2003; **362**: 903–08.

17 Epping-Jordan JE. Integrated approaches to prevention and control of chronic conditions. *Kidney Int Suppl* 2005; 86–88.

18 Klein JD, Dietz W. Childhood obesity: the new tobacco. *Health Aff (Millwood)* 2010; **29**: 388–92.

19 Katz IJ, Hoy WE, Kondalsamy-Chennakesavan S, et al. Chronic kidney disease management—what can we learn from South African and Australian efforts? *Blood Purif* 2006; **24**: 115–22.

20 Thompson JE. Poverty, development, and women: why should we care? *J Obstet Gynecol Neonatal Nurs* 2007; **36**: 523–30.

21 The Bangkok Charter for health promotion in a globalized world. In: Tang KC, Beaglehole R, de Leeuw E, eds. 6th Global Conference on Health Promotion. Bangkok: Oxford University Press, 2005: 10–14.

22 Hallan SI, Stevens P. Screening for chronic kidney disease: which strategy? *J Nephrol* 2010; **23**: 147–55.

23 Wilson JMG, Jungner F. Principles and practice of screening for disease. Public Health Papers, 1968. http://whqlibdoc.who.int/php/WHO_PHP_34.pdf (accessed Feb 13, 2013).

24 Imai E, Yamagata K, Iseki K, et al. Kidney disease screening program in Japan: history, outcome, and perspectives. *Clin J Am Soc Nephrol* 2007; **2**: 1360–66.

25 Fink HA, Ishani A, Taylor BC, et al. Screening for, monitoring, and treatment of chronic kidney disease stages 1 to 3: a systematic review for the U.S. Preventive Services Task Force and for an American College of Physicians Clinical Practice Guideline. *Ann Intern Med* 2012; **156**: 570–81.

26 Perico N, Bravo RF, De Leon FR, Remuzzi G. Screening for chronic kidney disease in emerging countries: feasibility and hurdles. *Nephrol Dial Transplant* 2009; **24**: 1355–58.

27 Raffle AE, Gray JAM. What screening is and is not. In: Raffle AE, Gray JAM, eds. Screening: Evidence and Practice. USA: Oxford University Press, 2007: 288.

28 Hoerger TJ, Wittenborn JS, Zhuo X, et al. Cost-effectiveness of screening for microalbuminuria among African Americans. *J Am Soc Nephrol* 2012; **23**: 2035–41.

29 Manns B, Hemmelgarn B, Tonelli M, et al. Population based screening for chronic kidney disease: cost effectiveness study. *BMJ* 2010; **341**: 5869.

30 Sharma SK, Zou H, Togtokh A, et al. Burden of CKD, proteinuria, and cardiovascular risk among Chinese, Mongolian, and Nepalese participants in the International Society of Nephrology screening programs. *Am J Kidney Dis* 2010; **56**: 915–27.

31 Boulware LE, Jaar BG, Tarver-Carr ME, Brancati FL, Powe NR. Screening for proteinuria in US adults: a cost-effectiveness analysis. *JAMA* 2003; **290**: 3101–14.

32 de Jong PE, Curhan GC. Screening, monitoring, and treatment of albuminuria: public health perspectives. *J Am Soc Nephrol* 2006; **17**: 2120–26.

33 Zhang L, Wang F, Wang L, et al. Prevalence of chronic kidney disease in China: a cross-sectional survey. *Lancet* 2012; **379**: 815–22.

34 Perico N, Codreanu I, Schieppati A, Remuzzi G. The future of renoprotection. *Kidney Int Suppl* 2005; **97**: S95–101.

35 Rucker D, Hemmelgarn BR, Lin M, et al. Quality of care and mortality are worse in chronic kidney disease patients living in remote areas. *Kidney Int* 2011; **79**: 210–17.

36 Eggers PW. Has the incidence of end-stage renal disease in the USA and other countries stabilized? *Curr Opin Nephrol Hypertens* 2011; **20**: 241–45.

37 WHO. Global status report on noncommunicable diseases 2010. http://www.who.int/nmh/publications/ncd_report_full_en.pdf (accessed Oct 31, 2012).

