CHAPTER - 2
REVIEW
OF
LITERATURE
2.1 Diabetes Mellitus

American Diabetes Association, ADA (2005) described the Diabetes mellitus that it is a common metabolic syndrome resulting from defects in insulin secretion or action or both and characterized by hyperglycemia often accompanied by glycosuria, polydipsia, and polyuria.

Gupta et al. (2008) stated that Diabetes mellitus is a major global metabolic disorder of current century and characterized by excessive sugar in the blood (Hyperglycemia) due to deficiency in production of insulin by the pancreas or by the ineffectiveness of the insulin produced. Diabetes affects almost every cell in the body and essential biochemical processes that cause severe effects on health.

Sharma et al. (2010) delineated that Diabetes mellitus is a syndrome, which is characterized by hyperglycemia, lipo protein abnormalities, raised basal metabolic rate, defect in enzymes and high oxidative stress induced damage to pancreatic beta cells.

2.2 Type 1 Diabetes Mellitus

Chatterjea and Shinde (2007) acknowledged about type 1 Diabetes Mellitus that this is an Insulin dependent Diabetes Mellitus (IDDM) since there is almost deficiency of insulin found due to destruction of pancreatic β cells, so require insulin therapy. It is also known as “Juvenile onset Diabetes” and featured by lesser occurrence frequency, commences usually before 15 years of age, more susceptible to male than females, susceptibility is associated with particular HLA (Human leukocyte antigen) phenotype, two to three times more risk in those who are HLA phenotype B8 or BW15, speedy progression to keto-acidosis and coma, underweight and thin body.

2.3 Type 2 Diabetes Mellitus

Chatterjea and Shinde (2007) detailed about type 2 Diabetes mellitus that this is Non Insulin dependent Diabetes Mellitus (NIDDM), since insulin response to glucose metabolism is reduced due to decrease in insulin receptors on target cells and that’s why not require insulin therapy and control by oral hypoglycemic agents and exercise. It is also known as Maturity Onset Diabetes (MOD) because its occurrence frequency is more common in middle aged individuals (above 35 years). This type is featured by over eating with low activity, mildly precede, keto-acidosis,
normal or even raised plasma insulin level, fatigue, nausea, frequent urination/polyuria, thirst, unusual weight loss, blurred vision, frequent infections, slow healing of wounds or sores.

2.4 Gestational Diabetes

Gupta (2007) and Raghuram et al. (2003) stated that it occurs during normal pregnancy in about 2-5% of pregnant women, where insulin sensitivity is reduced due to the action of placental hormones and low glucose tolerance. In normal women prominent insulin resistance develops, mostly by second half of pregnancy. Repeated pregnancy may increase the risk of developing irreversible Diabetes, particularly in obese women.

2.5 Indian scenario of Type-2 Diabetes mellitus

Sinclair (2003) reported that in the developing countries the younger age groups are affected more than elderly people, whereas in industrialized (developed) countries increase in the number of cases of Diabetes mellitus type 2 occurs among elderly people.

Trout and Teff (2004) reported that women of developing countries have more difficulty in maintaining normal blood sugar in comparison to men especially around the time of their menses, and are more susceptible to Diabetes mellitus type 2.

Boon et al. (2006) gave a view about aging of population (elder peoples) which are more prone to affect by Diabetes mellitus type 2 and accounted that there are over 80 million people of age group between 45-64 years are suffering from Diabetes worldwide.

Mohan et al. (2007); Ramachandran et al. (2001) conducted a population based study by National Urban Diabetes Survey (NUDS) in six metropolitan cities across India and recruited 11,216 subjects aged 20 yr and above representative of all socio-economic strata. An oral glucose tolerance test was done using capillary glucose and Diabetes was defined using the WHO criteria. The study reported that the age standardized prevalence of Type-2 Diabetes was 12.1%. This study also revealed that the prevalence in the southern part of India to be higher i.e. 13% in Chennai, 12.4% in Bangalore, and 16.6% in Hyderabad; compared to eastern India (Kolkata), 11.7%; northern India (New Delhi), 11.6% and western India (Mumbai), 9.3%. The study also suggested that there was a large pool of subjects with impaired glucose tolerance (IGT), 14% with a high risk of conversion to Diabetes.

Agarwal, (2007a) reported that over 13% adults in urban India suffer from Diabetes mellitus type 2 whereas 5% in the countryside. In India, roughly 43% of all Diabetic cases were found to be up to 40 years of age.
Ramachandran and Snehalatha (2009) reported about prevalence of diabetes in different parts of India not only in urban populations, but also in rural populations as a result of the urbanization of lifestyle parameters. The prevalence of pre-diabetes is also high. They found a rapid conversion of impaired glucose tolerance to diabetes in the southern states of India, where the prevalence of diabetes among adults has reached approximately 20% in urban populations and approximately 10% in rural populations. Because of the considerable disparity in the availability and affordability of diabetes care, as well as low awareness of the disease, the glycemic outcome in treated patients is far from ideal.

Diamond (2011) reviewed that in India, a wide range of outcomes for different groups is buried within the average diabetes prevalence of 8%. Prevalence is only 0.7% for non-obese, physically active, rural Indians. It reaches 11% for obese, sedentary, urban Indians; and it peaks at 20% in the Ernakulam district of Kerala, one of India’s most urbanized states. Among lifestyle factors predicting the incidence of diabetes in India, some are familiar from the West, whereas others turn expectations upside down.

Whiting et al. (2011) reported that the most anticipated increase in Diabetes mellitus cases will come from developing countries upcoming next few decades. India is the number one danger zone of Diabetes in the world. In year 2011 there were 366 million people found with diabetes globally, and this is expected to rise to 552 million by the year 2030.

2.6 Diagnosis of Type-2 Diabetes Mellitus

When Diabetes is suspected, it may be easily confirmed by determining blood sugar levels, urine sugar concentration and glycosylated haemoglobin content. According to WHO (1999), Fasting plasma glucose (FPG) more than 126mg/dl (7.0mmol/L), 2 hour post load plasma glucose (PPG) more than 200mg/dl (11.1mmol/L) hypertension (BP ≥ 140/90mmHg), high density lipoprotein cholesterol (≤ 35mg/dl or less), Triglyceride level (≥250mg/dl or more) are the diagnostic criteria for diabetes.
<table>
<thead>
<tr>
<th>Venous plasma glucose concentration Condition</th>
<th>Normal</th>
<th>Impaired Fasting Glycemia</th>
<th>Impaired Glucose Tolerance</th>
<th>Diabetes Mellitus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fasting (mmol/L or mg/dl)</td>
<td>&lt; 6.1</td>
<td>&gt; 6.1 &amp; &lt; 7.0</td>
<td>&lt; 7.8</td>
<td>&gt; 7.8</td>
</tr>
<tr>
<td></td>
<td>( &lt; 110 )</td>
<td>&gt;110 &amp; &lt; 126 )</td>
<td>( &lt; 140 )</td>
<td>( &gt; 140 )</td>
</tr>
<tr>
<td>2 hours after Glucose intake (mmol/L or mg/dl)</td>
<td>&lt; 8.9</td>
<td>&lt; 7.8</td>
<td>&lt; 11.1</td>
<td>&lt; 11.1</td>
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<tr>
<td></td>
<td>( &lt;160 )</td>
<td>&lt; 140</td>
<td>( &lt; 200 )</td>
<td>( &gt; 200 )</td>
</tr>
</tbody>
</table>

Table 2.1: Diagnostic criteria for oral glucose tolerance test, (WHO, 1999).

Gupta (2007); ADA (2013) accounted that when the blood sugar level elevates beyond renal threshold (180mg/dl) and the glucose is excreted in the urine, this conditioned is called diabetic glycosuria and could also be monitored by diastex or Benedict’s test. As blood sugar (glucose) increases, more of it gets binds to haemoglobin; this combined molecule is estimated as glycosylated haemoglobin. In normal population concentration varies from 4-7% while in diabetes, it ranges 8-18%, depending on blood sugar level.

American Diabetes Association, ADA (2013) clarified that some patients cannot be clearly classified as Type-1 or Type-2 Diabetes. Clinical presentation and disease progression vary considerably in both types of diabetes. Occasionally, patients who otherwise have Type-2 Diabetes may present with ketoacidosis. Similarly, patients with Type-1 Diabetes may have a late onset and slow (but relentless) progression of disease despite having features of autoimmune disease. Such difficulties in diagnosis may occur in children, adolescents, and adults. The true diagnosis may become more obvious over time.

2.7 Factors affecting Type-2 Diabetes Mellitus

Meyer et al. (2001) reported that all types of dietary fat (except essential 3 fatty acids) may have an adverse effect on insulin sensitivity, particularly among obese individuals. Saturated fats may have the greatest adverse effect on insulin sensitivity, especially as compared with monounsaturated fats. Increased intake of polyunsaturated fat, in the context of appropriate total energy intake for weight management, may reduce the risk of Type-2 Diabetes.

Wilson et al. (2007) observed that insulin resistance and the clustering of cardio vascular disease (CVD) risk factors appear to confer much of the risk for diabetes commonly associated with
excess adiposity. Metabolic syndrome increases risk of diabetes by about a factor of 7 and accounts for about 50% of Diabetes cases on a population basis. Indeed, metabolic syndrome appears synonymous with prediabetes and can be used as an efficient tool for predicting diabetes.

Meigs et al. (2007b) also reported that insulin resistance is a potent and complex risk factor for diabetes. Whereas insulin resistance alone triples the risk of diabetes, when it occurs in the setting of a cluster of risk factors for cardiovascular disease (CVD) called metabolic syndrome (typically including hyperglycaemia, hypertension, dyslipidemia, and central adiposity), it is an especially common and powerful risk factor for type 2 diabetes.

Gupta et al. (2008) also comprised the hereditary background, aging, obesity, dietary imprudence, endocrine imbalance, psychic stress, reduction in physical labour and discriminated social structure as important factors, for exploding the prevalence Diabetes mellitus in India and other countries.

2.8 Induction of Type 2 Diabetes mellitus by Alloxan (2, 4, 5, 6-tetraoxypyrimidine)

2.8.1 Alloxan: Mechanism of action

Lenzen and Panten (1988a) suggested the mechanism of Alloxan. In the presence of thiols, generate free radicals as reactive oxygen species (ROS) in a cyclic reaction with its reduction products, dialuric acid. Autoxidation of dialuric acid generates superoxide anion, hydrogen peroxide molecules and finally hydroxyl radicals. These hydroxyl radicals are ultimately responsible for death of beta cells. As a thiol reagent, Alloxan selectively inhibits glucose induced insulin secretion through its ability to specifically inhibit the glucokinase through oxidation of functionally essential thiol groups in the protein, thereby impairing oxidative metabolism and the glucose sensor function of this signaling enzyme of the beta cell.

Iwase et al. (1991) reported that Alloxan and Streptozotocin, both are used for Diabetes induction in experimental rats for in vivo studies but there are some disadvantages to use of streptozotocin in chronic experiment due to spontaneous recovery from high blood glucose levels by the development of functioning insulinoma, high incidence of kidney, liver and pancreatic tumors and its solution instability toward slight change of pH, temperature, light and solvent medium.

