Review of Literature
Diabetes mellitus - Early search for the etiology and cure

The glucose appearing in the urine was a mystery in olden days. In 1857, Claude Bernard described glycogen as a product of glucose metabolism in liver and set forward the concept that altered glucose metabolism is the cause of diabetes. In 1869, Paul Langerhans discovered the Islets of Langerhans. A major conceptual breakthrough came from Minkowski and co-workers (Von Mering et al., 1890), whose studies demonstrated that the removal of the pancreas led to diabetes mellitus indicating that the secretions of the pancreas were involved in the etiology of diabetes. In 1921, Banting and Best succeeded in producing a pancreatic extract through duct ligation technique that was pure enough to consistently lower blood glucose and reduce glucose loss in the urine (Banting et al., 1922). They demonstrated that this extract could improve the clinical condition of depancreatized dogs and provided the information on the different modes of administration. With this discovery insulin was established and considered as the only anti-hyperglycemic hormone in the body.

Insulin - a key player in etiology of diabetes mellitus

Insulin is a peptide hormone secreted by β cells of Islets of Langerhans of pancreas. It is a polypeptide containing two chains of amino acids (A chain containing 21 amino acids and B chain containing 30 amino acids) linked by disulfide bridges. The peptide segment connecting the A and B chains is
Pyruvate transporter

Glucose transporter

Glucose

Fatty acids

Pyruvate dehydrogenase

Lipase

FAT CELL

MUSCLE CELL

Glycogen synthetase

Glycogen

Glucose

Alanine

Pyruvate kinase

Glycogen synthetase

Phosphorylase

CO₂
Fig. II

RESPONSE TO INSULIN

- Insulin receptor
- Glucose transporter
- DNA synthesis
- Activation and inhibition of enzymes
- Protein phosphorylation and dephosphorylation

Pre-proinsulin → proinsulin → insulin → insulin-responsive cell
subunits and two β-subunits linked by disulphide bonds. The alpha subunits are entirely extra cellular and house insulin binding domains, while the linked beta subunits penetrate through the plasma membrane (fig. II).

The insulin receptor is a tyrosine kinase. In other words, it functions as an enzyme that transfers phosphate groups from ATP to tyrosine residues on intracellular target proteins. Binding of insulin to alpha subunits causes the beta subunits to phosphorylate themselves (autophosphorylation), thus activating the catalytic activity of the receptor. The activated receptor then phosphorylates number of intracellular proteins, which in turn alter their activity, thereby generating a biological response. Insulin, other hormones, exercise, food and other factors affect the number of insulin receptors and their affinity. Exposure to increased amount of insulin decreases the receptor concentration (down regulation) and exposure to decreased insulin levels increases the affinity of the receptors.

**Role of insulin in carbohydrate metabolism**

Dietary carbohydrates such as starch liberate glucose by hydrolysis in the small intestine, which is absorbed, then resulting in the elevation of the blood glucose levels. This in turn stimulates the pancreatic β cells to release insulin, which acts on different cells throughout the body to stimulate the uptake of glucose. A large amount of glucose is absorbed in the small intestine after a meal and is immediately taken up by the hepatocytes, which store it in the form of glycogen so that glucose can be released to maintain the blood glucose level
during fasting condition. In insulin dependent tissues, insulin increases the uptake and utilization of glucose. These actions of the insulin on glucose metabolism vary depending on target tissue. In the absences of insulin most of the cells (except brain, testis, RBC) are unable to take up glucose and begin to switch for alternative fuels like fatty acid for energy.

The increase in blood glucose in diabetes is mainly due to the result of the impairment in liver and peripheral tissue to metabolize glucose and the activation of glycogenesis in the liver and kidneys (Pilkis et al., 1988). Insulin deficiency impairs the ability of liver to synthesize glycogen by altering the activity of enzymes involved in glycogen metabolism (Hartmann et al., 1987). It was reported that the insulin deficiency causes decreased expression of genes for key enzymes in glucose and ketone bodies metabolism in the liver of diabetic rats (Valera et al., 1993).

**Role of insulin in fat (lipid) metabolism**

The metabolic pathways for utilization of fats and carbohydrates are highly intertwined. Hence, insulin also has important effects on lipid metabolism.

