COMPARISON BETWEEN ANTIPROTEINURIC EFFECTS OF CILNIDIPINE AND AMLODIPINE AS ADD ON THERAPY IN HYPERTENSIVE PATIENTS WITH CHRONIC RENAL DISEASES

DISSERTATION SUBMITTED TO

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IN

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CERTIFICATE

This is to certify that the dissertation entitled **"COMPARISON BETWEEN ANTIPROTEINURIC EFFECTS OF CILNIDIPINE AND AMLODIPINE AS ADD ON THERAPY IN HYPERTENSIVE PATIENTS WITH CHRONIC RENAL DISEASE"** presented herein by **Dr. Y. NISHA MAHESWARI** is an original work done by her in the Department of Pharmacology, Tirunelveli Medical College, Tirunelveli for the award of the Degree of Doctor of Medicine in Pharmacology during the academic period of 2012 – 2015.

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DECLARATION

I, **Dr. Y. NISHA MAHESWARI** declare that, I carried out this work on **"COMPARISON BETWEEN ANTIPROTEINURIC EFFECTS OF CILNIDIPINE AND AMLODIPINE AS ADD ON THERAPY IN HYPERTENSIVE PATIENTS WITH CHRONIC RENAL DISEASES"** at the Department of Pharmacology, Tirunelveli Medical College and I also declare that this bonafide work or a part of this work was not submitted by me or any others for any award, degree, or diploma to any other University, Board, either in India or abroad.

This is submitted to the Tamilnadu Dr.M.G.R Medical University, Chennai in partial fulfillment of the rules and regulations for the M.D Degree examination in Pharmacology.

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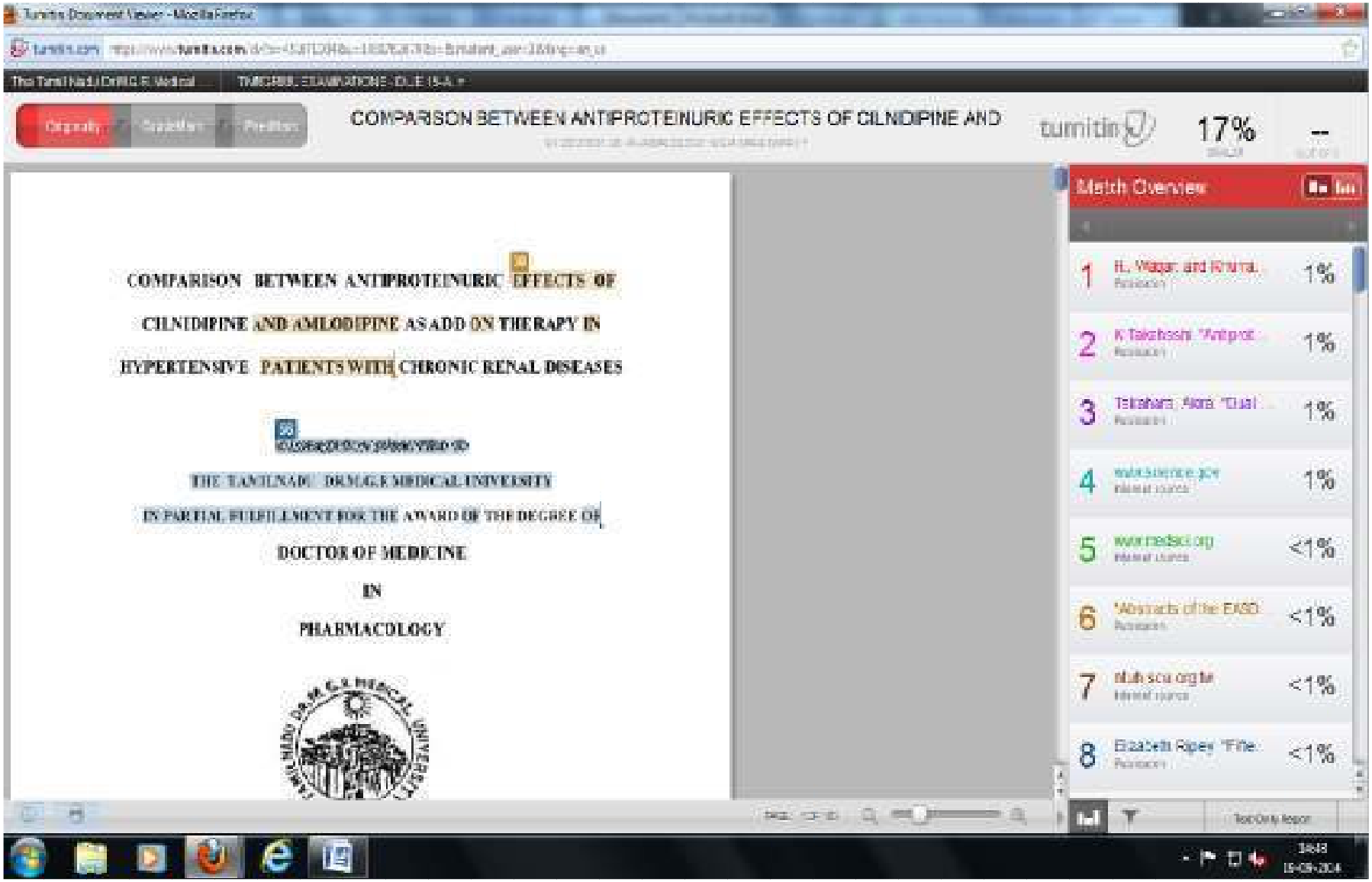
CONTENTS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ABSTRACT

COMPARISON BETWEEN ANTIPROTEINURIC EFFECTS OF CILNIDIPINE AND AMLODIPINE AS ADD ON THERAPY IN HYPERTENSIVE PATIENTS WITH CHRONIC RENAL DISEASES

AIM: To compare the efficacy and safety of cilnidipine and amlodipine as add on therapy in chronic kidney disease patients who are on losartan (Angiotensin receptor blocker) for > 2 months.

METHODS: In this prospective, single centered, open labeled, randomized study, the antiproteinuric effects of cilnidipine (L/N type calcium channel blockrer) and amlodipine (L type calcium channel blocker) were examined in diabetic chronic renal disease patients with hypertension (BP ≥ 130/80 mmHg) who are already under treatment with T. Losartan 50 mg OD. Antiproteinuric effects were assessed by reduction in spot urine protein creatinine ratio from baseline.

RESULTS: Patients received cilnidipine (n=46) or amlodpine (n=50) for 6 months. Cilnidipine and amlodipine reduced systolic and diastolic blood pressure equally. The spot urine protein creatinine ratio values for cilnidipine and amlodipine were 1.94±1.22 g/g and 1.38±0.98 g/g respectively before treatment and 1.09±0.72 g/g and 1.40±0.65 g/g respectively after treatment. The mean serum creatinine concentration gradually increased in both the groups

and attained statistical significance at the end of 6 months. Estimated GFR was maintained by both the drugs throughout the study period. Distribution of CKD stages were also similar between the two groups before and after treatment. None of the produced reflex tachycardia.

CONCLUSION: In conclusion, cilnidipine has antihypertensive effect equivalent to amlodipine but addition of cilnidipine rather than amlodipine to losartan decreased urine protein excretion in diabetic chronic kidney disease patients. Therefore combination therapy with cilnidipine and RAS inhibitor may be more beneficial and renoprotective in patients with diabetic chronic kidney disease.

KEY WORDS: L/N type calcium channel blocker, L type calcium channel blocker, Angiotensin receptor blocker, Diabetic chronic kidney disease/chronic renal disease, Hypertension, Urine protein creatinine ratio, antiproteinuric effects.

INTRODUCTION:

Chronic kidney disease (CKD) comprises of a spectrum of different pathophysiological processes associated with abnormal kidney function and progressive decline in glomerular filtration rate. Diabetic nephropathy is the most common cause of chronic renal failure worldwide. It is mainly due to epidemic increase in obesity, metabolic syndrome and type II diabetes mellitus. Hypertension is the major consequence of chronic renal disease which develops early during the course of the disease[1]. Uncontrolled hypertension and proteinuria are the most crucial risk factors for rapid progression of kidney disease and development of extra renal complications such as cardiovascular disease and stroke[2]. Thus strict control of blood pressure and suppression of proteinuria are the essential goals of antihypertensive therapy in patients with chronic renal disease. The National kidney foundation clinical practice guidelines recommend a blood pressure goal of < 130mmHg systolic and < 80 mmHg diastolic for all patients with chronic renal disease[3].

Renin angiotensin inhibitors such as ACE (Angiotensin converting enzyme) inhibitors and ARB (Angiotensin receptor blockers) are the widely recognized renoprotective agents. These agents effectively reduce proteinuria than any other antihypertensive agents[4]. According to

Japanese society of hypertension guidelines they are recommended as first choice treatment for hypertensive patients with CKD[5]. But sometimes it is difficult to achieve satisfactory decrease in proteinuria and blood pressure with these agents alone[6]. Combination therapy with two or more antihypertensive agents are often required to reduce blood pressure to target levels in these patients[7].

Dihydropyridine calcium channel blockers are one of the main candidate for combination with RAS (Renin Angiotensin System) inhibitor because they reduce BP even in patients who are unresponsive to other antihypertensive agents[8]. But the effect of these drugs on proteinuria is inconsistent. Traditional CCB (Calcium channel blocker) like amlodipine act by blocking L type calcium channel leading to dilatation of afferent arteriole with no effect on efferent arteriole. Ultimately renal blood flow and glomerular pressure increase accelerating proteinuria[9]. Recently developed CCB, cilnidipine is a dual blocker of L type and N type calcium channel and thereby dilates both afferent and efferent arteriole. Hence renal blood flow increases without increase in glomerular pressure thereby reducing proteinuria[6].

Renoprotective effects of L type blockers are considered less than the dual L/N type blockers. There is still lack of clinical trials

comparing the renoprotective effects of various calcium channel blockers. Thus the present study was designed to compare the antiproteinuric effects of cilnidipine and amlodipine as add on therapy to losartan in hypertensive chronic kidney disease patients.

DEFINITION:

Chronic kidney disease/ Chronic renal disease is defined as structural or functional abnormalities of the kidney for ≥ 3 months manifesting either as

1. Kidney damage with or without decreased GFR as defined by
    a) Pathological abnormalities
    b) Markers of kidney damage including abnormalities in composition of blood and urine or abnormalities in imaging tests

or

2. GFR < 60 ml/min/1.73m^2 with or without kidney damage[10].

A cutoff of 60 ml/min/1.73m^2 is selected because

a) It represents a reduction to approximately half of normal renal function.

b) It avoids classification of older individuals who may have mild reductions in their glomerular filtration rate[11].

Chronic renal failure represents a process of continuing significant irreversible reduction in nephron number and corresponds to CKD stages 3–5.

End stage renal disease corresponds to stage of CKD that results in uremic syndrome due to accumulation of toxins, fluid and electrolytes which are normally excreted by kidney

RECOMMENDED EQUATIONS FOR ESTIMATION OF GFR:

1. Equation from the modification of diet in renal disease study:

Estimated GFR = $1.86 \times (P_{cr})^{-1.154} \times (age)^{-0.203}$

Multiply by 0.742 for women

2. Cockcroft – Gault equation:

$$\text{Estimated GFR} = \frac{(140 - \text{age}) \times \text{body weight (Kg)}}{72 \times P_{cr}\,(\text{mg/dl})}$$

Multiply by 0.85 for women[1].

INCIDENCE AND PREVALANCE:

The estimated prevalence of CKD is 8 -16 % worldwide[12]. In United states , 6% of adult population are in stages 1 and 2 and 4.5% are in stages 3 and 4[1]. Age adjusted incidence rate of ESRD in India is 229 per million population and greater than 1,00,000 new patients enter renal replacement therapy every year[13]. Prevalence of chronic renal disease in India was estimated to be 0.78%[14]. But SEEK (Screening and Early Evaluation of Kidney disease), a community based voluntary screening program in India has reported very high prevalence

of about 17.2%. Prevalence of stage 1,2 ,3,4,5 CKD was found to be 7%, 4.3%,4.3%, 0.8% and 0.8% respectively. The most frequent cause of CKD is diabetic nephropathy often secondary to type 2 diabetes mellitus which contributes to 30% - 40% of these patients[15]. From Indian CKD registry it was found that 70% were males , 73.6% were in stage 4 – 5 and only 20 % of patients with ESRD were in some forms of renal replacement therapy[16].

CLASSIFICATION:

Classification of CKD is based on GFR as proposed by the Kidney Disease Outcomes Quality Initiative (KDOQI) Guidelines[17].

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STAGE	DESCRIPTION	GFR (ml/min/1.73m^2)
O	Normal or increased GFR with risk factors for CKD	≥ 90
1	Normal or increased GFR with demonstrated kidney damage reflected by microalbuminuria, proteinuria, hematuria or histologic findings	≥ 90
2	Mild reduction in GFR	89 – 60

3	Moderate reduction in GFR	59 – 30
4	Severe reduction in GFR	29 – 15
5	Renal replacement therapy required in the form of dialysis or transplantation to sustain life	< 15

NATURAL HISTORY OF CHRONIC RENAL DISEASE:

Rate of progression of kidney disease varies according to individual patients and underlying pathology. Patients with stage 3 – 5 CKD eventually progress to end stage renal disease. Rate of decline in GFR is found to be rapid in patients with diabetic nephropathy averaging about -10ml/min/year. Effective control of blood pressure slows the rate of progression to – 5 ml/min/year. Patients in whom both glycemia and hypertension are optimally controlled further improvement can be expected (-1 to -2 ml/min/year)[18]. In CKD GFR loss averages about 4 to 10ml/min/year. For every 1g reduction in proteinuria GFR decline is slowed by 1- 2ml/min/year[19]. Thus effective control of proteinuria is mandatory.

RISK FACTORS:

A) SUSCEPTIBILITY FACTORS:

Factors that increase susceptibility to kidney damage after exposure to initiation factor.

Genetic predisposition

Alterations or polymorphism of genes coding for putative mediators including RAS, NOS, kallikrein, cytokines (IL – 1 & TNF – α) , growth factors (PDGF, TGF-β), plasminogen activator inhibitor – 1 and complement factors have been associated with increased risk of CKD.

Maternal – fetal factors:

Maternal undernutrition and the ensuing fetal malnutrition may contribute to the development of hypertension, metabolic syndrome, diabetes mellitus and CKD in adult life. Low birth weight causes reduction in nephron numbers (oligonephronia) thereby the ability of nephrons to handle increased solute and salt load is reduced. Oligomeganephronia characterized by reduced nephron number together with glomerular hypertrophy has been implicated in the pathogenesis of CKD[18].

Socio - economic factors:

Low socio economic status has been associated with the risk of CKD. This is due to linkage of poverty with diabetes and hypertension. Thus lower income and social deprivation leads to development of macroalbuminuria, progressive renal function loss and ESRD. They also have less access to renal replacement therapy[20].

B) INITIATION AND PROGRESSION FACTORS:

1. NON MODIFIABLE:

Age and Gender:

Elderly patients are at risk of faster rate of decline in GFR. But in patients with type 1 diabetic nephropathy , young age at diagnosis is associated with rapid progression of the disease. Males are more prone for CKD than females.

Race:

In United States, incidence and prevalence of CKD is higher in the African – Americans and in the Hispanic Americans than the Caucasians. Similarly disease progression is also faster in them. In United Kingdom, the Indo Asians show a faster rate of GFR decline than Caucasians. This racial predisposition has been attributed to increased susceptibility to other diseases like diabetes mellitus and hypertension.

Genetics:

Insertion/Deletion polymorphism in ACE gene has been linked to CKD susceptibility and progression as well as responses to antihypertensive drugs.

Loss of renal mass:

The threshold for natural progression of the disease appears to be crossed when reduction in nephron function exceeds 50%. Threshold is also lowered by various co – morbid conditions like hypertension, obesity, diabetes and dyslipidemia. Diffuse glomerular damage carries higher risk of rapid progression than segmental damage.

2. MODIFIABLE:

Hypertension:

Systemic hypertension can be directly transmitted to glomerular capillaries . This leads to development of glomerular hypertension which can initiate glomerulosclerosis and CKD.

