An Update of Pathogenesis and Treatment in Patients with Chronic Kidney Disease (CKD) and Cardio–Renal Syndrome

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Chronic kidney disease (CKD) is a major cause of end–stage kidney disease (ESKD). The growing body of evidences has demonstrated that even minor renal dysfunction and/or albuminuria are associated with increased risks for cardiovascular events, and therefore the concept ‘cardio–renal syndrome’ is well recognized. Recently, over 300,000 Japanese patients had maintenance dialysis therapy. The diabetic kidney disease, IgA nephropathy, nephrotic syndrome and hypertension are major factors driving the progression of CKD to ESKD. The last decade has seen an evolution and ongoing refinement of a disease–oriented approach to CKD that offered an advantage of aligning patient’s comorbidities. In this review, we summarize and discuss the evolution of disease–oriented approach for CKD and the underlying mechanisms for cardio–renal syndrome, focused on prevention, early diagnosis, and treatment of individual disease.

Key words: diabetic kidney disease, IgA nephropathy, nephrotic syndrome, cardio–renal syndrome, biomarker

Introduction

The definition and classification of chronic kidney disease (CKD) guidelines were introduced by the National Kidney Foundation (NKF) Kidney Disease Outcomes Quality Initiative (KDOQI) in 2002. CKD includes urinary excretion of albumin >30 mg/day and/or reduction in kidney function defined as a decrease in estimated glomerular filtration rate (eGFR) <60 ml/min/1.73 m² for a period longer than three months, in the presence of kidney damage verified by imaging or histologic methods. The major cause of CKD is diabetes, inflammatory disease (glomerulonephritis, interstitial nephritis), hypertension, arteriosclerosis and polycystic kidney disease. In early stages of CKD, patients usually have no symptoms, however there are changes in presence of albuminuria, increase of serum creatinine, estimated GFR (eGFR) reduction, especially in patients at risk. Determining the grade of renal impairment is important because of different approaches to treatment, monitoring, expected complications, and patient education. Due to improved diagnostic methods and population aging, CKD is diagnosed ever more increasingly. Recently, CKD is a public health priority worldwide in all racial and ethnicity. As reported by the Japanese Society of Nephrology, 13.0 million people are diagnosed as CKD, and CKD is a major cause of end–stage kidney disease (ESKD). Basic principles of screening and diagnosis of CKD as well as better understanding of pathogenesis of cause of CKD provide appropriate care to prevent ESKD.

Diabetic kidney disease (Lecturer: Tomohito Gohda)

In classical diabetic nephropathy (DN), patients with diabetes develop microalbuminuria (MA),
which leads to macroalbuminuria and then to progressive decline of GFR and eventually ESKD. However, some patients with diabetes start renal function decline without remarkable increase of albuminuria by advance in treatment of diabetes and/or older age. Therefore, it becomes a challenge to distinguish between classical DN and nephrosclerosis with concomitant diabetes. The term diabetic kidney disease (DKD) generally includes patients with not only classical DN, but also CKD as a result of nephrosclerosis and other causes, although the official definition of DKD is under discussion. The concepts structure of CKD, DKD, and DN is shown in Figure-1.

