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NOVEL TARGETS FOR DIABETIC NEPHROPATHY

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ABSTRACT

Diabetic nephropathy (DN) is the leading cause of end-stage renal disease. In addition to the renin pathway,certain bioactive molecules are involved in its pathogenesis. Strategies to interrupt pathophysiological pathways are on the horizon. Protein kinase C over expression is blocked by ruboxistaurin. Pentoxifylline and m-TOR inhibitors are anti-inflammatory agents in the pathogenesis of diabetic nephropathy. Inhibitors of advanced glycation, oxidative stress have proved useful in animal models of diabetic nephropathy. Avosentan, an endothelin antagonist, decreases urinary albumin. Such targeted therapies have opened up avenues for researchers to develop agents that can halt and may even reverse the progression of diabetic nephropathy.

INTRODUCTION

Diabetes has become the most common single cause of end-stage renal disease (ESRD) in the country like India. Diabetes and obesity are the two things which are interlinked with each other; Diabetes is mainly characterized by hyperglycemia, polyuria, polydepsia and polyphasia. Diabetes is rapidly increasing prevalence, resulting in the profound socioeconomic effects in both developed and under developed countries [1-2] Diabetes is becoming an important chronic disease in India. In 2010, 45.5 million individuals had diabetes. By 2020, the number of prevalent diabetes cases will increase to 69.7 million.[3] Diabetes has number of complications such as Diabetic Gastropathy, Diabetic cardiomyopathy, Diabetic foot ulcer, Diabetic encephalopathy, Diabetic retinopathy, Diabetic neuropathy and Diabetic nephropathy etc.

DIABETIC NEPHROPATHY

Diabetic nephropathy is also known as Kimmelstiel Wilson syndrome and it was discovered in 1936 by Clifford Wilson and Paul Kimmelstiel. It has been reported that among 4837 patients with chronic renal failure seen over a period of 10 years, the prevalence of DN was 30.3% followed by chronic interstitial nephritis (23.0%) and chronic glomerulo nephritis (17.7%).[4] Diabetic Nephropathy (DN) is a microvascular complication affecting patients with both type 1 and type 2 diabetes. It has become a leading cause of End stage renal disease (ESRD). Diabetic nephropathy is mainly characterized by structural abnormalities such as:-

1) Hypertrophy of both glomerular and tubular elements. 2) Increase in the thickness of the Glomerular Basement Membrane (GBM) 3) Progressive accumulation of extracellular components.4) Increase in GFR, Intra glomerular B.P, Subsequent proteinuria 5) Eventual loss of renal function.Diabetic nephropathy is a spectrum of progressive renal lesions secondary to diabetes mellitus ranging from renal hyperfiltration to end stage kidney disease. A diabetic nephropathy consists of few stages from hyperfilteration to renal disease as stated below,

Hyperfilteration

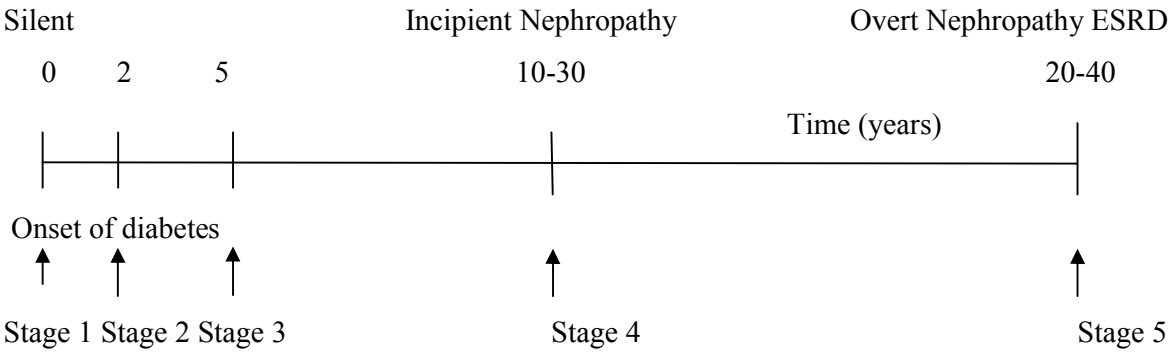


Figure 1. Natural history of diabetic nephropathy

STAGES OF DIABETIC NEPHROPATHY[5]

- Stage 1 (hyperfilteration) Glomerular hypertension and hyperfiltration, Normal albuminuria: urinary albumin excretion rate (AER) <20 μg/min, Raised GFR, normal serum creatinine.
- Stage 2 (Silent phase) Normoalbuminuria Structural changes on biopsy but no clinical manifestations. Basement membrane thickening and mesangial expansion.
- Stage 3 (incipient phase) Microalbuminuria: AER 20 – 200 μg/min, Normal serum creatinine, there may be increased blood pressure.
- Stage 4 (overt nephropathy) Overt "dipstick positive" proteinuria (macroalbuminuria) AER > 200 μg/min, Hypertension, Increase in serum creatinine with progression of Nephropathy.
- Stage 5 (End stage renal failure) GFR has fallen to <10 ml/min , renal replacement therapy (i.e., haemodialysis, peritoneal dialysis, kidney transplantation) is needed.

CELLULAR AND MOLECULAR MECHANISMS IMPORTANT IN THE DEVELOPMENT OF NEPHROPATHY

Diabetic nephropathy occurs as a result of an interaction between hemodynamic and metabolic factors. Hemodynamic factors that contribute to the development of diabetic nephropathy include increased systemic pressure, activation of vasoactive hormone pathways including the renin angiotensin system and endothelin. Glucose dependent pathways are also activated within the diabetic kidney and result in enhanced oxidative stress, renal polyol formation.[6]

HEMODYNAMIC PATHWAYS

The hemodynamic pathways activate intracellular second messengers such as protein kinase C (PKC), Mitogen-activated protein(MAP kinase), nuclear transcription factors such as NF-kB and various growth factors such as the prosclerotic cytokine, TGF-β and the permeability enhancing growth factor, vascular endothelial growth factor, VEGF. Any alteration in these cytokinines may leads to diabetic nephropathy.