38 Rodriguez-Iturbe B, Correa-Rotter R. Cardiovascular risk factors and prevention of cardiovascular disease in patients with chronic renal disease. *Expert Opin Pharmacother* 2010; **11**: 2687–98.

39 Keith DS, Nichols GA, Gullion CM, Brown JB, Smith DH. Longitudinal follow-up and outcomes among a population with chronic kidney disease in a large managed care organization. *Arch Intern Med* 2004; **164**: 659–63.

40 Bayliss EA, Bhardwaja B, Ross C, Beck A, Lanese DM. Multidisciplinary team care may slow the rate of decline in renal function. *Clin J Am Soc Nephrol* 2011; **6**: 704–10.

41 Devins GM, Mendelssohn DC, Barre PE, Binik YM. Predialysis psychoeducational intervention and coping styles influence time to dialysis in chronic kidney disease. *Am J Kidney Dis* 2003; **42**: 693–703.

42 Richards N, Harris K, Whitfield M, et al. The impact of population-based identification of chronic kidney disease using estimated glomerular filtration rate (eGFR) reporting. *Nephrol Dial Transplant* 2008; **23**: 556–61.

43 Thanamayooran S, Rose C, Hirsch DJ. Effectiveness of a multidisciplinary kidney disease clinic in achieving treatment guideline targets. *Nephrol Dial Transplant* 2005; **20**: 2385–93.

44 Wu IW, Wang SY, Hsu KH, et al. Multidisciplinary predialysis education decreases the incidence of dialysis and reduces mortality—a controlled cohort study based on the NKF/DOQI guidelines. *Nephrol Dial Transplant* 2009; **24**: 3426–33.

45 Gaede P, Vedel P, Larsen N, Jensen GV, Parving HH, Pedersen O. Multifactorial intervention and cardiovascular disease in patients with type 2 diabetes. *N Engl J Med* 2003; **348**: 383–93.

46 Levin A, Hemmelgarn B, Culleton B, et al. Guidelines for the management of chronic kidney disease. *CMAJ* 2008; **179**: 1154–62.

47 Fox CS, Larson MG, Leip EP, Culleton B, Wilson PW, Levy D. Predictors of new-onset kidney disease in a community-based population. *JAMA* 2004; **291**: 844–50.

48 Stengel B, Tarver-Carr ME, Powe NR, Eberhardt MS, Brancati FL. Lifestyle factors, obesity and the risk of chronic kidney disease. *Epidemiology* 2003; **14**: 479–87.

49 Abbate M, Zoja C, Remuzzi G. How does proteinuria cause progressive renal damage? *J Am Soc Nephrol* 2006; **17**: 2974–84.

50 El-Ghoul B, Elie C, Sqalli T, et al. Nonprogressive kidney dysfunction and outcomes in older adults with chronic kidney disease. *J Am Geriatr Soc* 2009; **57**: 2217–23.

51 Senior PA, MacNair L, Jindal K. Delivery of multifactorial interventions by nurse and dietitian teams in a community setting to prevent diabetic complications: a quality-improvement report. *Am J Kidney Dis* 2008; **51**: 425–34.

52 Barsoum RS. Chronic kidney disease in the developing world. *N Engl J Med* 2006; **354**: 997–99.

53 Fabian J, Naicker S. HIV and kidney disease in sub-Saharan Africa. *Nat Rev Nephrol* 2009; **5**: 591–98.

54 Eggers PW, Kimmel PL. Is there an epidemic of HIV infection in the US ESRD program? *J Am Soc Nephrol* 2004; **15**: 2477–85.

55 Szczech LA, Grunfeld C, Scherzer R, et al. Microalbuminuria in HIV infection. *AIDS* 2007; **21**: 1003–09.

56 Cravedi P, Sharma SK, Bravo RF, et al. Preventing renal and cardiovascular risk by renal function assessment: insights from a cross-sectional study in low-income countries and the USA. *BMJ Open* 2012; published online Sept 22. DOI:10.1136/bmjopen-2012-001357.