Robert et al. (2002) reported another mechanism in which diabetogenic agent Alloxan also interferes with the process of O-glycosylation in beta cells. Because Alloxan is derived from
uracil and Oxy-N-acetylglucosamine transferase, or O-GlcNAc transferase (OGT) uses UDP–GlcNAc as a substrate, so Alloxan may interfere with O-GlcNAcylation by being an inhibitor of OGT, possibly through an interaction of Alloxan with the UDP-binding domain in OGT. Since OGT is required for cell viability and so abundant in beta cells, the inhibition of OGT by Alloxan might explain how this drug is relatively specific for beta cells. Furthermore, in order to show definitively that Alloxan was inhibiting OGT activity, recombinant OGT was incubated with 0–10 mM Alloxan, and OGT activity was measured directly by quantitating UDP-[3H]-GlcNAc incorporation into the recombinant protein substrate, nucleoporin p62. Under these conditions, OGT activity was completely inhibited by 1 mM Alloxan with half-maximal inhibition achieved at a concentration of 0.1 mM Alloxan. It shows that Alloxan is an inhibitor of OGT, and as such, is the streptozotocin OGT inhibitor described).

Lenzen (2008) reported that chemically induction of Diabetes in experimental animals is widely used as diabetic model in the studies on the complication caused by this disease. The cytotoxic glucose analogues Alloxan and streptozotocin are the most prominent diabetogenic chemical agents in experimental Diabetes research. Both are selectively toxic to pancreatic beta cells because they preferentially accumulates in beta cells as glucose analogues through uptake via GLUT2 glucose transporter but their mechanism of cytotoxic action are different.

**2.8.2 Alloxan: Dose for induction of Type-2 Diabetes Mellitus**

Dave and Katyare (2002) used Male and female albino rats of the Charles Foster strain weighing between 200 ± 20 g for diabetic model. After fasting for 24 h the animals were given a single subcutaneous injection of freshly prepared Alloxan solution using saline (0-9% (w/v) NaCl) as vehicle, at a dose of 120 mg Alloxan/1kg body weight. Symptoms of Diabetes like loss of body weight, polyuria, glycosuria, polydipsia, polyphagia and increased blood glucose levels were observed within a week of Alloxan injection.

Sindhu et al. (2010) induced Type-2 Diabetes into female albino rats by a single intraperitoneal (i.p) injection of Alloxan monohydrate (120 mg/kg b.wt) in sterile normal saline (0.9%) and diabetic state was determined after 3 days of alloxination by high blood glucose level tested by one touch glucometer.

Kumar et al. (2010) induced diabetes in Charles Foster strain normal rats by a single intraperitoneal injection of Alloxan monohydrate 150 mg/kg body weight in standard vehicle. After 2 weeks, rats shown fasting blood glucose level 80-367 mg/dl were considered to be diabetic.
Dallak and Bin-Jaliah (2010) induced diabetes in male wistar albino rats by intraperitoneal administration of Alloxan monohydrate (150 mg/kg body weight), dissolved in normal saline and rats were treated with 30% glucose solution orally at different time intervals after 6 h of Alloxan induction to prevent hypoglycaemia. After 10 days, rats with diabetes mellitus having glycosuria (indicated by Benedict’s test) and hyperglycaemia, with blood glucose range of 250–375 mg/dl.

Lanjhiyana et al. (2011) induced diabetes by single dose intraperitoneal injection of Alloxan monohydrate (120 mg kg$^{-1}$ BW) in 0.9% w/v NaCl solution to overnight fasted Charles Foster strain normal rats. Blood glucose level was checked by using one-touch glucometer and Diabetes was confirmed after 72 hr of alloxaonisation. Rats shown FBG > 250 mg/dl were considered to be diabetic.

2.9 Pathophysiology of Type-2 Diabetes Mellitus

Virella-Lopes and Virella (2003) reported that chronic hyperglycaemia during Diabetes causes gyration of body proteins that in turn leads to secondary complications affecting eyes, kidneys, nerves and arteries. Along with hyperglycaemia and abnormalities in serum lipids, Diabetes is associated with microvascular and macrovascular complications which are the major causes of morbidity and death in diabetic subjects. Also, Diabetes mellitus leads to many complications, such as cataract formation, loss of sensivity of receptors, formation skin lesion, gangrene formation and pulmonary tuberculosis.

Raghuram et al. (2003) reported that Diabetic patients with long standing impaired vision, cataract, renal failure, sensory loss, gastrointestinal problems, foot ulcers, hardening of blood vessels and stroke, are recognized as chronic complications under pathophysiology.

Murata et al. (2005) conferred that important Diabetic complications are recognized by hyperglycemia, hypoglycemia and ketoacidosis. The patients using excessive insulin or oral drugs develop rapid and severe lowering of blood sugar below certain critical limits (below 45-55 mg/dl), resulting hypoglycemia that may cause coma.

Boon et al. (2006) presented a view about carbohydrate metabolic products during Diabetes i.e. when body can’t use carbohydrate as fuel for energy, it utilizes large amount of fats and proteins. This results in over production of metabolic product ketones. The increase amount of ketones in blood stream cause ketoacidosis and patients may enter into coma.
Agarwal (2007b) disclosed that Diabetic patients who have high blood sugar levels are at increased risk of formation of blood clots. This is due to their stickier platelet cells which cause several abnormalities.

2.10 Diabetes Mellitus of Type-2 and Biochemical Changes

2.10.1 Diabetes with Hyperglycemia and Dyslipidemia

Lyons (1991); Fang et al. (2002) observed that Diabetes produces disturbances of lipid profiles, especially an increased susceptibility to lipid peroxidation, which is responsible for increased incidence of atherosclerosis.

American Diabetes Association, ADA (2005) reported that Type-2 Diabetic dyslipidemia is associated with elevated plasma triglycerides, very low-density lipoprotein (VLDL)-triglycerides and decreased high-density lipoprotein (HDL) cholesterol levels that are related to the risk for cardiovascular disorder ie.CHD. Up to 80% death within this high risk population are due to associated cardiovascular disease. Lipid management aimed at lowering LDL cholesterol, raising HDL cholesterol, and lowering triglycerides has been shown to reduce macrovascular disease and mortality in patients with Type-2 Diabetes, particularly those who have had prior cardiovascular events. In studies using HMG (hydroxymethylglutaryl) CoA reductase inhibitors (statins), patients with Diabetes achieved significant reductions in coronary and cerebrovascular events. Glycemic control can also beneficially modify plasma lipid levels. Particularly in patients with very high triglycerides and poor glycemic control, glucose lowering maybe necessary to control hypertriglyceridermia.
In Diabetes mellitus all the biochemical changes are resulted from cellular metabolic changes as given below-

MacDonald et al. (2009) studied the pancreatic islets isolated from rodent models of Type-2 Diabetes have shown decreases in certain mitochondrial enzymes, such as mitochondrial glycerol phosphate dehydrogenase (mGPD), the key enzyme of the glycerol phosphate shuttle, and pyruvate carboxylase (PC), an anaplerotic enzyme. The activities of the mitochondrial enzymes, glycerol phosphate dehydrogenase, pyruvate carboxylase (PC) and succinyl-CoA: 3-ketoacid-CoA transferases (SCOT) were found decreased by 73%, 65% and 92%, respectively, in the diabetic compared with the non-diabetic islets. ATP citrate lyase, a cytosolic enzyme of the mitochondrial citrate pyruvate shuttle, was decreased 57%. Activities of propionyl-CoA carboxylase, NADP-isocitrate dehydrogenase, cytosolic malic enzyme, aspartate aminotransferase and malate dehydrogenase were not significantly different from those of the control.

Esposito et al. (2009) reported that within the small intestine, Diabetes is associated with numerous changes, including hyperplasia and hypertrophy of epithelial cells, increased
absorption of sugars and amino acids and increased endogenous cholesterol synthesis. Inflammatory cytokines are also increased in diabetes. It is believed that increases in human histocompatibility antigens (HLA) class II, HLA-DR and HLA-DP, found in structurally normal intestine in diabetic individuals, may result from secretion of inflammatory cytokines such as interferon gamma (IFN-γ).

Williams et al. (2011) reported that hyperglycemic hyperosmolar state (HHS) in Type-2 Diabetes is characterized by profound hyperglycemia, dehydration, change in mental status, and the absence of ketoacidosis. Decreased insulin causes muscle and the liver to increase plasma glucose. Hyperglycemia leads to loss of glucose and water in the urine, which lowers plasma glucose but leads to dehydration. In Diabetic ketoacidosis (DKA) or in hyperglycemia without HHS, thirst increases with dehydration. This thirst increases the intake of water to offset dehydration and allows continued loss of glucose in the urine. Patients with HHS have an inappropriately low thirst. As dehydration becomes fast, the ability to lose glucose in urine is diminished because blood flow to the kidneys is decreased and creates other kidney function abnormalities.

2.10.2 Diabetes and malfunctioning of Liver

Everhart (1995) reported that elevated serum activity of the two aminotransferases i.e. aspartate aminotransferase (AST) and alanine aminotransferase (ALT) are the most frequently measured indicators of liver disease in Diabetes mellitus type 2 than in the general population.

Erbey et al. (2000) conferred that liver pathology among diabetic patients is similar to that of alcoholic liver disease, including fatty liver (steatosis), steatohepatitis, fibrosis, and cirrhosis.

Wong et al. (2004) reported that nearly 70 to 80% of the diabetic subjects have hepatic fat accumulation, referred to as non alcoholic fatty liver (NAFL). NAFL leads to non alcoholic steato hepatitis (NASH), a progressive fibrotic disease, which can result in cirrhosis or liver related death.

Hanley et al. (2004) observed the association of liver disease with Diabetes mellitus type 2 by the insulin resistance atherosclerosis study (IRAS), which showed that liver function markers like the aspartate aminotransferase (AST) and alanine aminotransferase (ALT) are predictors of incident Diabetes mellitus.
Medina et al. (2004) reported that there are not much therapeutic options for non alcoholic fatty liver except correction of obesity with hypo caloric diets and physical exercise and controlling hyperglycaemia with diet, insulin, or oral hypoglycaemic agents.

Sattar et al. (2004); Wannamethee et al. (2005); Nakanishi et al. (2005) reported about major role of liver in the regulation of carbohydrate metabolism during Diabetes mellitus type 2, as it uses glucose as a fuel and has the capability to store glucose as glycogen and also synthesize glucose from non-carbohydrate sources. This key function of liver makes it vulnerable to diseases in subjects with metabolic disorders, particularly Diabetes. Increased activities of liver enzymes such as aspartate aminotransferase (AST), alanine aminotransferase (ALT) and γ-glutamyltranspeptidase (GGT) are indicators of hepatocellular injury. Increased activity of these markers is associated with insulin resistance metabolic syndrome and Type-2 Diabetes mellitus.

2.10.3 Diabetes and malfunctioning of Kidney

David et al. (2005) reported that blood pressure (BP) control is undeniably important in delaying progression of kidney disease in hypertensive Diabetic patients. Attention to other potentially remediable factors may be more important in delaying progression once BP is effectively treated. Antiproteinuric agents (such as ACE inhibitors and ARBs) may exert beneficial actions beyond those due to BP control. Hypoalbuminemia is independently associated with progression of CKD due to Type-2 Diabetes after adjustment for the effects of proteinuria suggests the possibility that inflammation (and/or malnutrition) may be a progression promoter.

