REVIEW ON RECENT ADVANCES IN THE MANAGEMENT OF DIABETIC NEPHROPATHY

Sriram S²*, Suresh Damodaran¹ and Jipin Thomas Jacob²

¹Consultant Diabetologist, Sri Ramakrishna Hospital, Coimbatore.
²College of Pharmacy, Sri Ramakrishna Institute of Para Medical Sciences, Coimbatore.

ABSTRACT

Diabetes mellitus and chronic kidney disease have become two of the fastest growing pathologies worldwide while diabetic kidney disease is still the leading cause of CKD and end stage renal disease. Population ageing and increase in prevalence of many interrelated comorbidities suggest that these numbers will worsen in the near future. Diabetic nephropathy is the leading cause of chronic renal disease in patients starting renal replacement therapy. DN has been classically defined as an increased protein excretion in urine. Early stage is characterized by a small increase in urinary albumin excretion, also called microalbuminuria or incipient DN and advanced disease is defined by the presence of macroalbuminuria or proteinuria or overt DN. The two main risk factors for DN are hyperglycemia and arterial hypertension and also because of smoking, dyslipidemia, proteinuria, glomerular hyperfiltration and dietary factors. For the prevention and treatment of DN, it is important to know the stage of DN which is the target of intervention and the outcome of interest and also to know the risk factors. Classical nonspecific measures include glycemic control, blood pressure control, weight loss, protein restriction, and smoking cessation and treatment measures include use of renin-angiotensin aldosterone blockers, vitamin D analogues like paricalcitol, nonspecific phosphodiesterase inhibitors like pentoxifylline, endothelin receptor antagonists, antifibrotic and anti-inflammatory agents and new methods like use of novel biomarkers, micro RNA and drug discoveries with targets like NOX, PKC-β etc.