Proteinuria/Albuminuria:

Threshold for natural progression of the disease is crossed when proteinuria exceeds 500mg/day. The risk of rapid decline in GFR is high with non selective proteinuria than with highly selective proteinuria. Also urinary albumin creatinine ratio is directly proportional

to the presence and severity of CAD and CVD. Even low grade albuminuria is associated with this risk[18].

Metabolic syndrome

It consists of constellation of abnormalities like abdominal obesity, dyslipidemia, hypertension, insulin resistance, hyperfiltration, prothrombotic and proinflammatory states. The prevalence of CKD is high in patients with 2 or more components than patients with zero or 1 component. Thus there exist a linear relationship between number of components of MS and presence of CKD[21].

Metabolic factors:

a) Glycemia :

The risk of CKD as well as its progression can be minimized by excellent glycemic control. The recommended values are pre prandial blood sugar to be maintained between 90 – 130 mg/dl and HbA1c less than 7 %[1].

b) Dyslipidemia and Obesity:

Both contributes to worsening of CKD. Weight reduction lowers obesity related renal hemodynamic changes and CKD associated proteinuria[18]. Also patients with high triglyceride and low HDL levels are prone for rapid decline in GFR[21].

c) Hyperuricemia:

Hyperuricemia causes hypertension and renal injury by crystal independent pathways possibly by activating RAS[18]. Hyperuricemia also decreases nitric oxide production provokating endothelial dysfunction, promotes fibrosis and release of proinflammatory cytokines[22].

Miscellaneous factors:

a) Smoking and alcohol:

Cigarette smoking and alcohol consumption (exceeding 2 drinks per day) affect renal hemodynamics leading to ESRD[18].

b) Analgesics:

Analgesic nephropathy is a slowly progressing renal disease characterized by chronic nephritis and renal papillary necrosis[23]. Patient presents with decreased concentrating capacity of renal tubule and sterile pyuria. Risk factors are chronic use of high doses of combination of NSAIDs and frequent urinary tract infections. NSAIDs cause loss of prostaglandin induced inhibition of both reabsorption of chloride and action of ADH (leads to sodium and water retention)[24]. It also suppress PGE_2 mediated compensatory vasodilation in response to norepinephrine and angiotensin II [23].

COMMON ETIOLOGIES OF CHRONIC KIDNEY DISEASE:

1. Primary glomerular diseases:

It includes idiopathic cresentic glomerulonephritis, primary focal segmental glomerulosclerosis and primary mesangiocapillary glomerulonephritis.

2. **Tubulointerstitial disease:**

- Chronic heavy metal poisoning like lead, cadmium and mercury
- Chronic hypercalcemia as with vitamin D intoxication and primary hyperparathyroidism
- Chronic potassium depletion from prolonged use diuretics without potassium supplementation in patients with ascites or chronic heart failure.

3. Renal vascular disease:

a. <u>Main renal artery disease:</u>

Renal artery stenosis which may be due to atherosclerosis (common in elderly males) or fibromuscular dysplasia (common in middle aged females).

b. <u>Renal vein disease:</u>

Bilateral renal vein thrombosis seen in patients with nephrotic syndrome.

C. Small renal vessel disease:

Nephrosclerosis secondary to long standing systemic hypertension, polyarteritis nodosa or malignant hypertension.

4. Chronic UTI:

Chronic pyelonephritis which may be due to tuberculosis, E.coli.

5. Chronic urinary tract obstruction:

a. Upper urinary tract obstruction:

Bilateral ureteric or renal stones, ureteric stricture or neoplasm.

b. Lower urinary tract obstruction:

Bladder stones, bladder tumour, senile prostatic enlargement, stricture urethra.

6. Collagen diseases:

- Systemic lupus erythematoses
- Systemic sclerosis
- Polyarteritis nodosa
- Rheumatoid arthritis

7. Metabolic diseases:

- Diabetic nephropathy
- Gout leads to CKD either directly or secondary to use of NSAIDs

- Renal amyloidosis as a complication of familial mediterranian fever or chronic suppuration like osteomyelitis[25].

MECHANISMS OF PROGRESSION IN CKD:

Three main mechanisms are involved in disease progression.

1. Glomerulosclerosis
2. Tubulointestitial fibrosis
3. Vascular sclerosis

GLOMERULOSCLEROSIS:

It is initiated by adaptive changes that occur in normal glomeruli of diseased kidney. In response to an insult there is a reduction in renal mass. Compensatory hypertrophy of remaining glomeruli occurs in order to maintain renal function. Glomerular hypertrophy is associated with hemodynamic changes like increase in glomerular blood flow, hyperfiltration and transcapillary pressure [26]. Hyperfiltration is predominantly mediated by preglomerular vasodilation leading to enhanced transmission of systemic BP to glomerular capillaries. The resulting impairment of autoregulation contribute to glomerular hypertension[27]. All these changes leads to the following events.

- Endothelial injury characterized by loss of anticoagulant and anti inflammatory properties and acquisition of procoagulant and

proinflammatory poperties. This leads to attraction and activation of platelets and microthrombus formation.

- Initiation of glomerular microinflammation and formation of glomerular tufts by inflammatory cells (particularly monocytes).
- Infiltrating monocytes stimulates the proliferation of mesangial cells either by direct cell to cell or through release of mitogens leading to mesangial hypercellularity.
- Under the influence of fibrogenic growth factors like TGF – β activated mesangial cells gets converted to myofibroblast expressing α - SMA (Smooth muscle actin) and synthesize ECM (extracellular matrix components) leading to ECM deposition[18].
- Epithelial injury - Podocytes do not have the ability to replicate in response to an insult. This leads to decrease in podocyte number. The remaining podocytes stretch along the GBM (Glomerular basement membrane) to maintain the filtration barrier exposing areas of denuded GBM that would interact with parietal epithelial cells forming capsular adhesions. This phenomenon contributes to the development of proteinuria and segmental glomerulosclerosis.

TUBULOINTERSTITIAL FIBROSIS:

It is the major contributor for progression of CKD. Thus the decline in renal function has a better correlation with extent of tubulointerstitial damage than with the severity of glomerular injury. Proteinuria cause direct injury and activation of tubular cells. The activated tubular cells in turn express adhesion molecules and elaborate proinflammatory cytokines, chemokines and growth factors that contributes to interstitial fibrosis[26].

VASCULAR SCLEROSIS:

Vascular sclerosis is the key feature of renal scarring. Renal scarring is associated with

- Loss of peritubular capillaries that directly correlates with fall in renal expression of proangiogenic VEGF (Vascular endothelial growth factor).
- Overexpression of thrombospondin which perpetuate microvascular deletion and ischemia.
- Ischemia stimulates tubular cells to produce ECM components leading to vascular sclerosis[18].

PATHOPHYSIOLOGY:

A. DISTURBANCE IN WATER HOMEOSTASIS:

In early stage of CKD, kidney loses its ability to concentrate urine leading to polyuria and nocturia. Nocturia worsens uremic symptoms particularly nausea and vomiting by causing volume depletion. This phenomenon is called as morning sickness of uremia. But in late stage, there will be loss of renal ability to dilute urine leading to fluid overload[28].

B. DISTURBANCE IN SODIUM EXCRETION:

CKD is associated with disruption of glomerulotubular balance (excretion matches intake) of sodium such that dietary intake of sodium exceeds its excretion leading to sodium retention and ECFV expansion. This expansion can contribute to hypertension. As long as water intake does not exceed its clearance, this ECFV expansion will be isotonic and plasma sodium concentration remains normal. If glomerulotubular balance of water is disturbed, water retention occurs and leads to hyponatremia. Thus hyponatremia is not commonly seen in CKD but if present it responds to water restriction[1]. Salt losing nephropathy may be present in some patients. When they develop fluid loss from extrarenal cause they

are prone for ECFV depletion causing hypotension, dehydration and hypovolemia. This contributes to acute on chronic kidney failure[25].

C. ALTERATIONS IN POTASSIUM HOMEOSTASIS:

Potassium excretion is mainly mediated by aldosterone dependent secretory events in the distal segments. Thus decline in GFR is not accompanied by decline in potassium excretion. Hyperkalemia in CKD may be contributed by increased dietary intake of potassium, protein catabolism, Hemolysis/Hemorrhage/Transfusion of red blood cells, metabolic acidosis and drugs like ACE inhibitors, β blockers and aldosterone antagonist[1].

D. ACID BASE DISORDERS:

Metabolic acidosis is common in patients with CKD. In early stages it is due to decreased ammonia production leading to non anion gap metabolic acidosis[1]. In late stages due to decrease in GFR there will be decrease in excretion of titratable acid causing anion gap acidosis[25].

E. DISTURBANCE IN CALCIUM – PHOSPHATE METABOLISM:

Retention hyperphosphatemia:

Kidney is the main route of phosphate elimination. When GFR falls below 30ml/min phosphate gets accumulated causing hyperphosphatemia.

Hypocalcemia:

Serum calcium and phosphate are always in dynamic equilibrium. Hypocalcemia occurs with any increase in phosphate levels. Hyperphosphatemia also causes decreased activation of vitamin D in PCT which also contributes to hypocalcemia.

Hyperparathyroidism:

Secondary hyperparathyroidism occurs in response to hypocalcemia. Hyperphosphatemia increases parathyroid hormone secretion independent of calcium levels by mechanisms like stimulation of parathyroid cell growth, inducing calcitriol resistance in parathyroids and direct activation of PTH secretion leading to tertiary hyperparathyroidism.[29]

Vitamin D metabolites:

CKD is associated with progressive decline in activity of 1 α hydroxylase activity leading to decrease in 1, 25 dihydoxyvitamin D_3 (active form) levels and decrease in concentration of VDR (Vitamin D receptor). This leads to secondary hyperparathyroidism, decreased intestinal absorption of calcium, defective mineralisation and growth retardation[30].

F. RETENTION OF UREMIC TOXINS:

These are responsible for most of the uremic symptoms. Uremic toxins recognized are

1. Small, water – soluble , non – protein – bound compounds, such as urea.
2. Small, lipid – soluble , protein – bound compounds, such as phenols
3. Larger or middle – molecules , such as beta2 – microglobulin and parathyroid hormone[31].

G. ACTIVATION OF RENIN ANGIOTENSIN ALDOSTERONE SYSTEM:

In CKD plasma renin activity is high inspite of hypervolemia and sodium retention. The main mechanism involved is due to luminal narrowing of preglomerular vessels because of vascular sclerosis, baroreceptors in JG apparatus will measure low perfusion pressures falsely leading to increased renin release[30].

CLINICAL MANIFESTATIONS:

1. GASTROINTESTINAL MANIFESTATIONS:

Mouth:

In saliva urea is broken down to ammonia that leads to dysguesia and uremic fetor (urine like smell in breath).

Stomach :

Gastritis, peptic disease, mucosal ulcerations occur due to high concentration of urea in gastric juice that cause chronic irritation of gastric mucosa. Gastrointestinal bleeding also occurs.

Intestine:

Constipation is more common in CKD due to dehydration. But diarrhea also occur due to deposition of urea in colonic mucosa which leads to ulceration that is liable to superadded infections[25].

2. NEUROLOGIC MANIFESTATIONS:

CNS effects:

They become evident in stage 3 CKD. Early symptoms are mild disturbances in concentration, memory and sleep. In advanced stage neuromuscular irritability including hiccups, muscle twitching, fasciculations occur. If untreated patient develops asterixis, myoclonus, seizures and coma. It may be due to

- Disruption of balance between excitatory and inhibitory neuronal pathways by organic substances.
- Guanidine compounds will act as agonist at NMDA receptors and antagonist at $GABA_A$ receptors leading to cortical excitability.

- In patients with CKD asymmetrical dimethyl arginine is increased which inhibits endothelial nitric oxide synthase[32].

Peripheral neuropathy:

Peripheral neuropathy is commonly seen in patients with stage 4 CKD. Initially sensory involvement occurs later it becomes mixed. Also distal parts are affected more than proximal and lower extremities more than upper extremities[1]. CKD is often associated with persistant hyperkalemia that cause reverse activation of Na^{+} / Ca^{2+} exchanger. This leads to increased influx of calcium which causes continuous membrane depolarization. All these changes contributes to axonal degeneration which leads to peripheral neuropathy.

Autonomic neuropathy:

Autonomic neuropathy may be due to

- Sympathetic overactivity caused by increased renal afferent signaling, tonic arterial chemoreceptor activation and reduced renalase production (newer monoamine oxidase that metabolises catecholamines)
- Parasympathetic hypoactivity due to impaired baroreceptor sensitivity Induced by arterial stiffness and vascular calcification[33].

- Autonomic dysfunction results in orthostatic hypotension, sweating abnormalities, impotence and abnormal response to valsalva maneuver[34].

Restless leg syndrome:

It is due to decrease in dopaminergic modulation of intracortical excitability with decreased supraspinal inhibition and increased spinal cord excitability. It is an unpleasant creepy sensations in extremities and a compulsive need to move the limbs[32].

Sleep disorders:

- Disruption of sleep wake cycle leading to excessive daytime sleepiness and insomnia
- Some patients develop sleep apnea due to pharyngeal narrowing that predisposes to upper airway occlusion. It can be precipitated by volume overload and uremic neuropathy that causes impaired upper airway muscle tone.
- Periodic limb movements of sleep (PLMS) characterized by repetitive and sudden jerking movements of lower limbs during sleep[35].

3.HEMATOLOGICAL MANIFESTATIONS:

Anemia:

Patients usually develop normochromic normocytic anemia. The most common cause is relative erythropoietin deficiency caused by

- ✓ Transformation of peritubular fibroblast to myofibroblast leading to decreased EPO gene expression[36].
- ✓ Diseased kidney adapts to an increased single nephron sodium load by attenuating tubular sodium absorption. This leads to decreased oxygen consumption and improves oxygenation in outer medulla and thereby reduces stimulus for EPO deficiency.
- ✓ Neutralisation of EPO by soluble EPO – R which is induced by inflammatory mediators like IL – 6 and TNF – α.
- ✓ Inactivation of EPO by desalylation mediated by proteases which is increased in uremic patients[37].

Abnormal homeostasis:

Bleeding tendency is increased due to impaired platelet aggregation and adhesion to endothelium caused by defect in expression and activation of glycoprotein adhesion receptors, reduced quantities of ATP, selective defect in the pool of deposited serotonin and decreased synthesis of thromboxane A2[36]. Thrombophilic tendency may be increased

in certain patients. After activation platelets release platelet microparticles which expose procoagulant proteins, possess membrane receptor for coagulation factor V and contains high amounts of negatively charged phospholipids on outer membrane that act as a high catalytic surface for prothrombinase reaction[38].

4. CARDIOVASCULAR MANIFESTATIONS:

Hypertension:

It is the most common complication in CKD. Hypertension is either due to

- ✓ High renin secretion
- ✓ Activation of baroreceptors and chemoreceptors in kidney in response to pressure changes or metabolites from ischemia leading to increased renal nerve firing that contributes to centrally mediated hypertension by activating sympathetic nervous system
- ✓ Increase in ET – A receptor expression in vascular smooth muscle cells that causes vasoconstriction and decreased expression of ET – B receptor in endothelial cells causing loss of negative feedback inhibition of endothelin – 1.
- ✓ CKD is associated with elevated vasopressin levels which act on V_{1a} receptor on vascular smooth muscle causing vasoconstriction and

V_2 receptor that promotes water reabsorption by inserting aquaporin channels in collecting duct[39].

Left ventricular hypertrophy:

Left ventricular hypertrophy is characterized by increased left ventricular mass together with increase in size of individual cardiomyocytes and cytosolic calcium . Initially it was thought to be an adaptive response to increased afterload (concentric hypertrophy) or volume overload (eccentric hypertrophy)[36]. Eventually it becomes maladaptive with an imbalance between energy expenditure and production causing chronic energy deficit and myocyte death. In CKD, LVH is also associated with development of intramyocardial fibrosis exacerbated by factors like male gender, ischemia, aldosterone, angiotensin II and catecholamines. Eventually, the deleterious effects of hypertrophy, increased LV chamber pressure and fibrosis dominate leading to the development of cardiomyopathy and LV failure[40].

Dyslipidemia:

Dyslipidemia often occurs in stage 3 CKD. It is characterized by accumulation of partially metabolized VLDL and disturbance in maturation of HDL. Laboratory findings are increased triglycerides and HDL[38]. LDL levels are normal or elevated.