Lesley et al. (2006) reported about importance of creatinine level in kidney functions. Creatinine is an amino acid derivative with a molecular mass of 113 D that is freely filtered by the glomerulus. Many studies support the similarity of creatinine clearance to GFR and its reciprocal relationship with the serum creatinine level. Creatinine is secreted by proximal tubular cells as well as filtered by the glomerulus; thus, the creatinine clearance exceeds the GFR. The generation of creatinine is determined primarily by muscle mass and dietary intake. Extra renal elimination of creatinine may be increased at low levels of GFR and serum creatinine level probably accounts for malfunctioning for kidney. The reciprocal relationship between GFR and serum creatinine levels makes it difficult for clinicians to appreciate the level and rate of change in GFR by simply monitoring serum creatinine levels for kidney dysfunctions.

Carmine et al. (2006); Elizabeth et al. (2008) suggested that serum uric acid (UA) is a relevant and independent risk factor for cardiovascular and renal disease, particularly in patients with hypertension, heart failure, or diabetes on the basis of extensive epidemiologic and experimental
evidence. The association of elevated serum uric acid, blood urea and serum creatinine and higher urinary albumin excretion rate with advanced impaired renal function prompts an examination of its role in early renal function decline in patients with Type-1 Diabetes before proteinuria develops.

**Salim (2010)** reported that serum uric acid, blood urea, and serum creatinine are significantly increased in Type-2 Diabetic patients in compared with Type-1 Diabetes and with healthy individuals. Increase of uric acid in a non-significant pattern in Type-1 Diabetic patients in compared with healthy individuals, but blood urea and serum creatinine are increased significantly. Serum calcium and serum albumin are decreased in Type-2 Diabetic patients compared with healthy individuals.

**American Diabetes Association, ADA (2013)** reported that Diabetic nephropathy occurs in 20–40% of patients and is the single leading cause of end-stage renal disease (ESRD). Persistent albuminuria in the range of 30–299 mg/24 h (microalbuminuria) has been shown to be the earliest stage of Diabetic nephropathy in Type-1 Diabetes and a marker for development of nephropathy in Type-2 Diabetes. Reduced kidney function is also assessed by a GFR < 60 ml/min/1.73 m². Normal values, which are related to age, sex, and body size, are approximately 130 ml per minute per 1.73 m² in young men and 120 ml per minute per 1.73 m² in young women.

### 2.10.4 Diabetes and malfunctioning of Pancreas

**Nsien (1992)** found that three patients with diabetic ketoacidosis (DKA) had striking elevations of serum lipase levels and less striking elevations of amylase and trypsinogen. All three had nausea and vomiting, but none had objective evidence of abdominal pain, and computed tomography scans of the pancreas were unremarkable. These cases suggest the association of asymptomatic enzyme elevations with DKA.

**Lee and Lee (2001); Heo et al. (2009)** reported that the enzymes secreted by the pancreas, like α-amylase and lipase, are known to break down dietary polysaccharides and lipids into monosaccharides and free fatty acids, which represent some of the major fuels needed to maintain human metabolic energy.

**Aughsteen and Mohammed (2002); Rizvi (2003)** reviewed that although most of the research so far conducted on diabetes has focused on dyslipidemia as a major risk factor for cardiac, cerebral, and renal complications. Several studies have recently showed an impairment of pancreatic exocrine functions in type-1 and type-2 diabetes. The analysis of serum/plasma
pancreatic enzymes was suggested to provide additional informative parameters of pancreatic functions for the assessment of the chronicity and progress of the illness as well as of the response to therapy.

Mooren *et al.* (2003); Frulloni *et al.* (2005) reported that an increase in the serum concentration of pancreatic enzymes (amylase and lipase) is commonly an expression of inflammatory or neoplastic pancreatic disease. However, an elevation of pancreatic enzymes, generally mild, may be a non-specific phenomenon without any clinical implication. A rapid Ca$^{2+}$ release from the intracellular stores in response to hormonal stimuli is a signalling mechanism which regulates exocrine pancreatic secretion. The pathological mechanism is probably related to a disruption of pancreatic acini or to an alteration of the normal exocytosis process, with the secretion of the zymogen contents at the basolateral side of the acinar cells. The pancreatic enzymes are therefore released into the interstitial space and later reabsorbed directly or via the lymphatic into the bloodstream.

Haddad *et al.* (2004) postulated that pathogenesis of serum pancreatic enzyme elevations in metabolic disorders (diabetic ketoacidosis, acidemia) remains unclear and results from direct injury to the pancreas with enzyme leakage from the acini and decreased renal clearance, but other Authors have suggested a possible role of acidosis in the pancreatic and extra pancreatic secretion of amylase and lipase. Some others have postulated that, in patients with dyslipidemia, particularly hypertriglyceridemia or, hypercholesterolemia or both, there may be an accumulation of fat inside the pancreatic acinar cell, disturbing exocytosis.

Quiros *et al.* (2008) reported that elevation of pancreatic enzymes is common in children with DKA, but clinical pancreatitis is rare. Pancreatic enzyme levels reach a peak 12-24 hrs after initiation of treatment for DKA. Pancreatic enzyme elevation is associated with increased BUN concentrations at presentation but is not associated with abdominal pain.

**2.11 Diabetes Mellitus Type-2 and Oxidative Stress**

Halliwell and Gutteridge (1989) reported about diabetic oxidative stress, as the mechanism underlying diabetes and diabetic complications. Enhanced oxidative stress and changes in antioxidant capacity, observed in both clinical and experimental diabetes mellitus, are thought to be the etiology of chronic diabetic complications.
2.11.1. Diabetes and Production of free radicals as ROS and RNS

De-Fronzo and Ferrannini (1991); Toborek and Henning (1994) postulated that insulin resistance are associated with elevated fasting plasma non-esterified fatty acid (NEFA) concentration. Fatty acids cause an increase in oxidative stress in cultured endothelial cells and an initial decrease in reduced glutathione concentrations after 6h of exposure to the incubation medium. Thus non-esterified fatty acid associated with Insulin resistance induced oxidative stress in diabetes mellitus.

Schmidt et al. (1994) reported that non-enzymatic protein glycation is a spontaneous chemical reaction between glucose and the amino groups of proteins in which reversible Shift bases and more stable Amadori products are formed. Advanced glycation end products (AGEs) are then produced by auto-oxidation of Amadori product.

![Diagram](Figure 2.2: Generation of oxidative stress due to hyperglycaemia.)
Ceriello et al. (1996) conferred a view that Diabetes mellitus is characterized by increased generation of glycoxidation products associated with the advanced oxidative stress. The presence of higher glucose or glycated protein concentration enhances lipid peroxidation and reversely, lipid peroxides may increase the extent of advanced glycation end-products. Hyperglycemia is a widely known cause of enhanced plasma free radical concentrations and occurred via at least four different routes; increased glycolysis, intercellular activation of sorbitol (polyol) pathway, auto oxidation of glucose and non-enzymatic protein glycation.

Ceriello et al. (1996) reported that glucose can be auto-oxidized in a cell-free system under physiological conditions via enediol tautomer formation which generates hydrogen peroxide; reactive intermediate such as hydroxyl and superoxide radicals, and ketoaldehydes. Transition metals such as iron are believed to be of crucial importance in the cascade of these reactions, as they catalyze auto-oxidation of glucose.

Wautier et al. (1996) conferred the mechanism by which AGEs elicit their cellular effects by binding to specific cellular receptors, one of which, RAGEs (receptor for AGEs), has been identified on endothelial cells, monocytes/macrophages, mesangial cells, neurons and smooth muscle cells. Interaction of AGEs with endothelial surface RAGEs generates intracellular oxidative stress and therapy modulates cellular functions, even in the presence of intact antioxidant mechanisms. This process is probably enhanced and amplified when antioxidant defense mechanisms are reduced.

Cameron et al. (1997) proposed the mechanism of polyol pathway involving increased glycolysis that is a consequence of hyperglycemia and closely related to an increase in NADH / NAD+ ratio due to impaired oxidation of NADH to NAD+. In sorbitol (polyol) pathway glucose is reduced to sorbitol by aldose reductase (AR), coupled with oxidation of NADH/NAD+. Sorbitol is then oxidized to fructose coupled with reduction of NAD+ to NADH by sorbitol dehydrogenase (SDH).

Evans et al. (2002); Robertson et al. (2004) reported that a phagocyte-type oxidase operates in the kidney as an oxygen sensor and it is known as kidney-specific NADPH oxidase or renox (now renamed Nox4). The targets of ROS action have been poorly characterized and may comprise members of the classical mitogen-activated protein kinase cascade as well as the c-Jun N-terminal protein kinase pathway. Moreover, tyrosine phosphatases and the small guanosine 5′-triphosphate–binding proteins Ras and Rac1 are targeted by ROS in a redox-dependent manner.

Singh et al. (2009) reported that both ROS and RNS are involved in the etiopathogenesis of Diabetes mellitus Type-2. When the concentration of ROS produced exceeds the cellular
capacity to cope with them, oxidative stress results. ROS are generated most prominently by xanthine oxidase, cyclooxygenases, lipoxygenases, cytochrome P450 oxidases, NOS, the mitochondrial respiratory chain, and NADPH oxidases.

$$\text{O}_2 \rightarrow \text{O}_2^- \rightarrow \text{H}_2\text{O}_2$$

Figure 2.3: Correlation between generation of ROS and Diabetes mellitus type-2.

The above composite diagram shows different sources leading to enhanced generation of ROS in diabetes (Singh et al., 2009).
2.11.2 Diabetes and Enzymatic Antioxidative Markers

Kazuhiro et al. (1989); Matkovics et al. (1998) postulated that Glutathione reductase (GR) reduces the oxidized glutathione and release reduced glutathione for further catabolising $\text{H}_2\text{O}_2$. A decrease in the activity of glutathione reductase (GR) was elucidated in erythrocyte hemolysates of streptozotocin induced diabetic rats and attributed this decrease to the enzyme glycation by the uncontrolled hyperglycemia.

Yan and Harding (1997) conferred about the catalase (CAT) enzyme activity that it decomposes hydrogen peroxide ($\text{H}_2\text{O}_2$) to molecular oxygen ($\text{O}_2$) and water ($\text{H}_2\text{O}$) molecules ($2\text{H}_2\text{O}_2 \rightarrow 2 \text{H}_2\text{O} + \text{O}_2$). It also exhibits peroxidative activity and catalyses the oxidation of various hydrogen donors in the presence of relatively lower concentrations of hydrogen peroxide. In hyperglycaemic condition during diabetes, a decrease in CAT activity has been observed which may be due to glycation of enzyme.

Anuradha and Selvam (1993); Mohan et al. (2011) reported a decrease in the activity of these antioxidant enzymes (SOD, CAT, GSH-PX, GST and GR) in liver, kidneys and serum of alloxan induced diabetic rats.