METABOLIC PATHWAY

The glucose dependent pathway is also termed as metabolic pathway, The high glucose concentration in chronic diabetes mellitus induces oxidative stress by generating Reactive oxygen species (ROS) ROS through an activation of number of enzymatic and non-enzymatic sources in the body. The major sources of ROS in diabetes include polyol pathway, advanced glycation and uncoupling of NADPH oxidases. By understanding the pathomechanism of DN. It has helped in designing a rational approach for optimal therapy and prevention of DN.

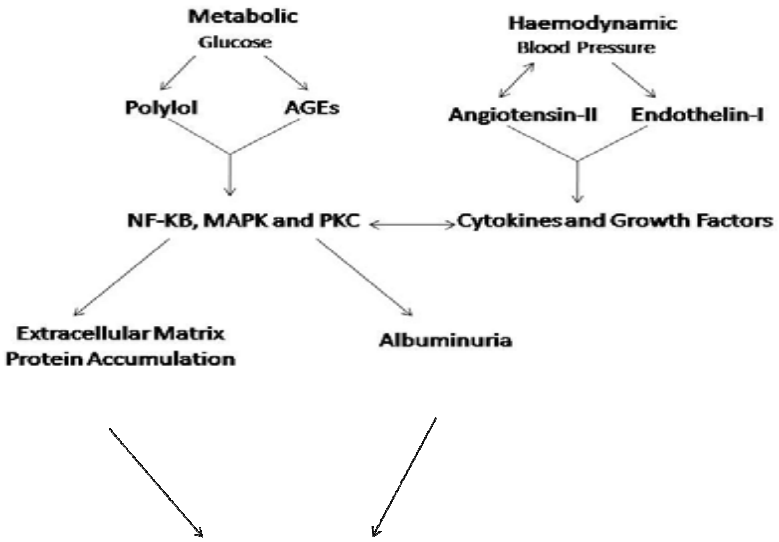


Figure 2. Interaction of hemodynamic and metabolic pathway, cytokines and intracellular signaling molecules mediating diabetic nephropathy

TARGET AND TREATMENT OPTIONS [7]

Based on its pathogenesis, the initial treatment of DN should be strict control of hyperglycemia, hypertension, dyslipidemia, proteinuria, and obesity and cessation of smoking. The beneficial role of thiazolidinediones (TZDs), 3-hydroxy-3-methylglutaryl coenzyme A (HMGCoA) reductase inhibitors, angiotensin-converting enzyme inhibitors (ACEIs), and angiotensin receptor blockers (ARBs) in DN is already established. Over the past few years our understanding of the complexity of the pathogenesis of DN has grown as new mechanisms, components, and regulatory steps have been identified. Also, the potential of targeted therapies has been explored, resulting in many novel drugs for the treatment and prevention of DN.

NOVEL TARGETS: CYTOKININS

- PROTEIN KINASE C (PKc) Protein kinase c is a family of serine-threonine kinase that are multifunctional isoenzymes acting as an intracellular signal transduction system for many hormones. There are 11 known isoenzymes that are classified in to following groups [8]

Group A: - Conventional (cPKCs) Ca2+dependent α, β1, β2 and γ

(Activated by phosphatidylserine (PS) and secondary messanger DAG)

Group B: - Novel (nPKCs) Ca2+-independent δ, ε, η and β etc

(Activated by phosphatidylserine (PS) and secondary messenger DAG)

Group C: - Atypical (aPKCs) Ca2+Independent ζ and λ [9-10]

In diabetes mellitus, PKC can be activated by several mechanisms, including increased DAG levels by de novo synthesis or inhibition of DAG kinase.PKC α, β1, β2 and γ were observed in cultered mesangial cells and glomeruli .In certain studies all of these isoforms have been activated by high glucose level or by diabetes mellitus in mesangial cells [11] Hyperglycemia either directly or indirectly leads to activation of the protein kinase c pathway which ultimately causes activation of both β and δ isoforms and causes release of fibrotic factors such as CTGF, TGF- β and vaso- active substances such as Angiotensin, endothelin – 1 and ultimately leads to albuminuria which is initital symptom of nephropathy decipeted as below.

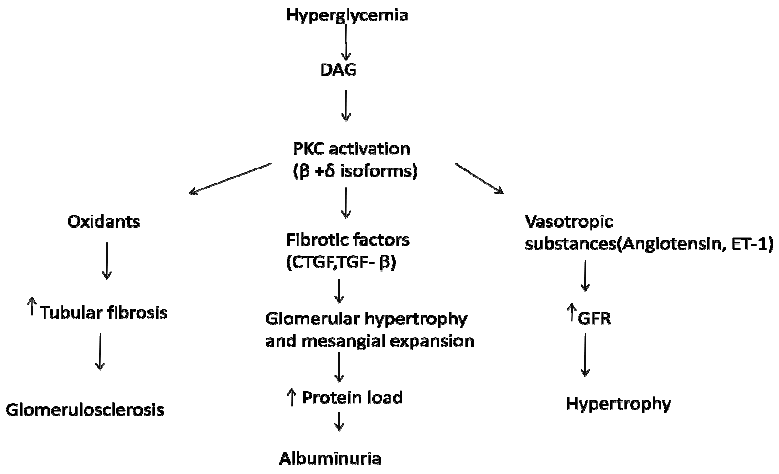


Figure:3 Protein Kinase C Pathway and Renal Disease

Among all PKC isoforms,PKC- β isoforms may be the most sensitive to change in the DAG level.In diabetic rats,chronic hyperglycemia predominantly activates PKC- β_2 isoform in the glomeruli,aortas,retina and heart.[12]

Ruboxistaurin (RBX) Ruboxistaurin, an orally active selective inhibitor of the β-isoform of PKC, reduces the actions of vascular endothelial growth factor (VEGF) and attenuates the progression of diabetic retinopathy. Animal studies have shown it to normalize glomerular hyperfiltration and reduce TGF-β levels and proteinuria. Excess matrix, hypertrophy, and apoptosis are features of DN, and ruboxistaurin has been shown to attenuate histological injury, functional decline, and expression of the profibrotic and proapoptotic growth factor TGF-β.