KEYWORDS: Diabetic Nephropathy, CKD, Hyperglycemia, Proteinuria.
Diabetic nephropathy is the leading cause of chronic kidney disease and a major cause of cardiovascular mortality. The persistent rise in the proportion of chronic dialysis patients resulting from diabetic kidney disease in Asian countries, including Japan, over the past 20 years has been associated with higher mortality. The prevalence of DN varies according to ethnicity: it is higher in African-Americans, Asians and Native-Americans than in Caucasians. African-Brazilians are more susceptible to progress to end-stage renal disease than people of European ancestry, but there appears to be a similar prevalence of micro- or macroalbuminuria. In the early 1980s, seminal studies from Europe revealed that small amounts of albumin in the urine, not usually detected by conventional methods, were predictive of the later development of proteinuria in type 1 and type 2 diabetic patients. The cumulative incidence of microalbuminuria in patients with type 1 diabetes was 12.6% over 7.3 years according to the European Diabetes (EURODIAB) Prospective Complications Study Group and 33% in an 18-year follow-up study in Denmark. In patients with type 2 diabetes, the incidence of microalbuminuria was 2.0% per year. According to Prospective Diabetes Study (UKPDS), proteinuria occurs in 15–40% of patients with type 1 diabetes, with a peak incidence around 15–20 years of diabetes. In patients with type 2 diabetes, the prevalence is highly variable, ranging from 5 to 20%. Compared with Western countries, the proportion of patients with diabetes receiving a recommended medical evaluation, such as an annual urinary albumin measurement, has increased from 21.7% in 2000 to 27.2% in 2006. Viberti et al. reported microalbuminuria in 12.6% of the 87 type 1 diabetics, from whom 88% progressed to severe albuminuria when followed for 14 years. Similar progression rates from microalbuminuria to macroalbuminuria by Mogensen and Christensen were found among 43 studied 1969-1976 and re-evaluated in 1983. Hovind et al. reported a decline in prevalence of proteinuria from 1965-1969 to 1979-1984, 31.2% and 13.7% respectively. Diabetic nephropathy is a distinct phase clinical syndrome with partial or complete loss of kidney function. The structural changes in kidney including thickening of basement membrane, mesangial expansion, glomerular hypertrophy, fibroblast proliferation, matrix deposition, glomerulosclerosis and tubular necrosis are generally observed in the patients of diabetic nephropathy. Various functional abnormalities of kidney such as persistent elevated albuminuria, elevated arterial blood pressure, declined glomerular filtration rate (GFR) and fluid retention are also associated with diabetic nephropathy. The down regulation of endothelial nitric oxide synthase (eNOS), peroxisome proliferator-activated receptor-γ (PPAR-γ) has been noted to be involved in pathogenesis of diabetic nephropathy. The elevated levels of several pathological substances such as vasoactive peptide like angiotensin-
II, endothelin-1 and growth factors like vascular endothelial growth factor (VEGF), transforming growth factor-β1 (TGF-β), advanced glycation end (AGE) products and lipid mediators such as 5-lipoxygenase derived substances like 12-hydroxyeicosatetraenoic acid (12-HETE) and 20-hydroxyeicosatetraenoic acid (20-HETE) have been implicated in the pathogenesis of diabetic nephropathy. According to UAE values, DN has been categorized into stages: Normoalbuminuria, microalbuminuria and macroalbuminuria. Although microalbuminuria is considered a risk factor for the development of macroalbuminuria, not all patients progress to this stage, and some may regress to normoalbuminuria. The initial studies suggested that about 80% of type 1 diabetic patients with microalbuminuria would progress to proteinuria over a period of 6 to 14 years. More recent studies suggest that only 30 to 45% of microalbuminuric patients will progress to proteinuria over 10 years of follow-up. In fact, some of them will present regression to normoalbuminuria. This might be the result of more intensive glucose and BP control strategies employed in the last decade than in the initial studies. This regression of microalbuminuria is more frequent among subjects with short duration of microalbuminuria, glycosylated hemoglobin A1c (HbA1c) below 8%, systolic BP <115 mm Hg, and favorable lipid profile (serum total cholesterol <198 mg/dl and triglycerides <145 mg/dl). On the one hand, not all subjects will progress to overt DN, and some might even regress as stated before. On the other hand, subjects in the upper-normal range of albuminuria seem to be at high risk for complications. In the microalbuminuric stage, no decline in GFR is expected. Once the subject has developed macroalbuminuria, the expected GFR decline is 1.2 ml/min/month in type 1 DM, this could be decreased by BP treatment. In type 2 DM, the rate of GFR decline is less predictable. A mean decline of approximately 0.5 ml/min/month has been described, but in some patients GFR may remain stable for long periods of time.

<table>
<thead>
<tr>
<th>Stage</th>
<th>Urine with marked time (µg/min)</th>
<th>24 hr urine (mg/24 hr)</th>
<th>Random urine sample</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Albumin conc (mg/l)</td>
</tr>
<tr>
<td>Normoalbuminuria</td>
<td>&lt;20</td>
<td>&lt;30</td>
<td>&lt;17</td>
</tr>
<tr>
<td>Microalbuminuria</td>
<td>20-199</td>
<td>30-299</td>
<td>17-173</td>
</tr>
<tr>
<td>Macroalbuminuria</td>
<td>≥200</td>
<td>≥300</td>
<td>≥174</td>
</tr>
</tbody>
</table>

