ABSTRACT:
Diabetic nephropathy is the most common cause of end stage renal failure. In addition to renin pathway, other bioactive molecules are involved in its pathogenesis. Strategies to interrupt pathophysiological pathways are on the horizon. Aliskiren is the renin inhibitor, blocking the first step in renin pathway. Protein kinase C overexpression is blocked by ruboxistaurin. Pentoxifylline and m-TOR inhibitors are anti-inflammatory in the pathogenesis of diabetic nephropathy. Inhibitors of advanced glycation, oxidative stress and plasminogen activator inhibitor-1 have proved useful in animal models of diabetic nephropathy. Avosentan, an endothelin antagonist decreases urinary albumin. Such targeted therapies would halt and rather may even reverse progression of diabetic nephropathy.

INTRODUCTION
Diabetic nephropathy (DN) is the leading cause of end stage renal failure worldover. Indicators of worsening of nephropathy are progression from stage of microalbuminuria to macroalbuminuria. So the goal of advanced nephropathy is to decrease albuminuria by atleast 30%. (1) This has well been demonstrated in numerous clinical trials by using renin-angiotensin-aldosterone system (RAAS) blockade and others like diuretics and calcium channel blockers. But are they enough or do we need something more in management of DN.

PATHOGENESIS OF DIABETIC NEPHROPATHY
Classical factors contributing to the pathology of diabetic nephropathy, e.g., hypertension, hyperglycemia, hyperinsulinemia, and hyperlipidemia, are now amenable to treatment. Current therapies however do not fully prevent its renal complications. RAAS and endothelins are associated with intraglomerular hypertension. In response, transforming growth factor β (TGF-β) is upregulated. (2)

Recent studies, mainly performed in experimental animals, have identified newer culprits in the pathogenesis, such as hypoxia, advanced glycation, oxidative stress, and other bioactive molecules like cytokines, growth factors and metalloproteinases. Animal experiments highlight the fact that renoprotection is not necessarily linked to hemodymanic (blood pressure) or metabolic (glycemic and lipid controls) alterations but appears rather associated with an improved hypoxia, oxidative stress, and/or advanced glycation.

NOVEL MOLECULES
Aliskiren-Direct renin inhibitor
Aliskiren is an orally active non-peptidic renin inhibitor that is highly specific for renin.

It acts by binding to the active site of renin, thereby inhibiting catalytic activity. As renin acts at the rate-limiting step in the RAAS cascade, the blockade of renin with aliskiren may cause more complete inhibition of the RAAS, and reduce the feedback effects when compared with ACEis and ARBs. In addition, the blockade of renin may reduce activity at the (pro)renin receptor (P)RR. Therefore, aliskiren may provide superior renoprotection.

Azizi et al. showed that in healthy human volunteers maintained on a high sodium diet a single 300mg dose of aliskiren caused a reduction in PRA and Ang I and II levels within 1 h of administration. In contrast, the ARB, valsartan, increased each of these parameters. (3) In streptozotocin-induced diabetes mellitus in transgenic Ren(2) rats, a model of advanced diabetic nephropathy in humans, aliskiren reduced albuminuria and glomerulosclerosis to a similar extent as the ACE inhibitor, perindopril. This effect was seen despite the fact that aliskiren did not lower blood pressure as much as perindopril. Reduction in tubulointerstitial fibrosis was significantly greater in aliskiren-treated diabetic rats compared with the perindopril-treated animals. (4)

Results of the first published clinical trial to consider the renoprotective effects of aliskiren were recently reported by Parving et al. in the New England Journal of Medicine. This multicenter, international trial studied 599 patients who had type 2 diabetes mellitus, hypertension, and diabetic nephropathy, defined as an early morning albumin-to-creatinine ratio of 4300mg/g, or 4200mg/g in patients taking medications that antagonized the RAAS. Combination therapy with maximal doses of losartan (100mg daily) and aliskiren (150mg daily for 3 months, then
300mg daily for a further 3 months) was compared with losartan and placebo. Treatment with aliskiren and losartan was superior to losartan and placebo, as evidenced by a 20% reduction in albuminuria compared with placebo (P<0.001). This effect seemed to be independent of blood pressure. (5)

The results of these animal and human studies are very encouraging and results of a large outcome trial in diabetic nephropathy, ALTITUDE (Aliskiren Trial in Type 2 Diabetes Using Cardio-Renal Endpoints) is awaited. The primary objective of the ALTITUDE trial is to determine whether aliskiren 300 mg once daily, reduces cardiovascular and renal morbidity and mortality compared with placebo when added to conventional treatment (including ACEi or ARB). ALTITUDE is an international, randomized, double-blind, placebo-controlled, parallel-group study. Primary outcome measure is time to first event for the composite endpoint of cardiovascular and renal death, resuscitated death, myocardial infarction, stroke, unplanned hospitalization for heart failure, onset of end-stage renal disease or doubling of baseline serum creatinine concentration. Secondary endpoints include a composite CV endpoint and a composite renal endpoint. (6)