A specific PKC- β isoforms inhibitor,has been shown to ameliorats many functional and structural features of experimental diabetes.[13-14] RBX inhibits PKC β_1 and β_2 at nanomolecule concentrations and shows greater affinity for PKC isoforms as compared to other isoforms. RBX inhibited PKC activity,arachidonic acid release and PGE_2 production and normalized Na^+-k^+- ATPase activity.[15]

- TRANSFORMING GROWTH FACTOR-BETA (TGF-B)

The TGF- β superfamily encompasses three isoforms, TGF- β l, β 2, and β 3, each encoded by a distinct gene, and all expressed in the kidney. In normal rats TGF- β 1 mRNA was detected in glomeruli and all segments of renal tubules,predominantly in distal tubules.[16] TGF- β1 is known to be involved in renal fibrogenesis, which potentially leads to diabetic nephropathy.[17] TheTGF- β 1 membrane receptor complex, consists of two families of proteins with serine–threonine kinase activity, namely TGF- β RII and TGF- β RI.TGF- β 1 activates its receptors through a combination of two pathways. In the first pathway, TGF- β 1 combines with TGF- β RIII and then transfers to TGF- β RII. In the second pathway, TGF- β 1 combines directly with TGF- β RII . The two pathways ultimately lead to phosphorylation of TGF- β RII after its activation to a (TGF- β 1)–(TGF- β RII) complex. The complex recruits and phosphorylates TGF- β RI after which phosphorylated TGF- β RI continues to phosphorylate its downstream functional proteins, Smad2, Smad3 etc. Phosphorylation of Smad2 and Smad3 leads to their translocation into the nucleus and subsequently to tissue fibrosis. [18-19] TGF- β RII plays a key role during phosphorylation and activation of TGF- β RI receptors and small mothers against decapentaplegic (Smads)Smad transcriptional regulators. In the absence of TGF- β RII, TGF-b has no affinity for TGF- β RI. Therefore, inhibiting the phosphorylation of TGF- β 1 and its receptors is one way to prevent renal fibrosis.

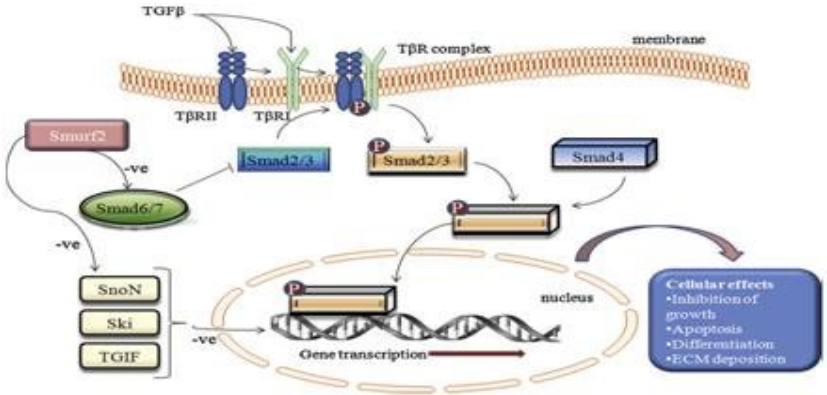


Figure 4. Schematic depicting TGF-b1 signalling.

TGF-b binds to its type II serine/threonine kinase receptor and instigates autophophorylation permitting the recruitment of the type I receptor. This interaction forms an activated heteromeric complex and facilitates phosphorylation of the receptor-regulated Smad2/3 small mothers against decapentaplegic (SMADs), promoting interaction with common Smad4. The active Smad2/3/4 complex translocates to the nucleus where it regulates the transcription of TGF-b1

target genes. TGF-b1 signalling is stringently regulated, and depends on both inhibitory Smads6/7 and transcriptional co-repressors that include SnoN, Ski, TGIF that help modify net cellular effects. Various approaches have been proposed either to block TGF- β receptor or TGF- β anti-body as summarized as below.

1) Nicousamide, a potent inhibitor of phosphorylation by TGF-b receptor II [20]

Renal fibrogenesis is related to the development of diabetic nephropathy.TGF- β receptor II (TGF- β RII) plays a vital role during renal fibrogenesis by phosphorylation and activation of type I receptors and downstream regulators. Nicousamide is a class of drug, which can inhibit renal fibrosis in animal models of diabetic nephropathy.

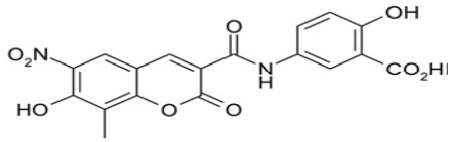


Figure 5. Chemical structure of nicousamide (C18H12N2O9 _H2O).

Studies in rat models of 5/6 nephrectomy and streptozotocin (STZ) induced diabetic nephropathy have shown that nicousamide can alleviate renal failure and reduce glomerulosclerosis and interstitial fibrosis by inhibiting renin and phosphorylation of TGF- β RII. In this respect it is better than benazepril and equipotent with losartan.

2) Decorin

Decorin belongs to the family of small leucine-rich proteoglycans (SLRPs). It is overexpressed in diabetic kidneys and has been suggested to act as a protective factor. [22] Decorin is capable of forming complexes with all three isoforms of TGF- β, leading to inhibition and/or sequestration of this cytokine within the extracellular matrix. [23]

3) Antibody TGF- β therapy

Diabetic nephropathy can be targeted by antibody TGF- β therapy. Early administration of pan-anti-TGF-β antibody to mice with streptozotocin-induced diabetes prevented glomerular enlargement and suppressed expression of genes encoding extracellular matrix components. [24] Antibody therapy almost completely prevented and sometimes even reversed the established lesions of diabetes and preserved renal function in db/db mice. [25]

PLASMINOGEN ACTIVATOR INHIBITOR-1

Plasminogen activator inhibitor-1 (PAI-1) prevents conversion of tissue plasminogen activator and urokinase plasminogen activator (uPA) to plasminogen.[26]. Plasmin, the active form of plasminogen, is a broad-spectrum protease that degrades fibrin clots and extracellular matrix (ECM) proteins. In diabetic nephropathy, accumulation of ECM proteins in the mesangium

leads to glomerulosclerosis, the hallmark of diabetic nephropathy. In normal human kidney, PAI-1 levels are undetectable, but in diabetes, PAI-1 expression is up-regulated in renal glomeruli and arteries.[27] Several experimental and clinical studies support the role for PAI-1 in the renal fibrogenic process that occurs in chronic glomerulonephritis, DN etc. Inhibition of PAI-I activity or of PAI-I synthesis by specific antibodies, peptidic antagonists, antisense oligonucleotides, or decoy oligonucleotides has been obtained in vitro, but needs to be evaluated in vivo regarding the prevention or treatment of renal fibrosis.[28,29] Disruption of the PAI-I gene protects mice against DN. Some novel, orally active, small molecule substances— TM-5001, TM-5007, TM-5275—were identified. In vitro, inhibited PAI-I activity and formation of a PAI-I–tissue plasminogen activator (t-PA) complex, and they enhanced fibrinolysis. [30, 31]

