CHAPTER 2

Literature survey

2.1 Epidemiology and prevalence of diabetes mellitus

2.1.1 Epidemiology of DM:

India has been projected by WHO as the country with the fastest growing population of diabetic patients. It is estimated that as of 2010, an estimated 285 million diabetic patients were there globally, among them about 90% are T-2 by 2030, this incident may be doubled. DMs occur globally, but T-2 is more common in the grown up countries. Asia and Africa are the greatest expecting in increasing DMs by 2030 [2] due to urbanization and changing in lifestyle as "Western-style" diet [109, 210].

India

India has been projected by WHO as the country with the fastest growing population of diabetic patients. It is estimated that between 1995 – 2025 diabetic patients in India will increase by 19.5%.

In the recent time India has more DMs than China according to IDF; 50 million Indians and more affects ie, 7.1% of adults and kills about 1 million/year. The average onsetage is 42 years.

Australia

Indigenous have a higher prevalence and increasing incidence than non-indigenous people in the worldIn Australia, the self-reported diabetes in indigenous Australians are almost four times that of non-indigenous Australians.
China

1/10 Chinese adults with DMs. >92 million Chinese adults have the DMs (2010), the incidences of DMs with another 150 million are with early symptoms. 2009 study found a 30% may increase in 2016-17.

United Kingdom

About 3.8 millions have DMs in UK, but the CDUK, U.K. predicted that could be more as 6.2 million by 2035-36.

United States

Diabetes is a fastly growing in U.S. In 1993, ~7.8 millions were diagnosed and confirmed DMs.. This is about of 3.1% of country’s population, even, rates ranges from ~1.3% to 10.4% (30 to 65 years). The prevalence & rate are growing in the country since 1958 [2].

2.1.2 Prevalence of DM:

Due to increase in the growth of the population, ageing, urbanization and increased obesity prevalence, inactivity of physical exercises and fast-food habits, the number of DMs is increasing. The world wide DMs prevalence for all age group was estimated to be 300 million by 2025 [109, 210]. T-2 DMs epidemic in India is a result of social influence and change in life styles. The epidemiological studies show that

1960 - 1970: Urban 1-4%, Rural 1.2%,
1990: Urban 5-15%, Semi urban-4-6%, Rural- 2.5%,
2000: Urban 5.4%,
Chennai - 12.3-15.5% and Jaipur 12.3-16.8 % [70].
Table: 2.1 Estimation of top ten countries for DMs

<table>
<thead>
<tr>
<th>COUNTRY</th>
<th>1995(millions)</th>
<th>2025(millions)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indian</td>
<td>19.4</td>
<td>57.2</td>
</tr>
<tr>
<td>Chinist</td>
<td>16.0</td>
<td>37.6</td>
</tr>
<tr>
<td>USA</td>
<td>13.9</td>
<td>21.9</td>
</tr>
<tr>
<td>Russians</td>
<td>8.9</td>
<td>14.5</td>
</tr>
<tr>
<td>Japanis</td>
<td>6.3</td>
<td>12.4</td>
</tr>
<tr>
<td>Brazilian</td>
<td>4.9</td>
<td>12.2</td>
</tr>
<tr>
<td>Indonesian</td>
<td>4.5</td>
<td>11.7</td>
</tr>
<tr>
<td>Pakistanies</td>
<td>4.3</td>
<td>11.6</td>
</tr>
<tr>
<td>Mexicos</td>
<td>3.8</td>
<td>8.8</td>
</tr>
<tr>
<td>Ukraines</td>
<td>3.6</td>
<td>8.8</td>
</tr>
<tr>
<td>Other places</td>
<td>49.7</td>
<td>8.5</td>
</tr>
<tr>
<td>SUM</td>
<td>135.3</td>
<td>300</td>
</tr>
</tbody>
</table>
2.2 Insulin Regulation

2.2.1 Synthesis and insulin secretion:

In rough endoplasmic reticulum pre proinsulin (precursor insulin) is produced

Precursor insulin is transported to the Golgi bodies

Then undergoes proteolytic cleavage to proinsulin then to insulin

This fragment is called as C-peptide.