57 Plata R, Silva C, Yahuita J, Perez L, Schieppati A, Remuzzi G. The first clinical and epidemiological programme on renal disease in Bolivia: a model for prevention and early diagnosis of renal diseases in the developing countries. *Nephrol Dial Transplant* 1998; **13**: 3034–36.

58 US FDA. CDER approval times for priority and standard NDAs and new BLAs: Calendar Years 1993–2008. http://www.fda.gov/downloads/Drugs/DevelopmentApprovalProcess/HowDrugsareDevelopedandApproved/DrugandBiologicApprovalReports/UCM123957.pdf (accessed Oct 31, 2012).

59 Karara AH, Edeki T, McLeod J, Tonelli AP, Wagner JA. PhRMA survey on the conduct of first-in-human clinical trials under exploratory investigational new drug applications. *J Clin Pharmacol* 2010; **50**: 380–91.

60 Kimmelman J. Ethics at phase 0: clarifying the issues. *J Law Med Ethics* 2007; **35**: 727–33, 514.

61 Braun MM, Farag-El-Massah S, Xu K, Cote TR. Emergence of orphan drugs in the United States: a quantitative assessment of the first 25 years. *Nat Rev Drug Discov* 2010; **9**: 519–22.

62 Kretzler M, Cohen CD. Integrative biology of renal disease: toward a holistic understanding of the kidney's function and failure. *Semin Nephrol* 2010; **30**: 439–42.

63 Miyata T, Kikuchi K, Kiyomoto H, van Ypersele de Strihou C. New era for drug discovery and development in renal disease. *Nat Rev Nephrol* 2011; **7**: 469–77.

64 Baxter K, Terry SF. International Rare Disease Research Consortium commits to aggressive goals. *Genet Test Mol Biomarkers* 2011; **15**: 465.

65 Bamshad MJ, Ng SB, Bigham AW, et al. Exome sequencing as a tool for Mendelian disease gene discovery. *Nat Rev Genet* 2011; **12**: 745–55.

66 Wheeler GN, Brandli AW. Simple vertebrate models for chemical genetics and drug discovery screens: lessons from zebrafish and Xenopus. *Dev Dyn* 2009; **238**: 1287–308.

67 Harakalova M, Mokry M, Hrdlickova B, et al. Multiplexed array-based and in-solution genomic enrichment for flexible and cost-effective targeted next-generation sequencing. *Nat Protoc* 2011; **6**: 1870–86.

68 Swinney DC, Anthony J. How were new medicines discovered? *Nat Rev Drug Discov* 2011; **10**: 507–19.

69 Wagner E, Frank MM. Therapeutic potential of complement modulation. *Nat Rev Drug Discov* 2010; **9**: 43–56.

70 Welsh GI, Saleem MA. The podocyte cytoskeleton—key to a functioning glomerulus in health and disease. *Nat Rev Nephrol* 2012; **8**: 14–21.

71 Keller BJ, Martini S, Sedor JR, Kretzler M. A systems view of genetics in chronic kidney disease. *Kidney Int* 2012; **81**: 14–21.

72 He JC, Chuang PY, Ma'ayan A, Iyengar R. Systems biology of kidney diseases. *Kidney Int* 2012; **81**: 22–39.

73 Redlich CA, Sparer J, Cullen MR. Sick-building syndrome. *Lancet* 1997; **349**: 1013–16.

74 Norback D. An update on sick building syndrome. *Curr Opin Allergy Clin Immunol* 2009; **9**: 55–59.

75 Ravnskov U, Forsberg B, Skerfving S. Glomerulonephritis and exposure to organic solvents. A case-control study. *Acta Med Scand* 1979; **205**: 575–79.

76 Martineau T, Decker K, Bundred P. "Brain drain" of health professionals: from rhetoric to responsible action. *Health Policy* 2004; **70**: 1–10.

77 Mullan F. The metrics of the physician brain drain. *N Engl J Med* 2005; **353**: 1810–18.

78 Mudur G. Indian doctors criticise health ministry's plan to curb medical "brain drain". *BMJ* 2012; **344**: e3143.

79 Dodani S, LaPorte RE. Brain drain from developing countries: how can brain drain be converted into wisdom gain? *J R Soc Med* 2005; **98**: 487–91.