Muller (2000); Sozmen et al. (2001) reported that superoxide ($\text{O}_2$) ions are the primary ROS produced in the course of oxygen metabolism and referred as highly reactive cytotoxic ROS. SOD acts as first line of defense against ROS-mediated injury and catalyzes the disproportionation of superoxide ($\text{O}_2$) to molecular oxygen and peroxide ($2\text{O}_2^- + 2\text{H}^+ \rightarrow \text{H}_2\text{O}_2 + \text{O}_2$). Thus it is critical for protecting the cell against the toxic products of aerobic respiration. Reduction in plasma and cellular SOD activity has been observed during diabetes and probably due to inactivation of SOD by $\text{H}_2\text{O}_2$ or by glycation of enzyme.

Muller (2000); Anuradha and Selvam (1993) reported that Glutathione peroxidase (GSH-PX) enzyme with selenium plays a primary role in minimizing oxidative damage. Glutathione peroxidise and Glutathione-s-transferase (GST) works together with glutathione in the decomposition of $\text{H}_2\text{O}_2$ or other organic hydroperoxides to non-toxic products at the expense of reduced glutathione ($2\text{GSH} + \text{H}_2\text{O}_2 \rightarrow \text{GS}--\text{SG} + 2\text{H}_2\text{O}$). Reduced activities of GSH-PX may result from radical–induced inactivation and glycation of the enzyme.

Soon and Tan (2002) reported that oxidative stress in the pathogenesis of diabetes is occurred, not only by oxygen free-radical generation, but also due to alteration in antioxidant enzymes like the superoxide dismutase (SOD), catalase (CAT), Glutathione peroxidase (GSH-Px), Glutathione reductase (GR), Glutathione-S transferase (GST) and Guaiicol peroxidase (GPX) whose activities
contribute to eliminate ROS and RNS like superoxide, hydrogen peroxide and hydroxyl radicals. The decreased activity of antioxidant enzymes along with elevated lipid peroxide levels in diabetic rats could probably be associated with oxidative stress and/or decreased antioxidant potential.

Andallu and Varadacharyulu (2003) observed the reduced activity of GST in the diabetic state that may be due to the inactivation caused by reactive oxygen species.

Sathishsekar and Subramanian (2005) reported that Glutathione is a substrate for glutathione peroxidase and glutathione-S transferase enzymes. Increased levels of GSH enhances the activity of GSH-POX and GST to scavenge free radicals in diabetic rats while low GSH content indicates low GSH-POX activity, which may produce increased oxidative stress propensity. Reduced activities of GSH-POX and GST in the liver and kidney have been observed during diabetes and this may result in a number of deleterious effects due to the accumulation of toxic products.

2.11.3 Diabetes and Non Enzymatic Antioxidative Markers

Like enzymatic antioxidants, non enzymatic antioxidants play a vital role in protecting cells from oxidative damages. These non-enzymatic antioxidants detoxify free radicals directly and also interact in recycling process to engender reduced forms of the non-enzymatic antioxidants. Vitamins also scavenge ROS and up regulate the activities of antioxidant enzymes. Among them vitamin A, E & C have important antioxidant property. A significant decrease of these vitamins has been observed in Diabetes mellitus type-2. So enhancement of these vitamins in vivo system by the supplement of some other herbal sources plays an important role in reduction of oxidative stress Diabetes mellitus type-2.

Jain and McVie (1994); Davi et al. (2005) reported that Glutathione (a tripeptide of γ-Glu-Cys-Gly and present in millimolar concentrations in all the cells) acts as important non enzymatic antioxidant. Reduced glutathione normally plays the role of an intracellular radical scavenger and is the substrate of many xenobiotic elimination reactions. There is a negative correlation between GSH and HbA1c in diabetic patients which confirms the link between hyperglycemia and GSH depletion.

Paolisso et al. (1995) observed in a placebo-controlled study that the supplementation with 500 mg vitamin C twice daily for 4 months reduces the plasma levels of LDL, TC, TG and insulin significantly in type 2 diabetes patients.
Ciuchi et al. (1996) reported a marked decreased level of reduced glutathione (GSH) in the plasma & erythrocytes of diabetic patients, as a result of decreases in activities of the enzymes involved in GSH synthesis (such as γ-glutamylcystein synthetase) or in the transport rate of oxidized glutathione (GSSG) from erythrocytes and enhanced sorbitol pathway.

Tuitoek et al. (1996) reported that insulin-dependent diabetes mellitus (IDDM) or, STZ induced diabetes, is associated with an impaired metabolic availability of vitamin- A. Abnormal metabolism of vitamin-A has been described with decreased circulating level along with decreased carrier protein, i.e. retinol binding protein (RBP).

Kajanachumpol et al. (1997) reported that lipid peroxidation is the primary cellular damage resulting from free radical reactions in diabetic state. In this state the structure changes are oxidative in nature due to peroxidative deterioration of unsaturated fatty acids of cellular membrane phospholipids, via intermediate radical reactions with a result of producing lipid hydro peroxides (LHP). The net effect of these combined reactions is the generation of highly toxic peroxy radicals (ROO⁻) which generate new lipid hydro peroxides because of their close proximity in bio membranes to other lipids. Extra cellular lipid hydro peroxides are transported in the systemic circulation by low- and high-density lipoproteins. Consequently, mechanisms in the formation of lipid hydro peroxides and biologically active metabolites, together with their effect on cellular structure and function are becoming of increasing importance to the study of diabetogenesis.

Stohs et al., (1984); Helen and Vijayamal (1997) observed that vitamin-A is lipid soluble antioxidant that inhibits oxidation of biomolecules and regulates endogenous activities of scavenging enzymes in cigarette smoke-induced or, TCDD-induced oxidative stress.

Datta and Lianos (1999) postulated that vitamin- A inhibits iNOS gene transcription in vascular smooth muscle cells, endothelial cells, cardiac myocytes, mesangial cells and thus prevent radical induced cytotoxicity.

Carr et al. (2000) reported that vitamin- E inhibits ROS-induced generation of lipid peroxyl radicals, thereby protecting cells from peroxidation of PUFA in membrane phospholipids, from oxidative damage of plasma very low-density lipoprotein, cellular proteins, DNA, and from membrane degeneration.

Fang et al. (2002) conferred a view about reduced glutathione (GSH) that it is a major component of the cellular antioxidant system can be partly absorbed from the small intestine and can be synthesized de novo. GSH can react with a variety of xenobiotic electrophilic compounds
in the catalytic reaction of glutathione-S-transferase. GSH effectively scavenges ROS (e.g., lipid peroxyl radical, peroxynitrite, and H_2O_2) directly and indirectly through enzymatic reactions. GSH can conjugate with NO, resulting in the formation of S-nitrosoglutathione adduct, which is dissociated by the thioredoxin system to release GSH and NO. GSH interacts with glutaredoxin and thioredoxin (thiol-proteins), which play an important role, in regulation of cellular redox homeostasis.

**Ardekani and Ardekani (2007); Osman et al. (2010)** observed a decreased basal vitamin- C level in diabetic patients and thus decreased plasma lipid peroxide levels, GSH and enzymic antioxidants. A supplementation with 1000 mg/day of vitamin C in addition to the normal diet and treatment schedule showed significant reduction in serum FBS, LDL, HbA1c as well as serum fasting insulin in patients with type 2 diabetes. It was also observed that vitamin- C reduces plasma lipid peroxide levels; increase GSH and enzymic antioxidants in case of type 2 diabetes mellitus.

**2.12 Pharmacological interventions of Diabetes mellitus Type-2 by Plant’s product vs. synthetic chemical drugs**

The prevalence of Type-2 Diabetes mellitus is increasing worldwide at alarming rates. Several therapeutic strategies are currently available for the treatment of this chronic metabolic disorder, including the stimulation of endogenous insulin secretion, enhancement of insulin action at the target tissues, inhibition of dietary starch and lipid degradation, and treatment with oral hypoglycemic agents. The limitations associated with those therapeutic strategies have led to a determined search for more efficient and cost-effective alternatives.

**2.12.1 Regulation of Type-2 Diabetes Mellitus with Synthetic Chemical Drugs**

**Ovalle and Bell (1998)** reported that patients with Diabetes mellitus type 2 remain uncontrolled based on current recommendations of synthetic drug treatments. Further improvement in glycemic control may thus require higher insulin doses and/or the addition of a third oral synthetic agent. Weight gain is a concern in patients with type 2 diabetes while treated with insulin. However, for patients receiving metformin the addition of inhaled human insulin (INH) did not cause significantly more weight gain than the addition of a sulfonylurea.

**Cook et al. (2005)** reported that Diabetes is a progressive disease which may require insulin therapy with oral agents like metformin, glibenclamide to achieve a level of glycemic control. These oral agents some time do not give proper effects or give other unwanted responses.
Ramachandran et al. (2010) reported that oral antidiabetic drug (OAD) is the first line of drug treatment for type 2 diabetes. However, the progressive nature of type 2 diabetes usually requires a combination of two or more oral agents in the long term, often as a prelude to insulin therapy. Both OADs and insulin treatment increased the risk of hypoglycaemia. Weight gain was significantly higher in the intensive group with a sulphonylurea (SU) (chlorpropamide, glibenclamide or glipizide) or with insulin than in the conventional group with diet.

Rabbani et al. (2010) reported that Glibenclamide is a known sulfonylurea drug which is effective in moderate diabetic state and ineffective in severe diabetic animals where pancreatic β-cells are almost totally destroyed. Several studies indicated that it enhance the level of antioxidant enzymes besides reducing the lipid peroxidation in diabetic animals.

Bhoyar et al. (2011) reported about different approaches to the treatment of diabetes, like insulin treatment in type 1 diabetes: Sul-phonylureas, which release insulin from pancreas by blocking the ATP-sensitive potassium channels; Biguanides, which decrease the insulin resis-tance; Thizaolinediones, which increase the insulin sensitivity; alpha-glucodase inhibitors like acarbose, which decrease glucose absorption from intestine, the-reby decreasing postprandial hyperglycemia; metigli-nides like repaglimide and nateglamide, which are insu-lin secretogogues. But synthetic oral antidiabetic and antioxidant drugs have long term use and safety problems with minor or major side effects and only efficient with life style modification like dieting.

2.12.2 Regulation of Type-2 Diabetes Mellitus with Plants’ Products

Mansi and Lahham (2008) shed light on herbal medical plants important roles in the management of diabetes mellitus especially in developing countries where resources are meager. Over the two decades, data from controlled investigations in animal models and patients have validated the therapeutic value of numerous phytotherapies for diabetes. Phytotherapies and their combinations demonstrate multiple beneficial anti-diabetic mechanisms including modulation of carbohydrate metabolism, restoration of beta-cell integrity and function, insulin-releasing activity, improvements in glucose uptake/utilisation, antioxidant properties and a reduction in the risk of cardiovascular disease.

Rohman et al. (2010) conferred that antioxidants have already been found in plant materials and supplements. Due to their natural origin, the antioxidants obtained from plants are of greater benefit in comparison to synthetic ones which induces side effects.
Pandey and Rizvi (2009) discussed that polyphenols are secondary metabolites of plants and are generally involved in defense against ultraviolet radiation or aggression by pathogens. In the last decade, there has been much interest in the potential health benefits of dietary plant polyphenols as antioxidant. Epidemiological studies and associated meta-analyses strongly suggest that long term consumption of diets rich in plant polyphenols offer protection against development of cancers, cardiovascular disease, diabetes, osteoporosis and neurodegenerative diseases.