- **m-TOR INHIBITON**

A serine/threonine kinase, mTOR plays a pivotal role in mediating cell size, mass, proliferation, and survival. mTOR has also emerged as an important modulator of several forms of renal disease. Renal enlargementis due to the hypertrophy of existing glomerular cells.[32] A number of studies have shown that activation of mTOR plays a pivotal role in physiologic and pathologic forms of hypertrophy in the kidney and other organs, including the renal hypertrophy characteristic of DN. Hyperglycemia stimulates Mtor, mTOR is activated within the kidney in DN. Hyperglycemia activates PI3K and Akt and inhibits AMPK. The activation of Akt and inhibition of AMPK lead to activation of mTORC1. Activation of mTORC1 contributes to the renal changes characteristic of DN, including glomerular hypertrophy, glomerular basement membrane (GBM) thickening, and the accumulation of mesangial matrix.[33]

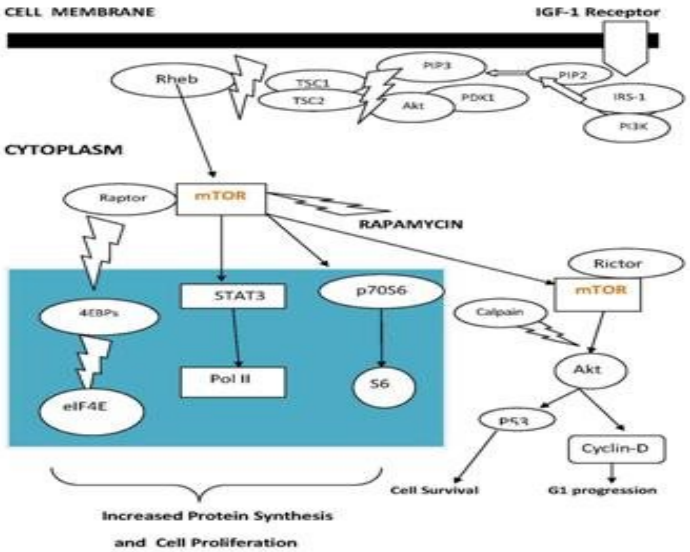


Figure 6. Hyperglycemia, mTOR activation and action of rapamycin.

Intracellular effects of Rapamycin through mammalian target of rapamycin pathway (mTOR). mTOR-Raptor complex is sensitive to Rapamycin however mTOR-Rictor complex is resistant to effects of Rapamycin. eukaryotic initiation factor 4EIGF-1; Insulin-like growth factor 1, PI3K; Phosphotidylinositol 3 kinase, PDK1; Phosphotidylinositol-dependent-kinase-1, Akt; protein kinase B, TSC1,2; Tuberosclerosis complex 1,2 Rheb; Rashomolog- enriched in brain act as Ras-related small GTPase Raptor; Rapamycin-sensitive adaptor protein, eIF4E; eukaryotic initiation factor 4E, 4EBP; eukaryotic initiation factor 4E binding protein, Pol II; polymerase II.

Sirolimus

Sirolimus (Rapamycin, RapamuneR) is a macrolide, product of the fermentation of an actinomycete, Streptomyces hygroscopicus**,** isolated (1975) from a soil sample in Rapa Nui (Easter Island)**,** having a structure similar to tacrolimus (TaC) and to macrolide antibiotics.[34] The potential therapeutic role of mTOR inhibition in patients with autosomal dominant polycystic kidney diseases is being evaluvated in clinical trials.[35]

VASOACTIVE SUBSTANCES

- **ENDOTHELIN – 1** Endothelin-1 (ET-1) is a potent vasoconstrictor peptide produced by vascular endothelium from big ET-1 via specific cleavage by endothelium converting enzyme (ECE). ET-1 produces its actions by acting on endothelin ETA and ETB receptors. [35, 36] ETB receptors on endothelial cells mediate vasodilation through the production of nitric oxide and prostacyclins.[37] ET-1 is involved in the pathogenesis of cardiovascular disorders such as hypertension and heart failure including diabetic nephropathy. It was shown that diabetes-induced elevated level of renal ET-1 may induce glomerular hyperperfusion and damage.[37] It has been documented that ET-1 activates a variety of signalling systems to induce contraction, hypertrophy in mesangial cells. ET-1 is also a potent proinflammatory and profibrotic mediator.

1)Darusentan (LU135252)

It has indeed been shown that prolonged application of the endothelin A (ETA) receptor antagonist darusentan (LU135252) was able to prevent the progression of diabetic nephropathy and to improve endothelium-dependent relaxation in mesenteric microvessels. [39, 40, 41]

2) Bosentan

It has been observed that bosentan also completely prevented the development of hypertension and renal vasoconstriction, and largely prevented the development of proteinuria and renal structural injury in wistar rats.[42]

3) Avosentan

Avosetan significantly decreased albuminuria in clinical study and found to be effective in the reduction of miroalbuminuria. Avosentan is a nonpeptidergic, once-daily, orally available endothelin-A (ETA) antagonist that is currently in clinical development for the treatment of DN. Endothelin antagonists have shown anti-inflammatory effects in experimental studies. [43, 44]

UROTENSIN

Human urotensin-II (U-II) is a cyclic peptide of 11 amino acids, with a molecular weight of ~1,388. U-II has been identified as an endogenous ligand for G-protein coupled receptor 14 (GPR14), which is now known as UT receptor [45]. Both U-II and UT receptor are expressed in different tissues, including the cardiovasculature, brain, kidney, atherosclerotic plaques in the coronary and carotid arteries, and abdominal aortic aneurysms.[46] U-II is the most potent vasoconstrictor identified to date, with a potency 1–2 order of magnitude greater than that of endothelin-1 (ET-1), and UII has been shown to play an important role in the pathogenesis Of pulmonary artery hypertension and remodeling. The main sources of circulating U-II are thought to be the heart, liver, kidney, vascular endothelium, and lymphocytes [47, 48].