Ischemic heart disease

CKD patients are more prone for accelerated atherosclerosis that leads to ischemic heart disease. Risk factors are

- ✓ Hypertension, dyslipidemia
- ✓ Increased homocysteine levels
- ✓ Microinflammation reflected by increased concentration of high sensitivity c – reactive protein[36] with a corresponding fall in negative acute phase reactants like serum albumin and fetuin[1]
- ✓ Propensity of coronary plaques to calcify under the influence of high phosphate concentration[36].

Congestive heart failure:

It can be a consequence of diastolic dysfunction in association with LVH or systolic dysfunction caused by dilated cardiomyopathy or ischemia.

Arrythmia:

Both atrial and ventricular arrhythmias can occur. The precipitating factors are LVH, CAD, electrolyte disturbances and dialysis associated hypotension (more common in patients undergoing hemodialysis)[41].

5. MUSCULO - SKELETAL MANIFESTATIONS:

Bone disease/ Renal osteodystrophy is common in advanced cases.

Osteitis fibrosa cystica:

It is due to secondary hyperparathyroidism and stimulation of osteoclast by PTH. It is a high bone turn over disease characterized by subperiosteal lesions and osteoclastic bone resorption. It causes proximal muscle weakness, bone pain[42] and formation of brown tumors[1] (bone cyst with hemorrhagic elements).

Adynamic bone disease:

It is a low bone turn over disease caused by iatrogenic suppression of PTH , downregulation of PTHR1 leading to skeletal resistance to PTH actions and inhibition of PTH secretion by proinflammatory cytokines. It is more common in diabetics and possess high linkage with malnutrition inflammation complex syndrome (MICS)[43].

Osteomalacia:

Osteomalacia is characterized by lack of bone mineralization. It is due to aluminium toxicity (chronic ingestion of aluminum containing phosphate binders/Excess aluminium in impure dialysate water) or hypovitaminosis D[42].

Vascular calcification:

It is an active process characterized by de differentiation of vascular smooth muscle cells to osteoblast or chondrocyte like cells. Later these cells lay down extracellular matrix of collagen and non collagenous proteins and create matrix vesicles or apoptotic bodies that attach to ECM initiating mineralization[44]. It is of 2 types

- ✓ One involves intimal layers of arteries and is associated with atherosclerotic plaques which is patchy in distribution and causes target organ ischemia from lumen obstruction or plaque rupture.
- ✓ Other type involves medial wall of arteries which is diffuse that leads to increased vessel stiffness and decreased vascular compliance contributing to LVH[43].

Calciphylaxis:

It is also called calcific uremic ateriolopathy. It is a form of vascular calcification presenting with painful nodules that advances to ischemic necrosis. The main etiology is deficiency of matrix gla protein (MGP) which normally inhibits extraskeletal calcification by modulating the activity of Bone morphogenetic protein – 2 (induces vascular calcification) and low plasma levels of fetuin A. MGP activity is inhibited by warfarin leading to calciphylaxis[44].

Myopathy:

Muscle wasting occurs mainly in the proximal area of lower limbs leading to waddling gait[25]. It is mainly due to metabolic acidosis that activates ubiquitine sensitive proteasome pathway and branched chain amino acid dehydrogenase in skeletal muscle cells causing protein breakdown[45].

6. ENDOCRINE MANIFESTATIONS:

Thyroid hormones:

In CKD total circulating T3 concentration is reduced due to impaired peripheral conversion of T4 to T3 and loss of thyroid binding globulin. But clinical hypothyroidism is extremely rare in these patients[46].

Insulin:

Since kidneys are the major route of elimination of insulin, plasma half life of exogenously administered insulin is increased. Thus in diabetics there is fall in the requirement of insulin as kidney function declines progressively[47]. In CKD, patients are prone for insulin resistance. Insulin binding to its receptor and receptor density remains unchanged. But the contributing factors are

- ✓ High levels of adipose tissue derived hormones like leptin, resistin and adiponectin.
- ✓ Presence of unidentified nitrogenous products[48].

Sex hormones:

Males:

In stage 5 CKD prolactin levels are elevated that leads to galactorrhoea. Patients are prone for testicular failure characterized by erectile dysfunction, decreased testosterone and reduced spermatogenesis.

Females:

In stage 4 – 5 pituitary ovary axis is disturbed. LH levels are raised but preovulation surge is absent. So the cycles will be anovulatory and irregular[49].

Growth hormone:

CKD patients are prone for growth failure that mainly occurs due to GH insensitivity characterized by decrease in GH receptors, post GH receptor defect and decreased IGF – 1 syntheis[48].

Appetite:

Appetite is suppressed in CKD due to increased leptin levels and increased des acyl GHrelin levels[46].

7. IMMUNOLOGICAL MANIFESTATIONS:

Infection is another leading cause of death in patients with CKD stage 5. CKD is a state of chronic immunosuppression with both cellular and humoral immunity defects like

- ✓ increased levels of granulocyte inhibitory protein I and II that inhibits

  chemotaxis, uptake of deoxyglucose, oxidative metabolism and intracellular killing by polymorphonuclear leucocytes.
- ✓ Increased degranulation inhibitory protein I and II that inhibits spontaneous and stimulated polymorphonuclear leucocytes degranulation. These mechanisms increases susceptibility to infections[50].
- ✓ Antibody responses to immunization is poor. Thus in CKD to increase the chance of seroconversion Hepatitis B vaccine should be given as early as possible[38].

8. NUTRITIONAL ABNORMALITIES:

Protein energy malnutrition is more common in patients with stage 4 – 5 CKD. It is an indicator for initiation of renal replacement therapy[1]. The main causes are anorexia, acidosis, insulin resistance, inflammation and urine protein loss[38].

9. DERMATOLOGIC MANIFESTATIONS:

In CKD, patients develop cutaneous pigmentation due to deposition of retained pigmented metabolites or urochromes[1]. Recently nephrogenic fibrosing dermopathy has been reported in many patients

which is characterized by progressive subcutaneous induration in arms and legs. It can be precipitated by magnetic resonance contrast agent gadolinium. Patients are also prone for uremic frost characterized by yellowish tinge on skin due to evaporated urea rich sweat[51]. Pruritis can occur from skin dryness or irritation of the cutaneous sensory nerves by calcium deposits or by parathormone[25].

10. PULMONARY MANIFESTATIONS:

The most common manifestations are uremic lung and uremic pleuritis. Uremic lung characterized by batwing perihilar infiltrates represents pulmonary edema and is caused by volume overload or myocardial dysfunction[51].

EVALUATION OF PATIENTS WITH CHRONIC KIDNEY DISEASE:

I. History:

- ✓ Family history should be elicited. Familial renal diseases may be monogenic (Polycystic kidney disease, Alport's disease, Medullary cystic disease) or polygenic (Diabetes, Hypertension, Obesity)
- ✓ Evaluation of appetite, recent changes in weight, mental acuity, memory, mood, any change in sleep pattern should be done.

Changes in these parameters are consistent with presence of uremic syndrome.

- ✓ Analysis of all systems should be done – urinary symptoms (polyuria, nocturia, frequency), Cardiovascular system (H/O MI/CHF), Peripheral vascular disease (claudication, peripheral ulcers), Musculoskeletal complaints (muscle wasting), osteodystrophy (bone and joint pain) should be elicited.
- ✓ Medication history to rule out drug induced chronic kidney disease. History of intake of drugs like calcineurin inhibitors, lithium, pamidronate, chemotherapeutic drugs, analgesics are to be elicited.

II. Physical examination:

- ✓ Assessment of vital signs – supine and upright pulse and BP
- ✓ Determine the volume status

A) To rule out volume depletion orthostatic pulse is a better marker than orthostatic hypotension because orthostatic hypotension is present in 10% of subjects normally. Orthostatic pulse increase of more than 30 beats/min indicates less severe volume depletion. Other signs are dry axilla, dry mucous membrane, furrowed tongue and flat neck veins.

B) To rule out volume overload the signs are hypertension, peripheral edema, pleural effusion, pulmonary rales.

- ✓ Generalized muscle wasting and sallow appearance of skin are signs of advanced CKD.
- ✓ Evaluation of target organ damage

A) Cardiac examination to reveal LVH or decompensation

B) Carotid pulse for bruits to rule out atherosclerosis[52]

C) Fundoscopic examination - can demonstrate microaneurysms and proliferative retinopathy diagnostic of diabetic retinopathy or arteriolar narrowing, arteriovenous nicking, hemorrhage and exudates which are characteristics of Hypertensive retinopathy.

D) Skin changes - pallor, scratches and excoriations from pruritis and uremic frost.

E) Perforation of nasal septum diagnostic of Wegener's granulomatosis or cocaine use

F) Abdominal examination - palpable kidneys observed in polycystic kidney disease, palpable bladder suggestive of chronic urinary outlet obstruction and flank mass that might indicate retroperitoneal fibrosis or tumours[53].

III. Laboratory test: Blood

- ✓ Complete blood count, absolute reticulocyte count, serum ferritin, transferrin saturation

- ✓ Blood urea and creatinine levels - usually elevated
- ✓ Serum electrolytes - Hyperkalemia is common
- ✓ Serum calcium and phosphate levels - Hypocalcemia and Hyperphosphatemia are common
- ✓ Markers of bone formation and resorption - total alkaline phosphatase levels and bone specific ALP levels.
- ✓ Lipid panel for dyslipidemia
- ✓ Serum uric acid levels for hyperuricemia
- ✓ Sedimentation rate and C – Reactive protein to assess patient's inflammatory stage
- ✓ Serological testing for SLE or other collagen vascular diseases, HIV panel, Hepatitis B/C[52].

Urine analysis:

- ✓ Urine specific gravity is low and fixed at 1010.[25]
- ✓ Hematuria and proteinuria can be seen. Presence of oval fat bodies that signifies high grade proteinuria and dysmorphic red cells points towards glomerular disease[53]. Low grade proteinuria will be seen in interstitial disease and polycystic kidney disease[52].

IV. Assessment of progression of disease:

a) Estimation of glomerular filtration rate:

It is the measure of number of functioning nephrons. It indicates degree of kidney injury and rapidity of progression. It is estimated from Cockcroft and Gault or MDRD (Levey) formulas which are based on creatinine clearance.

Creatinine is the metabolic product of creatine and phosphocreatinine that are found in muscle. It is freely filtered by glomerulus and a small amount is also secreted. Thus creatinine levels are influenced by muscle mass and gender. Hence recently new endogenous compounds have been evaluated for better estimation of GFR.

Serum cystatin C is a basic protein of cystatin superfamily. It is synthesized by all nucleated cells at a constant rate. It is freely filtered, not secreted and not influenced by muscle mass, gender and inflammatory process[54]. In a study it was found that when GFR levels fell to 88ml/min/1.73m^2 cystatin levels rose but creatinine levels did not rise until GFR dropped to 75 ml/min/1.73m^2. Hence cystatin is considered as a better endogenous marker of GFR[55].

b) Quantification of proteinuria:

Proteinuria occurs with glomerular damage. It can precede elevations of serum creatinine and so it is considered as an early marker of kidney damage.

- ✓ Proteinuria – total protein excretion rate > 300mg/24hr. It includes albumin and other low molecular weight proteins like globulin and apoproteins
- ✓ Microalbuminuria – albumin excretion rate 30 – 300 mg/day. Assessment of albuminuria is a better indicator than proteinuria as it is more specific for glomerular damage.

Timed collection:

24 hour collection of urine is the best method for quantification but timed sample overnight is more reliable since protein excretion vary throughout the day and with postural changes.

Untimed collection:

Spot urine samples are used and the values are expressed as protein or albumin creatinine ratio. It is more convenient and not affected by posture and hydration status. It is preferably done in first morning urine samples (as it correlates better with 24 hr protein excretion) but random samples are also accepted[56].

Diagnostic criteria:

Total protein excretion rate:

Classification	24 hour collection (mg/day)	Spot urine dipstick (mg/dl)	Spot urine protein creatinine ratio (mg/g)
Normal	<300	<30	<200
Clinical proteinuria	>300	>30	>200

Albumin excretion rate

Normal	<30	<3	<17(males) <25(females)
Microalbuminuria	30 – 300	>3	17 – 250 (males) 25 – 355 (females)
Albuminuria	>300	NA	>250 (males) >355 (females)

V. Imaging studies:

✓ Renal ultrasonography - B/L small kidneys suggestive of CKD with scarring. Sometimes kidney size might be normal as with diabetic CKD, amyloidosis and HIV nephropathy. Kidney size is increased in polycystic disease. Discrepancy in size of more than 1 cm between the kidneys indicates unilateral disease process[1]. Increase echogenicity is also suggestive of chronic medical renal disease[57].

✓ Doppler sonography to rule out renovascular disease

✓ Voiding cystogram for diagnosing reflux nephropathy

✓ Echocardiogram to assess cardiac size, LV function, regional wall motion abnormalities, pulmonary pressures and valvular function

✓ Radiological bone studies to assess renal osteodystrophy

radiographic contrast imaging should be avoided in CKD for fear of contrast induced renal failure[1].

VI. Renal biopsy:

It is indicated for the following reasons in CKD

✓ To find out if the underlying cause is reversible and treatable

✓ To help clinical decisions like further investigations, selection of renal replacement therapy

✓ To assess prognosis[58]

- ✓ Unexplained proteinuria associated with progressive renal dysfunction[57].

It is contraindicated in bilateral small kidneys because of too much scaring underlying disease may not be apparent and associated with increased chance of bleeding[1].

TRIPLE MARKER APPROACH FOR DETECTION OF CKD:

This approach has been recently introduced and the parameters assessed are Cystatin C based measure of GFR, serum creatinine based GFR estimation and urine albumin creatinine ratio.

CHRONIC KIDNEY DISEASE BIOMARKERS:

Plasma Asymmetric DiMethyl Arginine (ADMA):

It is an endogenously generated methylated arginine that possess inhibitory activity against NOS. ADMA levels are high in patients with chronic kidney disease and is associated with carotid intima – media thickness, left ventricular hypertrophy, cardiovascular complications, progression of renal disease and mortality in ESRD.

Fibroblast Growth Factor 23 (FGF 23):

FGF 23 is secreted by osteoblast. Physiologically it regulates the activity of sodium dependent phosphate cotransporters at the brush border membrane of proximal tubule and helps in maintanence of phosphate

homeostasis. CKD is associated with phosphate retention and decrease in calcium and activated vitamin D levels. FGF 23 is elevated in CKD and act as a prognostic indicator for cardiovascular disease.

Urinary Monocyte Chemoattractant Protein – 1(MCP – 1) :

Podocytes and tubular cells produce MCP – 1 in response to proinflammatory cytokines, high glucose levels and advanced glycosylation end products. Thus urinary levels of MCP – 1 is increased in diabetic CKD[59].

MANAGEMENT :

Step 1. Establishing chronicity:

Factors suggesting chronicity are absence of severe symptoms inspite of very high urea and creatinine levels, anemia of chronic disease, sexual dysfunction, skin disorders, neurological complications and small kidneys on renal imaging[60].

Step 2. Attenuation of progression of CKD:

Smoking cessation:

Smoking is an important risk factor for development of microalbuminuria and overt proteinuria. Risk is increased with high daily consumption (> 20 cig/day), long duration (> 40 yrs) and high cumulative

dose (> 30 pack years). Smoking cessation reduces the risk of development of CKD and progression of existing CKD.

Weight loss:

Weight loss as little as 10 lbs (4.5 kg) results in reduction of blood pressure and glycemic markers. Also each 1 kg loss is associated with corresponding 110 mg decrease in proteinuria and 1.1 mg reduction in microalbuminuria independent of BP changes[61].

Dietary sodium intake:

High salt intake causes activation of tissue renin angiotensin aldosterone system leading to renal and myocardial fibrosis. It also overrides the antihypertensive and antiproteinuric effects of drugs. Thus restriction of sodium intake is advised. Recommended sodium intake in CKD is $<1.5 g Na^{+}$/day (65 mmol/day)[19].

Control of protein intake:

Protein mediated hyperfiltration leads to rapid decline in renal function. Thus protein restriction is recommended. Dietary protein intake advised is 0.6 – 0.75g/kg/day and it should be of high biological value. In stage 5 CKD, spontaneous protein intake is reduced and the patients become prone for protein energy malnutrition. So the protein intake can be increased upto 0.9g/kg/day[1].