Rice-Evans et al. (1997) reported that antioxidative properties of polyphenols arise from their high reactivity as hydrogen or electron donors, and from the ability of the polyphenol-derived radical to stabilize and delocalise the unpaired electron (chain-breaking function), and from their ability to chelate transition metal ions.

Arora et al. (2000) postulated another mechanism underlying the antioxidative properties of phenolics is the ability of flavonoids to alter peroxidation kinetics by modification of the lipid packing order and to decrease fluidity of the membranes.

Dai and Mumper (2010) reported that plants originated antioxidants can change oxidative damages by the sterically hinder diffusion of free radicals and restriction of peroxidative reactions. Further they scripted that phenolics and flavonoids are considered as great antioxidants and proved to be more effective than Vitamin C, E and carotenoids.

Zapolska-Downar et al. (2006) postulated that flavonoids are able to inhibit aldose reductase enzyme (that converts sugars to sugar alcohols) and is implicated with phenolic acids for Antidiabetic activity. A different flavonoid, Quercetin (QE), used in doses of 15–50 mg/kg body mass was capable of normalizing blood glucose level, augmenting liver glycogen content and significantly reducing serum cholesterol and LDL concentration in alloxan induced diabetic rats.

Abdelmoaty et al. (2010) observed that quercetin treatment (at the dose of 15 mg/kg BW) significantly increased the antioxidant enzyme activities and shown to be normalizing blood glucose level in STZ induced diabetic rats.

Ghosh et al. (2009); Rao et al. (1997) detailed that flavonoids, steroids/terpenoids, phenolic acids are known to be bioactive antidiabetic principles. Flavonoids are also known to regenerate the damaged beta cells in the alloxan induced diabetic rats and acts as insulin secretagogues.

Willcox et al. (2004); Kris-etherton et al. (2002) studied the clinical roles of carotenoids, which are major pigment of plants origin having polyene chains and responsible for their characteristic absorption spectra and specific photochemical properties. Among the carotenes, only alpha, beta and epsilon carotenes possess vitamin A activity and out of them β-carotene is the most active.
Natural β-carotene is the precursor of vitamin A and has preventive action against eye diseases and cancer. Carotenes enhance immune response and protect skin cells against UV radiations. They help to lower the risk of cardiovascular diseases, age related vision disorders, diabetic complications and oxidants activities.

Lee et al. (2001); Kim and Lee (2004) reported that ascorbic acid (vitamin- C) consists of a 6-carbon lactone ring with 2, 3-enediol moiety and shows antioxidant activity due to enediol group. It is a leading natural antioxidant that can scavenge ROS and has anticarcinogenic effects. The antioxidant mechanism of ascorbic acid is based on hydrogen atom donation to lipid radicals, quenching of singlet oxygen and removal of molecular oxygen.

2.13 Antidiabetic and antioxidative properties of plants of Oxalidaceae family

2.13.1 Oxalis corniculata L.

Raghvendra et al. (2006) scripted that Oxalis corniculata L., commonly known as creeping wood sorrel, belongs to the family Oxalidaceae, is a sub-tropical plant and originated from India. The plant having most diverse 4 genus and consist of about 900 species. It is distributed as a weed in damp shady places, roadsides, plantations, lawns, nearly all regions throughout the warmer parts of India. It is a good source of vitamin C and is used as an anti ascorbutic in the treatment of scurvy. In the folk medicines, the juice of the plant is given in stomach trouble; decoction of roots is useful for worms, used to clean rusted vessels. The extract of the plant is applied in case of scorpion sting; fresh leaves of Oxalis corniculata are crushed and are used to stop bleeding from wounds. The raw fresh leaves are crushed and directly applied on skin to treat eczema.

Kathiriya et al. (2010) screened phytochemicals of Oxalis corniculata and observed the presence of oxalic acid, tannins, palmitic acid, a mixture of various fatty acids, calcium, fiber, calcium oxalate, flavones (acacetin and 7,4'-di-O-Me apigenin), glycoflavones(4'-O-Me vitexin, 4'-O-Meiso-vitexin and 3',4'-di-O-Me orientin), flavonols (3',4'-di-OMe quercetin) and phenolic acids such as p hydroxybenzoic, vanillic and syringic acids. Traditional uses of this plant were enlisted as an antiscorbutic in the treatment of scurvy, in stomach trouble, in case of scorpion sting, to stop bleeding from wounds, to get relief from aphthae, to treat giddiness, diarrhoea and dysentery. Apart from, various pharmacological investigations enumerate its antimicrobial, antifungal, wound healing, antiimplantation, abortifcient, cardiorelaxant and nematocidal activities.
**Kumar and Kapoor (2010)** scrutinized the increasing effects of antioxidant enzymes levels such as superoxide dismutase (SOD), glutathione peroxidise (GSH-POX), glutathione reductase (GR) and catalase (CAT) in rat brain after methanolic extract of *Oxalis corniculata* (MEOC) treatment at the doses of 200 & 400mg/kg BW. Inversely lipid peroxidation (LPO) decreased in MEOC treated rats. Hence the antioxidant properties of MEOC extract delays the generation of free radical in MES & PTZ induced epilepsy.

**Kathiriya et al. (2010)** reported that ethanolic extract of *Oxalis corniculata* (EEOC) had significant antitumor and antioxidant activities in Ehrlich Acsites Carcinoma (EAC) bearing mice. The dose dependent reduction in body weight, tumour volume, packed cell volume, tumour cell counts and increase in median survival time (MST) and percentage increase in life span were observed. They also found significant increase in RBC count; haemoglobin content, total protein and albumin content, but decrease in total WBC count, AST, ALT and ALP contents in EEOC treated animals. Also a significant decrease in liver MDA levels and increase in catalase and reduced glutathione levels were observed in EEOC treated animals.

**Sakat et al. (2010)** studied antioxidant and anti-inflammatory activity of methanolic extract of whole plant of *Oxalis corniculata* L. in Male Sprague-Dawley rat model. Antioxidant activity was assaying using 1, 1-Diphenyl-2-Picrylhydrazyl (DPPH) and nitric oxide radical scavenging activity, while anti-inflammatory activity was evaluated using albumin denaturation assays, membrane stabilization assay and proteinase inhibitory activity at different concentrations. Aspirin was used as a standard drug for the study of anti-inflammatory activity. Results showed that, the extract exhibited significant DPPH and nitric oxide radical scavenging activity with IC50 value of 302.93±4.17 and 73.07±8.28 μg/ml respectively. Extract also showed *in vitro* anti-inflammatory activity by inhibiting the heat induced albumin denaturation and Red Blood Cells membrane stabilization with the IC50 values of 288.04±2.78 and 467.14±9.56 μg/ml respectively.

**Vhuiyan et al. (2010)** studied antioxidant and membrane stabilizing activities of ethanolic extract of the whole plant of *Oxalis corniculata* using various methods including free radical, hydrogen peroxide, nitric oxide scavenging and phosphomolybdenum antioxidant assay. It was revealed that the methanolic extract of *O. corniculata* had moderate antioxidant activity and significant membrane stabilizing property.

**Jyothi et al. (2011)** screened different solvent extracts of *Oxalis corniculata* with two other plants was tested for α-amylase inhibition in order to evaluate their inhibitory potential on porcine pancreatic α-amylase that regulate postprandial hyperglycemia (PPHG) which is of
major concern in Type -2 diabetes. Results showed that aqueous extract of *Oxalis corniculata* exhibited 89.87% (100μg/ml, IC₅₀ = 68.08±0.06) inhibition of α-amylase activity. The other extracts of the plants showed inhibition, but not statistically significant. Thus, this extract showing potent inhibition might prove to be efficient sources for the extraction of natural α-amylase inhibitors.

Imran *et al.* (2012) showed that ethanolic extract of Oxalis corniculata attenuated anxiety parameters in the open-field and plus-maze tests and also inhibited foot shock-induced fighting behavior, which supported its medicinal behavior.

2.13.2 *Averrhoa bilimbi* L.

Pushparaj *et al.* (2000) applied an oral glucose tolerance test (OGTT) in both normoglycaemic and streptozotocin-induced diabetic rats and observed an optimal hypoglycaemic effect at a dose of 125 mg/kg of ethanolic extract obtained from *Averrhoa bilimbi* L. (bilimbi) leaves. Repeated administration (twice a day) of a dose of 125 mg/kg further reduced glycaemia in diabetic rats by 50% and blood triglyceride by 130% when compared with vehicle (water).

2.13.3 *Averrhoa carambola* L.

Chau *et al.* (2004) reported that fruits of *A. carambola* L. possess several insoluble fiber-rich fractions (FRFs) including insoluble dietary fiber, alcohol-insoluble solid, and water-insoluble solid dietary fibers which adsorb glucose and thus retard glucose diffusion, postpone the release of glucose from starch, and inhibit the activity of α-amylase to different extents and help control postprandial serum glucose. It also possessed hypocholesterolaemic and hypolipidaemic activities. He also observed hypoglycemic effects of these insoluble FRFs were significantly (P <0.05) stronger than that of cellulose.

Shui and Leong (2006) studied the residue of star fruit, which was found to contain much higher antioxidant activity than the extracted juice using several methods for assessing antioxidant activity. Under optimized extraction conditions, the residue accounted for around 70% of total antioxidant activity (TAA) and total polyphenolic contents, however only contributed 15% of the weight of whole fruit. Freeze-dried residue powder, which accounted for around 5% of total weight, had total polyphenolic content of 33.2 ± 3.6 mg gallic acid equivalent (GAE)/g sample and total antioxidant activity of 3490 ± 310 and 3412 ± 290 mg L-ascorbic acid equivalent antioxidant capacity (AEAC) or 5270 ± 468 and 5152 ± 706 mg trolox equivalent antioxidant capacity (TEAC) per 100 g sample obtained by 2,2'-azino-bis-(3-ethylbenzthiazoline-6-sulfonic acid) free radical (ABTS(·+)) and 1,1-diphenyl-2-picryl-hydrazyl (DPPH(·)) scavenging assays,
respectively. It was also found to have $510.3 \pm 68.1$ mol ferric reducing/antioxidant power (FRAP) per gram sample. The residue extract also shows strong antioxidant activity in delaying oxidative rancidity of soya bean oil at 110 degrees C. Antioxidant activity and polyphenolic profile of residue extracts were compared with extracts of standardized pyconogenol. The high content of phenolics and strong antioxidant activity of residue extracts indicate that residue powder may impart health benefits when used in functional food products and that residue extracts should also be regarded as potential nutraceutical resources in future.

2.13.4 Biophytum sensitivum L.

Puri and Baral (1998) revealed the hypoglycaemic effect of leaves extract of *Biophytum sensitivum* in alloxan induced diabetic male rabbits. They observed fall in fasting plasma glucose level and improvement in the OGTT after giving single dose and prolonged administrations in alloxan induced diabetic male rabbits. There was fall in 1 and 2.5 h glucose values by 26 % and 27 % found respectively in the sub diabetic rabbits and by 37 % and 38 % in the mildly diabetic rabbits after single dose administration. More significant improvements occurred following one week of the above treatment.