Urotensin II (UII) is initially isolated from the caudal neurosecretory system of teleost fish. Plasma concentrations of UII are elevated in patients with hypertension , congestive heart failure, chronic renal failure and diabetes mellitus. UT is expressed in glomerular arterioles, thin ascending limbs, and inner medullary collecting ducts . High-affinity binding of human 125IUII has been reported in the human kidney. [49]

Palosuran

A selective UT receptor antagonist, palosuran (ACT-058362; Actelion Pharmaceuticals Ltd., Allschwil, Switzerland) was found to be an effective in diabetes. Preliminary data from a series of three clinical proof-of-concept studies of palosuran in diabetic nephropathy patients do not suggest a major efficacy, at the doses used, the duration of treatment applied, for the patients examined. [50,51]

VASOPRESSIN

Vasopressin plays an important role in the cardiovascular and renal diseases. It is previously known as anti-diuretic hormone. It is secreted from posterior pituitary lobe when osmolarity increases.[52] It's renal effects are mediated through the V_{1a}- receptor localized in the mesangial cells, efferent arterioles, vasa recta and medullary interstitial cells, which induce an increase in glomerular filtration rate and the V_2 receptors localized in collecting duct which prevents water and sodium loss.[53] By acting through these V_{1a}- receptors vasopressin causes vaso constriction,

proliferation and hypertrophy of mesangial cells leading to decrease in the filtration rate and ultra-filtration coefficient. Therefore various approaches have been made to block or antagonize V_{1a}- receptors as mentioned below.

1) OPC 21268

It is recently developed an orally non-peptidic AVP V1 antagonist. It causes a decrease in urinary albumin excretion due partly to decrease the intraglomerular pressure .This molecule is under clinical trials and results regarding their effects on patients is still awaited. [54]

2) YM 218

The V1a-receptor antagonist, YM218, protects against the early progression of renal injury caused by a reduction in nephron number. Where as its effectiveness seems limited in established renal damage caused by deteriorating function of previously healthy nephrons. [55, 56]

VASOPEPTIDASE INHIBITORS

Vasopeptidase inhibitors simultaneously inhibit the enzymes angiotensin-converting enzyme (ACE) and neutral endopeptidase (NEP).[57] Neutral endopeptidase (NEP) is cell membrane associated zinc metalloprotease, NEP is the major enzyme responsible for the degradation of the natriuretic peptides, and its inhibition leads to increases in the levels of these vasorelaxant, diuretic and natriuretic peptides.It is particularly abundant in membranes of brush border epithelial cells of intestine and kidney.[58] While ACE inhibition prevents the formation of the vasoconstrictor, anti-natriuretic and trophic hormone angiotensin II. Part of the pharmacological action of ACE inhibitors is based on inhibition of bradykinin degradation, leading to increased stimulation of the bradykinin B2 receptor. As bradykinin is degraded by both ACE and neutral endopeptidase, simultaneous inhibition of both enzymes by the novel vasopeptidase inhibitors is supposed to increase renal bradykinin concentrations and protect the diabetic kidney even more effectively than selective ACE inhibition alone. However, the relative role of bradykinin in the therapeutic action of vasopeptidase inhibitors in diabetic nephropathy has not been investigated so far. Recent data indicate that indeed, vasopeptidase inhibitors possibly related to their greater potency in increasing tissue bradykinin concentrations. [59, 60]

1) Omapatrilat

The vasopeptidase inhibitor omapatrilat has shown superior nephroprotection over selective ACE inhibition in nondiabetic nephropathy.[61] Omapatrilat is a vasopeptidase inhibitor that causes significant inhibition of tissue ACE and NEP, Both the degree and site of tissue enzyme inhibition by omapatrilat may be relevant to effects on various substrates, as well as to end-organ protection and side-effect profiles.[62]

2) AVE7688

AVE7688 prevents nephropathy in the Zucker diabetic fatty (ZDF) rat, a type II diabetes animal model, when treatment is started early and reduces proteinuria when diabetes and nephropathy are established, chronic vasopeptidase inhibition with AVE7688 reduces albuminuria and morphological kidney damage in rats with established diabetes and nephropathy.[63,64]

OXIDATIVE STRESS

High glucose concentration in chronic diabetes mellitus induces oxidative stress by generating ROS through an activation of number of enzymatic and non-enzymatic sources in the body. The major sources of ROS in diabetes include polyol pathway, uncoupling of NADPH oxidases. [65]

- **NADPH OXIDASE PATHWAY [NADPH oxidase inhibitors]**

In diabetes, NADPH oxidase is a major source of generation of ROS. NADPH oxidase is located in plasma membrane of various renal cell types, including mesangial and proximal tubular cells, vascular smooth muscle cells, endothelial cells and fibroblasts.

Due to activation of NADPH oxidase enzyme, it generates superoxides known as reactive free radicals.[66] The NADPH oxidase complex comprises several isoforms, now designated as the nox family, particularly nox4 isoform, a 578-amino acid protein and a major source of ROS in the renal milieu and thus NADPH oxidase dependent overproduction of ROS play a key role in promoting hyperglycemia-induced oxidative stress.[67] The NADPH oxidase increase oxidative stress and finally results in development of diabetic nephropathy in rats. Thus, NADPH oxidase may be one of potential target deserving further investigation in the development of drug in the treatment of diabetic nephropathy.[68]

EX. **1) Apocynin** [69]

Apocynin attenuated diabetes-associated increases in albuminuria. Finally, renal extracellular matrix accumulation of fibronectin and collagen IV was decreased by apocynin.

- **Polyol pathway [Aldose reductase inhibitors]**

In cell, unused glucose in the cytosol is diverted to the polyol pathway, which involves two enzymatic reactions: the first is the reduction of glucose to Sorbitol by the action of aldose reductase, and the second oxidation of sorbitol to fructose by the action of Sorbitol dehydrogenase. Reduction of glucose to sorbitol uses NADPH and oxidation of sorbitol increases NADPH with a resultant rapid change in the cytoplasmic redox state and enhanced production of ROS. Hence, polyol pathway is considered as a major source of ROS generation in the pathogenesis of diabetic nephropathy. A number of studies have shown a decrease in urinary albumin excretion in animals administered aldose reductase inhibitors.