80 Iredale R. Tackling the roots of brain drain, 2012. http://unpan1.un.org/intradoc/groups/public/documents/APCITY/UNPAN022373.pdf (accessed Oct 31, 2012).

81 WHO. World Health Statistics 2011. http://www.who.int/entity/whosis/whostat/EN_WHS2011_Full.pdf (accessed Oct 31, 2012).

82 International Society of Nephrology. Global Outreach Program. http://www.theisn.org/isn-information/global-outreach-go-programs/itemid-451 (accessed Oct 31, 2012).

83 Thorsteinsdottir H, Melon CC, Ray M, et al. South-South entrepreneurial collaboration in health biotech. *Nat Biotechnol* 2010; **28**: 407–16.

84 Mohammadi NK, Zaree F, de Leeuw E, Emamjomeh M. Share of Nations in 37 International Public Health Journals: An Equity and Diversity Perspective towards Health Research Capacity Building. *Iran J Public Health* 2011; **40**: 129–37.

85 Guindon GE, Lavis JN, Becerra-Posada F, et al. Bridging the gaps between research, policy and practice in low- and middle-income countries: a survey of health care providers. *CMAJ* 2010; **182**: 362–72.

86 Ruggenenti P, Perticucci E, Cravedi P, et al. Role of remission clinics in the longitudinal treatment of CKD. *J Am Soc Nephrol* 2008; **19**: 1213–24.

87 Ronco C, Garzotto F, Ricci Z. CA.R.PE.DI.E.M. (Cardio-Renal Pediatric Dialysis Emergency Machine): evolution of continuous renal replacement therapies in infants. A personal journey. *Pediatr Nephrol* 2012; **27**: 1203–11.

88 Benigni A, Morigi M, Remuzzi G. Kidney regeneration. *Lancet* 2010; **375**: 1310–17.

89 Bussolati B, Bruno S, Grange C, et al. Isolation of renal progenitor cells from adult human kidney. *Am J Pathol* 2005; **166**: 545–55.

90 Sagrinati C, Netti GS, Mazzinghi B, et al. Isolation and characterization of multipotent progenitor cells from the Bowman's capsule of adult human kidneys. *J Am Soc Nephrol* 2006; **17**: 2443–56.

91 Benigni A, Morigi M, Rizzo P, et al. Inhibiting angiotensin-converting enzyme promotes renal repair by limiting progenitor cell proliferation and restoring the glomerular architecture. *Am J Pathol* 2011; **179**: 628–38.

92 Zhang Y, McNeill E, Tian H, et al. Urine derived cells are a potential source for urological tissue reconstruction. *J Urol* 2008; **180**: 2226–33.

93 Macconi D, Sangalli F, Bonomelli M, et al. Podocyte repopulation contributes to regression of glomerular injury induced by ACE inhibition. *Am J Pathol* 2009; **174**: 797–807.

94 Poulsom R, Forbes SJ, Hodivala-Dilke K, et al. Bone marrow contributes to renal parenchymal turnover and regeneration. *J Pathol* 2001; **195**: 229–35.

95 Morigi M, Imberti B, Zoja C, et al. Mesenchymal stem cells are renotropic, helping to repair the kidney and improve function in acute renal failure. *J Am Soc Nephrol* 2004; **15**: 1794–804.

96 Li J, Deane JA, Campanale NV, Bertram JF, Ricardo SD. The contribution of bone marrow-derived cells to the development of renal interstitial fibrosis. *Stem Cells* 2007; **25**: 697–706.

97 Cheng Z, Ou L, Zhou X, et al. Targeted migration of mesenchymal stem cells modified with CXCR4 gene to infarcted myocardium improves cardiac performance. *Mol Ther* 2008; **16**: 571–79.

98 Xinaris C, Morigi M, Benedetti V, et al. A novel strategy to enhance mesenchymal stem cell migration capacity and promote tissue repair in an injury specific fashion. *Cell Transplant* 2013; **22**: 423–36.