DKD develops approximately 30–40% of patients with diabetes, and is a major cause of morbidity and mortality in patients with both types of diabetes. Since 1998 the most common cause of ESKD has become diabetes instead of chronic glomerulonephritis according to the annual report by the Japanese society of dialysis therapy (JSDT). However, the annual hemodialysis induction rate attributed to diabetes (44%) has almost stopped increasing in the past 6 years. Can we explain this plateau (stabilized rate) by advance in diabetic care alone? In fact, various sorts of agents have been sold for patients with diabetes and its related diseases such as hypertension and dyslipidemia in the past. A quarter-century ago, sulfonylurea agent was only available oral glucose-lowering agent for the treatment of type 2 diabetes (T2D), because biguanide agent was not available due to risk of lactic acidosis at that time. Then, different kinds of glucose-lowering agents such as α-glucosidase inhibitors, thiazolidinediones, glinides, dipeptidyl peptidase-4 (DPP-4) inhibitors, and sodium-glucose cotransporter (SGLT) 2 inhibitors were developed in a rapid pace. These novel drugs allowed us to control of blood glucose easily unlike before. However, the issue seems to be irrevocably complex. Based on the data of National Health and Nutrition Examination (NHANES) in the United States, the overall prevalence of DKD did not change significantly, whereas the prevalence of increased albuminuria was decreased and the prevalence of reduced glomerular filtration rate (GFR) was increased. These results suggest that the core of DKD has changed from increased albuminuria to reduced GFR over the last 2 decades. The development of albuminuria has considered to be an initial clinical manifestation of DKD, and the levels of albuminuria are associated with the progression of DKD even in patients with diabetes and normoalbuminuria. However, a growing body of evidence is accumulating to demonstrate that albuminuria itself lacks the sensitivity and specificity to predict future renal function decline, and clinical course of DKD is diversified into various kinds, likely because of change in therapeutic approach [e.g. preferred renin–angiotensin–system (RAS) inhibitors usage] (Figure-2). Therefore, the development of specific biomarker as an alternative to albuminuria is urgently needed. We have demonstrated that circulating tumor necrosis factor (TNF) receptor 1 (TNFR1) and TNFR2 have predicted future GFR decline and ESKD in patients of a wide variety of stages and both types of diabetes. Note that these associations are independent of conventional relevant clinical covariates such as age, hemoglobin A1c, albuminuria, baseline GFR, blood pressure, and treatment with RAS inhibitors.

Current therapeutic strategies of DKD are basically dependent on early identification, management of hypertension by RAS-priority blood

![Conceptual diagram of diabetic kidney disease](image)
pressure-lowering agent, and thoughtful glycemic control (i.e. avoiding hypoglycemia). Regarding the blood glucose control, approximately 70% of Japanese patients are now treated with DPP-4 inhibitor. Because DPP-4 inhibitors appeared to be a safe (lower risk of hypoglycemia) and effective to improve glycemic control even in patients with diabetes and CKD. Saxagliptin, a selective DPP-4 inhibitor, have been shown to be beneficial in reducing albuminuria in patients with T2D, irrespective of baseline albuminuria stage. Recent large-scale, randomized controlled clinical trial showed that SGLT2 inhibitors might be beneficial preventing the progression of renal disease in addition to reduction of cardiovascular (CV) events in patients with T2D at high CV risk. The mechanisms by which SGLT2 inhibitors protect renal function are supposed to be derived from not only glucose-lowering effect but also their pleiotropic actions including (1) reduction of glomerular hyperfiltration; (2) osmotic diuretic, natriuretic, and uricosuric effects which reduce blood pressure, serum uric acid, and body weight; (3) reduction of fat mass due to lipolysis; (4) activation of hypoxia-inducible factor 1 followed by erythropoiesis; and so on. Combination therapy with RAS and SGLT2 inhibitors might be potential for improving the long-term prognosis for renal function in patients with T2D at high CV risk and normal renal function although more studies are needed whether this therapeutic approach can be also applicable in patients with T2D at low CV risk or reduced renal function.

IgA nephropathy (Lecturer: Hitoshi Suzuki)

IgA nephropathy (IgAN; nephropathy with mesangial IgA and IgG deposits, so-called Berger disease in France) is the most common type of primary chronic glomerulonephritis in the world and was first described by Berger and Hinglais in 1968. IgAN has a significant morbidity, culminating in end-stage of kidney disease (ESKD) in about 40% of patients within 20 years of the diagnosis. Histopathologically, IgAN is characterized by the expansion of glomerular mesangial matrices with mesangial cell proliferation and/or mononuclear cell infiltration. Glomeruli typically contain generalized diffuse granular mesangial deposits of IgA (mainly polymeric IgA1), IgG, and C3. Electron microscopy revealed electron-dense deposits in the glomerular mesangial areas. Therefore, this disease is generally considered to be an immune complex-mediated glomerulonephritis. The main production site of IgA is the mucosa of the respiratory tract and intestine, and the onset and exacerbation of IgAN are often related with infections of the upper respiratory tract and gastrointestinal tract. IgA secreted from the mucosa is mainly dimeric or polymeric form, and the IgA deposited in the
glomeruliof IgAN patients has the same properties, suggesting that mucosal immunity plays a central role in the pathogenesis of this disease. It is postulated that many antigenic substances, i.e., viruses, bacteria, fungi, or food, may stimulate the immune complex formation in patients with IgAN.