EX. **1) Epalrestat** [70]

Epalrestat (Ono Pharmaceutical, Co. Ltd., Osaka, Japan) is one of the carboxylic acid derivatives which inhibit aldose reductase, an enzyme of the sorbitol (polyol) pathway. Epalrestat, prevents the progression of diabetic nephropathy in rats.

2) Zopolrestat [71]

Zopolrestat restored the hyporesponsiveness of diabetic rats to antigen provocation, in parallel with impairment of alloxan-induced mast cell depletion and hypercorticolism, indicating that polyol pathway activity seems to play an important role in diabetes induced nephropathy.

ADVANCED GLYCATION END PRODUCTS (AGE) INHIBITORS

Advanced glycation end products (AGEs) are diverse group of molecules and are well known heterogenous compounds formed non-enzymatically through an interaction of reducing sugar with free amino group of proteins, lipids and nucleic acids. Reducing sugars can react non-enzymatically with the amino groups of proteins to form reversible Schiff bases. These early glycation products undergo further complex reactions such as rearrangement, dehydration and condensation to become irreversibly cross-linked, heterogeneous fluorescent derivatives termed AGEs. The AGE9 receptor has been recently described. Its activation by the AGE has been shown to play an important part in the pathogenesis of DN and other nephropathies .

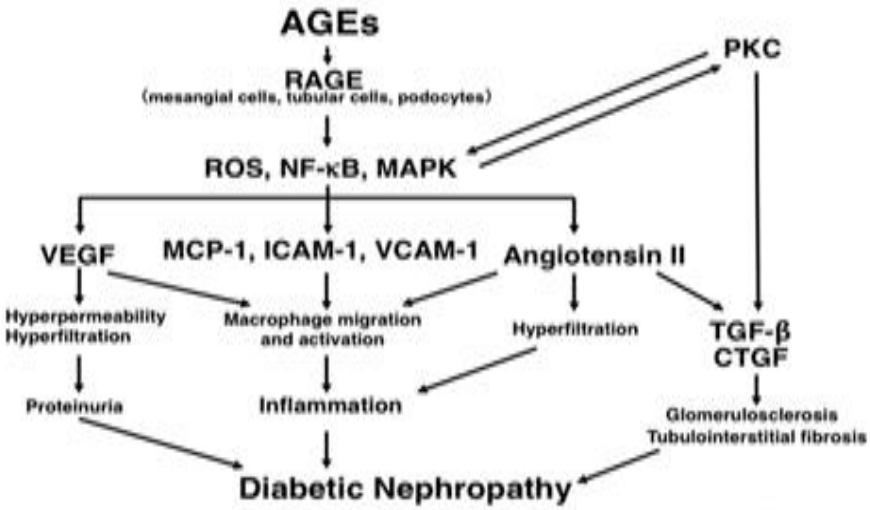


Figure 7.Pathophysiological role of the AGE-RAGE axis in diabetic nephropathy

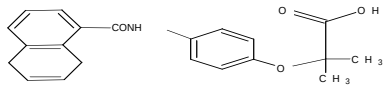
AGE's are mainly the component of interest of which receptor for AGE is mainly targeted for nephropathy as summarized below.

EX.

A: pyridoxamine [72] pyridoxamine (PM), and aminoguanidine protects against renal structural lesions, proteinuria and renal function loss in experimental diabetes.

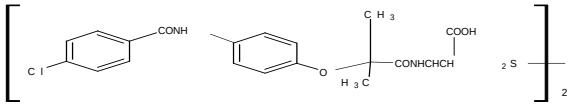
B: Novel AGE-Inhibitors

1) LR-9, 4-(2-naphtylcarboxamido) phenoxyisobutyric acid



LR-9

2) LR-20, L-bis-4[-(4-chlorobenzamidophenoxyisobutyryl) cystine



C: Novel AGE-Breakers

I. LR-20, L-bis-4[-(4-chlorobenzamidophenoxyisobutyryl)cystine
II. LR-23,4-(3,5-dichlorophenylureido)-phenoxyisobutyryl-l-amidocyclohexane-1-carboxylic acid
III. LR-99, 4-[(3,5-dichlorophenylureidophenoxyisobutyryl]-4-aminobenzoic acid)]
IV. LR-102, 1,4-benzene-bis [4-methyleneaminophenoxyisobutyric acid].
V. SMR-5, 5-aminosalicylic acid (5-ASA)
VI. SMR-12, dimethylbiguanide (metformin)

REFERENCES

1. David Savage, Understanding Diabetic Nephropathy – Is There a Genetic Susceptibility? Reader in Molecular Medicine, Centre for Public Health, Queen's University Belfast,[2] Professor of Renal Medicine, Regional Nephrology Unit, Belfast City Hospital 2009: 52-56
2. Ray JA, Curr Med Res Opin, 2005;21:1617–29
3. Epidemiology: Diabetes in India - a country faced with a mammoth diabetes epidemic . Published by Data monitor Published on 16 Mar 2011 Product code HC00132-001 :45-47
4. Vijay Viswanathan, Type 2 diabetic and diabetic Nephropathy in India- Magnitude of problem,Nephrol dial transplant 1999: 2805-2807
5. Scottish Intercollegiate Guidelines Network. SIGN 11: Management of diabetic renal disease – a National Clinical Guideline recommended for use in Scotland. March 1997.
6. Atul Arya, Harlokesh Narayan Yadav, pathogenesis of Diabetic Nephropathy, International Journal of Pharmacy and Pharmaceutical Sciences, 2010;2(4): 24-29.
7. Amitabh Dash, Treatment for Diabetic Nephropathy,Hong Kong J Nephrol 2011;13:1:19-26
8. Yuzuru ohshrio,Yvonne lee and George l king , Mechanism of diabetic nephropathy: role of protein kinase c activation, advance study in medicine 2005:5;1A:S10- S19