Insulin and C-peptide are stored in granules in β-cells and are normally co-secreted by exocytosis in together with smaller amounts of proinsulin.

There is a steady basal release of insulin and also a response to control in blood glucose [163].

The response to an increase in blood glucose

Initial rapid phase reflects the release of hormone.

A slower, delayed phase reflects continued release of hormone and new synthesis.

β- Cells respond (Insulin) to both the absolute glucose concentration and also to the rate in change of BG. Glucose enters β-cells via Glut-2 cellcoat transporter, subsequent metabolism through glucokinase and glycolysis. This blocks ATP dependent K⁺ channel (KATP) causes depolarization of membrane and voltage dependent Ca channels opening, encourages the Ca²⁺ influx.

This Ca²⁺ signal induces secretion of insulin hormone, only in the presence of amplifying messengers DAG, non esterified arachidonic acid (which enhance few more Ca²⁺ entry) and 12 lipoxygenase products of arachidonic acid (mainly 12S-hydroxyeicosatetraenoic acid (12-S-HETE). This activity is driven by ATP for insulin secretion [163]
Figure: 2-1 Factors Regulating Insulin Secretion

The islets of Langerhans contain four types of main cell:

i) B-(or -β) cells secrete insulin

ii) A-cells secrete glucagon

iii) D-cells secrete somatostatin

iv) PP cells secrete pancreatic polypeptide.

The each islet core contains the predominant β-cells covered by A-cells and interspersed with D-cells or PP cells.

β - Cells secrete, insulin, a peptide known as islet amyloid polypeptide or amylin, by stimulating glycogen breakdown in striated muscle which delays gastric emptying and opposes insulin.
Glucagon opposes insulin, increasing blood glucose and stimulating protein breakdown in muscle.

The secretion of insulin and of glucagon are inhibited by Somatostatin

- Widely distributed outside the pancreas
- Released from hypothalamus,
- Inhibiting the release of growth hormone from the pituitary [163,164].

2.2.2 β- Cell Regeneration and Insulin releasing activity:

Insulin is the hormone produced by the β cells of the islets of Langerhans (Pancrease) and released into systemic circulation, Insulin circulates through the body and acts on specific insulin receptor sites to stimulate glucose transportation into the cells for its growth and development, this process is called as “facilitated diffusion”.

1) Insulin stimulates the synthesis of glycogen (store of glucose)
2) Insulin helps the conversion of lipids into fat stored in the form of adipose tissue.
3) Insulin synthesis of needed proteins from amino acids.
4) Insulin is released after a mealie, when the BG rises
5) Glucose metabolism is affected by the circulating insulin, leads to either store/use of nutrients from the meal.
6) As a result insulin release, BG falls and insulin release drops off.
7) Sometimes, an insuffiency of insulin is released; May be due to enough insulin is not produced by pancreas [163].
2.2.3 Regulation of insulin secretion [161]:

1) Hormonal regulation
2) Neural regulation
3) Chemical regulation.

**1) Hormonal regulation**

**Figure: 2-2 Hormonal regulation**

**KEY NOTE -1**

<table>
<thead>
<tr>
<th>α cells</th>
<th>D cells</th>
<th>β cells</th>
</tr>
</thead>
<tbody>
<tr>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>
Ketone bodies are seen in type one.

RBC, Brain and Cells of liver (hepatocytes) take up glucose independent of Insulin.

Insulin moves glucose into cells

Glucagon = Glucose Gon out of cells. KEY NOTE -2

2) Neural regulation

a) Vagal & Sympathetic nerve supplies to islets

β cells

- + by inhibiting adenycyclase of β cells

Releases

B2 stimulation

Insulin

By stimulating adenylcyclase of β cells

b) Vagal stimulations / NM stimulations by Ach.

↓

IP3/DAG stimulations

↑ intracellular ca^{2+} in β cells

↓

Insulin secretion
WBC and medullary cells are independent of insulin. Insulin has insulin sparing effect.