Renal biopsy is required for the definite diagnosis of IgAN. However, the findings may differ according to the timing of renal biopsy during the 20-years clinical course of IgAN. Moreover, renal biopsy is not frequently performed because of procedural risks, and renal biopsy provides only a snap-shot of the disease status in the assessment of disease activity. Poor prognosis of IgAN is partly a result of delayed diagnosis. The strategies for early diagnosis leading to early and effective medical intervention are urgently needed. Taken together, useful methods for activity assessment other than renal biopsy and urinalysis are needed.

To determine disease activities, it is necessary to develop new biomarkers specific for IgAN. Several recent studies suggest that galactose-deficient IgA1 (Gd-IgA1) is essential in the pathogenesis of IgA nephropathy. Gd-IgA1 has been identified as one of the key effector molecules in the pathogenesis of IgAN, although the underlying molecular mechanisms remain under investigation. Recently, a multi-hit hypothesis regarding the pathogenesis of IgAN was proposed. In this hypothesis, three major steps are required for the onset and progression of IgAN: overproduction of Gd-IgA1 and autoantibodies against Gd-IgA1, formation of immune complexes, and deposition of those immune complexes in glomeruli (Figure-3).

Recent studies demonstrated that increased Gd-IgA1 levels associated with progression of proteinuria and a greater risk of deterioration of renal function in IgAN. In addition, the combination of high serum Gd-IgA1 levels and circulating levels of advanced oxidation protein products were correlated with progression of renal injuries in IgAN. Berthoux et al. reported that serum levels of IgG and IgA autoantibodies (specific for Gd-IgA1) at the time of renal biopsy were significantly associated with clinical progression of IgAN towards dialysis or death. Moreover, our recent study revealed that improvement of urinary abnormalities was well correlated with decreased serum levels of Gd-IgA1 and Gd-IgA1 containing immune complexes. Those findings confirmed the multi-hit hypothesis for the disease mechanism of IgAN, and indicate the possibility that evaluation of not only serum levels of the autoantigen (Gd-IgA1) but also those of autoantibodies against Gd-IgA1 or immune complexes should be required as disease markers for IgAN.

Depend on this pathogenetic hypothesis, we establish a robust and stable enzyme-linked immunosorbent assay (ELISA) method that uses a specific monoclonal antibody (KM55 mAb) to recognize human Gd-IgA1. The KM55 mAb bridges the independent status of Gd-IgA1 to support a multi-hit hypothesis. Importantly, KM55 mAb (Gd-IgA1-specific monoclonal antibody) can detect glomerular IgA specifically in IgAN. Although tonsillectomy combined with steroid therapy is effective for patients with IgAN, an early and timely manner is associated with higher likelihood of clinical remission. The noninvasive and real-time testing with KM55 mAb for the

Figure-3 Multi-hit model of pathogenesis of IgAN
Three major steps are required for the onset and progression of IgAN: i.e., overproduction of Gd-IgA1 (Hit 1) and autoantibodies against Gd-IgA1 (Hit 2), and formation of immune complexes (Hit 3), resulted in deposition of those immune complexes in glomeruli.
measurement of serum levels of Gd–IgA1 can pave the way for a more convincing diagnosis and activity assessment of IgAN.