9. Idris I, Gray S,Donnelly R, Protein kinase c activation isozymes specific effects on metabolism and cardiovascular complications in diabetes.Diabetologia 2001;44:659-673.
10. Mellor H, Parker PJ. Extended protein kinase c super family.biochem j.1998;332:281-292.
11. Way KJ,Chou E,King GL. Identification of protein kinase c isoforms-specific biological actions using pharmacological approaches. Trends pharmacol Sci.2000;21:181-187.
12. Inoguchi T, Preferential elevation of protein kinase c isoforms beta 2 and diacylglycerol level in the aorta and heart of diabetic rats:differential reversibility to glycemic control by islets cells transplantation proc Natl Acad sci USA 1992;89:11050-11063.
13. Jirousek MR, (s)-13[(dimethylamino)methyl]-10,11,14,15-tetrahydro-4,9:16,2]-dimetheno-1H,13-Hdibenzo[e,k]pyrrolo[3,4h][1,4,13]oxadiazocyclohex decene-1,3[2H]-D ione (LY333531) inhibitors of protein kinase β.J Med chem..1996;39;2664-2671.
14. K.R.Tuttle, The Effect of Ruboxistaurin on Nephropathy in Type 2 Diabetes Diabetes Care, 2005;(11):28;11:2686-2690
15. Koya D,Jirousek MR,Lin YW,Characterisation of protein kinase c β isoforms on gene expression of TGF-B,extracellular matrix components and prostanoids in the glomeruli of diabetic rats.j clin invest.1997;100:115-126.
16. Elena Gagliardini, Ariela Benigni Role of anti-TGF-b antibodies in the treatment of renal injury Cytokine & Growth Factor Reviews 2006; 17 : 89–96.
17. Ando T, Okuda S, Tamaki K, Yoshitomi K, Fujishima M. Localization of transforming growth factor-beta and latent transforming growth factor-beta binding protein in rat kidney. Kidney Int 1995; 47:733–9.
18. Iwano M EMT and TGF- β in renal fibrosis front.Biosci (sci ed) 2010;2:229-238.
19. Wang W, Koka V,Lan HY,Transforming growth factor – β and Smad signaling in kidney diseases.Nephrology (caltron) 2005;10:48-56.
20. Hai-jing Zhang, a potent inhibitor of phosphorylation by TGF- β receptor II Acta Pharmaceutica Sinica B 2011;1(3):160–165.
21. Schaefer L, Iozzo RV. Biological functions of the small leucine-rich proteoglycans: from genetics to signal transduction. J Biol Chem 2008; 283: 21305-21309.
22. Williams KJ, Qiu G, Usui HK, et al. Decorin deficiency enhances progressive nephropathy in diabetic mice. Am J Pathol 2007; 171: 1441-1450.
23. Border WA, Noble NA, Yamamoto T, et al. Natural inhibitor of transforming growth factor-b protects against scarring in experimental kidney disease. Nature 1992; 360: 361-364.

24. Sharma K, Neutralization of TGF-b by anti- TGF-b antibody attenuates kidney hypertrophy and gene expression in STZ-induced diabetic mice. Diabetes 1996;45:522–30.
25. Ziyadeh FN, Long-term prevention of renal insufficiency, excess matrix gene expression, and glomerular mesangial matrix expansion by treatment with monoclonal antitransforming growth factor-b antibody in db/db diabetic mice. PNAS 2000; 97:8015–20.
26. Loskutoff D,Regulation of PAI-1 gene expression in vivo.Throm Haemostasis70:135,1993
27. Xu Y, Different expression of the plasminogen activation system in renal thrombotic microangiopathy and the normal human kidney. Kidney Int 50:2011–2019, 1996.
28. Chen Y-Q, Sp-1 sites mediate activation of the plasminogen activator inhibitor-1 promoter by glucose in vascular smooth muscle cells. J Biol Chem 273:8225–8231, 1998.
29. Rerolle JP, Hertig A, Nguyen G, et al. Plasminogen activator inhibitor type 1 is a potential target in renal fibrogenesis. Kidney Int 2000;58:1841–50.
30. Izuhara Y, Inhibition of plasminogen activator inhibitor-1: its mechanism and effectiveness on anticoagulation and anti-fibrosis. Arterioscler Thromb Vasc Biol 2008; 28:672–7.
31. Izuhara Y, A novel inhibitor of plasminogen activator inhibitor-1 provides antithrombotic benefits devoid of bleeding effect J Cerebral Blood Flow Metab 2010;30:904–12.
32. Wilfred, The Role of the Mammalian Target Of Rapamycin (mTOR) in Renal Disease Journal of the American Society of Nephrology 2009;20: 2493–2502
33. J. A. Sánchez-Plumed, first Tor inhibitor Nefrología 2006 ;2:21-32
34. Kultigin Turkmen The Protective Effect of mammalian Target of Rapamycin (mTOR) in Cisplatin Induced Nephropathy Iranian Journal of Medical Hypotheses and Ideas 2009,3:1-7
35. Xu, D., Emoto , N., Giaid , A., ECE-1: a membrane-bound metalloprotease that catalyzes the proteolytic activation of big endothelin-1. Cell.1994;78:473–485.
36. Haynes , W.G.,Webb, D.J.Venoconstriction to endothelin-1 in humans: role of calcium and potassium channels. Am J Physiol.1993;265:H1676- H1681.
37. Verhaar, Endothelin-A receptor antagonist-mediated vasodilatation is attenuated by inhibition of nitric oxide synthesis, Circulation.1998; 97:752–756.
38. Huang, S., Chen M, Li Y, Tian Q. Expression of ET-1 and Pr ET-1 mRNA in kidney of early diabetic rats. Hua Xi Yi Ke Da Xue Xue Bao.1999;30: 293–295.
39. Hocher B, Schwarz A, Reinbacher D, Effects of endothelin receptor antagonists on the progression of diabetic nephropathy. Nephron. 2001; 87:161–9.
40. Dhein S, Hochreuther S, Aus Dem Spring C, et al. Long-term effects of the endothelin(A) receptor antagonist LU 135252 and the angiotensinconverting enzyme inhibitor trandolapril

on diabetic angiopathy and nephropathy in a chronic type I diabetes mellitus rat model. J Pharmacol Exp Ther. 2000;293:351–9.