(c) Hypothalamus (primary site of insulin regulation)

Ventro Medial Nuclei Stimulation  Ventro lateral Nuclei stimulation

Inhibition of insulin release  Insulin release
3) **Chemical regulation**

Oral glucose

\[ \downarrow \]

Incretin release

\[ \downarrow \]

Glucose sensing mechanism $\beta$ cells

\[ \downarrow \]

Increasing intra cellular $\text{Ca}^{2+}$ (↓ efflux, ↑ influx and release from intra cellular stores)

\[ \downarrow \]

Exocytic release of insulin occurs

Oral glucose $\rightarrow$ incretin release (VIP, GIP, Gastrin, Secretin, Glucagon)

Glucagon $\rightarrow$ ↑ cAMP in $\beta$ cells

\[ \downarrow \]

Insulin releases

---

**2.2.4 Mechanism Action of insulin** [163]:

Site of Action: Cell Membrane. Insulin has, i) Short action

ii) Intermediate

iii) Long term action.
Mechanism – 1

ATP dependent translocation of glucose across cell membrane

INSULIN

- Increase glucokinase synthesis → G. 6. P
- Phosphorylase → Glycogenolysis in Liver
- Glycogen synthetase → Glycogen synthesis in liver fat and muscle

Mechanism- 2

Transcription of vascular endothelial lipoprotein lipase → clearance of VLDL & chylomicrons

INSULIN

- Lypolysis and stimulation of triglyceride synthesis .
- Amino acid entry into the cells → Protein Synthesis
- Synthesis of phosphoenol pyruvate carboxy kinase .
- Gluconeogenesis from protein in liver.
2.2.5 Stimulation and Inhibition of insulin release:

A) *Stimulation of insulin release*

1) Glucose is the stimulus for synthesis & release of insulin
2) Leucine
3) Vagal stimulation & sulfonyl release drugs

B) *Inhibition of insulin release*

1) Ca (α-Sympathomimetic)
2) Somatostatin
3) Phenytoin
4) Thiazide
5) Diazoxide
6) Colchicine
7) Vinblastine

2.2.6 Over all effects of insulin:

Insulin reduces plasma glucose level by

1) Increasing glucose transport across cell membrane
2) Increasing glucose conversion to glycogen
3) Inhibiting FFA release from adipose tissues
4) Inhibiting Lypolysis + glycogenolysis [161].
2.3 Pathophysiology of diabetic mellitus

Figure: 2-3 Pathogenesis of Diabetes mellitus
2.3.1 Role of environmental factors:

The independent risk factors of diabetes are

- Ageing
- Fatigue & Lack of exercise
- less calorie consumption
- Alcoholism, Tobacco like substances, etc.,
- Obesity:

  Inactive physical exercise
  \[\downarrow\]
  Loss of Muscle mass
  \[\downarrow\] causes (leads to)

  Insulin Resistance [37, 57].

It occurs rapidly in the middle and high-aged patients. The *obesity* and causes of deterioration of *glucose tolerance* is due to changes in nutritional energy sources particularly,

- High fat intake
- More consumption of simple sugars
- Less starch intake
- Less dietary fibre intake.

Even less obesity (BMI < 24.99) leads 4 to 5 times increase in the risk of developing diabetes due to increase in visceral fat mass [121].

2.3.2 Insulin resistance (IRc):

Insulin exerts insufficient action in proportion to its blood glucose concentration. The impairmental action of insulin majorly on liver and muscle leads to T2 diabetus IRc increases before onset the disease. [37, 57]
The molecular mechanism for insulin action clarifies the relation of IRc and genetic factors & environmental factors.

**Genetic factors:**

- Insulin receptor (IR)
- Insulin receptor substrate (IRS) genetic polymorphs affect signals of insulin directly.
- Polymorphisms of thrifty genes associate with organ obesity and stimulates IRc.
- Gluco-lipotoxicity and inflammation mediators are also important for IRc Insulin signaling impairments.

Recent attention has focused on the involvement of adipocyte-derived bioactive substances (adipokines) in IRc. While TNF-α, leptin, resistin and free fatty acids act to increase resistance, adiponectin improves resistance [37, 57].