Control of blood pressure and proteinuria:

The goals of therapy are blood pressure < 130/85 (<125/80 if proteinuria is >1gm/day), reduction in proteinuria below 300mg/24hr and a loss of GFR less than 2ml/min per annum[62].

A. Angiotensin Converting Enzyme inhibitors: (ACE inhibitors)

ACE Inhibitors inhibit the cleavage of angiotensin-I to angiotensin-II (potent vasoconstrictor). They also inhibit breakdown of bradykinin (which is a potent vasodilator by stimulating release of nitric oxide and prostacyclin). Both these mechanism contribute to hypotensive action[63]. They decrease both arterial and venous pressures thereby reduce cardiac afterload and preload[64].

ACE inhibitors dilate both afferent and efferent arteriole. Thus provide improved intrarenal hemodynamics such as decrease in glomerular efferent arteriolar resistance and reduction in intraglomerular capillary pressure[63]. Angiotensin II stimulates growth factors and inflammatory cytokines leading to glomerular and interstitial sclerosis and also induces glomerular heparanase causing loss of glomerular permselectivity. ACE inhibitors attenuate these angiotensin II mediated intrarenal pathways leading to renoprotection.

ACE inhibitors also reduce proteinuria by improving the permeability selectivity of filtering membrane and thereby ameliorate the deleterious effect of protein exposure on tubular cells, podocytes and mesangium[65]. The antiproteinuric effects are independent of blood pressure lowering effect. Thus they are recommended in diabetics even in the absence of hypertension[63]. The renoprotective effect of ACE inhibitors are directly proportional to baseline proteinuria. Thus patients with high grade proteinuria benefit more. But patients with less severe proteinuria also benefit as they also attenuate the progression of incipient nephropathy (manifesting with microalbuminuria) to overt nephropathy. They also induce regression towards normoalbuminuria from microalbuminuria. Thus they are effective in all stages of renal failure in diabetic patients[65].

Some of the side effects of ACE inhibitors are non productive cough, hyperkalemia, reduction in GFR in patients with atherosclerotic renovascular disease and angioneurotic edema[63].

B. Angiotensin II receptor blockers: (ARB) Losartan:

Mechanism of action:

It acts by causing competitive inhibition of AT_1 receptor but the reaction is insurmountable (maximum response to angiotensin II

cannot be restored in the presence of ARB regardless of concentration of angiotensin II). The mechanisms for insurmountable antagonism are

- Slow dissociation kinetics of drug from AT_1 receptor
- ARB induced receptor internalization
- Alternate binding sites for ARB on AT_1 receptor

Thus insurmountable antagonism leads to sustained receptor blockade even with increased levels of endogenous ligand. It has high affinity for AT_1 receptor than AT_2 receptor.

ARB and ACE inhibitors differ in certain aspects.

- ARBs reduce activation of AT_1 receptor more effectively than ACE inhibitors because ACE inhibitors do not block non ACE angiotensin II generating pathways
- ARBs permit activation of AT_2 receptors. Most of the biological effects of angiotensin II like contraction of vascular smooth muscle, rapid and slow pressor responses, aldosterone secretion, release of adrenal catecholamines, enhanced noradrenergic neurotransmission, increased sympathetic tone, changes in renal function, cellular hypertrophy and hyperplasia is mediated via AT_1 receptor. AT_2 receptor counterbalances the actions of AT_2 receptor. AT_2 receptor possess antiproliferative, proapoptotic, vasodilatory, natriuretic and

antihypertensive effects. ARBs stimulate renin release and so increased amounts of angiotensin II is available to activate AT_2 receptor.

- ACE inhibitors increase Ang (1-7) levels more than ARBs because Ang (1-7) is cleared by angiotensin converting enzyme.
- ACE inhibitors increase levels of many ACE substrates like bradykinin which is responsible for cough and angioneurotic edema. ARBs are less prone for these adverse effects[66].

Renal effects of AT_1 blockade:

- ✓ Renal hemodynamics - decrease renal blood flow, decrease renal vascular resistance, variable effects on GFR depending on BP, improved autoregulation, decreased efferent arteriolar resistance, reduction in intraglomerular capillary pressure.
- ✓ Renal tubular function – reduction of sodium and fluid reabsorption, normalizes acidification and bicarbonate excretion, blocks stimulation of aquaporin channels[67].
- ✓ Glomerular permselectivity – improves charge selectivity of glomerular basement membrane , reduction in mesangial uptake and clearance of macromolecules. These effects contribute to reduction in proteinuria[68].

- ✓ Renal fibrosis – blocks ECM deposition in the mesangium, attenuates fibroblast proliferation and transformation.
- ✓ Inflammation – blocks proliferation of leukocytes and upregulation of adhesion molecules, decreased generation of reactive oxygen species[67].

Pharmacokinetics:

Oral bioavailability of losartan is 33%. It undergoes first pass metabolism with CYP450 2C9 and 3A4 and gets converted to active metabolites[66]. Carboxylic acid metabolite EXP 3174 is one among them which is more potent than the parent drug and a reversible non competitive inhibitor of AT_1 receptor. Losartan achieves peak plasma concentration in 1 – 3 hrs. The plasma t1/2 of losartan and EXP 3174 are 2.5 and 6-9 hrs respectively. Elimination is by renal (40%) and biliary (60%) excretion[67]. Thus its plasma concentration is affected by hepatic and not by renal insufficiency. It is administered at a dose of 25 – 100mg once or twice daily[66].

Adverse effects:

- ✓ ARBs have long onset of action of about 4 - 6 wks. This avoids first dose hypotension and rebound hypertension seen with other drugs[68].

- ✓ Cough and angioedema less than ACE inhibitors
- ✓ Acute renal failure in patients with renal artery stenosis
- ✓ Hyperkalemia (in renal disease/combined with K^+ supplements or K^+ sparing diuretics)
- ✓ Post marketing reports of anaphylaxis, hepatitis, neutropenia, leucopenia, agranulocytosis, pruritus, urticaria and vasculitis[66].

ARBs versus ACE inhibitors:

Hypertensive patients with normal or impaired renal function treated with ARBs exhibit renal responses similar to ACE inhibitors. Antiproteinuric effects are also similar. Both the dugs exhibit antiproteinuric effects in diabetic and non diabetic kidney disease and also in renal transplant recipients. Maximal antiproteinuric effects are obtained in 3 – 4 weeks. Thus studies in CKD have found no differences in the efficacy of ACE inhibitors or ARBs[68]. Few randomized trials have shown that ACEI are renoprotective in type 1 diabetes, type 2 diabetes with microalbuminuria and nondiabetic CKD. But ARBs are more renoprotective in type 2 diabetes with microalbuminuria and overt nephropathy [69] . When these drugs are given in doses higher than recommended maximum it results in greater lowering of urine protein levels without further reduction in BP[22] . One additional property seen

with losartan that is not seen with other drugs is induction of uricosuria. Uricosuric effect is independent of RAAS inhibition and is not associated with risk of nephrolithiasis. This reduction in serum uric acid levels contribute to additional renoprotective effect[68].

C. Calcium channel blockers(CCB):

Amlodipine:

Mechanism of action:

It is a third generation dihydropyridine type of CCB with stable pharmacokinetics and less cardioselective[70] . Voltage gated L – type calcium channel is the dominant type found in cardiac and smooth muscle. It is comprised of α_1 (larger pore forming subunit) , α_2 , β, γ and δ subunits. Amlodipine act by binding to one site on α_1 subunit. The drug acts from inner side of the membrane and bind effectively to open and inactivated channels. This binding leads to reduction in the frequency of opening of channels in response to depolarization. This results in marked decrease in transmembrane calcium current through these channels leading to decrease in total amount of calcium reaching the intracellular store.

Pharmacological effects:

Vascular smooth muscle:

Vascular smooth muscles are highly dependent on transmembrane calcium influx to maintain normal resting tone and for

contractile responses. Amlodipine by blocking transmembrane calcium influx causes long lasting smooth muscle relaxation that leads to reduction in peripheral vascular resistance and fall in blood pressure. Arterioles are more sensitive to hypotensive action than venules. It also has a greater ratio of vascular smooth muscle effects relative to cardiac effects[71].

Cardiac muscle:

Due to gradual onset of action and long duration effects it causes less activation of sympathetic nervous system and hence heart rate is unaffected. In general amlodipine does not cause any change to sympathetic nervous system although it appears to suppress baroreceptor mediated activation of sympathetic nervous system but this sympatholytic activity is less effective than cilnidipine[72]. They also dilate coronary arteries leading to increase in coronary blood flow[68].

Renal effects:

L- type calcium channels are abundant in afferent arteriole but not in efferent arteriole. Amlodipine dilate afferent arteriole directly than efferent arteriole resulting an increase in intraglomerular pressure. Filtration fraction which parallels the change in intraglomerular pressure is also increased. Now amlodipine is thought to blunt glomerular hypertension

due to their systemic BP lowering effect as systemic blood pressure is directly transmitted to afferent arteriole. But this effect is inconsistent and varies with patients[73]. Long term effects have shown that only few patients exhibit no change in GFR but others exhibit exaggerated increase in GFR and renal plasma flow. Proteinuria is not decreased but paradoxically increased in some cases due to increased glomerular capillary pressure. It also exhibits natriuretic effect mediated by direct inhibition of renal tubule sodium and water absorption[74].

Pharmacokinetics:

It has high oral bioavailability (60-80%). Onset of action is gradual due to intermediate rate of drug absorption. Peak plasma concentration is achieved in 6-8 hrs. Its t1/2 is 30 – 50 hrs[71]. It is extensively plasma protein bound and eliminated via liver[75].

Cilnidipine :

Mechanism of action:

It is a unique dihydropyridine classified as fourth generation calcium channel blocker. It is a dual blocker of L type and N type calcium channel. N type is widely distributed in nervous system. Cilnidipine by blocking N type channels blocks the release of sympathetic neurotransmitter. Thus it possess antisympathetic property in additional.

Pharmacological effects:

Vascular smooth muscle:

Cilnidipine relaxes vascular smooth muscle by calcium channel antagonism and activation of eNOS that leads to production of nitric oxide. It exhibits slow onset and long lasting antihypertensive action. Pure L – type calcium channel blockers due to their potent vasodilatory property prompts a rapid compensatory increase in sympathetic nervous activity mediated by N type calcium channel. This reflex leads to release of norepinephrine that produces vascular contraction, tachycardia and activation of RAAS. Cilnidipine by blocking N type channel counteracts these responses and reduces the risk of cardiovascular disease. Thus cilnidipine exhibits significant hypotensive action without causing reflex tachycardia.

Cardiac effects :

It is a more potent coronary vasodilator . During schemia and reperfusion periods, cilnidipine leads to reduction in myocardial infarct size and incidence of ventricular premature beats by decreasing myocardial interstitial norepinephrine levels. It also causes abbreviation of abnormally prolonged ventricular repolarization, decrease in BNP, LV mass index, heart rate and cardiothoracic ratio. As it possesses high vascular

selectivity it does not depress cardiac functions like contractility, SA node automaticity and AV node conductivity like previous CCBs.

Antioxidant property:

All CCBs act as lipophilic chain breaking antioxidant. Lipophilicity of cilnidipine is very much greater than amlodipine such that it suppresses the oxidative stress independent of N type blockade. Cilnidipine also inhibits NADPH oxidase derived superoxide formation in kidneys whereas amlodipine does not. N type calcium channels in podocytes play an important role in angiotensin II induced superoxide formation. Cilnidipine counteracts this and elicits podocyte protection.

Renal effects:

The renal nerve stimulation leads to release of norepinephrine that activates adrenoceptors in the vascular vessels, renal tubular cells and granular cells of the juxtaglomerular apparatus and thereby induces renal vasoconstriction, anti-natriuresis and renin secretion respectively. These effects can be suppressed by cilnidipine. Sympathetic nerves are distributed in both afferent and efferent arteriole. Cilnidipine dilates both afferent and efferent arterioles leading to reduction in glomerular capillary pressure, afferent and efferent arteriole resistance, and glomerular volumes. Filtration fractions remain unchanged or decreased. It also suppresses

glomerular hypertrophy and interstitial fibrosis. Amelioration of glomerular hypertension and antioxidant property both contribute to antiproteinuric effects. Thus cilnidipine retards the progression of renal disease.

Other effects:

- ✓ Cilnidipine decreases the production of aldosterone from adrenocortical cells and reduces plasma levels of angiotensin II and aldosterone.
- ✓ Downward shift of the lower limit of autoregulation for cerebral blood flow, reduction of cerebral infarction size and increase in cerebral blood flow.
- ✓ Improves insulin resistance.
- ✓ Cilnidipine also causes reduction in uric acid levels. This also contributes to renoprotection .
- ✓ Also causes attenuation of platelet activation as catecholamines induce platelet activation via α_2 receptor[76].

Adverse effects of calcium channel blockers:

- ✓ Peripheral edema - results from increased hydrostatic pressure in lower extremities due to precapillary dilation and reflex post capillary constriction . Cilnidipine does not cause peripheral edema as it dilates both precapillary and post capillary beds.

- ✓ Headache, nausea, dizziness, flushing - related to vasodilation (less with amlodipine and cilnidipine)
- ✓ Gingival hyperplasia
- ✓ Worsening of gastroesophageal reflux disease – due to inhibition of contraction of lower esophageal sphincter
- ✓ Rash, constipation
- ✓ Elevated liver enzymes.[75]

Advantages of calcium channel blockers in clinical use:

CCBs are generally well tolerated. They do not cause electrolyte derangements, alteration in glycemic or lipemic control, sexual dysfunction and orthostatic hypotension (as venoconstriction is intact). Most of the CCBs, especially long acting agents reduce heart rate, improve myocardial oxygen demand, conserve contractility and improve ventricular filling. Thus they are mainly used as antihypertensive agents in CKD with metabolic disorders like diabetes, peripheral vascular disease and stable IHD. They are also ideal agents for elderly patients as they tend to lower the risk of stroke. Newer CCBs have additional effect of reducing proteinuria , maintaining GFR and retarding progression of CKD[74].

D. Direct renin inhibitors (DRI) :

DRI inhibits RAAS at its rate limiting step (conversion of angiotensinogen to angiotensin I) and therefore achieve complete blockade of RAAS[61]. They do not cause reactive increase in renin and so plasma levels of angiotensin II is not elevated. They cause dose dependent decrease in systolic and diastolic blood pressure, also reduce proteinuria and reverse left ventricular hypertrophy. Thus provide comparable renoprotection as an ACE inhibitor and ARB[74].

E. Aldosterone antagonist:

Aldosterone is an important mediator of progressive renal injury through hemodynamic and profibrotic actions[61]. Aldosterone antagonist blocks sodium and water retention leading to reduction in blood pressure and also reduces proteinuria independent of BP lowering effect[68]. Their actions are generally additive to ACE inhibitors or ARBs and monotheray with these drugs are not preferred. But they increase the risk of hyperkalemia when co administered with these drugs[61].

F. Beta blockers:

They act via attenuation of sympathetic stimulation through competitive antagonism of catecholamines action at β adrenergic receptors. They cause reduction in GFR and suppression of plasma renin activity.

They do not have significant action on proteinuria. They are mainly used as secondary therapy in patients with CKD with specific comorbidities like heart failure, post myocardial infarction and angina. It is also found that reduction in major cardiovascular events associated with beta blockers is not seen in elderly patients. Hence they are not recommended as first line agents in elderly patients.

E. Diuretics:

Diuretics are usually employed in CKD to control blood volume and to reduce edema formation. When serum creatinine reaches 2 mg/dl volume reduction is better with loop diuretics than with thiazide diuretics. The renal clearance of loop diuretics fall in parallel with GFR because of accumulation of organic acids that compete for proximal secretion and reduced renal mass. But the maximal increase in sodium excretion is maintained due to increased expression of $NA^+/K^+/2\ Cl^-$ cotransporters. Thiazide diuretics are ineffective if creatinine clearance is <35ml/min[74]. Thiazides when used with loop diuretics in patients with GFR 5 – 15 ml/min/1,73m^2 significantly reduces the dose of loop diuretics. For this metalazone is usually preferred[77].