99 Boor P, Ostendorf T, Floege J. Renal fibrosis: novel insights into mechanisms and therapeutic targets. *Nat Rev Nephrol* 2010; **6**: 643–56.

100 Ross EA, Williams MJ, Hamazaki T, et al. Embryonic stem cells proliferate and differentiate when seeded into kidney scaffolds. *J Am Soc Nephrol* 2009; **20**: 2338–47.

101 Song JJ, Guyette JP, Gilpin SE, Gonzalez G, Vacanti JP, Ott HC. Regeneration and experimental orthotopic transplantation of a bioengineered kidney. *Nat Med* 2013; published online April 14. DOI:10.1038/nm.3154.

102 Xinaris C, Benedetti V, Rizzo P, et al. In vivo maturation of functional renal organoids formed from embryonic cell suspensions. *J Am Soc Nephrol* 2012; **23**: 1857–68.

Diabetic nephropathy: are there new and potentially promising therapies targeting oxygen biology?

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The multipronged drug approach targeting blood pressure and serum levels of glucose, insulin, and lipids fails to fully prevent diabetic nephropathy (DN). Recently, a broad range of anomalies associated with oxygen biology, such as hypoxia, oxidative stress (OS), and dyserythropoiesis, have been implicated in DN. This review delineates the cellular mechanisms of these anomalies to pinpoint novel therapeutic approaches. The PHD–HIF system mitigates hypoxia: HIF activates a broad range of reactions against hypoxia whereas PHD is an intracellular oxygen sensor negatively regulating HIF. The Keap1–Nrf2 system mitigates OS: Nrf2 activates cellular reactions against OS whereas Keap1 negatively regulates Nrf2. Clinical trials of PHD inhibitors to correct anemia in patients with CKD as well as of a Nrf2 activator, bardoxolone methyl, for DN are under way, even if the latter has been recently interrupted. A specific PHD1 inhibitor, a Keap1 inhibitor, and an allosteric effector of hemoglobin may offer alternative, novel therapies. Erythropoietin (EPO) is critical for the development of erythroid progenitors and thus for tissue oxygen supply. Renal EPO-producing (REP) cells, originating from neural crests, but not fibroblasts from injured tubular epithelial cells, transdifferentiate into myofibroblasts and contribute to renal fibrosis. Agents restoring the initial function of REP cells might retard renal fibrosis. These newer approaches targeting oxygen biology may offer new treatments not only for DN but also for several diseases in which hypoxia and/or OS is a final, common pathway.

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Numerous factors have been implicated in the development of diabetic nephropathy (DN). Their actual significance has been documented in several animal and human studies by the demonstration that their inhibition slowed the progression of DN. Still, despite a multipronged drug approach targeting blood pressure, serum levels of glucose, insulin, lipids, obesity, and so on, full prevention of DN remains elusive. Newer culprits thus remain to be identified. Besides hemodynamic and metabolic abnormalities, a broad range of abnormalities associated with oxygen biology, such as hypoxia, oxidative stress (OS), and dyserythropoiesis, have emerged in our understanding of DN.

All mammalian organs require a supply of oxygen to fuel various biometabolic processes. A decreased oxygen supply, that is, hypoxia, induces not only acute disorders such as ischemic heart disease but also chronic disorders such as renal fibrosis. OS during hypoxia may sound paradoxical. Yet, it may be induced not only by a rise but also a fall in oxygen tension. Hypoxic cells rely on anaerobic glycolysis to generate adenosine-5′-triphosphate but their residual low oxygen supply supports some level of oxidative production of adenosine-5′-triphosphate through the tricarboxylic acid cycle and electron transport chain. Electrons leaking from the mitochondrial electron transport chain generate an excess of reactive oxygen species (ROS), that is, OS. Thus, hypoxia and OS are closely linked. Reoxygenation or high oxygen levels following severe hypoxia further exaggerate ROS generation, a concept validated by the clinical benefits accruing from the use of agents able to scavenge ROS or the prevention of their formation in hypoxic lesions.[1]