**2.3.3 Reactive Oxigen Species (ROSp) induced oxidative damages:**

Depends on the reality, ROSp (for e.g. OH radicals) reactions with lipids, proteins and DNA which produce different types of secondary radicals like

Lipid radicals

Sugar and base derived radicals

Amino acid radicals

Thiyl radicals.

- These are converted to peroxy radicals in presence of oxygen.

- Chain reactions (Induced by the Peroxy radicals).

- The bio-implications of Chain reactions depends on
2.4 Types, Risk factors, Complications and Signs & symptoms of diabetes [9, 187]

2.4.1 Types of diabetes:

i. Type-I (T-1): Insulin-dependent diabetes. β - Cell destruction- Insulin deficiency [139].
   A) Immune-mediated.
   B) Idiopathic.

ii. Type-II (T-2): Non-insulin-dependent diabetes, Insulin Resistance [37, 113].

iii. Gestational diabetes mellitus

   Diabetes diagnosed in pregnancy includes pre-existing DMs and develops during pregnancy.

iv. Others,

   a. Defective of β-cell function (Genetically) e.g. MODY syndrome.
   b. Defective of Insulin action (Genetically) e.g. leprechaunism.
   c. Exocrine pancreatic Diseases
      Pancreatitis
      Neoplasia
      Cystic fibrosis
      Hemochromatosis.

   d. Endocrinopathies:
      Acromegaly
      Cushing’s syndrome
Pheochromacytoma
Glaucoma
Somatostatinoma.

e. Drug/chemical induced:

Nicotinic acid
Glucocorticoids
Thiazides
Vacor
Pentamidine
Diazoxide
Phenytoin

f. Infections:

Congenital rubella
Cytomegalovirus

g. Other genetic syndromes:

Downs syndrome
Klinefelters syndrome
Turner syndrome
Wolfram syndrome
Friedreich's ataxia
Myotonic dystrophy.

2.4.2 Major risk factors of T-2 DMs:

- History of Family (Parents siblings with DMs).
- Race / ethnicity.
- Obesity (20% ODBW or BMI >24Kg/m$^2$).
- Habit of physical inactivity.
- Fasting glucose Impairement (IFG) or impaired tolerance blood glucose (IGT).
- Bood pressor (BP) $\geq$ 140/90 mm of Hg in adults.
- TGL level $\geq$ 250mg/dL and /or HDL of <35mg/dl.
- gestational diabetes (as History) /or baby weighing = 4Kg during delivery
- Polycystic ovary syndrome (POS) [13].

2.4.3 Complications of DM:

Diabetic complications are due to poorly controlling of DMs, much more common in T-2. At the time diagnosis approximately 50% of patients suffering from one or more complications. By keeping FBG levels as normal through aggressive management, slows the onset and progression of eye, kidney and nerve disease caused by DMs [9,130]. Diabetic complications are divided as acute and chronic complications.

**Acute Complications**

i) Diabetic Keto-acidosis (DKA)

DKA is common in T-1diabetes than T-2 diabetes (ketONE in type ONE). It is an acute and dangerous complication. Clinically it is absolute insulin deficiency with hyperglycemia (glucose levels >200mg/dl) with higher lipolysis, ketone production and acidosis (pH ≤ 7.3). Patients in DKA is with,

Dehydrated
Rapid and deep breathing
Abdominal pain
Consciousness is typically normal until late in the process Lethargy may progress to coma
Causes hypotension, shock and death.
Hyper osmolar Non-Ketotic Coma (HNC).
ii) Hypoglycaemia

If there is high level of insulin in the body than blood sugar (non homeostasis) or blood sugar falls below normal leads to hypoglycemia. This occurs mostly in T-1 than T-2 since in the T-1, insulin is directly injected.

On the administration of higher dose of insulin / a person misses a meal cause’s hypoglycemia, the signs are mild hunger, followed by dizziness, sweating, palpitations, mental confusion and eventual loss of consciousness [45].