Step 3: Management of complications:

A. Treatment of hyperkalemia:

Reduction in serum potassium is required if it exceeds 7mmol/l due to risk of cardiac arrest. Emergency treatment is started with 10 ml of 10% calcium gluconate over 10 mins intravenously. Furthermore intravenous glucose (50 ml of 50% dextrose) combined with 10 U of short acting regular insulin can be given for greater decline in potassium levels. Hyperkalemia can also be corrected using β_2 adrenergic agonist (salbutamol 0.5 mg in 100ml of 5% dextrose over 15 mins infusion). Less severe cases can be treated with slowly acting ion exchangers (sodium/calcium polystyrene sulphonate), dietary potassium restriction and prompting diuresis using loop diuretics[60].

B. Treatment of metabolic acidosis:

HCO_3^- concentration should be maintained above 20 mEq/l. This can be achieved with modest amounts of alkali (1 – 1.5 mEq/kg/day). Alkali replacement prevents deleterious effect of prolonged positive H^+ balance like progressive catabolism of muscle and loss of bone. Both sodium citrate and sodium bicarbonate are equally effective[78]. But sodium citrate enhances the absorption of aluminium and so should not be given to patients taking aluminium containing phosphate binders. Also sodium

retention and hypertension is less when sodium is given with bicarbonate[60].

C. Treatment of anemia:

Erythropoiesis Stimulating Agents (ESA):

The target goal is to keep Hb level between 11-12 g/dl. To achieve this ESA are used. It has been shown that correcting anemia with ESA is associated with improvements in left ventricular mass index and electrophysiologic markers of cognitive function. First generation agent is epoetin α. It can be given subcutaneously or intravenously. Sustained levels of the drug is required to trigger erythroid differentiation and to avoid neocytolysis (premature phagocytosis of nascent RBC in the absence of circulating EPO). Subcutaneous administration provides more sustained levels and so less frequent dosing is required than intravenous route[37]. Epoetin α is given at a dose of 80 – 120 U/Kg SC thrice weekly. It leads to gradual rise in hematocrit over 2 – 4 months. Hematocrit beyond 36% is not recommended as it is associated with high incidence of myocardial infarction and death. The most common adverse effect is aggravation of hypertension. Recently darbepoetin α has been introduced which is classified as second generation agent. It has a better pharmacokinetic

profile and is administered at a dose of 0.45 μg/kg subcutaneous once weekly[79].

ESA hyporesponsiveness:

It is defined as continued need for recombinant human erythropoietin agents at doses of 450 U/kg per week intravenous EPO or 300 U/kg per week subcutaneous or 1.5 mcg/kg per week of darbepoetin alfa subcutaneous[31]. The most common causes are inflammation and absolute iron deficiency. MIC syndrome (Malnutrition and inflammation complex) in CKD is associated with increased cytokines that blocks the effect of rHuEPO on erythroid differentiation[37]. Absolute iron deficiency may be due to external blood loses or exhaustion of iron stores due to increased erythropoiesis from EPO therapy[31].

Iron supplementation:

Per oral treatment is ineffective due to dialysis associated loses of iron and high levels of hepcidin in CKD that blocks duodenal uptake of iron. But still it is easy to administer, cheap and associated with less adverse effects. In parenteral therapy, ferric gluconate is better than iron sucrose and dextran as it donates iron more readily to apotransferrin and increases the efficiency of erythropoiesis. In addition to this iron sucrose

can cause injury to proximal tubular cells and thereby increase the incidence of albuminuria[37].

D. Treatment of bleeding diathesis:

The main goal is to restore bleeding time less than 6 mins. This can be achieved using infusion of vasopressin (0.3µg/kg in 100 ml 0.9% saline in 30 mins). A more prolonged effect (up to 14 days) can be obtained by intravenous oestrogens in a dose of 0.6 mg/kg daily for 5 days. This can be also corrected by hemodialysis[60].

E. Treatment of renal osteodystrophy:

The targets are to reduce PTH < 150 pg/ml, phosphate concentration < 4.5 mg/dl and vitamin D 30 ng/ml[80].

Phosphate binders:

Hyperphosphatemia is common in CKD. Hence phosphate binders are used. Calcium containing phosphate binders are cheap but can cause hypercalcemia and promote vascular calcification. Sevelamer and lanthanum are used as they are highly potent and cause less hypercalcemia. This treatment should always be combined with dietary phosphate restriction to reduce pill burden.

Vitamin D:

Vitamin D therapy is aimed at achieving adequate levels of 25 (OH)D, reducing PTH, restoring bone histology to normal. Vitamin D reduces the production of inflammatory cytokines from monocytes. Also local extra renal tissue conversion of 25 (OH) D to 1,25 $(OH)_2$ D is needed for regulation of immune responses, oxidative stress and blood pressure. Nutritional vitamin D therapy is less effective in reducing PTH and restoring bone histology. Ergocalciferol is usually used to achieve the aims. If despite adequate 25(OH) D level, PTH remains elevated active vitamin D therapy in the form of calcitriol or vitamin D analogs are used. They lower PTH and improve bone histology. But they cause hypercalcemia and hyperphosphatemia in a dose dependent manner.

Calcimimmetics:

They allosterically regulate calcium sensing receptor and by sensitizing parathyroid calcium receptor to extracellular calcium inhibit PTH secretion. Cinacalcet is the only agent available in this class. It is used as an add on therapy to usual treatment (active vitamin D or phosphate binders) to reduce calcium and phosphate levels. It lowers phosphate levels by causing PTH reduction, reduced phosphate translocation from bone but has no effect on intestinal phosphate

reabsorption (differs from vitamin D). it is used only in stage 5 CKD and not recommended before due to hypocalcemia and paradoxical increase in serum phosphate levels and for phosphate binders.

Adynamic bone disease:

Limiting calcium load and active vitamin D treatment are the best measures to prevent and treat ABD. Calcium is the most potent suppressor of PTH release. Patients treated with calcium containing phosphate binders exhibit high rates of ABD. Sevelamer and lanthanum are preferable phosphate binders in high risk patients. Exposure to high calcium dialysate also suppress PTH. Thus ABD is common in patients undergoing peritoneal dialysis than hemodialysis. Low doses of active vitamin D is beneficial because excessive dose can cause greater reduction in PTH[43].

F. Intervention for minimizing cardiovascular diseases:

Control of hyperlipidemia:

The common abnormalities seen in CKD are hypertriglyceridemia and elevated LDL levels. Current guidelines recommend statin therapy for all patients as they reduce the risk of cardiovascular events and prevent accelerated loss of renal function. The target goal is to keep LDL < 100mg/dl and triglyceride < 180 mg/dl[60]. Statins reduce total cholesterol,

LDL, apo B and TG effectively. Effect on HDL is variable. They also cause dose dependent inhibition of receptor mediated endocytosis of protein in proximal tubular cells and thus exhibit antiproteinuric effects. Lp (a) levels are not reduced. But combination therapy of nicotinic acid derivative and statins reduce Lp (a) levels[65].

Antiplatelet therapy:

Aspirin at low dose 75 – 150 mg/day inhibits production of TXA_2 (inducer of platelet aggregation) by acetylating serine residue near the active site of platelet COX1. Since platelets do not synthesize new proteins aspirin inhibition on platelets are permanent. Thus it blocks platelet aggregation. Aspirin thus reduces risk of cardiovascular events in high risk patients like CKD. Even at this low dose it can cause serious GI bleeds in these patients. Hence clopidogrel is used which is an irreversible ADP receptor antagonist and associated with less bleeding incidence[24].

Blood pressure control:

All 5 category of drugs (ACEI, ARBs, CCB, Beta blockers, thiazides) produced significant reduction in blood pressure from pretreatment values. For each initial blood pressure 10 mmHg higher the reduction was 1mmHg (systolic BP) and 1.1 mmHg (diastolic BP) greater. Also the

effects were additive. They are similarly effective in preventing CHD events and strokes but CCB had a greater preventive effect on strokes[61].

Treatment of CHF and IHD:

Loop diuretics are the main stay of treatment for achieving and maintaining euvolemia in cardiac failure. ACEI and ARB improve symptoms, morbidity and survival. ACEI are indicated in symptomatic heart failure, asymptomatic heart failure with LVEF < 35% and in post MI patients with EF <40%. Beta blockers improve prognosis in patients with systolic dysfunction. Due to association of risk, digoxin is not recommended as first line therapy. The risks are toxicity due to impaired clearance and arrhythmias in association with hypokalemia. Diastolic dysfunction are usually treated with verapamil or diltiazem as they enhance left ventricular diastolic relaxation.

CKD patients with stable angina without infarct are treated with standard anti anginal drugs for symptomatic relief. In patients with infarct beta blockers and ACEI are to be considered as they reduce mortality[81].

G. Nutritional management:

- Supplements – low potassium and phosphate with high calorie content

- Anti inflammatory and antioxidants - inflammation and oxidative stress are associated with poor outcomes in CKD. Statins decrease CRP levels irrespective of lipid levels. ACEI exhibit anti inflammatory properties. Acetylcysteine possesses anti oxidant property and thereby reduces cardiovascular adverse events. Glitazones inhibit activation of inflammatory response genes and promote immune deviation from Th1 to Th2 pathway.
- Correction of anorexia – low dose megestrol acetate by downregulating proinflammatory cytokines relieves symptoms of anorexia – cachexia syndrome. Pentoxifylline downregulates proinflammatory cytokine mediated NOS pathway, inhibits TNF α production and decreases weight loss and muscle protein wasting. Hence used in MICS[45].

Step 4: Renal replacement therapy (RRT):

Referral to nephrologist:

Patients with CKD should be referred to a nephrologist early in the course of the disease, before the plasma creatinine concentration exceeds 1.2 and 1.5 mg/dl in women and men respectively or the eGFR is less than 60ml/min/1.732m^2. Renoprotective therapy has greatest impact if initiated at this stage.

Indications:

RRT includes dialysis (peritoneal dialysis or hemodialysis) and renal transplantation. The main indications are

- When GFR falls to 10 ml/min/1.73m^2 without uremic symptoms
- Pericarditis or pleuritis
- Progressive uremic encephalopathy or neuropathy with signs such as confusion, asterixis, myoclonus, wrist or foot drop or seizures
- A clinically significant bleeding diathesis attributable to uremia
- Fluid overload refractory to diuretics
- Hypertension poorly responsive to antihypertensive medications
- Persistent metabolic disturbances that are refractory to medical therapy. These include hyperkalemia, metabolic acidosis, hypercalcemia, hypocalcemia, and hyperphosphatemia.
- Persistent nausea and vomiting
- Weight loss or signs of malnutrition[31]

Prognosis:

The five year survival rate on dialysis depends on underlying disease which is 36% for diabetic kidney disease, 53% for glomerulonephritis. But the overall survival rate is 39%. Life expectancy of patients on dialysis is 3 – 5 yrs. The most common cause of death in

CKD is cardiac diseases (> 50%). Other causes are infection, CVA and malignancy[42].

RECENT ADVANCES IN MANAGEMENT OF CKD:

L and T type calcium channel blockers:

T type calcium channels are found in both afferent and efferent arteriole. L/T type blockers dilate both the arteriole and attenuate glomerular hypertension and reduce proteinuria. They also suppress mesangial cell proliferation by inhibiting activator protein – 1. The drugs in this class are benidipine, manidipine and efonidipine[82].

ETA (Endothelin A) Antagonism:

ET-1 acts on the ETA located in vascular smooth muscle cells, myocardium, fibroblasts, kidney and platelet . The ETA receptor stimulation produces vasoconstriction, fluid retention, proliferative effects, cardiac hypertrophy and releases norepinephrine and Ang II . ETA receptor stimulation also stimulates the release of cytokines and growth factor and facilitates platelet aggregation. Endothelin receptor A antagonist has been shown to reduce systolic and diastolic BP and to offer renoprotection[83].

Bone Morphogenetic Protein – 7 (BMP – 7):

It is an endogenous antagonist of TGF β and thereby inhibits epithelial to mesenchymal transition involved in interstitial fibrosis.

Pirfenidone:

It is an orally active antifibrotic agent that confers renoprotection in CKD[61].

ACE – 2:

It facilitates degradation of angiotensin II to angiotensin (1-7) which is a vasodilatory and antiproliferative mediator. So recombinant ACE 2 is tried for renoprotection[39].

CERA (Continuous Erythropoiesis Receptor Activator):

It is synthesized by integration of epoetin beta with 30 kDa methoxy polyethylene glycol polymer. It has a very long t1/2 of 130 hrs and hence administered every 2-4 weeks.

Peginesatide:

It is a pegylated synthetic dimeric peptide that activates EPO – R but bears no sequence homology with endogenous erythropoietin and hence it is unrecognizable by anti EPO antibodies. T1/2 is 14-60 hrs and given as monthly dosing.

HIF Prolyl Hydroxylase Inhibitors (PHI):

It is orally active oxoglutarate analogs that inhibits prolyl hydroxylase degradation (PHD) proteins which are responsible for hydroxylation, ubiquitination and degradation of Hypoxia inducible factor

α. Thus it leads to stabilisation of HIF α and thereby promote EPO release[37].

ARTICLES RELATED TO AMLODIPINE:

1. In a randomized trial conducted by Yasuhiko et al between losartan and amlodipine for 12 months in patients with CKD and hypertension, both the drugs exerted the same efficacy for blood pressure control; but losartan significantly reduced 24hr urinary protein excretion at months 3, 6, 12(20.7%, 35.2%, 35.85% reduction) whereas amlodipine did not change the amount of proteinuria during the study period[84].

2. In another randomized trial by Praga M et al between losartan and amlodipine conducted in patients with non diabetic proteinuric renal diseases, proteinuria decreased by 32.4% after 4 weeks of treatment and by 50.4% after 20 weeks of treatment in losartan group whereas no significant proteinuria changes were observed in amlodipine group. Also target blood pressure was achieved with initial dose of study medication in 76% in losartan group (50mg daily) and 68% in amlodipine group (5mg daily)[85].

3. A randomized trial by Abe et al between amlodipine and benidipine in patients with stage 3 – 5 CKD as add on therapy to ARBs showed that there were no differences between systolic BP(A –

133.4±1.3mmHg , B – 136.1±1.8mmHg) and diastolic BP(A – 75.2±1.6, B – 78.6±1.7) at the end of 6 months. Heart rate did not differ between the groups (A – 74.6± 2.3, B – 76.1±2.2). Serum creatinine increased and eGFR decreased significantly in both the groups. Urine PCR was significantly lower in B group than in A group (B – 2565±299.9 vs A – 3178±372.2 mg/g Cr , $p<0.05$) at 6 months[86].

4. A prospective study conducted by Agodoa et al between ramipril and amlodipine in patients with hypertensive nephrosclerosis showed that the mean decline in GFR was 2.07 and 3.22 ml/min/1.73m^2 per year in ramipril and amlodipine group respectively. Thus the mean decline was 1.15 ml/min/1.73m^2 per year or 36% slower in ramipril group. Proteinuria increased by 58% from 0.0997 to 0.1575 in amlodipine group and declined by 20% from 0.1147 to 0.0915 in ramipril group. The risk reduction for ramipril vs amlodipine groups in the clinical end points (ESRD, death, 50% decline in GFR) was 38%[87].

5. In a randomized trial by Tanaka et al between nifedipine CR and amlodipine as add on therapy to valsartan in CKD patients showed that urine ACR at 16 weeks (23±24 mg/g Cr) was significantly less than that at 0 weeks (49±63) in VN group but not in VA group (39±39 at 0 weeks and 50±95 at 16 weeks). Serum creatinine was significantly greater

at 16 weeks(1.03±0.34mg/dl) in VA group compared to 0 weeks(0.92±0.27) but not in VN group (0 weeks – 0.9±0.1 and 16 weeks – 0.9±0.2)[88].

ARTICLES RELATED TO CILNIDIPINE:

1. In a prospective study conducted by Nakatsu et al in asymptomatic non – diabetic hypertensive patients who received cilnidipine monotherapy showed that mean ± SD for morning systolic BP and urine albumin creatinine ratio decreased by 20.4 ± 11.4mmHg and 15.2±13.1mg/g creatinine respectively. The time constant for UACR reduction was significantly longer than that for BP reduction (43.5±22.9 vs 15.4±7.1 days)[89].