Erythropoietin (EPO) is essential for the proliferation and differentiation of erythroid progenitors and hence of tissue oxygen supply.[2] Recent studies have unraveled the cellular mechanism of renal EPO production and the sequential events leading to renal fibrosis, both of which are closely linked to each other.[3–6] In contrast to previous knowledge, fibroblasts originating from injured tubular epithelial cells do not play a major role in renal fibrosis, but renal EPO-producing (REP) cells, stemming from neural crests, do transdifferentiate into myofibroblasts upon long-term exposure to inflammatory conditions and

contribute to renal fibrosis.[6] Fortunately, to some extent, REP cells retain their plasticity: in experimental animals, some agents restore their initial function and retard renal fibrosis.[6] These observations provide the missing link in chronic kidney disease (CKD) between anemia and renal fibrosis.

We review the cellular mechanisms of various abnormalities associated with oxygen biology, such as hypoxia, OS, and dyserythropoiesis, with an emphasis on the genesis of DN. Eventually, we propose novel and potentially promising therapeutic approaches for DN.

OXIDATIVE STRESS

OS results from the accumulation of ROS and disrupts cellular function. Its existence and its possible localization in diabetes have been disputed. Williamson *et al.*[7] demonstrated an increased cellular nicotinamide adenine dinucleotide ratio ($NADH/NAD^+$) and suggested that diabetes is a state of 'reductive stress' and 'pseudo-hypoxia' rather than OS. On the contrary, OS was postulated in diabetes on the basis of indirect evidence including increased nicotinamide adenine dinucleotide phosphate ratio ($NADP^+/NADPH$) and of oxidized to reduced glutathione.[8–10] Still, Wells-Knecht *et al.*[11] argued against a 'generalized' OS in diabetes: the age-adjusted levels in skin collagen of two oxidized amino acids, *ortho*-tyrosine and methionine sulfoxide, proved virtually identical in diabetics and nondiabetics.

In contrast, we demonstrated a 'local' OS in the human diabetic kidney.[12,13] Advanced glycation end products (AGEs), generated nonenzymatically with sugars on proteins, include two different classes of structures: OS-dependent molecules (pentosidine and N^{ε}-(carboxymethyl)lysine) and OS-independent molecules (pyrraline). Should tissue AGE formation depend solely on hyperglycemia, all AGE structures should be detected in the diabetic kidney. The identification of individual AGE structures established that this is not the case. Both pentosidine and N^{ε}-(carboxymethyl)lysine were present in diabetic glomerular lesions, together with other protein modifications derived from the oxidation of lipids (for example, malondialdehyde-lysine), whereas pyrraline was absent. The contention of a 'local' OS in DN was subsequently confirmed in diabetic vascular lesions by us and others[14,15] and is now supported by a large body of evidence gathered in *in vitro* experiments as well as in *in vivo* animal and human studies.[16,17]

The primary cause of local OS in DN remains debated as ROS are generated by numerous enzymatic and nonenzymatic sources,[18–22] for example, the activation of the renin–angiotensin system, of NADPH oxidase, of nitric oxide synthase, and so on.

A newer pathway for OS has recently emerged: the prolylhydroxylase-1 (PHD1)–hypoxia-inducible factor (HIF) system. Aragonés *et al.*[23] demonstrated in mice that the genetic disruption of PHD1, an intracellular oxygen sensor, lowers oxygen consumption in mitochondria of skeletal muscle, mitigates the OS, and enhances cellular survival during hypoxia.

HYPOXIA

Renal tissue hypoxia remains difficult to document directly from blood or urine analyses; however, recently, molecular imaging technologies have allowed an evaluation of renal oxygen levels. For instance, blood oxygen level–dependent magnetic resonance imaging performed in a healthy subject given 1 l water load after an 8-h water restriction documents a significant increase in the oxygen level of the renal outer medulla.[24] In addition, inhibition of sodium reabsorption in the outer medulla by furosemide should reduce oxygen consumption and, indeed, renal blood oxygen level–dependent magnetic resonance imaging reveals a rise in medullary oxygen level within 15 min after furosemide administration to a healthy subject.[24]

Tissue hypoxia in the streptozotocin-induced diabetic rat kidney has been visualized by Ries *et al.*[25] by blood oxygen level–dependent imaging, a finding confirmed later by Rosenberger *et al.*[26] by pimonidazole staining (a probe to detect hypoxia) and HIF.