1. When liver is saturated with glycogen, any additional glucose taken up by the hepatocytes is shunted in to pathways leading to the synthesis of fatty acids which are exported from the liver as lipoproteins. These lipoproteins in circulation provide free fatty acids for different tissues including adipocytes, which use them to synthesize triglycerides.
2. Insulin inhibits the intracellular lipase in adipose tissue, thereby preventing the breakdown of fat. So hydrolysis of triglycerides to release fatty acids is inhibited.

3. Insulin facilitates the entry of glucose into adipocytes and within these cells glucose can be used for the synthesis of glycerol. This glycerol with fatty acids, delivered from the liver is used to synthesize triglycerides within the adipocyte. By these mechanisms, insulin is involved in further accumulation of triglycerides in fat cells.

In DM, one of the most common complications is hyperlipidemia, which is seen in about 40% of diabetics (Jaiprakash, 1993). The marked hyperlipidemia that characterizes the diabetic state is due to the uninhibited actions of the lipolytic hormones on fat depots to release the fatty acids (Al-Shamaony et al, 1994) due to lack of insulin.

**Actions of insulin on protein metabolism**

Insulin stimulates the uptake of amino acids in different cells and thereby favors the protein synthesis. This anabolic effect of insulin is important for the positive nitrogen balance. In the absence of insulin, the gluconeogenesis is stimulated in spite of high glucose availability. Due to this the proteins breakdown, as they contain the amino acids required for gluconeogenesis, which leads to a negative nitrogen balance and weight loss. Enhanced catabolism of both liver and plasma proteins in diabetes increases the urea nitrogen production.
Metabolic abnormalities in the liver in uncontrolled diabetes

Fig. III

Activity of pathway

- Greater than normal
- Probably below normal
- Markedly impaired
- Increased gluconeogenesis
Insulin deficiency (and glucagon excess)

- Decreased glucose uptake
  - Hyperglycemia
  - Glycosuria, osmotic diuresis

- Increased Protein catabolism
  - Increased plasma amino acids, nitrogen loss in urine

- Increased lipolysis
  - Increased Plasma FFA, Ketogenesis, Ketonuria, Ketonemia

- Dehydration, acidosis

- Coma, Death

**Effects of insulin deficiency**
Increase in the muscle proteins catabolism elevates serum creatinine levels. In addition to catabolism, diabetes causes decreased protein synthesis in the liver and decrease in mRNA content of skeletal muscle (Jefferson et al., 1983).

**Other actions of insulin**

Insulin increases the permeability of potassium, magnesium and phosphate ions in many cells. This property of insulin is clinically used for temporary relief of hyperkalemia in patients with renal failure (Ganong, 2001). However, hypokalemia often develops when patients with diabetic acidosis are treated with insulin. Insulin activates the sodium potassium ATPase in many cells.

The metabolic derangements due to insulin deficiency and altered metabolism seen in diabetes mellitus are summarized in fig. III.

**Diabetes mellitus- New classification**

DM has been traditionally divided in to Insulin Dependent Diabetes Mellitus (Type 1) and Non-Insulin Dependent Diabetes Mellitus (Type 2). The roles of autoimmunity and genetics were also added their importance in the causes of diabetes after the classification in 1997 (Expert committee reports, 1997). It also becomes evident that, the old classification is too simple and diabetes is a much more heterogeneous disorder than it was thought so far. Therefore, a WHO consultation group proposed a new classification of diabetes in 1998 and introduced some new subgroups. About 10% of persons with adult onset
diabetes (type2) have slowly progressing form of type 1 diabetes, also called LADA (Latent Autoimmune Diabetes in Adults) (Tuomi et al., 1993).

1. **Type 1 Diabetes mellitus**

Type 1 Diabetes, formerly known as ‘juvenile diabetes’ or Insulin Dependent Diabetes Mellitus. People with type 1 diabetes are usually young and thin, although it can and it does occur in older adults. Type 1 diabetes is due to the synergistic effects of genetic, environmental and immunological factors leading to the destruction of the pancreatic beta cells. This results in an absolute deficiency of insulin secretion. The onset is usually acute, developing over a period of a few days to weeks. Over 95% of persons with type 1 DM develop the disease before the age of 25, with an equal incidence in both sexes and an increased prevalence is seen in the white population (National Diabetes Data Group, 1995). In the absence of insulin, blood glucose levels rise and result in polyuria, polydipsia, polyphagia, and weight loss. If it is not diagnosed and treated, marked hyperglycemia will occur and progress to ketoacidosis, coma and death. Current therapy for type 1 diabetes is to provide the patient with exogenous insulin to maintain the normal blood glucose levels and to prevent hyperglycemia, ketoacidosis, to prevent or delay chronic complications (Expert committee report, 1997). However, assessment of required dosage in these patients is a difficult task. This discrepancy some time causes hypoglycemia after insulin injection.
1.1. Immune mediated diabetes