2. In a study conducted by Kanaoka, Tamura et al with cilnidipine as add on therapy to renin angiotensin inhibitors for 24 weeks showed that left ventricular mass index was significantly decreased in cilnidipine group compared to control group (change in LVMI -12.4±23.7 vs 26.2±64.4; p=0.007). Also 24 hr and daytime systolic BP (from ambulatory BP monitoring) were significantly lower in cilnidipine group compared to control CCBs[90].

3. In a study where diabetic patients on other CCBs were switched over to cilnidipine, it was found that after substitution BP did not

significantly change, but heart rate decreased from 73.9±7.1 beats/min to 72±8.4 beats/min and the log transformed UACR decreased to 82.9±49.4% of baseline values[91].

4. In a comparative study between valsartan and valsartan plus cilnidipine in diabetic patients for 1 year it was found that UACR was markedly decreased in combination therapy group rather than monotherapy group (reduction rate 44±11% vs 9±7%)[92].

5. A prospective study between cilnidipine and benazepril showed that urine albumin excretion significantly decreased in both the groups with no significant difference between the groups. The levels of serum creatinine were unchanged throughout the study in both the groups[93].

AMLODIPINE VERSUS CILNIDIPINE:

1. In a study by Abe et al conducted in CKD patients who received either cilnidipine or amlodipine for 48 weeks, a significant and comparable reduction in systolic and diastolic BP was observed in both the groups. The percent reduction in urinary ACR and Liver type fatty acid binding protein (L – FABP) was significantly greater in cilnidipine group than amlodipine group. Plasma renin activity did not differ between the groups but plasma aldosterone levels was significantly decreased in cilnidipine group[94].

2. In a comparative study between amlodipine and cilnidipine for 12 months in renal disease patients , amlodipine showed significant increase in proteinuria (87% increase of baseline values) while the increase was suppressed in cilnidipine group(4% increase of baseline values). Also cilnidipine showed an increase in creatinine values (baseline vs 12 months 1.36±0.20 vs 1.5±0.23 mg/dl)[95].

3. In SAKURA trial conducted between L/N type and L type calcium channel blockers for 12 months showed that both the drugs equally decreased the BP. UACR for cilnidipine and amlodipine before treatment (111.5±138.97 and 88.29±63.45 mg/g) and after treatment (107.93±130.23 and 89.07±97.55) did not differ much. Thus there was similar changes in UACR, serum creatinine and eGFR. Cilnidipine effects were similar to amlodipine and did not offer additional benefit[96].

4. In a cross over study among 4 types calcium channel blockers in CKD patients showed that BP reduction was comparable among them. Baseline urinary albumin excretion was 69.4. UAE endpoints seen with the drugs were nifedipine CR 30.8, cilnidipine 33.9, efonidipine 51 and amlodipine 40.6. Angiotensin II was significantly lower in cilnidipine than amlodipine. Plasma aldosterone concentration was significantly lower in cilnidipine and efonidipine as compared to amlodipine[97].

Thus in many clinical studies it was found that cilnidipine had superior antiproteinuric effects and found to be more efficacious than amlodipine in renal diseases. It was also found that combination therapy of RAS inhibitors and CCBs were more effective in reducing proteinuria and BP levels than monotherapy with either of the drugs in CKD patients. With the above extensive literature review, this study was designed to compare the efficacy and tolerability of cilnidipine and amlodipine and to prove the advantages of cilnidipine over amlodipine as add on therapy to ARB (Losartan) in CKD patients.

AIM OF THE STUDY

To compare the efficacy and safety of cilnidipine and amlodipine as add on therapy in chronic kidney disease patients who are on losartan for > 2 months.

METHODOLOGY

STUDY TYPE:

Interventional clinical study

STUDY DESIGN:

Open label, randomized, prospective, comparative, parallel group study.

STUDY PERIOD:

April 2013 – May 2014

STUDY CENTRE:

It is a single centered study conducted in the out patient Department of Nephrology, Tirunelveli Medical College, Tirunelveli

SAMPLE SIZE:

100 (50 patients in cilnidipine group and 50 patients in amlodipine group).

ETHICAL CONSIDERATIONS:

This study was approved by Instituitional Ethical Committee of Tirunelveli medical college Hospital. Written informed consent was obtained in local vernacular language from every patient before enrollment.

INCLUSION CRITERIA:

- Patients of both sex
- Patients aged > 18 years and < 80 years
- Patients diagnosed as having CKD with type II diabetes mellitus with GFR < 90 ml/min/1.73m^2
- Patients should be on losartan 50 mg once daily for > 2 months before administration of cilnidipine or amlodipine
- Patients with systolic BP ≥130 and < 180 mmHg and diastolic BP ≥ 80 and < 110 mmHg
- Patients with urine protein creatinine ratio < 0.2 g/g creatinine (normal protein excretion) or ≥ 0.2 g/g creatinine (clinical proteinuria).

EXCLUSION CRITERIA:

- Patients less ≤ 18 or ≥ 80 years of age
- Hypertensive emergency (systolic BP ≥ 180 mmHg ; diastolic BP ≥ 110 mmHg.)
- Patients not taking losartan
- Earlier treatment with steroids or immunosuppressants
- Renovascular hypertension
- Patients on renal replacement therapy (dialysis and transplant recipient patients)

- ❖ Patients with uncontrolled diabetes mellitus or complications of diabetes such as DKA that required hospitalization
- ❖ Patients with the history of hypersensitivity to calcium channel blockers
- ❖ Patients with severe heart failure , arrythmia, angina and myocardial infarction within 6 months before starting the study
- ❖ Patients with the history of convulsions
- ❖ Patients with stroke, hepatic impairment within 6 months before starting the study.

WITHDRAWAL CRITERIA:

- ❖ Blood pressure ≥ 180/110
- ❖ Noncompliance with protocol
- ❖ Protocol deviation
- ❖ Request for withdrawal by the subject
- ❖ Adverse effects (decision about withdrawal from the study was made either by subject or investigator)

SCHEDULE OF STUDY VISIT:

a) Screening and recruitment:

The subjects were enrolled based on inclusion criteria after screening. During enrollment clinical assessment and the following baseline investigations were done.

- Blood sugar, serum creatinine, potassium and hematocrit were done in a random blood sample using automated analyser.
- Estimated glomerular filtration rate was calculated by Modification of diet in renal disease (MDRD) study equation as follows

$$\text{Estimated GFR} = 1.86 \times (P_{cr})^{-1.154} \times (\text{age})^{-0.203}$$

(Multiply by 0.742 for women)

- Spot urine protein creatinine ratio (the ratio of spot urine protein to creatinine expressed as g/g cr) was measured by automated assay. The urine samples were collected at anytime in the morning. Values represents the mean of 2 measurements done in 1 week period.

Urine PCR < 0.2 – NORMAL

Urine PCR ≥ 0.2 – CLINICAL PROTEINURIA

- Blood pressure was measured manually after 5 minutes of rest twice atleast 2 mins apart in right arm in sitting posture with the cuff at heart level using sphygmomanometer.
- Pulse rate was assessed by palpating the radial artery on radial side of the wrist with tips of index , middle and ring fingers after 5 minutes of rest.
- Categorizing the stages of CKD based on GFR levels

Stage 1 = ≥ 90 ml/min/1.73m^2

Stage 2 = 89 – 60 ml/min/1.73m^2

Stage 3 = 59 – 30 ml/min/1.73m^2

Stage 4 = 29 – 15 ml/min/1.73m^2

Stage 5 = < 15 ml/min/1.73m^2

b) Randomisation:

After enrollment subjects were randomized into 2 groups (group 1 and group 2) with the help of computer generated random table.

c) Treatment protocol:

The patients received the drugs as follows

Group 1: T. Cilnidipine 10 – 20 mg/day for 6 months

Group 2: T. Amlodipine 5 – 10 mg/day for 6 months.

After randomization group 1 patients were started on T.Cilnidipine 10 mg once daily and group 2 patients were given T. Amlodipine 5 mg once daily initially. The patients were reviewed every 2 weeks. If the magnitude of reduction in blood pressure was insufficient (systolic BP < 20mm Hg and diastolic BP < 10mm Hg) the dosage was increased to 10mg twice daily and 5mg twice daily for cilnidipine and amlodipine respectively. Both the drugs were given orally either once or twice daily for a duration of 6 months for each patient. Also the patients were given a diary to note down the adverse events. The tablets were provided for

15 days only. Then the patients were instructed to report to the out patient department after 2 weeks along with the diary and empty strips to collect the drugs. During this visit blood pressure was monitored, compliance was assessed by pill count method and adverse effects if any were recorded. Cilnidipine Tablets (Cilacar) 10mg were donated by J.B.Chemicals and Pharmaceuticals.

C) Follow up :

At the end of 1st , 3rd and 6th month, clinical examination including vital signs such as blood pressure, pulse rate and laboratory investigations such as blood sugar, serum creatinine, potassium, spot urine protein creatinine ratio were performed. Also estimated GFR was calculated and categorization of stages of CKD based on GFR was done.

EFFICACY PARAMETERS:

PRIMARY ENDPOINT:

- Changes in spot urine protein creatinine ratio from the baseline to the endpoint (at the end of 6 months).

SECONDARY ENDPOINT:

- Changes in blood pressure from the baseline to the endpoint
- Changes in serum creatinine from the baseline to the endpoint
- Changes in estimated GFR from the baseline to the endpoint

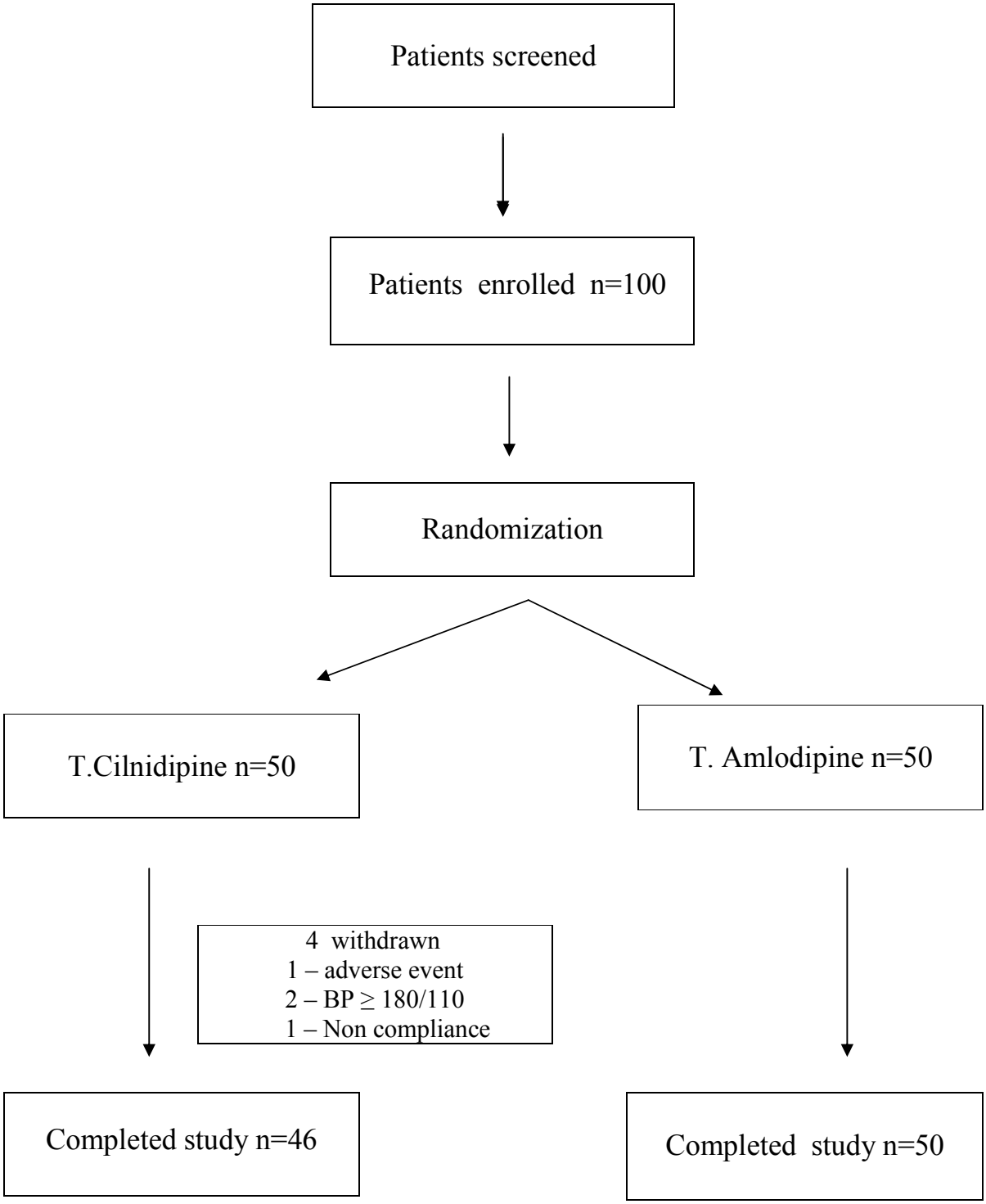
- Changes in pulse rate from the baseline to the endpoint
- Progression/Regression of CKD stages

STATISTICAL ANALYSIS:

1. The baseline characteristics of both the groups were expressed as descriptive statistics (mean, standard deviation). They were matched by unpaired student 't' test and Pearson's chi-square test.
2. For analysis and interpretation of variables within the group, student paired 't' test and wilcoxon signed rank test were used for normal and non normal data respectively.
3. For analysis of variables between the groups, unpaired 't' test and mann whitney u test were carried out for normal and non normal data respectively.
4. The categorical variables between the two groups were compared by chi square test of proportions.
5. Adverse events were expressed as percentages.

The above statistical analysis was done with the help of statistical package namely IBM SPSS Statistics 11 by adopting the following test of significance. The p value less than 0.05 was considered as significant in two tailed conditions.

PATIENT DISPOSITION: CONSORT DIAGRAM



RESULTS

In the period of 1 year from April 2013 to May 2014, 100 cases of diabetic chronic kidney disease attending the outpatient Department of Nephrology with eligibility criteria were included in the study. The patients were then randomly assigned into 2 groups receiving either T. Cilnidipine or T. Amlodipine . 4 patients in cilnidipine group were withdrawn (1 patient due to adverse event; 2 patients due to blood pressure elevation; 1 patient due to non compliance). In amlodipine group no one was withdrawn. Totally 96 patients completed the study and the results were statistically analysed.

TABLE 1: BASELINE CHARACTERISTICS

Baseline parameters		Cilnidipine (mean) (n – 46)	Amlodipine (mean) (n – 50)	p value
Age (SD)		59.19 (8.10)	55.16 (8.50)	0.019
Sex	Male	33	33	0.544
	Female	13	17	
Urine PCR		1.94	1.38	0.014
Serum creatinine		3.13	2.75	0.158
eGFR		25.28	29.6	0.142
Systolic BP		149.13	148.40	0.779
Diastolic BP		92.17	90.8	0.461
Pulse rate		79.7	78.82	0.525

Table 1: summarizes the baseline characteristics of patients enrolled in the study. There were no statistical difference in the baseline parameters between cilnidipine and amlodipine group except for age (p - 0.01) and urine protein creatinine ratio which was high in cilnidipine group (p - 0.014).