Localization of hypoxia within the kidney is hampered by the scarcity of methods that are able to identify and quantify tissue oxygenation at the cellular level. Tanaka *et al.*[27] relied on a new hypoxia-responsive reporter vector to generate a novel hypoxia-sensing transgenic rat. In this model, they identified 'diffuse cortical' hypoxia in the puromycin aminonucleoside-induced nephrotic syndrome and 'focal and segmental' hypoxia in the remnant kidney model. In both models, the degree of hypoxia was positively correlated with microscopic tubulointerstitial injury. Localization of tissue hypoxia may thus differ according to the type of renal disease, but remains precisely elusive in DN.

The causes of chronic hypoxia in DN are heterogeneous.[28–31] Glomerular efferent arterioles enter the peritubular capillary plexus to provide oxygen to tubular and interstitial cells. Lesions in efferent arterioles decrease the number of peritubular capillaries, which in turn impair oxygen diffusion to tubulointerstitial cells and lead eventually to tubular dysfunction and fibrosis. Dyserythropoiesis and anemia associated with chronic kidney disease further hinder oxygen supply.

Hypoxia not only causes local OS in DN, but also affects various biological reactions linked to oxygen metabolism,[32] including nitrosative stress,[33–35] advanced glycation and carbonyl stress,[36–38] and endoplasmin reticulum stress.[39,40] The interrelationship between these detrimental chain reactions is so complex that a single culprit unlikely accounts for the alterations of DN. Whatever the sequential events of diabetic renal injury, the consequences of hypoxia and the attendant impairment of oxygen metabolism is pivotal in the genesis and progression of DN. Therapies interfering with it may prove clinically useful.

DYSERYTHROPOIESIS

EPO production occurs mainly in the kidney and is reduced in CKD patients with an eventual anemia.[28] Plasma EPO concentration is dramatically reduced in a uremic animal

model.[41,42] Recombinant human EPO has been used for more than 20 years in CKD to compensate for the reduced endogenous EPO production.[3]

Recent studies have indicated that EPO administration improves kidney functions in CKD either directly or indirectly.[43] Low hemoglobin levels are associated with adverse outcomes such as renal and cardiac failure, the so-called cardio–renal anemia syndrome.[43,44] A broad array of cellular processes is modulated not only by the mitigation of hypoxia but also by the development of progenitor stem cell, cellular integrity, and angiogenesis.[43,44] The therapeutic benefits of EPO beyond the correction of anemia are still debated. It is noteworthy that recently evidence has been published on the pleiotropic effects of EPO on the central nervous and the cardiovascular systems as well as on the kidney.[45–47]

CELLULAR MECHANISMS

PHD–HIF pathway

Defense against hypoxia hinges upon the HIF[48,49] that activates a broad range of genes that stimulate erythrocytosis, angiogenesis, glucose metabolism, or cell proliferation/survival, and eventually protect hypoxic tissues. The level of HIF-α is determined by its oxygen-dependent degradation rate. In the presence of oxygen, it undergoes enzymatic hydroxylation by PHDs,[50,51] is recognized by the Hippel–Lindau tumor-suppressor protein (pVHL),[52,53] acting as an E3 ubiquitin ligase, and is rapidly degraded by the proteasome (Figure 1, upper panel).[54,55] During hypoxia, the nonhydroxylated HIF-α escapes interaction with Hippel–Lindau tumor-suppressor protein, is thus stabilized, and binds to its heterodimeric partner HIF-1β, mainly in the nucleus, to transactivate genes involved in the adaptation to hypoxic–ischemic stress.[56]

Three isoforms of the HIF-α subunit have been identified (that is, HIF-1α, HIF-2α, and HIF-3α).[57] HIF-1α and HIF-2α are structurally and functionally similar. In contrast, HIF-3α lacks the structures for transactivation present in the C-termini of HIF-1α and HIF-2α and might play an alternative role as a negative regulator of hypoxia-inducible gene expression.