(B) Chronic Complications

Damage of blood vessels occurs due to chronic elevation of blood glucose. The cell lining of endothelial blood vessels takes more glucose than normal, since they don’t depend on insulin. They form abnormal surface glycoproteins and cause the basement membrane to grow thicker and weaker.

In diabetes, the resulting problems are grouped under “Microvascular disease” (due to small blood vessels) and “Macro vascular disease” (due to damage to the arteries)

a) Micro vascular disease

The mostly affected sites are the retina, renal glomeruli and the nerves- heath leads to,

i) Retinopathy

ii) Nephropathy

iii) Neuropathy

b) Macro vascular Complications

These affect large vessels (arteries) in the body and are more likely in T-2 diabetes patients. Risk factors of macrovascular complications include hyperglycemia, obesity, dyslipidaemia, hypertension, smoking, lack of exercise. Eg: Arteriosclerosis.
Arteriosclerosis is, or “hardening of the arteries”, generally shows its effects first in legs and feet. Then narrowing stiffness (due to Deposition of Calcium in vessel) of arteries becomes closure of the blood vessel and leads to less elastic of the blood vessel and not allows greater blood flow when needed (during exercise).

(C) Other complications

i) Oral Complications (Periodontal disease leads to tooth loss).
ii) Infections (asymptomatic bacteraemia)
iii) Complications in pregnancy [45].

2.4.4 Signs and symptoms:

1) The signs of untreated diabetes,
   a) Weight loss
   b) Polyurea
   c) Polydypsia
   d) Polyphagia
   e) Pruritis valvae
   f) Balanitis.

2) In T-1 DMs Symptoms slowly develops and may be subtle or absent in T-2 DMs.

3) Chronic high blood glucose may increase absorption of glucose in the lens of the eye leads to
   a) Changes in lense shape
   b) Vision changes (Rapidly in T-1)
   c) Blurred vision

4) Type 2 changes are generally more gradual (suspected)

5) Skin rashes (dermadromes) [109].
2.5 Carbohydrate metabolism

2.5.1 Hyperlipidemia and Hyperglycemia:

The clinical state of DMs is often accompanied by elevated TC, Tgl and FFA in blood. This leads to deteriorating β cell function in diabetic patients (lipotoxicity hypothesis) [14, 40].

Insulin gene expression might be inhibited due to prolonged exposure of β cells to fatty acid. The glucose oxidation might decrease by elevated fatty acid oxidation.

<table>
<thead>
<tr>
<th>Over expression of diacylglycerol acyl-transferase</th>
<th>stimulates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tgl synthesis in islets,</td>
<td>inhibits</td>
</tr>
<tr>
<td>Glucose-induced insulin secretion after culture in high glucose,</td>
<td>but not in low glucose.</td>
</tr>
</tbody>
</table>

Some of the plants reported for their properties to alter the lipid profile in diabetic conditions are, *Averrhoa bilimbi*, *Plantago ovata L*, *Boerhavia diffusa*, *Hibiscus rosasinensis*, *Ficus religiosa*, *Coccinia indica*, *Gymnema montanum*, *Tinospora cordifolia*, *Cuminum cyminum*, *Helicere sisora*, *Salacia blonga*, *Syzigium cumini*, *Azadirachta indica*, *Ocimum sanctum*, *Trigonella foenum-graecum* [201]
2.5.2 Impairing of Carbohydrate Metabolism:

- Dietary sources are primary exogenous source of Carbohydrates (glucose)
- Glucose is the important energy/fuel for the organ functions, so continuous supply of glucose (energy) is required
- Mammals have sophisticated physiological systems to maintain normal serum glucose levels homeostatic mechanisms (during fasting and full diet) of the body.
- The glucose production by the liver and the peripheral uptake of glucose by skeletal muscle & heart muscle and fat meets the required quantity [154].
- During fasting, homeostasis of glucose is achieved by triggering expression of gluconeogenesis (synthesis of glucose from non carbohydrate sources) response to glucagon.
- During carbohydrate rich diet, the function is taken over by insulin for its uptake and utilization peripherally.
- The glucose metabolism impairment leads to physiological imbalance and warrants for proper control.
- Starting from the ingestion of carbohydrate, breaks into monosaccharide, sucrose and fructose by digestive enzymes for downhill utilization of glucose for energy production and to stimulate the β-cells of the pancreatic islets.
- If any variation occurs due to various causes in normal glucose metabolic pathway, which may lead to impairment of glucose metabolism furtherly causes hyperglycemia (DMs) [85].
- Longterm exposure of pancreatic islets to elevated glucose concentration can impair the glucose-stimulating insulin release.
- Glucose stimulation of pancreatic β-cells initiates a cascade of events resulting in insulin secretion and is dependent on an increase in intracellular Ca\(^{2+}\) [163].
- Glucose metabolism regulation is a key aspect of metabolic homeostasis and insulin is the dominant hormone to influence this regulatory system.
- One of the major roles of insulin is to enhance overall glucose disposal by stimulating the glucose uptake into the target tissues [161, 199].
2.5.3 Role of Carbohydrate Hydrolyzing Enzymes in Post-Prandial Hyperglycemia (PPHG):

- Diabetes fall into two broad etiopathogenic categories as T-1 and T-2.
- The management of T-2 diabetes demands multiple therapeutic approaches to treat and to decrease the PPHG.
- NOTE: PPHG plays a major role to develop of T-2 diabetes and complications associated with micro, macro-vascular and cardiovascular disease.
- This can be done by retarding the absorption of glucose by inhibiting the carbohydrate hydrolyzing enzymes (CHE) like α-amylase (α-A) and α-glucosidase (α-G).
- α-A and α-G participate in glucose digestion in the digestive tract hence considered as key enzymes that can control PPHG [154, 163].

In living organisms specific sequences of interrelated reactions convert the ingested food into the biochemical compounds required for growth and maintenance. These sequences are generally referred to as metabolic pathways. Those pathways change the ingested food into compounds that the organism must have in order to sustain life. Complex sequence of biochemical reaction may bring about the degradation of ingested foods or it may carry out the synthesis of required compounds. Sequences which bring about the degradation are referred to as catabolic pathways, while those which carry out biosynthesis are referred to as anabolic pathways [50].

Catabolic pathways are effectively oxidative pathways. The catabolism of carbohydrates involves the oxidation of carbon atoms that constitute these molecules, as a result of the oxidation of carbon atoms, useful chemical energy is made available. Anabolic pathways are effectively reductive pathways; biosynthesis of carbohydrates generally entails carbon reduction [50, 164].

Among the vast range of enzymes which involve in the carbohydrate metabolic pathway, Carbohydrate hydrolyzing enzymes play the vital role in the post parental hyperglycemia.
Carbohydrate hydrolyzing enzymes are those which hydrolyze the chemical bond in the ring structure of carbohydrates. The major enzymes under this class are Liquefying (dextrogenic) or α-amylases, Sacchrogenic or β-amylases, Phosphorylases, α-Glucosidases, β-Glucosidases and so on.

Inhibitors of these α-A and α-G enzymes delay digestion (and digestion time) carbohydrate causes a reduction in the rate of glucose absorption and consequently blunting the rise of postprandial plasma glucose level [120].

Such inhibitors which are available in clinical use are

Acarbose
Miglitol
Voglibose [20].

The continuous use of such synthetic agents may cause side effects such as

Flatulence
Abdominal cramp
Vomiting
Diarrhoea.

Management of any metabolic syndromes without side effects is challenge to the medical system [37].

Such drugs for novel hypoglycemic compounds from HMs have become an important aspect because of their efficacy in the human clinical trials with minimal side effects and extra ordinary sources for DMs [118].

2.6 Diagnosis and management of diabetes mellitus

2.6.1 Diagnosis of DM:

Diabetes mellitus is recurrent or persistent hyperglycemia, diagnosed by demonstrating as follows:

\[ FBG \geq 126 \text{ mg/dL} \]
Blood glucose $\geq 200$ mg/dL after Two hours with 75g oral glucose administration as in a GTT in human

Glycosylated hemoglobin (HbA$_1$C) $\geq 6.5$-$7.0\%$

The positive result, in the absence of unequivocal hyperglycemia, by repeating any of the above bio-marker tests on a different day can be confirmed.