TABLE 2

CHANGES IN MEAN PARAMETERS FROM BASELINE AT THE END OF 1ST, 3RD AND 6TH MONTH IN CILNIDIPINE GROUP

Parameter	Baseline (mean± SD)	1 month (mean± SD)	P value	3 months (mean± SD)	P Value	6 months (mean± SD)	P Value
n	46	46		46		46	
Urine PCR (g/g cr)	1.94±1.22	1.43±0.92	< 0.001*	1.12±0.75	< 0.001*	1.09±0.72	< 0.001*
Serum creatinine (mg/dl)	3.13±1.34	3.16±1.55	0.73	3.22±1.46	0.22	3.25±1.44	0.045*
eGFR (ml/min/1.73 m^2)	25.2±14.2	26±14.3	0.68	24.6±13.3	0.41	24.4±13.1	0.28
Systolic BP (mmHg)	149.13± 12.7	127.6± 14.3	< 0.001*	126.52± 13.6	< 0.001*	126.73± 12.1	< 0.001*
Diastolic BP (mmHg)	92.17±8.4	80.65± 10.2	< 0.001*	78.91±11	< 0.001*	79.35±7.7	< 0.001*

*p value < 0.05 statistically significant

- Table 2 : shows changes in mean parameters from baseline at the end of 1, 3 and 6 months in cilnidipine group .The results are expressed as mean ± SD.
- Change in UPCR from baseline was significantly lower in cilnidipine group at the end of 1st month itself. Thereafter PCR progressively regressed throughout the study period ($p<0.001$).
- The increase in creatinine levels did not change statistically at the end of 3 months (baseline vs 3 months: 3.13±1.34 vs 3.22±1.46; $p=0.22$) but it was statistically significant at the end of 6 months (3.13±1.34 vs 3.25±1.44; $p=0.04$).
- Estimated GFR increased at the end of 1st month of treatment. Thereafter GFR decreased gradually. Both the increase and decrease in eGFR from baseline was statistically insignificant. ($p = 0.68$; $p = 0.41$; $p = 0.28$).
- Both systolic and diastolic BP decreased significantly from baseline at the end of 1, 3 and 6 months ($p<0.001$).

TABLE 3

CHANGES IN MEAN PARAMETERS FROM BASELINE AT THE END OF 1ST, 3RD AND 6TH MONTH IN AMLODIPINE GROUP

Parameter	Baseline (mean± SD)	1 month (mean± SD)	P value	3 months (mean± SD)	P Value	6 months (mean± SD)	P Value
N	50	50		50		50	
Urine PCR (g/g cr)	1.38±0.98	1.41±0.83	0.47	1.30±0.71	0.83	1.40±0.65	0.21
Serum creatinine (mg/dl)	2.75±1.28	2.83±1.33	0.76	2.84±1.25	0.15	2.87±1.28	0.02*
eGFR (ml/min/1.73 m^2)	29.6±14.9	28.9±14.6	0.84	27.8±14	0.1	27.6±13.9	0.1
Systolic BP (mmHg)	148.4± 12.67	125.2± 13.5	< 0.001*	124.8± 11.1	< 0.001*	122.6± 14.5	< 0.001*
Diastolic BP (mmHg)	90.8±9.6	76.4±9.8	< 0.001*	78±7.8	< 0.001*	77.4±7.7	< 0.001*

*p value < 0.05 statistically significant

- Table 3: shows changes in mean parameters from baseline at the end of 1 , 3 and 6 months in amlodipine group. The results are expressed as mean ± SD.
- In amlodipine group there was no change in urinary PCR from baseline at the end of 1, 3 and 6 months statistically. (p=0.47; p=0.83; p=0.21).
- The mean serum creatinine concentration increased gradually and attained statistical significance at the end of 6 months (baseline vs 6 months: 2.75±1.28 vs 2.87±1.28; p=0.02).
- The estimated GFR decreased progressively but it was statistically insignificant with respect to baseline at the end of 1, 3 and 6 months

  (p = 0.84; p = 0.1; p = 0.1).
- Both systolic and diastolic BP decreased significantly from baseline at the end of 1, 3 and 6 months. ($p<0.001$).

TABLE 4

COMPARISON OF ANTIPROTEINURIC EFFECTS OF CILNIDIPINE AND AMLODIPINE

Duration	Groups	Mean difference of UPCR from baseline	SD	P value
1 month	Group 1	-0.50	0.63	< 0.001*
	Group 2	0.03	0.46	
3 month	Group 1	-0.83	0.88	< 0.001*
	Group 2	-0.08	0.56	
6 month	Group 1	-0.87	0.66	< 0.001*
	Group 2	0.01	0.6	

*p value < 0.05 statistically significant

- Table 4 : The mean difference of urine protein creatinine ratio from baseline at the end of 1, 3 and 6 months in both the groups were analysed and interpreted in the above table. The results are expressed as mean difference ± SD.

- In cilnidipine group urinary PCR decreased steadily from baseline during the study period. Amlodipine group showed a decrease at the end of 3 months but the decrease was not sustained thereafter. At the end of 6 months, in amlodipine group urine PCR returned to baseline values. Thus after 6 months of treatment urine protein creatinine ratio had decreased in cilnidipine group but not in amlodipine group. (-0.87±0.66 vs 0.01±0.6 ; p<0.001)

Figure 1

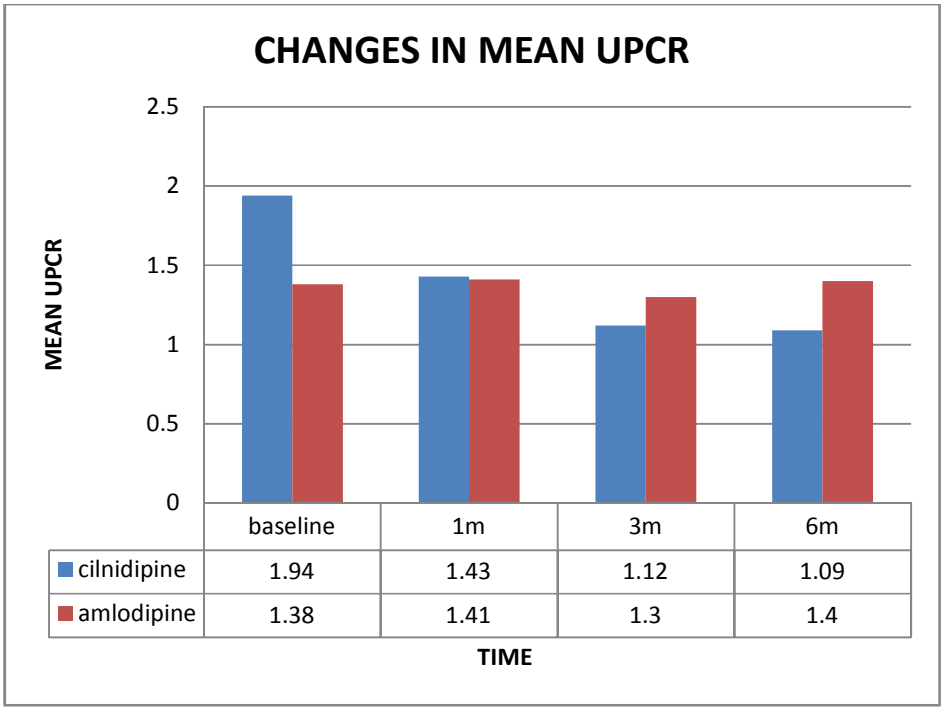


Figure.1: shows the pictorial representation of changes in mean urine protein creatinine ratio at the end of 1st, 3rd and 6th month in both the groups.

TABLE 5

PERCENTAGE REDUCTION OF URINE PCR IN PATIENTS WITH HIGH GRADE BASELINE PROTEINURIA

Duration	Cilnidipine	Amlodipine
3 months	45%	24%
6 months	46%	20%

- Baseline proteinuria levels were categorized into 2 groups
  a) High grade proteinuria – UPCR ≥ 1.5
  b) Low grade proteinuria – UPCR < 1.5
- Changes in urine protein creatinine ratio from baseline with respect to the above groups were calculated as percentage reduction and analysed at the end of 3 and 6 months.
- In patients with high grade proteinuria, cilnidipine group showed a reduction of 45% and 47% of urine PCR from baseline whereas amlodipine group showed 24% and 20% reduction at the end of 3 and 6 months respectively. The percentage reduction was high in cilnidipine group.
- In patients with low grade proteinuria, cilnidipine group showed a

decrease in urine PCR from baseline by 31% and 41% whereas amlodipine increased the urine PCR by 17% and 27% at the end of 3 and 6 months respectively.

TABLE 6

COMPARISON OF RENAL FUNCTION CHANGES BETWEEN CILNIDIPINE AND AMLODIPINE FROM BASELINE

Duration	Group	Serum creatinine			Estimated GFR		
		Mean difference from baseline	SD	P value	Mean Difference from baseline	SD	P value
1 month	Group 1	0.02	0.56	0.59	0.72	7.73	0.7
	Group 2	0.07	0.35		-0.64	6.8	
3 month	Group 1	0.08	0.47	0.98	-0.61	5.82	0.66
	Group 2	0.09	0.43		-1.72	7.29	
6 month	Group 1	0.12	0.39	0.98	-0.87	7.01	0.65
	Group 2	0.12	0.36		-1.92	6.71	

- Table 6: compares the renal function changes from baseline in both the groups at the end of 1, 3 and 6 months. The results are expressed as mean difference ± SD.

- The mean serum creatinine concentration increased from baseline in both the groups. The difference in increase from baseline were equal in the two groups at the end of 6 months (p=0.98).
- The estimated GFR gradually decreased in both the groups and the difference in decrease between the groups was statistically insignificant at the end of 1, 3 and 6 months (p = 0.7; p = 0.66; p = 0.65)

Figure 2

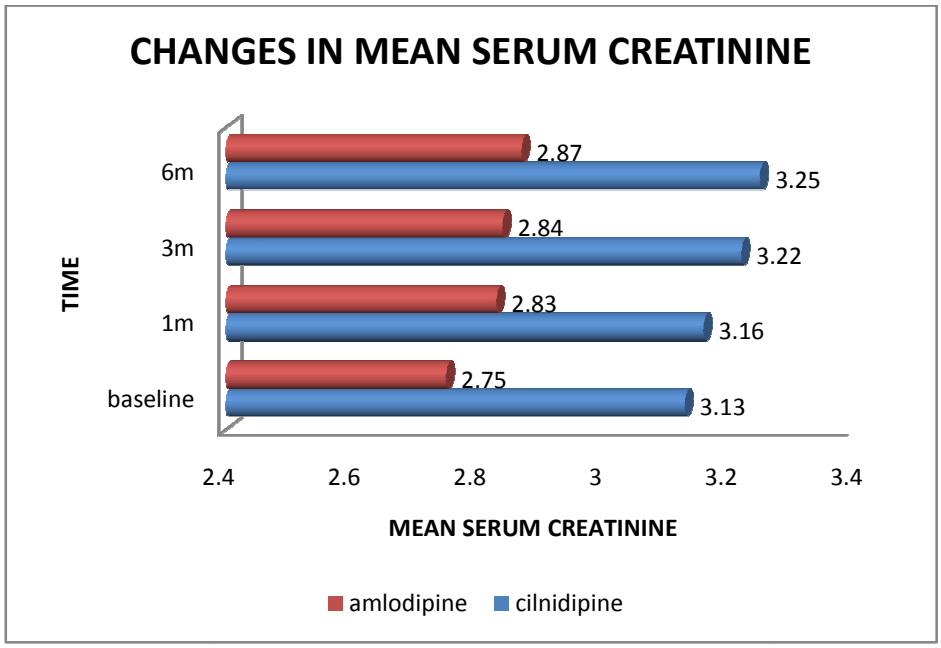


Figure 2: is a pictorial representation of changes in mean serum creatinine in both the groups at various intervals.

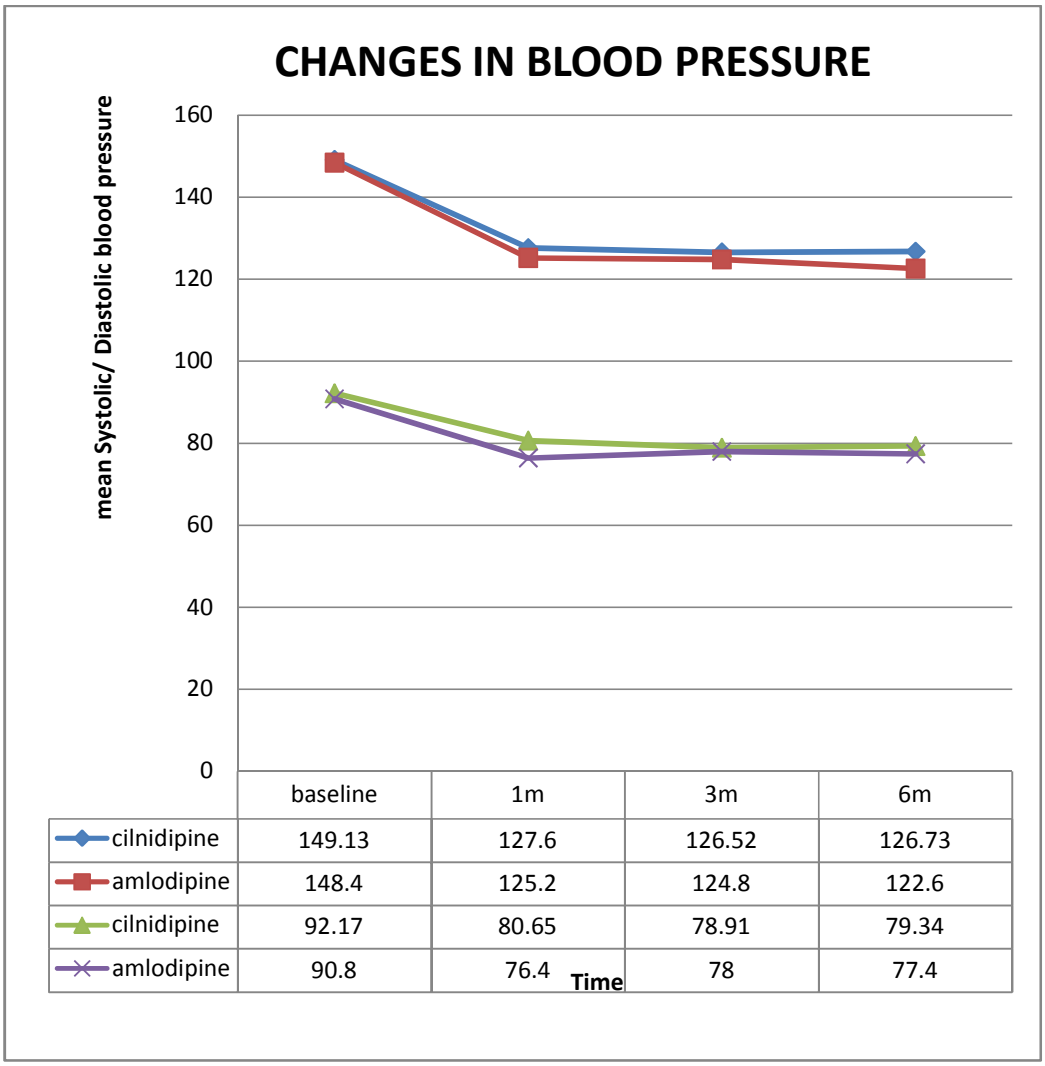
TABLE 7

COMPARISON OF BLOOD PRESSURE CHANGES BETWEEN 2 GROUPS FROM BASELINE

Duration	Groups	Systolic blood pressure			Diastolic blood pressure		
		Mean difference from baseline	SD	P value	Mean difference from baseline	SD	P value
1 month	Group 1	-21.5	9.88	0.39	-11.52	11.9	0.27
	Group 2	-23.2	9.57		-14.2	12.13	
3 month	Group 1	-22.6	14.21	0.69	-13.26	12.6	0.84
	Group 2	-23.6	10.25		-12.80	10.3	
6 month	Group 1	-22.39	11.38	0.21	-12.82	10.6	0.79
	Group 2	-25.4	12.32		-13.40	10.4	

- Table 7: compares the blood pressure changes at the end of 1 , 3 and 6 months in both the groups. The results are expressed as mean difference ± SD.
- The difference in systolic and diastolic BP from baseline was insignificant in both the groups at the end of 1, 3 and 6 months.

Figure 3



	baseline	1m	3m	6m
cilnidipine	149.13	127.6	126.52	126.73
amlodipine	148.4	125.2	124.8	122.6
cilnidipine	92.17	80.65	78.91	79.34
amlodipine	90.8	76.4	78	77.4

Figure 3: is a pictorial representation of trends in systolic and diastolic blood pressure changes in both the groups.

TABLE 8

CHANGES IN CKD STAGES AT THE END OF 6 MONTHS IN THE TWO GROUPS

Study group	Changes in CKD stages			P value
	Unchanged	Regression	Progression	
GROUP 1	40	3	3	0.19
GROUP 2	36	6	8	

Progression is defined as worsening by one stage and regression is improvement by one stage. The CKD stage was unchanged in 40 patients, advanced in 3 patients and regressed in 3 patients after treatment with cilnidipine and was unchanged in 36 patients, advanced in 8 patients and regressed in 6 patients in amlodipine treated group. Chi square test was done and it was found to be statistically insignificant (p=0.19).