Recent studies in mice, utilizing gene disruption of either HIF-1α or HIF-2α, disclosed that HIF-2α acts as a physiological regulator of EPO.[58] In humans, the *HIF2A* gene is responsible for familial erythrocytosis[59] and for comparatively high hemoglobin concentrations in polycystic kidney disease[60] (pericystic hypoxia leading to HIF-2 induction). In addition, it plays a crucial role in the defense against OS.[23,61]

PHDs belong to the Fe(II) and 2-oxoglutarate-dependent dioxygenase superfamily, which incorporates two atoms of molecular oxygen into their substrates:[57] the first, used in the oxidative decarboxylation of 2-oxoglutarate, yields succinate and carbon dioxide, whereas the second is incorporated directly into the proline residue of HIF-α. They are called 'oxygen sensors' as their activity rigorously depends on oxygen tension.[62]

PHD activity critically requires iron and is thus inhibited by transition metal chelators.[62] Cobalt chloride inhibits PHD activity through an intracellular depletion of ascorbate necessary for iron (reduced) activity.[63] Its erythropoietic effect is known in humans since the 1940s[64,65] and has been utilized in the 1970s to treat anemia associated with chronic renal failure.[66] Unfortunately, cobalt chloride proved too toxic and is no longer in clinical use.

Three different PHD isoforms have been identified (that is, PHD1, PHD2, and PHD3),[57] each of which has its own tissue and subcellular distribution.[67,68] PHD1 is exclusively nuclear, PHD2 is mainly cytoplasmic (but shuttles between nucleus and cytoplasm), and PHD3 is present in both cytoplasm and nucleus. PHD2 acts as a decisive oxygen sensor in the HIF degradation pathway.[69] Although hypoxia decreases overall PHD activity, upregulation of HIF-1α induces the expression of PHD2 and PHD3.[70] This HIF-induced PHD expression ensures rapid removal of HIF-α after reoxygenation. Feedback loops may thus exist during hypoxia signaling.[71,72]

Keap1–Nrf2 pathway

Nuclear factor-erythroid 2 p45-related factor 2 (Nrf2), a transcriptional factor, regulates the expression of several cellular antioxidant and cytoprotective genes[73,74] (Figure 1, lower panel). Upon exposure to OS and/or electrophiles, Nrf2 translocates into nuclei, heterodimerizes with a small Maf protein, eventually binds to the antioxidant/electrophile-responsive element, and activates the transcription of antioxidant genes, including heme oxygenase-1, glutathione peroxidase-2, NAD(P)H-quinone oxidoreductase 1, and glutathione *S*-transferase. Nrf2 thus causes a broad and coordinated set of downstream reactions against OS.

Nrf2-mediated transcriptional responses are protective in a variety of experimental animals models including oxidative lung injury and fibrosis, asthma, and brain ischemia–reperfusion damage.[75–77] For example, induction of renal ischemia followed by reperfusion in wild-type mice elevates Nrf2 levels and activates their downstream target genes in the kidney.[78] In contrast, Nrf2 deficiency enhances their susceptibility to both ischemic and nephrotoxic acute kidney injury.[79] Treatment of Nrf2 knockout mice with the antioxidants *N*-acetyl-cysteine or glutathione improves renal function. Furthermore, Nrf2 knockout mice with streptozotocin-induced diabetes progressively increase their urinary levels of nitric oxide metabolites (an indirect evidence of OS) and develop renal injury.[80] Upregulation of Nrf2 is thus a potential therapeutic target in order to mitigate OS-induced tissue injury.

The regulation of Nrf2 has been recently elucidated (Figure 1, lower panel). Nrf2 is ubiquitinated continuously through the Keap1–Cul3 system and degraded within the proteasome.[81,82] Its level depends on its rate of destruction. Keap1 is a sensor of OS and acts as a negative regulator of Nrf2.[83] Under OS, reactive cysteines within the Keap1 moiety undergo conformational changes, eventually leading to the detachment of Nrf2 from Keap1 and the inhibition of its