This form of diabetes was previously called Insulin Dependent Diabetes Mellitus, which is due to the cellular mediated autoimmune destruction of \( \beta \) cells of pancreas (Atkinson & Maclaren, 1994). It commonly occurs in childhood and adolescence, but can occur at any age.

1.2. Idiopathic diabetes mellitus

A few patients, usually those of African or Asian origin, have no antibodies against beta cells but have a similar clinical presentation. Consequently, this is called the idiopathic form of type 1 diabetes mellitus.

Major risk factors for type 1 Diabetes mellitus

- Family history of diabetes.
- Race/ethnicity (Whites)
- Age (being age of 20 or younger increases risk).
- Mother who had Preeclampsia (a condition characterized by a sharp increase in blood pressure during the third trimester of pregnancy)
- Family history of autoimmune diseases, including Hashimoto's thyroiditis, Graves' disease, Myasthenia gravis, Addison's disease, or Pernicious anemia
- Viral infections during infancy including mumps, rubella.
- Child of an older mother.
Comparison of Type 1 and Type 2 diabetes

<table>
<thead>
<tr>
<th>Features</th>
<th>Type 1</th>
<th>Type 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age at onset</td>
<td>Usually under 25</td>
<td>Usually over 40</td>
</tr>
<tr>
<td>% of all diabetics</td>
<td>Less than 10%</td>
<td>Greater than 90%</td>
</tr>
<tr>
<td>Family history</td>
<td>Uncommon</td>
<td>Common</td>
</tr>
<tr>
<td>Appearance of symptoms</td>
<td>Rapid</td>
<td>Slow</td>
</tr>
<tr>
<td>Obesity at onset</td>
<td>Uncommon</td>
<td>Common</td>
</tr>
<tr>
<td>Insulin levels</td>
<td>Decreased</td>
<td>Variable</td>
</tr>
<tr>
<td>Insulin resistance</td>
<td>Occasional</td>
<td>Often</td>
</tr>
<tr>
<td>Treatment with insulin</td>
<td>Always</td>
<td>Usually not required</td>
</tr>
<tr>
<td>Beta-cells</td>
<td>Decreased</td>
<td>Variable</td>
</tr>
<tr>
<td>Ketoacidosis</td>
<td>Frequent</td>
<td>Rare</td>
</tr>
<tr>
<td>Complications</td>
<td>Frequent</td>
<td>Frequent</td>
</tr>
</tbody>
</table>
2. **Type 2 Diabetes mellitus**

Type 2 Diabetes mellitus formerly called as Non Insulin Dependent Diabetes Mellitus or ‘adult onset diabetes’. It is characterized by peripheral tissues insensitiveness to insulin or defective insulin secretion from the beta cells. It is more common in women, especially women with a history of gestational diabetes. Insulin resistance and hyperinsulinemia eventually lead to impaired glucose tolerance. Many patients with type 2 diabetes have ‘insulin resistance syndrome’ (central obesity, hypertension, and hyperlipidemia) for many years. There are three major pathophysiological abnormalities in patients with type 2 diabetes. They are

1. Early loss of first phase insulin production associated with defective beta cells function.
2. Peripheral resistance to insulin appears primarily in muscle tissue and the liver.
3. Excessive hepatic glucose production as the disease progresses (DelPrato and Tiango, 2001).

**Major risk factors for type 2 Diabetes mellitus** (Singh et al., 2001)

- Family history of diabetes (parents or siblings with diabetes).
- Obesity (≥ 120% higher than desired body weight or body mass index).
- Race/ethnicity (African American, Hispanic, Native American, Asian American, Pacific Islander).
- Age ≥ 45 years.
- Previously identified impaired fasting glucose or impaired glucose tolerance.
- Hypertension (≥ 140/90 mmHg).
- High density lipoprotein cholesterol level ≤ 35 mg/dl or a triglycerides level ≥ 250 mg/dl.
- History of gestational diabetes mellitus or delivery of babies above 4.03 kg.