**RUBOXISTAURIN**

Protein kinase C (PKC) is overexpressed in diabetic nephropathy. It induces renal damage via activation of NADPH oxidase resulting in oxidative stress and upregulating TGF-β to induce extracellular matrix production. Ruboxistaurin is an oral PKC inhibitor. Animal studies have shown it to normalize glomerular hyperfiltration, reduce TGF-β and decrease proteinuria. Recently in a randomized study it was shown that patients of diabetic nephropathy who were treated with 32 mg daily ruboxistaurin compared to placebo had 24% greater reduction in albuminuria and a stable estimated glomerular filtration rate. (7)

**PENTOXIFYLLINE**

Metabolic and hemodynamic components are directly involved in pathogenesis of diabetic nephropathy. However, convincing data have shown that inflammation participates in the diabetic complications. In a prospective, randomized, double-blind, placebo-controlled study, the effect of pentoxifylline (PXF) 1200mg daily during 12 months, in 34 patients with incipient or established DN was evaluated. Evaluated parameters were inflammation, pro-inflammatory cytokines and urinary albumin excretion (UAE). PXF treatment had a renoprotective effect determined by a significant reduction in the urinary albumin excretion in both incipient and established (p<0.01) DN patient. This effect was attributed to a reduction in the C-reactive protein, interleukin-6, tumor necrosis factor-alpha and leptin serum levels (p<0.01). Thus, PXF treatment caused regression and prevented the progression of renal damage. (8)

**M-TOR INHIBITORS**

Studies have shown that m-TOR pathway is highly activated in progressive renal disorders including DN. Diabetic db/db mice which lack the leptin receptor signaling can be used as a model of ESRD associated with DN. It was demonstrated that p70S6-kinase was highly activated in mesangial cells in diabetic obese db/db mice. Furthermore, systemic administration of rapamycin, a specific and potent inhibitor of mTOR, markedly ameliorated pathological changes and renal dysfunction. Moreover, rapamycin treatment shows a significant reduction in fat deposits and attenuates hyperinsulinemia with few side effects. These results indicate that mTOR activation plays a pivotal role in the development of ESRD and that rapamycin could be an effective therapeutic agent for DN. (9) In another study in rats, Sirolimus (SRL) ameliorated renal inflammation, glomerular hypertrophy, and podocyte loss as indicated by morphometric and immunohistological analysis. SRL lowered expression and activity of glomerular TGF-β 1/2 and vascular endothelial growth factor, all of which are considered central cytokines in the pathogenesis of DN. (10)

**ADVANCED GLYCATION/OXIDATIVE STRESS INHIBITORS**

A novel ARB-derivative, R-147176 inhibits oxidative stress and advanced glycation, without binding to the angiotensin II type 1 receptor (AT1R) and has virtually no anti-hypertensive effect. The inhibition of advanced glycation end product formation, the AT1R affinity, and the pharmacokinetic characteristics of newly synthesized ARB-derivatives were assessed and R-147176 was eventually selected as it strongly inhibited advanced glycation but was 6700 times less effective than olmesartan in AT1R binding. Despite a minimal effect on blood pressure, it provided significant renoprotection in SHR/N worldly rats as well as in Zucker diabetic fatty rats. The renal benefits of ARB thus depend on the inhibition of AGEs and oxidative stress by their chemical structure. (11)

**PLASMINOGEN ACTIVATOR INHIBITOR-1 INHIBITORS**

The disruption of the plasminogen activator inhibitor-1 (PAI-1) gene protects mice against DN. (12) Two novel, orally active, small molecule substances, TM5001 and 5007, were identified. In vitro, they specifically inhibited PAI-1 activity and the formation of a PAI-1/tissue plasminogen activator (t-PA) complex, and enhanced fibrinolysis

Given to rats with Thy-1 nephritis, they reduced proteinuria and mesangial expansion, a benefit similar to that observed in the same model whose PAI-1 molecule had been mutated. Clinical benefits of PAI-1 inhibitors in DN remain to be demonstrated. If confirmed, these molecules might usefully expand our therapeutic armamentarium to prevent DN.

**SULODEXIDE**

It is an oral preparation of highly purified mixture of glycosaminoglycans (80% heparin sulphate and 20% dermatan sulphate). HPR-I is upregulated in hyperglycemic states and decreases proteoglycan content of glomerular basement.
membrane by degrading heparin sulphate proteoglycans. Sulodexide helps in restoring glomerular glycoproteins. It also suppresses overexpression of TGF-ß.