*values according to American Diabetes Association

[2] Nephropathy in type 2 DM is more likely to occur if the individual has a relative with diabetic nephropathy. If one parent has diabetic nephropathy, the prevalence of proteinuria is
14% among their offspring. This risk increases further if both parents have nephropathy. Diabetic renal disease is a complex genetic trait and cannot be attributed to a monogenic mutation. Genome wide association and linkage studies have identified several loci associated with renal disease in diabetes, including 3q, 7q, 10p, 14q, and 18q. Cardiovascular risk of diabetic kidney disease is high, Astrup et al. compared cardiovascular events and mortality in 199 types 1 DM with nephropathy to 192 with normoalbuminuria followed for ten years. Fatal and nonfatal cardiovascular events occurred in 40% of the nephropathy group and 10% in the normoalbuminuric group. Similarly, all-cause mortality, the majority cardiovascular, was much higher in the nephropathy group, 30% vs. 8%. An Italian cohort of 1538 type 2 DM patients were followed for 11 years to assess the relationship of GFR, albuminuria and mortality. Albuminuria (>200 μg/min) had a 2-fold increase in all cause and cardiovascular mortality compared to normoalbuminuria. In screening and diagnosis, the first step is to measure albumin in an isolated urine sample. The results of albuminuria in an isolated sample can be expressed as albumin concentration (mg/l) or as albumin/creatinine ratio (mg/g). Every abnormal albuminuria test should be confirmed in two of three samples collected at a three to six-months interval, due to the daily variability of UAE. Screening should not be performed under conditions that may increase UAE, such as hematuria, acute systemic diseases or fever, vigorous physical exercise, poor glycemic control, uncontrolled arterial hypertension and decompensated cardiac failure. Samples must be refrigerated if they are to be used the same day or the next day, and one freeze is acceptable before measurements. In situations in which UAE measurement is not available, semiquantitative dipstick measurements of albuminuria can be used, although these tests are less accurate. The quantitative methods most commonly used to measure albuminuria are immunoturbidimetry, immunonephelometry and radioimmunoassay. However, recently it has been observed that an appreciable quantity of albumin is not detected by routine immunoassay methods, defined as non-immunoreactive fraction, which results in an underestimate of UAE. On the other hand the HPLC method measures the immunoreactive and non-immunoreactive fractions which compose the total intact albumin, allowing the detection of earlier albumin elevations. DN screening must be performed when DM is diagnosed in patients with type 2 DM, since these individuals may have had a silent form of DM for some time already. For patients with type 1 DM, it is recommended that screening be performed beginning in the fifth year after DM diagnosis or earlier if the DM is chronically poorly compensated, or if the patient is an adolescent. In all cases, if albuminuria is normal, screening must be repeated annually.
The principles of prevention and treatment of DN are the same. However, the role of each factor could be different in each stage of disease. It is important to define the DN stage that is the target of intervention (microalbuminuria, proteinuria or GFR) and the outcome of interest. Both, ACE inhibitors and angiotensin receptor blockers (ARBs) seem to be effective reducing proteinuria and decreasing the creatinine doubling rate, but not decreasing mortality. In normo- or microalbuminuric subjects, the aim of treatment is to intervene at arterial hypertension, hyperglycemia, smoking habit and probably dyslipidemia. Clinical trials have demonstrated that intensive treatment of hyperglycemia is associated with a decreased risk for the development of DN in type 1 and type 2 diabetic patients. Clinical non specific measures include glycemic control, blood pressure control, weight loss, protein restriction and smoking cessation. DKD occurs in approximately 20% of diabetic patients, and it can appear despite a good glycemic control. Many important studies have demonstrated that a tighter glycemic control can delay the onset of DKD and slow its progression, beyond its wellknown cardioprotective effect. Glycemic control can be achieved through diverse pharmacological treatments. Some of them, such as incretin degradation inhibitors or glucagon-like peptide analogues, may have specific nephroprotective effects independent