Treatment goals for patients with type 2 diabetes include achieving and maintaining normoglycemia through weight reduction, exercise, diet and use of exogenous insulin, use of oral agents that stimulate the secretion of insulin or reduce the insulin resistance.

3. **Gestational diabetes mellitus (GDM)**

Gestational diabetes mellitus is defined as any degree of glucose intolerance that begins, or first recognised during pregnancy (Metzger and Coustan 1998). Approximately 7% of all pregnancies are complicated by GDM. For the mother, GDM increases the risk of preeclampsia, cesarian delivery, and future type 2 diabetes.

4. **Other specific types**

   A. **Genetic defects of β cell functions**

   B. **Genetic defects in insulin action**

   C. **Diseases of exocrine Pancreas**
D. Endocrinopathies
E. Drug or Chemical induced
F. Infections
G. Uncommon forms of immune mediated diabetes

H. Other genetic syndromes sometimes associated with diabetes

Diabetes mellitus - Complications

Diabetes results in the development of numerous complications due to hyperglycemia. The likelihood of developing complications, whether acute or chronic is ultimately a reflection of the levels of the blood glucose in these patients.

Acute complications

In addition to the common signs and symptoms like hyperglycemia, glycosuria, polyuria, polydypsia, loss of weight in spite of polyphagia, poor wound healing, diabetics are also susceptible to three major acute complications. They are

1. Hypoglycemia

The problem of hypoglycemia is more common in type 1 than type 2 diabetes. Taking too much insulin, missing a meal or over exercising can result in hypoglycemia.
2. Diabetic ketoacidosis

In type 1 diabetics, ketoacidosis is a consequence of insulin lack. If untreated, ketoacidosis can result in numerous metabolic problems including pH alterations and even coma.

3. Non-ketogenic hyperosmolar syndrome

With a mortality rate of over 50 %, non-ketogenic hyperosmolar syndrome constitutes a true medical emergency and it is due to severe dehydration secondary to deficient fluid intake.

Chronic complications

1. Atherosclerosis

Experts believe that micro vascular changes begin when the diabetic state becomes overt (i.e. fasting blood glucose is at or higher than 126 mg/dl) and macro vascular changes begin many years earlier (Qian & Eaton, 2000). DM is often referred to as a silent killer because it is a significant risk factor for early onset of atherosclerotic vascular disease and coronary heart disease (Singh et al., 2001; Alexander et al, 2000).

2. Diabetic retinopathy

Diabetics are twenty five times more prone to blindness than non diabetic subjects (Lloyd et al., 1981). 90 % of patients with type 1 diabetes and 65 % patients with type 2 diabetes will develop retinopathy 10 years after the onset of disease (Klein et al, 1994). Strict glucose control can delay the onset of
retinopathy and slow its progression (Reports of diabetes control research group, 1993).

3. Diabetic nephropathy

Diabetic nephropathy is the leading cause of end stage renal disease. It accounts for more than 25% of all cases of end stage renal disease (Krolewski et al., 1985). Diabetic nephropathy is characterized by albuminuria, hypertension and progressive renal insufficiency (Selby et al., 1990). Microalbuminuria, an important indicator of developing overt diabetic nephropathy is also considered to be an early stage of diabetic nephropathy (Allawi et al., 1988).

4. Diabetic neuropathy

Diabetic neuropathy is one of the commonest complications of type 2 diabetes. People with diabetes can overtime have damage to the nerves in the body. Neuropathies lead to numbness, pain and weakness in the hands, arms, feet and legs. Problems may also occur in any organ or system, including the digestive tract, heart and sex organs (Lehtinen et al., 1993). People with diabetes can develop problems at any time, but the longer a person has diabetes, the greater is the risk.

5. Diabetic foot ulcers

The foot is often the first part of the body to show the adverse effects of diabetic neuropathy. Most of the lower extremity ulcerations in diabetes are due to neuropathy, attributable to circulatory impairment (Boike, 2002). Impaired kidney
function can worsen the problem by causing increased swelling in the lower extremities.