IgAN is an autoimmune disease caused by the glomerular deposition of nephritogenic circulating immune complexes consisting of Gd–IgA1 (autoantigen) bound by anti-glycan autoantibodies. A better understanding of the multi-step process of the pathogenesis of IgAN and environmental contributing factors will lead to the development of specific biomarkers to identify patients with progressive disease who would benefit from a future disease specific therapy.

**Nephrotic syndrome (Lecturer: Teruo Hidaka)**

Nephrotic Syndrome is defined as large amount of proteinuria (3.5 g/day), low albumin (serum-alb <3.0 g/dl), edema, and lipid abnormality. It depends on various kidney diseases and systemic diseases. Here, we explain the mechanism of protein leakage in the kidney and new treatment.

The kidney filters a lot of blood on one day, and it treats 5,760 g/day of protein in the glomeruli and blocks 99.94% of them. Two barriers mechanisms of (1) charge barrier and (2) size barrier protect protein leakage, and nephrotic syndrome is thought to imply of these mechanisms.

Charge barrier is composed by glycocalyx, glomerular endothelial cells and glomerular basement membrane (GBM) and it exhibits negative charge. Because erythrocytes and albumin are negatively charged, it is thought that they prevent albumin leakage by the repulsive force of both.

Glycocalyx is made up of layers consisting of glycosaminoglycans, proteoglycans and complex carbohydrates that is existed on the endothelial cell luminal side, and it became possible to be visualized by recent fixation technique of electron microscopy. Although glomerular endothelial cells anatomically belong to fenestrated capillary, it is a special structure that does not have diaphragm unlike other things existing in other parts of the whole body. GBM is composed of laminin (α5β1γ1), type IV collagen (α345), perlecain, agrin, and endostatin. Laminin is required for podocyte adhesion and it is known that Alport syndrome develops due to abnormality of type IV collagen. Minimal change nephrotic syndrome (MCNS) is thought the disease of disrupt charge barrier.

Size barrier is a filtration barrier composed of podocyte slit–membrane, GBM and endothelial cells, which normally allows molecules of 1.8 nm or less to pass freely. 4.0 nm or more is completely blocked by this barrier. (Albumin is 3.6 nm, and this is blocked by charge barrier). The injuries of their
failures cause protein leakage and are applicable to kidney diseases except MCNS. Recent findings indicate that the mutation of genes coding nephrin\textsuperscript{24}, podocin\textsuperscript{25}, α-actinin 4\textsuperscript{26} and TRPC6\textsuperscript{27} exhibits nephrotic syndrome (Figure-4\textsuperscript{28}).

Nephrin was discovered at 1998 and this report had a great impact to the relevance of podocyte slit-diaphragm\textsuperscript{24}. Until then it was thought the nephrotic syndrome was mainly depend on GBM problem, after this report the attention to podocyte was increased. Up to now, more than 20 podocyte proteins have been discovered and it has been demonstrated that they are related to kidney disease tightly. Especially the phospholipase A2 receptor (at 2009)\textsuperscript{29} and thrombospondin type-1 domain-containing 7A (at 2014)\textsuperscript{30} were identified as the two major autoantigens in primary membrano-nous nephropathy.

Conventionally, steroid therapy was common as treatment of nephrotic syndrome. On the other hand nephrosis resistant to steroids and frequent recurrent nephrosis was difficult to treat with steroids. However, as Rituximab (RTX) emerges, it is taking a great deal of attention as a new treatment that turns into steroids with superior remission rate and few side effects, and it is already applied clinically\textsuperscript{31}. RTX is a chimeric monoclonal murine/human antibody, which is a glycosylated immunoglobulin including human IgG1 constant regions, murine light-chain and heavy-chain variable region\textsuperscript{30}. After administration of RTX the cell numbers of CD19/CD20 are significantly decreased for about 6 months, its effectiveness are correlate to the depletion of CD19/CD20 number\textsuperscript{31}.