In DiNAS, a randomized, double blind and placebo-controlled trial involving 223 patients with diabetes and micro/macroalbuminuria, sulodexide 50, 100, and 200 mg daily for 4 months was compared to placebo. After 4 months of treatment, albuminuria reduced by 30%, 49% and 74% respectively compared to placebo. The effect persisted even 4 months after drug stoppage. There were no significant adverse effects seen with the drug.

Another trial, sulodexide microalbuminuria trial (SUN-Micro-Trial) in 1000 patients with diabetes and persisting microalbuminuria on maximal RAAS blockade, failed to show reduction in urine albumin. Further studies are needed on this molecule before its utility for DN can be commented. (13)

PYRIDOXAMINE

It is an active inhibitor of AGES. It acts by inhibiting glycated proteins (Amadori products) breakdown, decreasing toxic effects of reactive oxygen species and scavenging reactive carbonyl compounds. In a phase 2 trial PYR-206, safety and efficacy of pyridoxamine was studied. 128 patients of DN received either 50 mg of pyridoxamine or placebo. In another, PYR-205/207 trial, 84 patients of type 1 or 2 diabetes were included. Patients were randomized to 250 mg pyridoxamine or placebo. Results were merged in these trials and published. Though there was no change in urine albumin excretion, pyridoxamine group had 48% reduction in serum creatinine and also reduction in TGF-ß. (14)

One must be cautious in interpreting these small studies. However they provide an insight to future.

AVOSENTHAN

The endothelin system regulates a number of renal functions and can induce proteinuria by various mechanisms. Plasma and urinary endothelin-1 (ET-1) levels are elevated in patients with diabetes and correlate with reduced renal function, increased BP and albuminuria, and severity and duration of diabetes. Antifibrotic effects of ERAs in experimental disease that reduce proteinuria, renal fibrosis, and survival are mainly ETA receptor mediated. Macrophage infiltration in renal tissue and urinary TGF-and prostaglandin E2 metabolites can be reduced using an ETA-selective antagonist.

Avosentan (SPP301) is a new, once-daily, orally available ETA antagonist in clinical development for the treatment of DN.

In a randomized trial the effect of avosentan (SPP301) on urinary albumin excretion rate (UAER) in patients with DN was studied. 286 patients with DN (UAER 0.2 to 5.6 mg/min), and BP <180/110 mmHg were randomly assigned to 12 week of avosentan (5, 10, 25, and 50 mg) or placebo. In addition to standard ACEI/ARB therapy. Relative to baseline, all avosentan dosages decreased mean relative UAER (-16.3 to -29.9%) compared to placebo (35.5%). Median relative UAER decreased with all avosentan dosages (-28.7 to -44.8%) compared with placebo (12.1%). Creatinine clearance and BP were unchanged at 12 wk. The main adverse events were peripheral edema (12%), mainly with high (25 mg) dosages of avosentan; significant increases in liver enzymes did not occur. Twenty-one (7.3%) patients experienced adverse events that led to withdrawal from study medication. In summary, the endothelin-A antagonist avosentan given in addition to standard ACEI/ARB treatment decreases UAER in patients with DN. (15)

PIRFENIDONE

Pirfenidone (PFD; 5-methyl-1-phenyl-2-(1H)-pyridone) is a low molecular weight synthetic molecule that exerts dramatic antifibrotic properties in cell culture and various animal models of fibrosis. In a study, PFD was given to 17-wk-old db/db mice for 4 wk. PFD treatment significantly reduced mesangial matrix expansion and expression of renal matrix genes but did not affect albuminuria. Using liquid chromatography with subsequent electrospray ionization tandem mass spectrometry, 21 proteins unique to PFD-treated diabetic kidneys were identified. Analysis of gene ontology and protein–protein interactions of these proteins suggested that PFD may regulate RNA processing. Immunoblotting demonstrated that PFD promotes dosage-dependent dephosphorylation of eukaryotic initiation factor, potentially inhibiting translation of mRNA. In conclusion, PFD is renoprotective in diabetic kidney disease and may exert its antifibrotic effects, in part, via inhibiting RNA processing. PFD can block TGF-ß production at the transcriptional and protein levels, inhibit TGF-ß–induced Smad phosphorylation and TGF-ß–induced gene transcription, and inhibit TGF—induced matrix stimulation in mesangial cells. (16)

CONCLUSIONS

The present established therapy of DN include RAAS blockade. However in context of new pathophysiologic mechanisms of DN, targeted molecules are in development. These novel agents interfering with several newer culprits should provide additional, well needed benefits. Probably we owe the new decade to their success in management of DN.

REFERENCES

Diabetic Nephropathy: Novel Mechanisms and Therapies in 2010