**Oral anti-hyperglycemic agents and their actions**

Currently, five different classes of oral anti-hyperglycemic agents are being used for treating DM. They are sulfonylureas, meglitinides, biguanides, alpha-glucosidase inhibitors and thiazolidinediones.

**Sulfonylureas**

Sulfonylureas (SU) have remained the mainstay of anti-diabetic therapy for almost three decades. The mode of action of SU is by the inhibition of K_{ATP} channels initiating insulin secretion (Chakrabarti et al., 2002). Thus this drug can be used only in patients with type 2 DM, having functional beta cells for endogenous insulin production. Overt hypoglycemia is the most worrisome side effect of SU. All SUs have been associated with weight gain. Thus, SU may not be optimal first choice for obese patients.

**Meglitinides**

The meglitinides stimulate the release of insulin from the pancreatic beta cells. However, this action is mediated through a different binding site on the sulfonylureas receptor of the beta cells. Unlike sulfonylureas the meglitinides have a very quick onset of action and a short half life. With this drug there is a greater decrease in postprandial glucose and a decreased risk of hypoglycemia. Because of quick onset of action, patients should take this drug immediately before meal (Chakrabarti, 2002).
Biguanides

Metformin is the most common drug in this class. It works by reducing the hepatic glucose production through inhibition of gluconeogenesis and to lesser extent by enhancing insulin sensitivity in hepatic and peripheral tissues (De Fronzo, 1999). This drug therapy has been associated with a lack of weight gain and is beneficial for obese patients. Worrisome side effect of this drug is that it rarely results in lactic acidosis.

Alpha-glucosidase inhibitors

Alpha-glucosidase inhibitors like acarbose delay the gastrointestinal absorption of glucose by inhibiting enzyme α-glucosidase, which break down ingested carbohydrates. These drugs are not recommended to the patients with renal dysfunction, liver cirrhosis and inflammatory bowel disease or history of bowel obstruction (Chakrabarti, 2002).

Thiazolidinediones

Thiazolidinediones increase insulin sensitivity in fat and muscle tissues and to a lesser extent inhibit hepatic glucose production. They are beneficial as they decrease triglycerides and there is no risk of hypoglycemia with thiazolidinediones monotherapy (Chakrabarti, 2002). Mild to moderate edema and increase in plasma volume has been reported with this drug which is of concern in patients with congestive heart diseases.
The limitations of currently available oral anti-diabetic agents either in terms of efficacy or safety have encouraged the researchers all over the world to discover an alternative drug to manage DM.

**Herbal medicine**

Herbal medicine has its own ancient history of usage and is common to all races, cultures and epochs. From the time immemorial, it is still extensively practiced. Today, many modern medicines are based on it and many drugs are still produced from plants (Balasubramanyam et al., 2002). The World Health Organization recognizes that nearly 80% of the world's population lives in developing countries and depends largely on traditional medicine, of which herbal medicine constitutes the most prominent part. Herbal medicine is still the mainstay of about 75-80% of the world population, mainly in the developing countries for primary health care because of better cultural acceptability, better compatibility with the human body and lesser side effects. However, the last few years have seen a major increase in their use in the developed world (Kamboj, 2000). The beneficial multiple activities like manipulating carbohydrate metabolism by various mechanisms, preventing and restoring the integrity and function of β cells, insulin releasing activity, improving glucose uptake and utilization, and the antioxidant properties present in the medicinal plants offer opportunity to develop them into novel therapeutics.
Herbal medicine in the treatment of diabetes mellitus

DM was known to ancient Ayurvedic physicians some 3000 years ago. The earliest known documentation of plant treatments for diabetes was found in the Ebers Papyrus of about 1550 BC. The association of polyuria with a sweet tasting substance in the urine was first reported in Sanskrit literature dating from 5th to 6th century AD at the time of two notable Indian physicians Sushrutha and Charak.

Anti-diabetic herbs are praised as ‘pharmaceutical foods’ and ‘biotechnologist’s flora’ (Balasubramanyam et al., 2002). More than 100 medicinal plants are mentioned in the Indian system of medicine, including folk medicines for the management of diabetes, which are effective either singly or in combination (Ajit Kar et al., 2003).

Following are some of the known anti-diabetic medicinal plants, which were studied scientifically and been documented by Ajit Kar et al., (2003).