41. Björn Lemmer, Effects of Endothelin A Receptor Antagonist Darusentan on Blood Pressure and Vascular Contractility in Type 2 Diabetic J Cardiovasc Pharmacol 2003; 41:890-896
42. Clozel M, Pharmacological characterization of bosentan, a new potent orally active nonpeptide endothelin receptor antagonist. J Pharmacol Exp Ther. 1994;270:228–235.
43. Pantelis, Antagonism for diabetic nephropathy, nature reviews nephrology 2010;6:447-449.
44. Wenzel R. R. Avosentan reduces albumin excretion in diabetics with macroalbuminuria. J. Am. Soc. Nephrol. 20, 655–664 (2009).
45. Takuya Watanabe, Human Urotensin-II Potentiates Mildly Oxidized Low-Density Lipoprotein on Hydrogen Peroxide Hypertens Res 2006; 29:821-831.
46. Douglas SA, Human urotensin-II, most potent mammalian vasoconstrictor identified to date, as a therapeutic target for cardiovascular disease. Trends Cardiovasc Med 2000;10:229–237.
47. Ames RS, Sarau HM, Cambers JK, et al: Human urotensin II is a potent vasoconstrictor and agonist for the orphan receptor GPR14. Nature 1999; 401: 282–286.
48. Watanabe T, Pakala R, Katagiri T, Benedict CR: Synergistic effect of urotensin II with serotonin on vascular smooth muscle cell proliferation. J Hypertens 2001; 19: 2191–2196
49. Ames RS, Human urotensin-II is a potent vasoconstrictor and agonist for the orphan receptor GPR14. Nature 1999;401:282–6.
50. Clozel M, Hess P, (2004) Pharmacology of the urotensin-II receptor antagonist palosuran (ACT-058362; 1-[2-(4-benzyl-4-hydroxy-piperidin-1- yl)-ethyl]-3-(2-methyl-quinolin-4-yl)-urea sulfate salt): first demonstration of a pathophysiological role of the urotensin system. J Pharmacol Exp Ther 311**:**204– 212.
51. Natish M,Suzuki H,Murukami M et.al effect of oral AVP antagonists OPC-21268 and OPC-31260 on CHF conscious dogs .Am J Physiol 1994;267:H2245-H2254.
52. Bankir L Anti-diuretic activity of Vasopressin: quantitative aspects and interaction between V_{1a} and V_2 receptor mediated effects cardiovasc res 2001;51:372-390
53. K Yamada,H Nakano,M Nishimura and Sho Yoshida The effect of AVP V_1 receptor antagonists on urinary albumin excretion and renal haemodynamics in NIDDM Nephropathy: role of AVP V_1 receptor J Diab Comp 1995;9:329-329
54. Willemijn AKM Windt, Early, but not late therapy with a vasopressin V1a-antagonist ameliorates the development of renal damage after 5/6 nephrectomy Journal of Renin-Angiotensin-Aldosterone System 2006;7:217–24

55. Atsuo Tahara, Masayuki Shibasaki Effects of YM218, a nonpeptide vasopressin V1A receptor-selective antagonist on vasopressin-induced growth responses in human mesangial cells European Journal of Pharmacology 2006;538: 32–38
56. Corti, R., Vasopeptidase inhibitors – A new therapeutic concept in cardiovascular disease? Circulation 2001; 104, 1856–1862
57. Gafford J T,R A Skidgel,E G Erdos and L B Hersh human kidney enkephalinase a neutral metaloendopeptidase that cleaves the active peptides Biochemistry 1983;22:3265
58. Linz, W. Specific B2-bradykinin receptor antagonist HOE 140 abolishes the antihypertrophic effect of ramipril. Br. J. Pharmacol., 1992.;105: 771–772
59. Wiemer, G., Contribution of kinins to the cardiovascular actions of angiotensin-converting enzyme inhibitors. Pharmacol. Rev., 1995; 47: 25–49.
60. Taal, M.W., Vasopeptidase inhibition affords greater renoprotection than angiotensin converting enzyme inhibition alone. J. Am. Soc. Nephrol., 2001; 12, 2051–2059
61. Eiji Kubota and Louise M. BURRELL Differential tissue and enzyme inhibitory effects of the vasopeptidase inhibitor omapatrilat in the rat Clinical Science (2003) 105, 339–345
62. Schafer S., Vasopeptidase inhibition prevents nephropathy in Zucker diabetic fatty rats. Cardiovasc. Res., 2003;.60:447–454.
63. Schafer S., Vasopeptidase inhibitor AVE7688 ameliorates Type 2 diabetic nephropathy. Diabetologia,2004; 47 :98–103
64. Pawan Krishan Diabetic nephropathy: Aggressive involvement of oxidative stress J Pharm Educ Res 2011; 2:35-41
65. Griendling KK, Angiotensin II stimulates NADH and NADPH oxidase activity in cultured vascular smooth muscle cells. Circ. Res. 1994; 74: 1141–1148
66. Taura M, A region N-terminal to the tandem SH3 domain of p47phox plays a crucial role in activation of the phagocyte NADPH oxidase. Biochem J. 2009; 419:329–338
67. Block K, Gorin Y, Abboud HE. Subcellular localization of Nox4 and regulation in diabetes. Proc. Natl Acad. Sci. USA. 2009; 106: 14385–14390.
68. Vicki Thallas-Bonke, Inhibition of NADPH Oxidase Prevents Advanced Glycation End Product–Mediated Damage in Diabetic Nephropathy Through a Protein Kinase C–Dependent Pathway Diabetes, 2008;57:460-469
69. S. Isogai, T. Inokuchi, K. Ohe Effect of an aldose reductase inhibitor on glomerular basement membrane anionic sites in streptozotocin-induced diabetic rats Diabetes Research and Clinical Practice 1995;30: 111- 116

70. Vinicius F. Carvalho, Marco A. Martins a, Zuleica Bruno Fortes , Patrícia M.R. e Silva Aldose reductase inhibitor zopolrestat restores allergic hyporesponsiveness in alloxan-diabetic rats European Journal of Pharmacology 2006;549: 173– 178.
71. Samuel Rahbar and James L. Figarola Novel inhibitors of advanced glycation end products Archives of Biochemistry and Biophysics 2003; 419: 63–79.