2.13.5 Biophytum condolleanum L.

Prakash et al. (2011) reported that whole plant ethanolic extract of the *B. condolleanum* Wight contains higher level of total tannis and flavonoids and showed its potential antioxidant activities against DPPH radicals with IC$_{50}$ value 43.10±7.20 µg/ml.

2.14 Antidiabetic and antioxidative properties of plants of Euphorbiaceae family

2.14.1 Phyllanthus fraternus L.

Rehman et al. (2004) reported that the genus Phyllanthus comprises 700 species. Only 10 species, *P. acidus*, *P. emblica*, *P. fraternus*, *P. maderaspatensis*, *P. parvifolius*, *P. reticulats*, *P. rotundifolius*, *P. urinaria* and *P. virgatus* are found with application in the folk medicine system for the treatment of jaundice and liver ailment. Several active chemical constituents have been isolated from different species of the genus Phyllanthus. The two flavonoids designated as FG-1 and FG-2, isolated from *P. fraternus*, exhibit oral hypoglycaemic activity in alloxan treated Diabetic rats. The mean reduction of blood sugar was found to be about 20% with FG-1 and FG-2.
Oudhia (2008) scripted that *Phyllanthus fraternus* Webster belonging to Family Euphorbiaceae commonly called as gulf leaf-flower, bhoomi amalaki, bhui-ama, Chanca piedra, quebra pedra, and stone breaker; probably originates from Pakistan and western India. Leaves contain the lignans, niranthin, nirtetralin and phyltetralin while other compounds isolated from the plant include alkamides (2, 4-octadienamide and 2, 4-decadienamide), a quinolizidine alkaloid (norsecurinine), the flavone tricin, triterpenoids (friedelin, epifriedelinol, kokoonol and sorghumol), the tetraterpenoid phyllanthusone, and waxes (octacosane, tetracosyl alcohol, tricosyl alcohol). Some of the alcohols are also present as esters, e.g. phyllanterpenyl ester and pentacosanyl ester. An alcohol extract of the root contained the seco-sterols phyllanthosterol, phyllanthosecosteryl ester, phyllanthostigmasterol and fraternosterol. The seed oil contains ricinoleic acid, linoleic acid and linolenic acid.

Matur et al. (2009) observed intrinsic antimalarial activity of *Phyllanthus fraternus* plant by its percentage chemo suppression and even curative ability compared to that of chloroquine which is the standard drug.

Garg et al. (2010) studied the effects of standardized *Phyllanthus fraternus* alcoholic extract at a single dose of 500 mg/kg BW for 21 days in alloxan induced albino rats. As a result, drug treatment has significantly improved the disturbed biochemical parameters at variable degrees when compared with standard drug tolbutamide at a dose of 200 mg/kg BW. The phytochemical studies conducted for standardization of the extract showed the presence of tannins and flavonoids as major phytoconstituents. The total phenolics content was found to be 37.51 mg/g of drug extract. Quantitative estimation carried out on two major flavonoids by HPTLC confirmed a concentration of 1.706% w/w rutin and 5.614% w/w of quercetin present in the alcoholic extract. In conclusion, owing to the positive potential activity against disturbed biochemical parameters associated with diabetes, *P. fraternus* can be used effectively in the management of this deadly disease.

Kushwah et al. (2010) observed that treatment with aqueous extract of *Phyllanthus fraternus* at the dose of 250 mg/kg BW to fructose fed induced hyperinsulinemic rats was found to significantly (p<0.01) preventive to hypertriglyceridemia, hyperglycemia, hyperinsulinemia and hypertension.

Koffuor and Amoateng (2011) evaluated the antioxidant and anticoagulant activity of an ethanolic extract of *Phyllanthus fraternus* (gulf leaf-flower) using *in vitro* and *in vivo* experimental models. The results obtained indicate that the ethanolic extract of *P. fraternus* exhibited antioxidant activity by significantly scavenging 2, 2-diphenyl-1-picrylhydrazyl
(DPPH) radicals, concentration-dependent reducing capacity and inhibition of lipid peroxidation. Detection of phenols in the extract gave preliminary evidence of its possible antioxidant activity which correlated with the total antioxidant capacity.

2.14.2 Phyllanthus amarus L.

Sharma et al. (1993); Joseph et al. (2011) reported that Phyllanthus amarus, a species of Phyllanthus genus have hepatoprotective, antiseptic, antitumor, antidiabetic, antilipidemic, antihypertensive, analgesic, anti-inflammatory and antimicrobial properties. It contains lignans like phyllanthine and hypophyllanthine, geraniin and 5 flavonoids (quercertin, astralgin, quercertrin, isoquercitrin and rutin). It also contains minor compounds like hydrolysable tannins like phyllanthusiin D, amariin, amarulone, amarinic acid and alkaloids like ent-norsecurinine, sobubbialine, nyrophyllin and a neolignan, phyllnirurin.

Srividya and Periwal (1995) performed a clinical trial on nine mild hypertensive patients of Diabetes Mellitus with a preparation of the whole plant extract of Phyllanthus amarus for 10 days. The observations indicated that Phyllanthus amarus preparation may act as a potential diuretic hypotensive and hypoglycaemic drug for human.

Sivaprakasam et al. (1995) observed in another clinical trial in which 25 patients of age group of 35 to 55 years with moderate and severe diabetic blood sugar level (250-400 mg/100 mL) showed statistically significant (p<0.05) lowering of blood sugar levels after intake of a preparation of whole plant of Phyllanthus amarus at a dose of 1 g thrice daily for a period of 3 months.

Lim and Murtijaya (2007) found that hot aqueous extract of Phyllanthus amarus had more antioxidant activity than dried plant materials. The antioxidant activity of fresh and dried Phyllanthus amarus were exhibited by the reduction in both free radical scavenging activity and Ferric Reducing Antioxidant Power (FRAP).

Karuna et al. (2009) observed significant decrease (11.9%) in plasma lipid per oxidation (LPO) and increase in plasma vitamin C (28.6%), uric acid (35.7%) and plasma GSH (41%) levels in normal rats after treatment with ethanolic extract of Phyllanthus amarus compared to control rats. Also there was found significant enhancement in the activities of plasma GSHPx (13.4%), CAT (28.4%) and SOD (25.18%) in ethanolic extract of Phyllanthus amarus treated animals compared to control animals.
2.14.3 *Phyllanthus urinaria* L.

Higashino *et al.* (1992) observed the decrease in elevated blood glucose level in streptozotocin induced diabetic rats after oral administration of methanolic extract of *Phyllanthus urinaria* L. at dose of 30mg/kg body weight. In the oral glucose tolerance test, the n-butanol fraction of this plant also inhibited the initial increase of blood glucose level. *P. urinaria* extract may act via the facilitation of glucose metabolism and/or the inhibition of glucose absorption in the gut like the action of biguanides.

2.14.4 *Acalypha torta* L.

Patricia *et al.* (2011) screened the phytochemicals in methanolic extract of *Acalypha torta* leaves and revealed the presence of alkaloids, flavonoids, tannins, resins, glycosides, saponins and carbohydrates. Further the crude methanolic extract was partitioned successively in n-hexane, ethyl acetate and butanol to give different fractions and toxicity test was carried out by Brine shrimp lethality test (cytotoxicity) which resulted the lethal doses: LC$_{50}$ of 6.9030 g/ml (hexane fraction), 45.0958 g/ml (ethyl acetate fraction), 0.7210 g/ml (butanol fraction) and 0.0002 g/ml (methanol), indicating their toxicity. The free radical scavenging activity of A. torta was determined by scavenging effect on 2,2-diphenyl-1-picrylhydrazyl radical (DPPH), hydroxyl radical and peroxide oxidation by ferric thiocyanate method. Comparison of the results obtained with the three antioxidant standards used in the assay revealed that the fractions possessed antioxidant activity. Butylated hydroxyl anisole (BHA), ascorbic acid and a-tocopherol were used as reference standards. The results obtained support the ethno medicinal applications of *Acalypha torta*.

Masih *et al.* (2011) studied the antidiabetic effects of the methanol and acetone (70:30) extract of *Acalypha indica* L. in normal and Alloxan induced diabetic model. Decreased blood glucose level of the test animals showed that the extract exhibit significant antidiabetic activity when compared to diabetic control group. The results also indicated the dose dependent effect. The antidiabetic activity produced by the extract may be due to increased uptake of glucose at the tissue level or by an increase in pancreatic beta cell function or due to inhibition of intestinal absorption of glucose. The present study supports the use of this herbal drug as antidiabetic.

2.14.5 *Croton cajucara* L.

Farias *et al.* (1997) isolated the trans-Dehydrocrotonin (t-DCTN), a 19-nor-clerodane diterpene chemical compound from the bark of *Croton cajucara* Benth. (Euphorbiaceae) and observed significant hypoglycemic activity in alloxan-induced diabetic rats but not in normal rats, at oral
doses of 25 and 50 mg/kg body weight. The drug also effectively lowered the blood sugar levels in glucose fed normal rats. The hypoglycemic effect of t-DCTN was almost comparable to that produced by glibenclamide (2 mg/kg), a clinically useful drug.

**Okokon et al. (2006)** evaluated the antidiabetic activity of ethanolic leaf extract of *Croton zambesicus* was evaluated using alloxan-induced (150mg/kg) hyperglycaemic rats. The activity of the ethanolic extract of leaves was compared with that of a reference drug Chlorpropamide. The Blood Glucose Levels (BGL) was measured using glucometer. The extract was found a significantly (P<0.01) reductive in BGL after a single dose of the extract and in prolonged treatment (for 7 days). The antidiabetic activity was comparable to that of the reference drug-chlorpropamide.

**2.14.6 Emblica officinalis L.**

Christi and Meona (2013) scripted that *Emblica officinalis* fruits (commonly known as ‘Amla’) are the natural source of vitamin C. The Ascorbic acid present in one fruit is equivalent to that present in two oranges. Its fruits may be used as cooling, refrigerant, diuretic, and laxative and also used for anemia, hepatopathy, jaundice, diarrhea, hemorrhages, leucorrhoea, cardiac disorder and antioxidant.