- *Coccinia indica* (Cucurbitaceae family).
- *Gymnema sylvestre* (Asclepiadaceae).
- *Pterocarpus marsupium* (Papilionaceae)
- *Trigonella foenum graecum* (Papilionaceae)
- *Moringa oleifera* (Moringaceae)
- *Eugenia jambolana* (Myrtaceae)
- *Tinospora cordifolia* (Menispermaceae)
- *Momordica charantia* (Cucurbitaceae)
Tinospora cordifolia

Natural view

Closer view of stem

Closer view of leaves
- *Ficus glomerata* (Moraceae)
- *Ficus bengahalensis* (Moraceae)
- *Ocimum sanctum* (Labiatae)
- *Zingiber officinale* (Zingiberaceae)
- *Terminalia bellirica* (Combretaceae)
- *Pterocarpus marsupium* (Leguminosae)

To have more effective drug potency, many of these herbs are used in combination. *Tinospora cordifolia* is one such herb. Though the anti-diabetic activities of TC were studied by many researchers, reports on mechanism of action of *Tinospora cordifolia* are lacking.

**Tinospora cordifolia**

*Tinospora cordifolia* (Willd.)(TC) is known as Giloya in Hindi, which is the Hindu mythological term that refers to the heavenly elixir that have saved celestial beings from old age and kept them eternally young (Singh et al., 2003). The stems of TC are rather succulent with long filiform fleshy aerial roots from the branches. The leaves are membranous and cordate. It is distributed throughout the tropical Indian subcontinent and China (Kirtikar and Basu, 1975).

**Chemistry**

Variety of constituents has been isolated from TC and their structures were elucidated. Since the composition of TC was well reported, no efforts were made
### Fig. IV. Constituents of TC

<table>
<thead>
<tr>
<th>Type of chemical</th>
<th>Active principle</th>
<th>Part in which present</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alkaloids</td>
<td>Tinosporin, Berberine, Palmatine, Isocolumnin, etc</td>
<td>Stem, Root</td>
</tr>
<tr>
<td>Glycosides</td>
<td>Furanoid diterpene glucoside, Tinoscordifolioside, Palmatosides, etc</td>
<td>Stem</td>
</tr>
<tr>
<td>Diterpenoid Lactones</td>
<td>Furanolactone, Tinosporon, Columbin etc</td>
<td>Whole plant</td>
</tr>
<tr>
<td>Steroids</td>
<td>β- sitosterol, Makisterone A, Giloinsterol, Ecdysterone, etc</td>
<td>Aerial part, Stem</td>
</tr>
<tr>
<td>Sesquiterpenoid</td>
<td>Tinocordifolin</td>
<td>Stem</td>
</tr>
<tr>
<td>Aliphatic compound</td>
<td>Octacosanol, Heptacosanol, etc</td>
<td>Whole plant</td>
</tr>
<tr>
<td>Miscellaneous compounds</td>
<td>Cordifol, Cordifelone, Giloin, Giloinin, Tinosporic acid</td>
<td>Whole plant</td>
</tr>
</tbody>
</table>
to redo the studies on the composition of TC extract. According to the reports from Singh et al., (2003) the different components in TC belong to different classes such as alkaloids, deterpenoid lactones, glycosides, steroids, phenolics, aliphatic compounds, sesquiterpenoid and polysaccharides. Constituents of TC are shown in fig IV.

Medicinal properties of *Tinospora cordifolia*

**Anti-diabetic properties**

TC is widely used in combination with other herbal preparations in Ayurvedic medicine for treating DM. Based on the research work, it was reported that the daily administration of either alcoholic or aqueous extract of TC decreased the fasting blood glucose levels, increased the glucose tolerance in rodents (Gupta et al., 1967; Grover et al., 2000) and has anti-hyperglycemic effects in diabetic rats. Stanely et al., (2000) reported that the oral administration of aqueous extract of TC roots in alloxan induced diabetic rats caused a significant reduction in blood glucose and brain lipids. Another study by Wadood, (1992) also revealed the significant hypoglycemic effects of leaves of TC in alloxan diabetic rabbits.

**On immune system**

TC was reported to benefit the immune system in a variety of ways. Atal et al., (1986) reported that the ethanolic extract of TC has immunomodulating effects as it improved phagocytic functions in mice. Further studies by Kapil et al.,
showed the immunopotentiating compounds from TC and immunomodulatory effect of both aqueous and alcoholic extracts of TC in mice.