of their glycemic impact. In case of blood pressure control, latest 2012 KDIGO guidelines maintain a tighter blood pressure recommendation for proteinuric patients, regardless of etiology. Overweight and obesity are frequent comorbidities to diabetes and play an important role in the pathogenesis of CKD. This may be due both to a further increase in hyperfiltration and to specific hormonal dysregulations related to adipokines. Weight loss in obese diabetic patients has been shown to markedly reduce albuminuria. Dietary advice in DKD patients is a complex issue: it compels carbohydrate consumption regulation, but the frequent concurrence of comorbidities also requires a low-salt diet for hypertension, fat-free for dyslipidemia, and hypocaloric intake for obesity. Cigarette smoking has been linked to the appearance and progression of DKD, probably due to oxidative stress stimulation, and the cessation of this habit has also been associated with slower progression of the nephropathy. The choice of anti-hypertensive agents to use is in some way not a problem in clinical practice, because to reach the BP goals the majority of patients will need several agents. However, due to the known renoprotective effect of ACE inhibitors and ARB, these agents should be used initially associated with a diuretic. The use of angiotensin converting enzyme inhibitors (ACEI) or angiotensin II receptor blockers (ARB) to reduce proteinuria is currently the first step strategy. This benefit is valid for both type 1 and type 2 diabetic patients, even with low-grade proteinuria and normal GFR. The aim of ACE inhibitors and ARBs use is not
only to diminish the risk for the development of micro and macroalbuminuria but also to
decrease the occurrence of cardiovascular events. RAS blockade with ACE inhibitors or ARB
confers an additional benefit on renal function. This renoprotective effect is independent of
BP reduction. These drugs decrease UAE and the rate of progression from microalbuminuria
to more advanced stages of DN. A meta-analysis of 12 trials in non-hypertensive
microalbuminuric type 1 diabetic patients showed that ACE inhibitors decreased the risk of
progression to macroalbuminuria by 60%, and increased the chances of regression to
normoalbuminuria. Therefore, the use of ACE inhibitors or ARB is recommended for all
microalbuminuric patients, even if normotensive. ARBs were also effective in reducing the
development of macroalbuminuria in microalbuminuric type 2 diabetic patients. Dual RAS
blockade with ACEi and ARB might be more effective in reducing proteinuria compared
with monotherapy in patients with diabetic kidney diseases. Based on the Ongoing
Telmisartan Alone and in combination with the Ramipril Global Endpoint Trial
(ONTARGET), although combination therapy with ramipril and telmisartan reduces
proteinuria than monotherapy, it worsens major renal outcomes including dialysis, doubling
of serum creatinine and death. Thus, combination RAAS blockade should not be used in
diabetic patients, especially elderly type 2 diabetic patients with normoand/or
microalbuminuria. First, ACEi or ARB should be used and their dosage should be increased
to obtain an optimal anti-albuminuricand/or proteinuric response. Since RAAS has been
implicated in potentiating tubulointerstitial fibrosis in kidney disease, inhibition of renin would
decrease angiotensin II levels further.2 Aliskiren, a direct renin inhibitor, has been found to be
effective in lowering BP and lower proteinuria. However study to combine ARB and
Aliskiren, resulted in early termination of the study to due higher risk of cerebrovascular
events. The combination therapy of ARB or ACE inhibition and Aliskiren is not deemed to
be safe. Aldosterone is a profibrotic agent for myocardium and kidneys. This hormone is able
to activate NADPH oxidase which increases oxidative species in the mesangial cells.
Through the activation nuclear factor kB, it potentiate the production of proinflammatory and
profibrotic agents such CTGF and TGFβ. To prevent further progression of diabetic
nephropathy, aldosterone antagonists have been used to lower proteinuria. In combination
with ARB and ACE inhibitors, further proteinuria reduction with aldosterone antagonists has
been documented. Spironolactone and eplerenone may show better long term results since the
cardiovascular literature has found these medications effective in lowering risk of
cardiovascular mortality in high risk congestive heart failure populations. Aldosterone
antagonists are associated with more episodes of hyperkalemia and serum creatinine rise.
Type 1 DM