It is preferable to measure a fasting glucose level because of the ease of measurement than OGTT (takes 2-3 hours to complete)

According to the new definition, two FBG measurements are above 126 mg/dL are considered diagnostic for DMs.

People with FBG levels from 110 - 125 mg/dL are considered to have impaired FBG.

Patients with plasma glucose at or above 140 mg/dL, but not more than 200 mg/dL, two hours after a 75 g oral glucose load are considered to be impaired glucose tolerance in human. In these states, the latter in particular is major risk factor for DMs and cardiovascular disease (CVD).

Glycosylated Hb is better than FBG to determine the risks of CVD and death due to any cause [211].
Table: 2.2 Simplified schemes for diabetic diagnosis

<table>
<thead>
<tr>
<th>Condition for diabetic Diagnosis</th>
<th>2 hr glucose (mg/dL)</th>
<th>Fasting glucose mg/dL</th>
<th>Hba1c (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>&lt;140</td>
<td>&lt;110</td>
<td>&lt; 6.0</td>
</tr>
<tr>
<td>Impaired FBG</td>
<td>&lt;140</td>
<td>≥110 &amp; &lt;126</td>
<td>6.0–6.4</td>
</tr>
<tr>
<td>Impaired OGTT</td>
<td>≥140</td>
<td>&lt;126</td>
<td>6.0–6.4</td>
</tr>
<tr>
<td>DMs</td>
<td>≥126</td>
<td>≥126</td>
<td>≥ 6.5</td>
</tr>
</tbody>
</table>

2.6.2 Management of DM:

(A) Treatment of T-1 DMs (IDDM)

Insulin administration is comprehensive DMs treatment plan that is necessary for T-1 DMs. Insulin treatment plans:

Intensive treatment
Standard (conventional) treatment

Intensive therapy is recommended for T-1 DMs. Types of insulin preparations are classified according to its source, duration of action.

Insulin preparations:

I) Sources: Bovine, Porcine Human (Semi-synthetic or DNA technology) [161].

II) Action:

Rapid-Acting (e.g.: Lispro, Aspart, Glulisine insulin)
Short-Acting (e.g.: Regular insulin)
Intermediate acting (e.g.: NPH, Lente).
Long-Acting (e.g.: ultra lente, Glargine insulin)
(A) Treatment of T-2 DMs (NIDDM) [50, 83].

Table: 2.3 Drugs Classification of oral anti-diabetic drugs [50]

<table>
<thead>
<tr>
<th>S.N0</th>
<th>Drugs</th>
<th>Mechanism</th>
<th>Uses</th>
<th>Adverse effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Sulphonylure (SUs) as (glyburide, glimepiride, glibenclamide, glipizide)</td>
<td>Stimulating insulin release by pancreatic β cells by inhibiting the KATP channel.</td>
<td>Type 2 Diabetes</td>
<td>Hypoglycemia mostly mild to moderate, can cause fatal complication, - Weight gain - GL disturbances - Hypersensitivity reactions</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Short-acting and safe</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Biguanides (Metformin, phenformin)</td>
<td>Acts on liver to cause decrease in IRc</td>
<td>Metformin is a drug of first choice in Obeic (more wt) patients. (With sulfonylurea).</td>
<td>Transient nausea, anorexia or diarrhoea, discomfort of abdomen and metallic taste. During chronic metformin therapy decreases intestinal absorption of vit B12 and folate</td>
</tr>
<tr>
<td>3</td>
<td>Alpha-glucosidase inhibitor (acarbose, miglitol, voglibose)</td>
<td>Decreases glucose absorbance on small intestine so decrease in synthesis of carbohydrates digesting enzymes</td>
<td>Predominately used in postprandial hyperglycaemia. Less potent, with mean HbA1c reduction.</td>
<td>Gastrointestinal effects Dose-related flatulence, diarrhoea, Abdominal bloating.</td>
</tr>
</tbody>
</table>
### 2.6.3 Treatment for hypoglycemia:

- Initially 10-20 g of glucose is given by mouth either as solution or granules.
- In acute insulin-induced hypoglycemia, Glucagon (1mg ie.,1unit by im) can be given but not for chronic hypoglycemia, If not effective within 10 min, intravenous glucose can be given.
Alternatively, 20% glucose (50 ml iv) infusion may be suggested into a large vein through without causing extravasation since it is an irritant at this concentration.

