CHAPTER 1. INTRODUCTION

There has been drastic change in lifestyle and food habits in last decades, especially in the developing countries like India. This has also resulted in changes in status of health and disease. Prevalence of metabolic disorders like diabetes mellitus and hypertension has risen to epidemic levels. India is about to become diabetes hub of the world (Snehalatha and Ramachnadaran, 2009). Against the voluminous increase in prevalence of diabetes there has been little progress in drugs used in management of diabetes mellitus (DM). Most drugs used in treatment of DM address the issue of hyperglycemia. However, the recent findings have shown that, many factors other than carbohydrate metabolism play important role in pathogenesis of DM (Permutt, 2005). Factors like chronic inflammation, activation of immune system, oxidative stress, derangement of protein and lipid metabolism all play important roles in disease progression and development of diabetic complications (Navale and Paranjape, 2013). Several studies have suggested that presently used antihyperglycemic agents do not sufficiently give protection against target organ damage caused by DM. As a result, DM in long term culminates in micro and macrovascular complications like, neuropathy, nephropathy, retinopathy, cardiomyopathy and coronary artery disease.

Diabetic neuropathy (DN) is the most common of all complications of DM, affecting as many as 50% of patients with type 1 (T1DM) and type 2 DM (T2DM). DN develops quite early in T2DM patients, while it may take many years to clinically manifest in T1DM. Early stages of DN may exert the symptoms of parasthesia and hyperalgesia progressing to thermal and chemical allodynia and then loss of sensation. High blood glucose may result in excessive entry of glucose in neuron. Here the excess glucose is shunted in polyol pathway, where it is converted to fructose and sorbitol by enzymes aldose reductase and sorbitol dehydrogenase. These polyols cannot leak out from the neuronal cell and exert a strong osmotic effect, which decreases nerve myoinositol level and reduces Na+/K+ - ATPase activity. This hampers the conduction through the nerve resulting in decrease in
nerve conduction velocity. Damage to autonomic neurons of gut result in reduced intestinal motility and constipation. Autonomic neuropathy also manifests as genitourinary dysfunction. Oxidative stress, Advanced Glycation End products (AGEs) and lipid peroxidation also play an important role in neuronal damage and impaired repair mechanisms (Nishimura-YABE, 1998).

Diabetes is the foremost cause of kidney associated mortality in chronic diabetes patients (Collins et al., 2007) and kidney disease can develop even if diabetes is under control (National Institute of Diabetes and Digestive and Kidney Diseases, 2008). Diabetes associated kidney disease develops in 30 to 40% diabetic patients (DeFronzo, 1995). Both T1DM and T2DM may result in development of nephropathy, which is pathophysiologically and clinically similar to major extent. Albuminuria is the gold standard to assess the renal complication in diabetic patients. Albuminuria results from glomerular and/or tubular damage of the diabetic kidney. Apart from this, diabetic nephropathy culminates with increased thickness of glomerular basement membrane, glomerulosclerosis and mesangial expansion. There is continued loss of functioning nephrons and deposition of fibrous tissue in kidney. This results in increase in relative kidney weight, reduced renal excretory capacity, increase in serum creatinine and blood urea nitrogen (BUN) levels (Navarro and Mora, 2006).

Cardiovascular diseases are the cause of death in nearly 65% diabetic patients. Long standing hyperglycemia, poorly controlled blood pressure, microvascular disease of kidney, dyslipidaemia, coronary atherosclerosis, myocardial protein damage by AGE or oxidative stress and autonomic neuropathy are various factors responsible for development of diabetic cardiomyopathy (DC). Hypertension, diastolic dysfunction, impaired heart rate variability, capillary basement membrane thickening, reduced efficiency of myocardium, left ventricular hypertrophy, changes in inner diameter of ventricles, leading to impaired filling and pumping of blood are some of the abnormalities associated with DC (Grundy et al., 1999).

Thus, from many years of clinical experience and by several research studies it is clear that diabetic complications are multifactorial in nature and addressing only one aspect of the pathogenesis cannot sufficiently prevent the disease progression and development of diabetic complications. This may be the probable explanation for failure of present antidiabetic agents in prevention of long term complications of DM.
Many plants have been used in medicine since ancient time. Although undermined by the development of modern medicine, herbal medicine is continued to be used in many parts of the world. This is due to many reasons, some of them being their effectiveness and better safety profiles. Moreover, investigation on traditional medicinal herbs has been recommended by WHO Expert Committee on diabetes (Bailey and Day, 1989). Many scientists propose the multiple mechanisms possessed by a herbal drug to be responsible for their beneficial effects in chronic diseases. Because of combination of chemical constituents present in plants, they may exert synergistic effect and may also possess multiple mechanisms like improving insulin sensitivity, beta cell protection, decreased hepatic glucose output, anti-oxidant activity, anti-inflammatory activity, anti hyperlipidemic activity etc. Such multiple effects of plants may prove beneficial in preventing debilitating complications if used in treatment of DM.

The plant, *Anogeissus acuminata* (AA) used in present study, belongs to the family Combretaceae. This plant is used in different parts of world by locales as medicine in different ailments including DM (Pullaiah and Naidu, 2003; Manosroi et al., 2011). Moreover, the plant is rich in phenolics, due to which it exerts varied pharmacological actions. AA extracts have demonstrated anti-inflammatory, anti-oxidant, analgesic, neuroprotective, HIV reverse transcriptase inhibitory activity in different studies (Hemamalini et al., 2010; Zaruwa et al., 2009; ArunaDevi et al., 2010; Hemamalini et al., 2011). It has also demonstrated hypoglycemic action in alloxan induced DM in mice (Manosroi et al., 2011). Thus, it was hypothesised that AA may be a good antidiabetic agent on virtue of its varied actions and constituents and may be able to prevent the development of diabetic complications on long term. Therefore, it was evaluated in the present study for its antidiabetic action in two types of experimentally induced DM and in models of diabetic complications like diabetic nephropathy, neuropathy and cardiomyopathy.

Further, attempt was made to evaluate the mechanism of action of AA in DM and its complications. Oxidative stress plays an important role in progression of DM and development of diabetic complications. A drug which is anti-oxidant may efficiently prevent development of diabetic complications, while controlling plasma glucose levels. Therefore, anti-oxidant activity of present drug was evaluated in *in vivo* and *in vitro* experiments.
Apart from this, insulin resistance is an important feature of T2DM. Insulin resistance results from abnormality in insulin receptor signaling. Recent findings have suggested that Protein Tyrosine Phosphatase 1B is a negative regulator of leptin and insulin receptor signaling pathway (He et al., 2014). Therefore, PTP1B inhibition is a potential drug target for treatment of type 2 diabetes and obesity (Combs, 2010). Phenolics have demonstrated potent PTP1B inhibitory activity (Jiang, Liang and Guo, 2012). AA being a plant rich in phenolics like flavonoids and tannins, is likely to have action on PTP1B enzyme. Hence, we attempted to assess the PTP1B inhibitory activity of AA in vitro.

In the present study, preliminary phytochemical evaluation was also done to determine major constituents present in the extract. Further, analysis of major constituents identified was done using chromatographic methods.

Thus, goal of the present study was to evaluate the antihyperglycemic activity of AA and to assess its potential benefit in prevention of complications of DM. The study was conducted with following objectives:

1. Evaluation of leaf and bark extracts of AA for antihyperglycemic effect on models of type 1 and type 2 DM


3. Evaluation of effect of AA extracts on oxidative stress parameters and insulin resistance to evaluate possible mechanism of action on diabetic complications.

4. Preliminary phytochemical evaluation of leaf and bark extracts of AA