**Cardio-renal syndrome (Lecturer: Seiji Ueda)**

In the last decade, numerous epidemiological studies have confirmed that even minor renal insufficiency could be associated with high risks of cardiovascular disease (CVD). Cardiovascular disease is the most common cause of death, responsible for 40% to 50% of all deaths in chronic CKD patients\textsuperscript{32}. The increase in cardiovascular risks in CKD may be ascribed, at least in part, to several factors such as hemodynamic alternations, anemia, dyslipidemia, a high prevalence of diabetes and hypertension, increased oxidant stress and uremic toxins, neurohormonal overactivity, endothelial dysfunction (ED) and inflammation\textsuperscript{33} (Figure-5).

On the other hand, in the setting of heart failure (HF), decreases in left ventricular systolic or diastolic function result in decreased cardiac output, stroke volume, and underfilling of the arterial beds. In HF patients with volume overload, low systemic pressures combined with increased central venous or pulmonary artery pressures can lead to compromise of renal perfusion pressure, which could

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**Figure-5** Pathophysiological interactions contributing to cardio-renal syndrome

The increase in cardiovascular risks in CKD may be ascribed, at least in part, to several factors such as hemodynamic alternations, anemia, dyslipidemia, a high prevalence of diabetes and hypertension, increased oxidant stress and uremic toxins, neurohormonal overactivity, endothelial dysfunction and inflammation. CKD: chronic kidney disease, CVD: cardiovascular disease, MBD: mineral bone disease.
worsen renal dysfunction. Indeed, renal insufficiency is reported to be present in more than one-third of patients with decompensated HF or with symptoms referred to as New York Heart Association class III and class IV. Accordingly, impairment of one organ could accelerate pathological processes in the other, which in turn accelerates the progression of failure of both. Although clinical studies hint at a specific bidirectional interaction between cardiovascular system and kidney, insight into the pathogenesis of cardio-renal syndrome (CRS) remains limited. One possible factor that could explain this link is ED. ED is a common feature in both CVD and CKD patients. In addition to the fact that ED is one of initiating factors for atherosclerosis in patients with hypertension, diabetes and CKD, accumulating evidence has suggested that ED is a risk factor for the progression of renal injury in CKD patients as well.

Reduced production and/or impaired bioavailability of nitric oxide (NO) are a characteristic feature of ED. NO is synthesized by oxidation of the terminal guanidine nitrogen of L-arginine via activation of the NO synthase (NOS). Guanidino-substituted analogues of L-arginine, such as asymmetric dimethylarginine (ADMA) can block the activation of the NO synthase (NOS). Guanidino-terminal guanidine nitrogen of L-arginine via the inhibition of the NOS active site. It has been demonstrated that plasma ADMA levels are increased in patients with HF, hypertension, diabetes, hyperlipidemia, and CKD. In addition, numerous epidemiological studies have revealed that its plasma levels could be a strong predictor for future cardiovascular events in these patients. Further, ADMA also could predict the development of renal injury in patients with CKD. For this, it has been previously demonstrated that inhibition of NOS accelerated renal damage in a remnant kidney model by eliciting peritubular capillary (PTC) loss. Therefore, ADMA-elicited decreased NO bioavailability in CKD may cause PTC loss and/or impaired PTC flow, which could contribute to tubulointerstitial ischemia and fibrosis in CKD. Indeed, we previously found that ADMA levels were correlated to PTC rarefaction, the degree of tubulointerstitial fibrosis and renal function loss in CKD model rats, all of which were attenuated by reduction of ADMA. Moreover, ADMA reduction prevented the tubulointerstitial ischemia in an animal model of early diabetic nephropathy by ameliorating impaired capillary flow. Therefore, increased ADMA in CKD patients causes impairment of PTC flow and/or PTC rarefaction, resulting in tubulointerstitial hypoxia and therefore contribute to the development of CKD. Taken together, ADMA is a potent causal factor for ED in CKD and its accumulation may contribute to the pathogenesis of cardio-renal syndrome. Therefore, it is under discussion that development of a specific therapy reducing ADMA levels in cardio-renal syndrome is an urgent issue.

**Conflict of interest statement**

The author declares that there are no competing interests.

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