**Nain et al. (2012)** evaluated the hypoglycemic and antioxidants effects of the hydro-methanolic (20:80) extract of leaves of *Emblica officinalis* Gaertn. (HMELEO) in streptozotocin induced diabetic rats. The hypoglycemic effect was measured by blood glucose and plasma insulin level while oxidative stress was measured in liver and kidney by assay of level of antioxidant markers i.e. lipid peroxidation (LPO), superoxide dismutase (SOD), reduced glutathione (GSH), glutathione peroxidase (GSH-POX) and catalase (CAT), and the others biochemical parameters like blood serum levels of creatinine, urea, SGPT, SGOT, ALP, total cholesterol and triglyceride levels were also observed in diabetic control and treated rats. Results showed that oral administration of the HMELEO at a concentration of 100, 200, 300 and 400 mg/kg BW daily for 45 days possesses a significant (P<0.05) decrease in fasting blood glucose and increase insulin level and all biochemical parameters (serum creatinine, serum urea, SGOT, SGPT and lipid profile) as compared with the diabetic rats. The treatment also resulted in a significant (P<0.05) increase in reduced glutathione, glutathione peroxidase, superoxide dismutase, catalase, and decrease LPO level in the liver and kidney of diabetic rats. Thus it is cleared that the hydro methanolic extract of leaves of *Emblica officinalis* may effectively normalize the impaired antioxidant status in streptozotocin induced diabetes at dose dependent manner than the glibenclamide-treated groups.
Mehta et al. (2009) screened antidiabetic activity of aqueous extract of Emblica officinalis Gaertn. (syn: Phyllanthus emblica L.) (Euphorbiaceae) seeds in Streptozotocin (STZ)-induced type 2 diabetes animal models. The standardized doses of 100, 200, 300, and 400 mg kg\(^{-1}\) body weight of the extract were administered orally to normal and diabetic rats in order to define its glycemic potential. The maximum fall of 27.3% (\(p < 0.001\)) in the blood glucose level of normal rats was observed at 6 h during fasting blood glucose studies, with the dose of 300 mg kg\(^{-1}\) identified as the most effective dose. The same dose produced a fall of 25.3% (\(p < 0.001\)) in the same models during the glucose tolerance test (GTT) at 3 h after glucose administration. However, the dose of 300 mg kg\(^{-1}\) of aqueous seed extract in sub- and mild-diabetic animals produced a maximum fall of 34.1 and 41.6% (\(p < 0.01\)), respectively, during the GTT at 3 h after glucose administration. This evidence clearly indicates that the aqueous extract of Emblica officinalis seeds has hypoglycemic potential as well as anti-diabetic property.

2.14.7 Euphorbia hirta L.

Kumar et al. (2010) studied the antidiabetic and in vitro free radicals scavenging effects of flower extract of Euphorbia hirta. The ethanolic and petroleum ether extracts (250 and 500 mg/kg) were orally tested for 21 days in alloxan induced diabetic mice and blood glucose level was measured with glucometer. Administration of extract resulted in significant reduction in serum cholesterol, triglycerides, creatinine, urea, alkaline phosphatase levels but high density lipoprotein levels and total proteins were found to be increased after treatments. Free radicals scavenging effect of ethanolic extract was also evaluated by 1, 1-diphenyl-2-picryl hydrazyl (DPPH) free radical scavenging activity, superoxide anion radical scavenging, nitric oxide scavenging, and reducing power assay. It was compared with standard antioxidants compounds such as butylated hydroxyl anisole and ascorbic acid. All the extracts showed good antioxidant activity in all the tested methods.

2.14.8 Euphorbia prostrate L.

Alarcon-Aguilar et al. (1998) studied the hypoglycaemic properties of Euphorbia prostrate using hyperglycaemic rabbits models and observed that Euphorbia prostrate have hypoglycaemic activity in hyperglycaemic rabbits.
2.15 Antidiabetic and antioxidative properties of plants of Cucurbitaceae family

2.15.1 *Trichosanthes cucumerina* L. (Vern. Chichinga or, Snake Tomato)

Adebooye (2008) scripted that *Trichosanthes cucumerina* L. (locally known as Snake Tomato) (Cucurbitaceae family) are usually consumed as a vegetable due to its good nutritional value. Its fruit pulp contains ascorbic acid, lycopene, phenolics, carotenoids, flavonoids as natural antioxidant power, that’s why it is recommended to use in curing of many of disease as folk medicine.

Kiran and Srinivasn (2008) reported that aqueous extract of *Trichosanthes cucumerina* significantly decreased the elevated blood glucose in NIDDM induced rats. The extract has significantly reduced the post prandial blood glucose of diabetic animals. Glycogen content of insulin dependent tissue such as liver and skeletal muscle was found to be improved by 62% and 58.8% respectively with *Trichosanthes cucumerina* as compared to NIDDM control.

Ojiako and Igwe (2008) reported that *Trichosanthes anguina* ripe fruits were found having nutritive and anti-hepatotoxic properties and also presence of saponins, flavonoids, cyanogenic, cardiac glycosides, vitamin A and vitamin C. Among different status of fruits such as raw fruits, cooked fruits, cooked fruits without seeds and cooked seeds, consumption of raw fruit had more weight loss, lowered enzymatic activity of ALP, AST and ALT than cooked fruit. It indicated that *T. anguina* fruits contain important nutrients and will not be hepatotoxic unless consumed raw or unprocessed.

Devendra et al. (2009) studied acute toxicity of whole plant ethanolic extract of *Trichosanthes cucumerina* L. and found no mortality, changes in the behaviour or any other physiological activities in treated mice. They recorded no mortality and significant differences in the body and organ weights between controls and treated rats in chronic toxicity studies. Hematological analysis showed no significant differences in any of the parameters examined (like RBC, WBC count and haemoglobin estimation). Further it also showed the antiovulatory activity by reducing female hormone in female albino rats.

Arawwawala et al. (2009) used hot aqueous extract of aerial parts of *Trichosanthes cucurmerina* to improve glucose tolerance and tissue glycogen in non insulin dependent diabetes mellitus induced rats. They observed that drug possessed antidiabetic activity with improvement in oral glucose tolerance and glucose uptake in peripheral tissues.
Sathesh et al. (2009) observed that methanolic extract of the whole plant of *Tricosanthes cucumerina* had good hepatoprotective activity against carbon tetrachloride induced hepatotoxicity.

2.15.2 *Trichosanthes lobata* L.

Rajsekaran and Periyasamy (2012) studied the hepatoprotective activities of ethanolic extract of *Trichosanthes lobata* against paracetamol-induced hepatotoxicity. Hepatotoxicity was induced in wistar male rats by oral administration, 2 g/kg body weight on 7th day after the administration of ethanolic extract of *Trichosanthes lobata* and silymarin (100 mg/kg). Ethanolic extract of *Trichosanthes lobata* was administered orally at doses of 200 mg/kg and 400 mg/kg body weight daily for 7 days. Several serum markers, aspartate transaminase, alanine transaminase, alkaline phosphatase, bilirubin, total protein was measured to assess the effect of the extract on paracetamol (acetaminophen)-induced hepatic damage. The study included histopathological examination of liver sections. Blood samples from rats treated with ethanolic extract of *Trichosanthes lobata* (200 mg/kg body weight and 400 mg/kg body weight) had significant reductions in serum markers in paracetamol administered animals, indicating the effect of the extract in restoring the normal functional ability of hepatocytes. Silymarin (100 mg/kg, p.o.) was used as a reference drug.

Jose et al. (2010) examined ethyl acetate, chloroform and methanol extracts of leaves of *T. cucumerina* for antibacterial activity against the different strains of bacteria and found to be quiet comparable with the standard antibiotics.

Devendra and Seetharam (2011) reported that *Trichosanthes cucurmerina* are widely used for curing jaundice, aphrodisiac, cardiotoic, skin disease, fever and other maladies. A number of pharmacologically important phytochemicals, such as cucurbitacins and sterols, have been isolated and seem to cure them.

2.15.3 *Trichosanthes dioica* L. (Vern. Parwal)

Sharma and Pant (1992) showed influence of alcoholic extract of whole fruit of *T. dioica* (commonly known as parwal) on blood sugar, serum lipids, lipoproteins and faecal sterols in normal albino rabbits.

Sharmila et al. (2007) observed cholesterol lowering activity of the aqueous extract of *Trichosanthes dioica* fruits in normal and streptozotocin diabetic rats.
**Rai et al. (2008)** reported that aqueous extract of *Trichosanthes dioica* leaves had good hypoglycemic potential in high antidiabetic profile when administered orally to normal and streptozotocin (STZ) induced sub- and mild-diabetic rats at variable doses of 250, 500, and 750 mg kg$^{-1}$ body weight. Further aqueous extract of *T. dioica* fruits showed decrease in levels of fasting blood glucose, postprandial glucose, aspartate amino transferase, alanine amino transferase, alkaline phosphatase, creatinine, urine sugar and urine protein whereas increase in total protein and body weight when administered at a dose of 1000mg/kg body weight daily once for 28 days to streptozotocin induced diabetes rats.

**Shivhare et al. (2010)** found significant anti-oxidant activity of aqueous extract of *Trichosanthes dioica* fruits against free radicals in different in-vitro methods and suggested this plant to be used in the management of antioxidant activity.

**Alam et al. (2011b)** also reported antioxidant properties as well as anti-inflammatory, anti pyretic properties of methanolic extracts and other soluble fractions of fruit of *T. dioica*, which was suggested to be due to presence of phenolics compounds and flavonoids in fruits.

**2.15.4 Momordica charantia L. (Vern. Karela)**

**Day et al. (1990)** observed that aqueous extract of *M. charantia* L. (Vern. Karela), reduce hyperglycaemia in diabetic mice after 1 h without altering insulin response. It was proposed that the hypoglycaemia activity of orally administered Karela extracts was independent of intestinal glucose absorption and involved in an extra pancreatic effect.

**Higashino et al. (1992)** reported that methanolic extract of *M. charantia* had lowering effect on increased blood glucose level after 3 hours of oral administration of glucose to streptozotocin-induced diabetic rats at a dose of 30 mg/kg BW. They also found that n-butanol soluble fraction from *M. charantia* extract was most effective in lowering BGL after intraperitoneal glucose load. Like the action of sulfonylureas, the *M. charantia* extract seems to act like insulin or via insulin secretion from the pancreas.

**Sarkar et al. (1996)** found that alcoholic extract of Karela fruit reduce the plasma glucose level in glucose loaded primed rats within 1 hour at dose of 500 mg/kg BW. Synthetic drug Tolbutamide produced the same effect under similar conditions at the dose of 100 mg/kg BW. This reduction in plasma glucose levels was not accompanied by increased insulin secretion. Further they also observed its significant improving effect of OGTT in streptozotocin-induced diabetic rats. The *M. charantia* extract caused a 4-5 fold increase in the rate of glycogen synthesis from U-14C glucose in the livers of normally fed rats.
Raza et al. (1996); Sitasawad et al. (2000) evaluated the effect *M. charantia* fruits juice on hepatic cytochrome P450 and glutathione S-transferase drug-metabolizing enzymes in streptozotocin induced diabetic rats. It was suggested that the changes in hepatic phase I and phase II drug-metabolizing enzyme activities in the diabetic animals may be associated with altered expression of different cytochrome P450 and glutathione S-transferase isozymes. In addition, Feeding of *M. charantia* fruit juice produced a reduction in hyperglycaemia in STZ-induced diabetic mice. It strongly reduced the STZ-induced lipid peroxidation in the pancreas of mice.

### 2.15.5 *Momordica cymbalaria* L.

Rao et al. (2001) reported that *Momordica cymbalaria* (Hook fruit) powder produced a significant blood glucose lowering effect in alloxan-induced diabetic rats, but not in normoglycaemic rats. The fruit powder was also found to reduce the level of cholesterol and triglycerides in diabetic rats.

### 2.15.6 *Coccina indica* L.

Hossain et al. (1992) observed that ethanolic extract of leaves of *Coccina indica* had decrement in glycaemia in normal-fed (21 %) and 48 h fasted rats (24 %). This effect was imparted due to the inhibition of the key gluconeogenic enzyme glucose-6-phosphatase.