ADVERSE EFFECTS:

Both the drugs were well tolerated. 1 patient was withdrawn from cilnidipine group at the start of the study due to non specific chest pain after 2 doses. ECG was normal in that patient. Out of the 46 patients who completed the study in cilnidipine group, 6 patients (13%) developed adverse effects. In amlodipine group all patients completed the study and no one was withdrawn due to adverse event. 4 patients (8%) out of 50 in amlodipine group developed adverse effects. All these adverse effects were mild and did not require discontinuation of drugs. There was no significant differences in adverse events between the groups.

Adverse effects	Cilnidipine	Amlodipine
Dizziness	1	2
Peripheral edema	0	2
Increased appetite	1	0
Increased blood sugar	2	0
Itching	1	0
Gastritis	1	0

DISCUSSION

Chronic kidney disease is a major risk factor for cardiovascular disease. In hypertensive patients with concomitant CKD, strict antihypertensive therapy is recommended. Antihypertensive therapy are primarily aimed at reducing proteinuria and blood pressure. The first line drugs in CKD are RAS inhibitors but most of the patients require multiple drugs to achieve blood pressure targets. In such cases, CCBs are more commonly used along with RAS inhibitors as they possess potent hypotensive effects[6]. Several clinical trials have also suggested that treatment with CCBs and ARB/ACEI combination was more effective in preventing cardiovascular events. Hence the present study was performed to compare the combined effects of cilnidipine or amlodipine with losartan in patients with chronic kidney disease.

Many clinical studies have demonstrated that proteinuria is an important predictor of subsequent progression of kidney disease. Proteinuria is also directly associated with the risk of development of CAD. Thus reduction in proteinuria protects against renal and cardiovascular failure. Recently, with regard to renal and cardiovascular outcomes in chronic kidney disease patients, antiproteinuric antihypertensive drugs are considered to be more beneficial[98].

Proteinuria was assessed by urine protein creatinine ratio in our study. Random spot urine samples were used to determine UPCR. In a study conducted by Gai et al, it was demonstrated that urine protein creatinine ratio in spot urine sample correlated well with daily urine protein excretion[99]. Either a first morning or a random spot urine sample can be obtained, as both are recommended by Kidney disease outcomes quality initiative of National kidney foundation[98].

In our study, we included patients with clinically proven diabetic chronic kidney disease on treatment with losartan. Baseline characteristics were comparable between cilnidipine and amlodipine group except for age (p - 0.01) and urine protein creatinine ratio which was high in cilnidipine group (p - 0.014). The mean age of the patients was 57.17 years. Majority of the patients were male and the distribution of males and females were similar between the groups.

In the analysis of primary endpoint, cilnidipine treated patients showed a steady decrease in urine protein creatinine ratio whereas in amlodipine group UPCR decreased at the end of 3 months but returned to baseline at the end of 6 months. The greater antiproteinuric effects of cilnidipine can be attributed to reduction in glomerular pressure caused by vasodilation of efferent arteriole due to blockade of N – Type

calcium channel. Amlodipine being a predominant L type CCB, does not have any effect on efferent arteriole due to absence of L type calcium channels and therefore glomerular pressure is not reduced[98]. These results were consistent with several other studies which reported that cilnidipine exhibited antiproteinuric effect greater than amlodipine[(6,95)].

In patients with high grade baseline proteinuria, urine PCR was reduced in both the groups after 3 and 6 months whereas in patients with low level proteinuria, cilnidipine group showed a reduction but in amlodipine group there was no change in UPCR levels. Thus high grade proteinuric patients responded better to both modalities of treatment. Based on antiproteinuric effects, it was found that cilnidipine and losartan had an excellent additive effects at all ranges of proteinuria whereas amlodipine and losartan exhibited additive effects only in patients with urine PCR greater than or equal to 1.5 (urine PCR $\geq$ 1.5).Thus it can be translated that cilnidipine may reduce cardiovascular events in CKD patients even with low grade proteinuria.

On analyzing the results of secondary endpoints, the serum creatinine increased in both the groups gradually. The difference in increase from baseline was equal in both the groups (p=0.98). Estimated GFR was decreased from baseline in both the groups but the decrease

was not statistically significant. Both cilnidipine and amlodipine maintained the renal function throughout the study period. Thus by maintaining GFR both the drugs are expected to improve long term prognosis in CKD patients.

Strict BP control is another important parameter to prevent the progression of renal disease. Lowering of BP is also associated with significant fall in cardiovascular event. Both systolic and diastolic BP decreased significantly from baseline in both the groups at the end of 1, 3 and 6 months. The difference in blood pressure reduction was equal in both the groups. Subjects with BP less than 130/80 accounted for 19% and 34% in cilnidipine and amlodipine groups respectively. Thus both the drugs were equally efficacious in decreasing the BP and maintaining the reduction. But the percentage of patients who achieved the target BP goal was high in amlodipine group. As systemic blood pressure is directly transmitted to glomerular capillaries, amlodipine by causing a greater reduction in systemic blood pressure is expected to decrease glomerular pressure which might be attributed to its antiproteinuric effects seen in some patients in our study.

Many studies have shown that higher heart rate is associated with long term risk of cardiovascular mortality independent of other risk

factors[100]. In our study pulse rate was maintained by both the drugs. None of them produced reflex tachycardia. This result was similar to CARTER study in which heart rate was unaffected by both the drugs[98.]

In majority of patients, CKD stages were unaltered by both the drugs. Nearly 6 patients (12%) improved in amlodipine group. Worsening of patients was less in cilnidipine compared to amlodipine group. But all these changes were statistically insignificant. Thus distribution of CKD stages were similar between the two groups before and after treatment (p=0.19).

Both the drugs were well tolerated. In cilnidipine group 1 patient developed chest pain after 2 doses and was withdrawn from the study. ECG was normal in that patient. Other adverse events reported were mild and did not require discontinuation of the drug. In amlodipine group , only a few adverse events were reported and no patients were withdrawn due to these adverse effects. No safety concerns were raised in this study.

There were certain limitations in our study. First, since baseline urinary protein excretion showed a significant difference between the two groups the rate of change in urine protein creatinine by cilnidipine might be overestimated. Second, 24 hour urine collection is the gold standard test for assessment of urine protein excretion. But it might be difficult for

the outpatients to give their co – operation and hence spot urine samples were used. Though spot urine sample may be a better alternative in clinical practice, still it carries some limitations . Third, it was an open label study and patients were followed up for short duration only. Hence long term renoprotective effects were not determined. Fourth, relationship between reduction in proteinuria and cardiovascular events were not assessed.

From the above results it was found that cilnidipine was equally efficacious to amlodipine with respect to blood pressure reduction and maintanence of renal function but based on antiproteinuric effects it was found to be more beneficial than amlodipine in diabetic chronic kidney disease patients who do not achieve desired treatment targets with ARBs alone.

CONCLUSION

In conclusion, cilnidipine has antihypertensive effect equivalent to amlodipine but addition of cilnidipine rather than amlodipine to losartan decreased urine protein excretion in diabetic chronic kidney disease patients. Therefore combination therapy with cilnidipine and RAS inhibitor may be more beneficial and renoprotective in patients with diabetic chronic kidney disease.

APPENDIX - I

ABBREVIATIONS

ADMA	:	Asymmetric Dimethyl Arginine
ACEI	:	Angiotensin Converting Enzyme Inhibitor
ARB	:	Angiotensin Receptor Blocker
ACE – 2	:	Angiotensin Converting Enzyme 2
ADH	:	Anti Diuretic Hormone
ALP	:	Alkaline Phosphatase
ABD	:	Adynamic Bone Disease
α SMA	:	Alpha Smooth Muscle Actin
BNP	:	Brain Natriuretic Peptide
BMP – 7	:	Bone Morphogenetic Protein 7
CKD	:	Chronic Kidney Disease
CRF	:	Chronic Renal Failure
CAD	:	Coronary Artery Disease
CVD	:	Cerebrovascular Disease
CVA	:	Cerebrovascular Accident
CHD	:	Coronary Heart Disease
CHF	:	Congestive Heart Failure
CCB	:	Calcium Channel Blocker

CERA	:	Continuous Erythropoiesis Receptor Activator
CRP	:	C – Reactive Protein
CR	:	Continuous Release
COX - 1	:	Cyclo – Oxygenase – 1
DKA	:	Diabetic Keto Acidosis
DRI	:	Direct Renin Inhibitors
eGFR	:	Estimated Glomerular Filtration Rate
ESRD	:	End Stage Renal Disease
EPO	:	Erythropoietin
EPOR	:	Erythropoietin Receptor
ET – A	:	Endothelin A
ET – B	:	Endothelin B
ESA	:	Erythropoiesis Stimulating Agents
eNOS	:	Endothelial Nitric Oxide Synthase
ECM	:	Extracellular Matrix
ECFV	:	Extracellular Fluid Volume
FGF – 23	:	Fibroblast Growth Factor
GBM	:	Glomerular Basement Membrane
GH	:	Growth Hormone
HIF α	:	Hypoxia Inducible Factor Alpha

HDL	:	High Density Lipoprotein
IGF	:	Insulin Like Growth Factor
LVH	:	Left Ventricular Hypertrophy
LVEF	:	Left Ventricular Ejection Fraction
LVMI	:	Left Ventricular Mass Index
LDL	:	Low Density Lipoprotein
Lp(a)	:	Lipoprotein (a)
LH	:	Leutenising Hormone
MS	:	Metabolic Syndrome
MICS	:	Malnutrition Inflammation Complex Syndrome
MGP	:	Matrix Gla Protein
MDRD	:	Modification of Diet in Renal Disease
MCP – 1	:	Monocyte Chemoattractant Protein
NSAID	:	Non Steroidal Anti Inflammatory Drug
PGE_2	:	Prostaglandin E_2
PCT	:	Proximal Convoluted Tubule
PTH	:	Parathyroid Hormone
PTHR	:	Parathyroid Hormone Receptor
PLMS	:	Periodic Limb Movements of Sleep
RAS	:	Renin Angiotensin System

RAAS	:	Renin Angiotensin Aldosterone System
rHuEPO	:	Recombinant Human Erythropoietin
RRT	:	Renal Replacement Therapy
TXA2	:	Thromboxane A2
TNF α	:	Tumour Necrosis Factor α
TG	:	Triglycerides
UPCR	:	Urine Protein Creatinine Ratio
UAE	:	Urine Albumin Excretion
UACR	:	Urine Albumin Creatinine Ratio
VEGF	:	Vascular Endothelial Growth Factor
VDR	:	Vitamin D Receptor
VLDL	:	Very Low Density Lipoprotein

INFORMED CONSENT FORM

Study Title : Comparison between anti proteinuric effects of cilnidipine & amlodipine as add on therapy in hypertensive patients with chronic kidney disease.

StudyNumber_____
Subject's Full Name _____
Date of Birth/Age_____
Address
_____
_____

1. I confirm that I have read and understood the information sheet dated for the above study and have had the opportunity to ask questions. **or** I have been explained the nature of the study by the Investigator and had the opportunity to ask questions
2. I understand that my participation in the study is voluntary and that I am free to withdraw at anytime, without giving any reason and without my medical care or legal rights being affected.
3. I understand that the sponsor of the clinical trial/project, others working on the Sponsor's behalf,the Ethics Committee and the regulatory authorities will not need my permission to look at my health records both in respect of the current study and any further research that may be conducted in relation to it, even if I withdraw from the trial. However, I understand that my Identity will not be revealed in any information released to third parties or published.
4. I agree not to restrict the use of any data or results that arise from this study provided such a use isonly for scientific purpose(s)
5. I agree to take part in the above study

Signature (or Thumb impression) of the Subject/Legally Acceptable Representative:

_____
Signatory's Name _____
Date _____
Signature of the Investigator _____
Date _____
Study Investigator's Name _____
Signature of the Witness _____ Date

_____
Name of the Witness

நோயாளிகளுக்கு அறிவிப்பு மற்றும் ஒப்புதல் படிவம்

மருத்துவ ஆய்வில் பங்கேற்பதற்கு

ஆய்வு செய்யப்படும் தலைப்பு **: சர்க்கரை மற்றும் உயர் ரத்த அழுத்தத்தினால் உண்டாகும் நாட்பட்ட சிறுநீரக பாதிப்புகளுக்கு சில்னிடிப்பின் அல்லது ஆம்லோடிப்பின் அளித்து ஓர் ஒப்புயர்வு ஆய்வு**

பங்கு பெறுபவரின் பெயர் **:**

பங்கு பெறுபவரின் வயது **:**

		பங்கு பெறுபவர் இதனை குறிக்கவும்
1.	நான் மேலே குறிப்பிட்டுள்ள மருத்து ஆய்வின் விவரங்களை நான் படித்து புரிந்து கொண்டேன். என்னுடைய சந்தேகங்களை கேட்கவும், அதற்கான தகுந்த விளக்கங்களை பெறவும் வாய்ப்பளிக்கப்பட்டுள்ளது என அறிந்து கொண்டேன்.	☐
2.	நான் இவ்வாய்வில் தன்னிச்சையாக தான் பங்கேற்கிறேன். எந்த காரணத்தினாலோ எந்த கட்டத்திலும், எந்த சட்ட சிக்கலும் உட்படாமல் நான் இவ்வாய்வில் இருந்து விலகி கொள்ளலாம் என்றும் அறிந்து கொண்டேன்.	☐
3.	இந்த ஆய்வு சம்பந்தமாகவோ, இதைச் சார்ந்து மேலும் ஆய்வு மேற்கொள்ளும் போதும் இந்த ஆய்வில் பங்கு பெறும் மருத்துவர் என்னுடைய மருத்துவ அறிக்கையை பார்ப்பதற்கு கொள்கிறேன். நான் ஆய்வில் இருந்து விலகிக் கொண்டாலும் இது பொருந்தும் என அறிகிறேன்.	☐
4.	இந்த ஆய்வின் மூலம் கிடைக்கும் தகவலையோ, முடிவையோ பயன்படுத்திக் கொள்ள மறுக்க மாட்டேன்.	☐
5.	இந்த ஆய்வில் பங்கு கொள்ள ஒப்புக் கொள்கிறேன். எனக்கு கொடுக்கப்பட்ட அறிவுரைகளின் படி நடந்து கொள்வதுடன் ஆய்வை மேற்கொள்ளும் மருந்து அணிக்கு உண்மையுடன் இருப்பேன் என உறுதியளிக்கிறேன். என் உடல் நலம் பாதிக்கப்பட்டாலோ, அல்லது எதிர்பாராக வழக்கத்திற்கு மாறான நோய்குறி தென்பட்டாலோ உடனே இதை மருத்துவ அணியிடம் தெரிவிப்பேன் என உறுதி அளிக்கிறேன்.	☐

பங்கேற்பவரின் கையொப்பம்/ _____இடம் _____ தேதி _____

பங்கேற்பவரின் பெயர் மற்றும் விலாசம்

ஆய்வாளரின் கையொப்பம்/ _____இடம் _____ தேதி _____

மையம் _____

கல்வியறிவு இல்லாதவர்க்கு (கைரேகை வைத்தவர்களுக்கு)இது அவசியம் தேவை

சாட்சியின் கையொப்பம்/ _____இடம் _____ தேதி _____

பெயர் மற்றும் விலாசம் _____

APPENDIX – III

PROFORMA

Group: OP. NO:

Random Number:

Patient's name: Date:

Age/Sex:

Address: Phone no:

Diagnosis:

Concomitant medications:

BP:

PR:

Investigations:

Complete blood count

Random blood sugar

Blood urea

Serum creatinine

Serum electrolytes

Routine urine analysis

Spot urine protein creatinine ratio

ASSESSMENT

PARAMETERS	BASELINE	1 MONTH	3 MONTHS	6 MONTHS
Spot urine protein creatinine ratio				
Serum creatinine				
Estimated GFR				
CKD stage				
Systolic blood pressure				
Diastolic blood pressure				
Pulse rate				
Dose of the drug				

ADVERSE EFFECTS (If any):

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