- End Stage (kidney and heart failure)

Type 2 DM

- Trandornapril

Microalbuminuria

- Lisnopril
- Imidapril

Macroalbuminuria

- Captopril

End stage renal disease

- Irbesartan
- Valsartan
- Telmisartan
- Irbesartan
- Losartan
- Aliskiren
Hyperglycemia

- Vasodilatory prostoglandins
- Hyperfiltration
- Abnormal response to Angiotensin II
- Atrial Natriuretic Peptide
- Abnormal endothelin/Nitric Oxide
- Growth hormone
- Hyperinsulinemia

Proteins

- Protein glycosylation AGE products
- Glomerular Hypertrophy
- Mesangial Matrix
- Glomerulosclerosis

ESRD
Among type 2 DM patients with macroalbuminuria, non-dihydropyridine calcium channel blockers, i.e. diltiazem and verapamil have shown to significantly reduce proteinuria. Bakris et al. compared ACE inhibitors, non-dihydropyridines and beta blockers on proteinuria reduction. The results were similar for the lisinopril or non dihydropyridine groups. However, atenolol did not reduce proteinuria despite equivalent blood pressure reduction. Amlodipine plus perindopril showed significant risk reduction in cardiovascular endpoints and new onset diabetes. Tighter control of blood pressure was achieved with captopril and atenolol in the UKPDS trial and both drugs were equally as effective in lowering risk of macrovascular and microvascular complications in type 2 diabetes. Since the purpose with the diuretics is to induce naturiesis, the combination of an ACE inhibitor or ARB with thiazides, allow better control of hyperkalemia. Hydrochlorothiazide, chlorthalidone and indapamide are diuretics used to treat BP and fluid overload states. Hydrochlorothiazides can induce metabolic derangements, hyperuricemia, hyperlipidemia and hyperglycemia, thus should be avoided in metabolic syndrome. Another class is AGE inhibitors where the areas of target is to inhibit the AGE activity, thus preventing the formation of AGE, inhibiting AGE cross linkage, and interaction with the AGE receptor. Aminoguanidine (Pimagedine), an AGE inhibitor, was tested in a randomized control trial for type 2 diabetes mellitus but was terminated due to other significant adverse events, such as glomerulonephritis and lupus like reactions. In diabetic nephropathy, endothelin 1 levels are increased and contribute to proteinuria and glomerulosclerosis, thus using endothelin antagonists. In animal models, vitamin D receptor activation resulted in lower albuminuria and less severe glomerulosclerosis. Vitamin D is also been shown to have an inhibitory effect on the RAAS., 25 OH Vitamin D directly suppresses plasma renin expression and activity. In humans, 25OH Vitamin D deficiency and insufficiency was associated with higher Angiotensin II and renin levels compared to sufficient Vitamin D 25OH individuals.

Recently there were many advancements in the field of management of diabetic nephropathy which includes biomarkers for repair of acute kidney injury which may be caused by diseases like diabetes, studies on gevokizumab, effect of SGLT-2 inhibitors on renal system, use of vasopressin V2 antagonists, role of microRNA’s, mesoblast cell therapy, drug discoveries targeting NOX, PKC-β, NF-κβ, CCL2 and TGF-β as targets etc. Acute kidney disease is a common clinical syndrome among hospitalized patients which may be due to reasons like diabetes, hypertension, obesity etc. In order to improve AKI management there is a critical need to develop a series of tests and biomarkers to detect renal function recovery and identify
patients with progressive kidney disease. Proteinuria and microalbuminuria are classical markers of chronic kidney disease progression and data on performance of other biomarkers of kidney repair and progression towards CKD are limited thus specifically the role of novel biomarkers including neutrophil gelatinase associated lipocalin, kidney injury molecule-1, and nephroneectin has been studied. In a recent animal model of ischemic reperfusion injury of the kidney, *Haverl* and *Lcn2* (genes that code for kidney injury molecule and neutrophil gelatinase associated lipocalin) were found to have the highest activity throughout the investigation period. Also urinary KIM-1 and NGAL may be surrogates for progressive kidney injury following IRI and the AKI-CKD transition. NPNT is a a8b1 integrin ligand involved in kidney morphogenesis and is found in the extracellular matrix of the Wolffian duct and the ureteric bud and its involvement with the recovery process and its timeline of expression may make it a reasonable target for clinical validation. Gevokizumab, a potent monoclonal antibody with unique allosteric modulating properties has shown significant decrease in proteinuria, a commonly seen abnormality in diabetic nephropathy, in a mouse model study of renal inflammation. It has also reduced inflammatory cytokine responses in blood and kidney cortex extracts as well as reduced development of fibrosis, the scarring of tissues that is central to the reduction of kidney function. It binds to IL-1 beta, a proinflammatory cytokine and modulates the cellular signaling events that produce inflammation. SGLT2 inhibitors, a new class of oral antidiabetes therapy, selectively target the SGLT2 protein and prevent renal sodium and glucose reabsorption in the kidney, a process known to be involved in the development of DN. Blocking the activity of SGLT2, which is almost exclusively expressed in the S1 segment of the renal proximal tubule, leads to substantial glucosuria and a reduction in plasma glucose levels. Diabetic nephropathy results from high glucose mediated inflammation and altered sodium handling that eventually results in fibrosis. There are both glomerular and tubulointerstitial damage, although the decline in renal function parallels more closely the degree of tubulointerstitial damage.