**Anti-stress activity**

Anti-stress activity of TC was clinically tested and it was found that it brought about good response in children with moderate degree of behavioral disorders and mental deficit. It also significantly improved the I.Q. levels (Singh et al., 2003). Sharma et al., (1996) showed that the roots of TC possess the anti-stress activity.

**Hepatoprotective action**

The hepatoprotective action of TC was reported by Mehrotra et al., (2000) in which, goats treated with TC have shown significant clinical and hemato-biochemical improvements in carbon tetra chloride induced hepatopathy. Similar observations in TC were also reported by Singh et al., (1984) for it’s hepatoprotective activity in rats.

**Anti-inflammatory effect**

The dried stem of TC produced significant anti-inflammatory effect in both acute and sub acute models of inflammation. The aqueous extract of TC exerted a significant anti-inflammatory effect on cotton pellet granuloma and formalin induced arthritis models. TC was found to be more effective than acetylsalicylic
acid in acute inflammation. However, in sub acute inflammation, the drug was inferior to phenylbutazone (Jana et al., 1999).

**Anti-oxidant activity**

The aqueous extract of roots of TC has shown the anti-oxidant action in alloxan induced diabetes rats (Stanely and Menon, 1999). This study showed that, the treatment with TC root extracts (for six weeks) leads to a decrease in plasma TBARS (Thiobarbituric acid reactive substances), ceruloplasmin, alpha-tocopherol and an increase in plasma GSH and vitamin C.

**Anti-cancer activity**

Jagetia et al., (1998) have found that the TC killed the HeLa cells very effectively *in vitro* and thus it indicated that TC was useful as an anti-neoplastic agent.

**Antipyretic activity**

Ethanolic extract of TC in rats showed antipyretic activity and was useful in treating fever. The extract produced results, comparable in efficacy to that of 200 mg/kg of aspirin and was reported to be safe (Vedavathy and Rao, 1991).

**Experimental induction of diabetes**

Experimental induction of diabetes in animal models for testing the new anti-diabetic drug is commonly practiced by scientific world. One of the known methods of producing experimental diabetes is to damage the β cells of Islets of Langerhans. The role of pancreas in diabetes mellitus was first identified by von
Mering and Minkowski in 1889, who demonstrated that the total pancreatectomy in dogs results in experimental diabetes. Due to surgical trauma and inconvenience, this method is not well suited for large number of animals. More convenient method, other than surgery is chemical destruction of β cells of Islets of Langerhans. Alloxan and streptozotocin and their related compounds have been widely used for inducing diabetes in experimental animals.

Streptozotocin (STZ)

Streptozotocin is a broad spectrum antibiotic isolated from *Streptomyces achromogenes* in 1959 and was originally isolated during a search for new antimicrobial agent (Herr et al., 1960). Rakieten and co-workers (1963) were the first to report that STZ, when given intravenously caused diabetes mellitus in rats and dogs. Chemically, STZ is a glucosamine-nitrosourea compound which has a chemical name of 2-deoxy-2-(3-methyl-3-nitrosoureido)-D-glucopyranose (C8 H15 N3 07). The structure is composed of a nitrosourea moiety with a methyl group attached at one end and a glucose molecule at the other end. The molecular weight is 265 g/mol (Weiss, 1982).

Studies by Junod et al., (1967) showed the histological changes in the β cells as early as one hour after STZ treatment and there was massive β cell degranulation and necrosis associated with decrease in serum insulin level and hyperglycemia after 7-10 hours. Recent experiments have proved that the main reason for the STZ induced β cell death is alkylation of DNA (Delaney et al.,
The alkylation activity of STZ is related to its nitrosourea moiety which leads to subsequent inhibition of insulin synthesis and secretion (Nukatsuka et al., 1990 b).

Because of poor stability, general toxicity and difficulty in predicting the extent of its diabetogenic effects, alloxan is disadvantageous than STZ and the range of STZ dosage is not as narrow as in the case of alloxan (Szkudelski, 2001). Based on these reports, STZ is used in the present study. Rats treated with STZ displayed many features seen in human subjects with uncontrolled diabetes mellitus including hyperglycemia, polydipsia and weight loss.