Kumar et al. (1993) reported that oral administration of the pectin isolated from the fruit of the *C. indica* at a dose of 200 mg/100 g BW/day produced a reduction in glycaemia and an increase in liver glycogen. It was probably due to significant increase in glycogen synthetase activity and reduction in phosphorylase activity was noted in the pectin-administered groups.

Balaraman et al. (2010) studied the antihyperglycemic and hypolipidemic effects of ethanol extract of aerial parts of *Melothria maderaspatana* (EEMm) and *Coccinia indica* (EECi) in STZ induced diabetic Sprague–Dawley rats. Rats were concurrently treated with 100 or 200 mg/kg BW p.o. for 14 days and changes in fasting blood glucose level and body weight were measured in 5 days interval. After 14 days, blood and liver samples were collected and biochemical estimation of plasma for glucose level, cholesterol, triglycerides, LDL, HDL, SGOT, SGPT and ALP were done. The liver glycogen content was estimated using standard procedure from homogenized liver sample. Administration of EEMm or EECi to STZ-diabetic rats caused significant antihyperglycemic and hypolipidemic effects (p< 0.001). The extracts were also found to be significantly effective (p< 0.001; p< 0.05) on recovery of altered biochemical
parameters and decreased body weight in treated animals. Glibenclamide (0.5 mg/kg BW) was used as standard.

2.15.7. *Bryonia alba* L.

Karagenzyan *et al.* (1998) observed that the administration of tri-hydroxy-octa-deca-dienoic acids obtained from the roots of the native Armenian plant *Bryonia alba* at a dose of 0.05 mg/kg/day for 15 days intra muscular, restored the disordered lipid metabolism of alloxan-diabetic rats and reduced thromboxane B2 generation with a corresponding increase in prostaglandin E2 release.

Singh *et al.* (2012) studied new avenues for the improvement of medicinal uses of *Bryonia alba* L. (Cucurbitaceae) for the selected area of diabetes. Alcoholic extracts of dried roots of *Bryonia alba* were subjected for hypoglycaemic activity in wistar rats (150-200 g). Blood sugar level was determined using digital glucometer. The oral administrations of extracts at doses of 200 mg / kg lead to a significant blood glucose reduction. This laid the foundation for further study the active compounds of such plants that are responsible for the hypoglycemic activities.

Vartanian *et al.* (1984) conducted an experiment on white rats with alloxan induced diabetes that have tri-hydroxy-octa-deca-diene acids isolated from *Bryonia alba* L. and screened for hypoglycemic action, activity of glycogen phosphorylase (a- and b-forms), phosphoprotein phosphatase and hexokinase in liver and muscle tissues of white rats with alloxan diabetes. One of the possible mechanisms of the hypoglycemic action of tri-hydroxy-octa-deca-diene acids was discussed. Results showed possibilities of this compound in improvement of above parameters.

2.15.8 *Citrullus colocynthis* L.

Abdel-Hassan *et al.* (2000) reported that *Citrullus colocynthis* Schrad fruits showed insulinotropic effect, when different extracts of its seeds perfuse for 20 min at 0.1 mg/ml. It was found immediately and significantly induced insulin secretion *in vitro* in the isolated rat pancreas and isolated rat islets in the presence of 8.3 mM glucose.

Jayaraman *et al.* (2009) studied the effect of petroleum ether extract of *Citrullus colocynthis* fruits on lowering the blood glucose levels and thiobarbituric acid reactive substances (TBARS) in streptozotocin induced diabetic albino rats. Oral administration of two different doses (300 & 500 mg/kg p.o) of *Citrullus colocynthis* fruit extract exhibited a significant reduction in blood glucose level in diabetic rats. In addition, extract significantly reduced TBARS levels when compared to diabetic control groups. Glibenclamide (0.5 mg/kg) was used as reference drugs.
Thus the petroleum ether extract of *Citrullus colocynthis* fruits might be promising for the development of a standardized phytomedicine for the treatment of diabetes mellitus.

**Agarwal et al. (2012)** observed the effect of root of *Citrullus colocynthis* on the biochemical parameters of normal and alloxan-induced diabetic rats. The blood glucose concentrations of the animals were measured at the beginning of the study and the measurements were repeated on 3rd, 5th and 7th day after the start of the experiment. On day 7, blood was collected by cardiac puncture under mild ether anesthesia. Aqueous extract of roots of *Citrullus colocynthis* showed significant reduction in blood sugar level (58.70%) when compared with chloroform (34.72%) and ethanol extracts (36.60%) (p < 0.01). The aqueous extracts showed improvement in parameters like body weight, serum creatinine, serum urea and serum protein as well as lipid profile and also restored the serum level of bilirubin total, conjugated bilirubin, serum glutamate oxaloacetate transaminase (SGOT), serum glutamate pyruvate transaminase (SGPT) and alkaline phosphatase (ALP).

**Salami et al. (2004)** reported that infusion of *Citrullus colocynthis* fruits are traditionally used as antidiabetic medication in Mediterranean countries and screened whether these fruits possess hypoglycemic effects or not. For this, suspension of *C. colocynthis* fruit was administered orally to normal 12 h fasting rats. Blood glucose was determined before and 1, 2, 3, 4, 6, 8, 12, and 24 hours after single oral administration of prepared drug at the doses of 10, 30, 90 mg/kg. Decreased level of BG were found by 18%, 24% at 4, 6 h, as compared with the control animals at the dose of 30 mg/kg. Such effects of drug were not pronounced in the doses of 10 mg/kg, 90 mg/kg at 0, 4, 6, and 12 h. It was concluded that *Citrullus colocynthis* fruit suspension have a hypoglycemic effect in the dose of 30 mg/kg, which could at least partially account for the antidiabetic activities of these fruits.

### 2.15.9 *Cucumis sativus* L.

**Karthiyayini et al. (2009)** screened the antidiabetic activity of powder fruit of *Cucumis sativus*. They observed different doses of ethanol extract of *C. sativus* fruit powder for their effects on serum glucose levels in streptozotocin-induced rats and lipid profile (biochemical parameters) in blood were also observed with histopathological studies. The oral administration of 200 and 400 mg/kg body weight of ethanol extracts of fruit of *Cucumis sativus* exhibited significant antidiabetic effects in streptozotocin induced rats as compared to standard drug. In the same study the action of the extracts on diabetes induced hyperlipidemia was analyzed where the extracts significantly lowered the elevated cholesterol as well as LDL level. The drug has the potential to act as antidiabetic as well as antihyperlipidemic at dose of 400 mg/kg body weight.
Minaiyan et al. (2011) studied the effect of hydro alcoholic and butanolic extract obtained from C. sativus seeds in a model of streptozotocin (STZ)-induced diabetic (type I) rats. They observed that C. sativus seeds extracts were not effective on reducing blood glucose levels (BGL) in normal and diabetic rats for initial phase of treatments. However, both hydro alcoholic (22.5-33.8 %) and butanolic (26.6-45.0 %) extracts were effective on diminishing BGL and controlling the loss of body weight in diabetic rats compared to controls after 9 days of continued daily therapy. Glibenclamide on the other hand, had hypoglycemic action in normal (27.8-31.0 %) and diabetic rats (36.0-50.0 %) after acute and prolonged treatments. They concluded that C. sativus seeds extracts (hydro alcoholic and butanolic) had a role in diabetes control probably through a mechanism similar to euglycemic agents.

Heidari et al. (2012) studied hepatoprotective effect of aqueous extract of Cucumis sativus fruit against cytotoxicity and reactive oxygen species (ROS) production, using accelerated cytotoxicity mechanisms screening (ACMS) techniques in isolated Sprague–Dawley rat hepatocytes as a cellular model. Fresh fruits of Cucumis sativus were cleaned, and then dried in shade at room temperature and aqueous extract of the fruit was obtained. Hepatocytes were obtained by collagenase perfusion of the liver and viability was assessed by plasma membrane disruption determined by trypan blue (0.2 w/ v) exclusion test. The rate of hepatocyte reactive oxygen species (ROS) generation was determined by induction of cumene hydroperoxide, the dichlorofluorescin diacetate (DCFH-DA) was added to the hepatocytes, which reacts with ROS to form the highly fluorescent dichlorofluorescein (DCF), which effluxes the cell. The fluorescence intensity of DCF was measured. Results showed that aqueous extract of Cucumis sativus acts as a hepatoprotective and antioxidant agent against CHP-induced hepatotoxicity suggesting that antioxidants and radical scavenging components of Cucumis sativus fruit extract can easily cross the cell membrane and cope with the intracellular ROS formation.

2.15.10 Cucurbita ficifolia L.

Acosta-Patino (2001) studied hypoglycemic activity of raw extract of Cucubita ficifolia against control in type 2 diabetic patients at a single oral dose of 4 ml/kg BW, in two different sessions at least separated by 1 week. Blood glucose levels were followed in both parts of the study by a period of 5 h. The patients had stopped their pharmacologic medication 24 h prior to each part of the study. He observed no significant changes on blood glucose after giving the vehicle; however, the oral administration of C. ficifolia was followed by a significant decrease in blood glucose levels, from 12.07+/−1.69 mM (217.2+/−30.4 mg/dl) to 9.42+/−1.96 mM (169.6+/−35.3 mg/dl) 3 h after and to 8.37+/−1.74 mM (150.8+/−31.3 mg/dl) 5 h after the extract administration.
The hypoglycemic action of *C. ficifolia* agrees with its effects previously observed in laboratory animals.

**Alarcon-Aguilar et al. (2002)** studied hypoglycemic effects of freeze-dried juice of *Cucurbita ficifolia* (commonly known as Chilacayote) fruits in healthy and alloxan-diabetic mice. *C. ficifolia* fruit was administered by intra peritoneal route, in a dose-dependent manner and observed a significant decrease of the glycemia in healthy mice. In addition, daily oral administration of this preparation at dose of 500mg/kg BW showed a highly significant reduction of the glycemia after 14 days of treatment. Freeze-dried juice caused acute toxicity when administered intraperitoneally and also when it was administered daily by the oral route.

**Roman-Ramos et al. (2012)** studied the influence of D-chiro-inositol-containing fraction from the *C. ficifolia* fruit (AP-Fraction) as hypoglycemic and on biomarkers of oxidative stress, as well as on the inflammatory cytokines in streptozotocin-induced diabetes. The AP-Fraction obtained from the mature fruit of *C. ficifolia* contained 3.31 mg of D-chiro-inositol/g of AP-Fraction. The AP-Fraction was administrated daily by gavage to normal mice for 15 days as a preventive treatment. Then these animals were given streptozotocin, and the treatments were continued for an additional 33 days. Pioglitazone was used as a hypoglycemic drug for comparison. Administration of the AP-Fraction significantly increased glutathione (GSH) and decreased malondialdehyde (MDA) in the liver without significantly affecting the levels in other tissues. The AP-Fraction reduced TNF-α and increased IL-6 and IFN-γ in serum. Interestingly, the AP-Fraction also increased IL-10, an anti-inflammatory cytokine. These results suggested that *C. ficifolia* might be used as an alternative medication for the control of diabetes mellitus and that it has antioxidant and anti-inflammatory properties in addition to its hypoglycemic activity.