High glucose is directly responsible for the changes seen in diabetic nephropathy, and improved glycaemic control has been demonstrated to slow the progression of the disease. However, there are two processes that contribute to the disorder of diabetic nephropathy that may be expected to be altered by SGLT2 inhibitors, independent of their effect on glucose lowering. The proximal tubular cell secretes inflammatory molecules and growth factors in response to high glucose. This results in activation of an inflammatory cascade and recruitment of macrophages with propagation of hypertrophy and interstitial fibrosis. The
most important mediator of this pathogenesis is TGFβ, which promotes fibrosis and epithelial to mesenchymal transdifferentiation. There are a host of other inflammatory mediators and growth factors involved in this complex process. It is possible that a reduction in glucose transit through the proximal tubular cells may reduce proximal tubular cell-induced inflammation and fibrosis in diabetic nephropathy. Indeed, our group has shown that SGLT2 inhibition in immortalized proximal tubular cell lines (HK2 cells) reduced high glucose induced Toll-like receptor 2 and 4 as well as nuclear factor kappa B (NFκB) and activator protein 1 (AP1) expression, which are important inflammatory and fibrotic mediators in diabetic nephropathy. Another important aspect of diabetic nephropathy is hyperfiltration-associated renal injury. Glomerular hyperfiltration associated with enhanced sodium and glucose reabsorption in the proximal tubule occurs quite early in the disease process and plays an important role in diabetic nephropathy. Increased proximal tubular sodium reabsorption results in decreased distal delivery of sodium to the macula densa, which regulates tubuloglomerular feedback.

By inhibiting sodium reabsorption in the proximal tubule and by thereby increasing sodium delivery to the juxtaglomerular apparatus, it might be expected that the glomerular hyperfiltration would be reversed. Indeed, phlorizin, a nonselective SGLT inhibitor, has been shown to abrogate the development of hyperfiltration in whole animal studies, and in single nephron studies animals treated with phlorizin have a reduction in sodium reabsorption and normalization of glomerular filtration rate (GFR), disproportionate to the improvement in plasma glucose. [7] Copeptin, the precursor of vasopressin, is an easily measurable surrogate marker of vasopressin. High plasma copeptin concentration was strongly associated with the risk of severe renal outcomes in patients with type 2 diabetes and albuminuria. This association was independent of relevant covariates such as age, duration of diabetes, blood pressure, and baseline levels of HbA1c, UAE, and eGFR. Thus vaptans (vasopressin receptor antagonists) could selectively target the V2 receptor mediated deleterious effects of vasopressin (Lixivaptan, Mozavaptan, Satavaptan, Tolvaptan). [8] MicroRNAs (miRNAs) are endogenously produced short noncoding RNAs of about 21–25 nucleotides that have been shown to play important roles in modulating gene expression, thus affecting almost every key cellular function. Several miRNAs are related to DN. Some of them take part in the pathogenesis and development of the disease while others serve as DN-killers or -preventers. Therefore, it would be wise to elevate the renal-protective miRNAs and reduce DN-inducing ones. By using miRNA mimics, miRNA expression vectors, miRNA-containing exosomes,
and miRNA-inducing natural agents, levels of renalprotective miRNAs can be restored and thus lead to the protection from DN. It is noted that, among these miRNAs, miR-21 and miR-195, let-7, miR-200a are all DN-related, which might provide hope for the treatment of DN. Mesoblast Limited of Newyork and Australia announced that results from the Company's Phase 2 trial in patients with diabetic nephropathy showed that a single infusion of its intravenously delivered allogeneic mesenchymal precursor cell (MPC) product candidate MPC-300-IV was safe, reduced damaging inflammation, and preserved or improved renal function over at least 24 weeks. NOX generate ROS that modulate redox-sensitive cellular responses and are essential mediators of normal cell physiology. However, excessive ROS production by an overactive NADPH oxidase system probably mediates constitutive activation of signaling pathways involved in the initiation and progression of DN. This occurs through the selective oxidation of specific signaling enzymes and/or proteins that are linked to processes such as activation of transcription factors, secretion of cytokines or altering signaling proteins such as protein kinases and phosphatases. Currently, the NOX family consists of seven members: the dual oxidases DUOX-1 and -2 as well as NOX-1–5. NOX-2, earlier called gp91phox, was first found in neutrophils and phagocytic cells, together with p22phox, a small regulatory subunit, modulating respiratory burst. Further studies showed that p67phox, p47phox, p40phox and the small GTPaseRac also have a vital role in activating the NOX family. Apparently, inhibiting the activity of the NOX family can significantly prevent oxidase stress, which is a more direct means to decrease the generation of ROS and one of the efficient therapeutic strategies of DN, thus comes the use of two non specific NOX inhibitors, diphenyleneiodonium (DPI) and apocynin. Activation of the PKC pathway appears to be a crucial mechanism in the occurrence and progression of DN. PKC enzymes are activated by signals such as increasing concentration of diacylglycerol (DAG) or calcium ions (Ca2+). Long-term hyperglycemia is likely to lead to overproduction of DAG, which then activates one or more PKC isoforms in different tissues. Although it remains an additional consideration, there is also evidence that the PKC-a and PKC-b isoforms are most closely involved in DN, which provides us with a new potential way to treat DN. Ruboxistaurin (RBX), was discovered and investigated by Eli Lilly as an inhibitor of PKC-β. Owing to insignificant or no side-effects in a large amount of preclinical trials to prevent retinal and renal diseases, RBX has been investigated in several clinical trials to evaluate its efficacy in diabetic complications such as diabetic retinopathy, diabetic neuropathy and DN. NF-kB regulating the expression of numerous genes plays a significant part in the inflammatory response during human and experimental kidney injury. As a result, the
downregulation of NF-kB will be of great value for the inhibition of inflammation. Bardoxolone methyl induced the activation of powerful Nrf2-dependent phase 2 inducers, which selectively reduced the DNA binding of NF-kB to inhibit the inflammatory process.\textsuperscript{[21]}

In addition, a common finding that the high level of CCL2, a downstream chemokine, in the urine of patients with DN has been detected. Increasingly, research is showing that CCL2 is closely associated with the process of glomerular and interstitial inflammatory cell recruitment, such as macrophage cells. Hence, the blockade of the CCL2/CCL2 receptor pathway could have considerable therapeutic potential for DN. CCX140-B thus was discovered as an orally delivered therapy for the treatment of DN.\textsuperscript{[21]} Also new techniques like blood oxygen level dependent MRI has been introduced,\textsuperscript{[19]} high doses of thiamine and its derivative benfotiamine have been shown to reduce the rate of microalbuminuria in experimental diabetic nephropathy probably due to decrease in activation of protein kinase, protein glycation and oxidative stress.

**REFERENCE**

2. Sameena Iqbal and Ahsan Alam, Renal Disease in Diabetes Mellitus: Recent Studies and Potential Therapies, J Diabetes Metabolism 2013.
4. Themis Zelmanovitz, Fernando Gerchman et al, Diabetic nephropathy, BMC, 2009; 1-17
5. Jorge L Gross, Mirella J De Azevedo, Diabetes Care 2005; 164-171
19. Dr. JunaidNazar, In focus: Treatment Options Available For Diabetic Nephropathy, JPMS Medical Blogs 2015.