Or 25 ml of 50% glucose (25 ml iv) infusion may be suggested though this concentration is viscous and difficult to administer.

Or 10 % can be given but a large volume is required.

Oral hypoglycemic drugs causing hypoglycemia must be taken to a hospital; it may persist for many hours [50].

2.7 Role of Free radicals in diabetes mellitus.

2.7.1 Free radicals (FRs):

Free radicals are the atoms or molecules with one or more loan pair of electrons are independent existence. The unpaired electrons that characterized an oxygen free radical confer a high level of instability and thus a high chemical reactivity on the molecule. FR causes Arthritis, aging, heart attack, cancer, atherosclerosis, trauma, hyperoxia, stroke, cataractogenesisis, asthma, dermatitis, vasospasm, periodontitis, sexual disfunction, liver injury, retinal damage [11, 13].

- FRs is responsible for many diseases, such as diabetes, CV disordes, pulmonary diseases and inflammation.
- FRs is involved in important physiological processes such as ageing [73].
- FRs are formed in human body constantly, but toxic if generates in excess or in natural antioxidant deficiency.

2.7.2 Formation of free radicals:

- A group of oxidants is known as ‘Reactive oxygen species (ROSp).

They are FRs or molecular species having capacity to produce FRs.
Superoxide (O$_2^-$) radicals and nitric oxide (NO) radicals generates ROS intracellularly

By normal physiological mitochondrial respiration and phagocytosis, nearly 2% of the oxygen is consumed by the body and converted into O$_2^-$

During infections, exercise, exposure to pollutants, UV light, ionizing radiation the ROS% will increase.

Nitric oxide synthase enzymes produces an endothelial relaxing factor and neurotransmitter, NO

NO and O$_2^-$ radicals (RO), proxy radicals (ROO), singlet oxygen ($^1$O$_2$) are produced by complex transformation reactions

Few radical species are converted to molecular oxidants such as hydrogen peroxide (H$_2$O$_2$), peroxynitrite (ONOO$^-$), hypochlorous acid (HOCl). Occasionally molecular species act as source of ROS [13].

For example, H$_2$O$_2$ is converted to OH radicals by Fenton reaction and HOCl through its reaction with H$_2$O$_2$ can be converted to O$_2$. ONOO$^-$ at physiological concentrations of carbon dioxide becomes a source of carbonate radical anion (CO$_3^{2-}$). Types free radicals:

Free radicals are 5 types. The first four types come from oxygen atoms and are called Reactive Oxygen Species (ROS), but the fifth type derives from nitrogen.

1) Superoxide ion (O)
2) Hydroxyl radical (OH)
3) Singlet oxygen
4) Hydrogen peroxide (H$_2$O$_2$)
5) Reactive Nitrogen Species (RNS).
2.7.3 Redox state and oxidative stress:

- Redox state is defined as ‘the total potential reduction or the reducing capacity of all redox couples such as GSSG/2GSH, NAD+/NADH, Asc•−/AcsH−, etc., present in biological fluids, organelles, cells or tissues.

- Steady state concentration of ROS is determined by the homeostasis of production and removal by various antioxidants this maintain the quality of life.

- The redox state of a cell is determined by concentration of concentration of reducing species like GSH, NADH, FADH, etc., which is stored in many cellular constituents.

Oxidative stress induced clinical damages [13]

- Kidney: Renal graft, Glomerulo nephritis
- Skin: Burn, Dermatis, Psoriasis
- Heart: Angioplasty, Keshan disease (selenium deficiency)
- Joint: Rheumatoid arthritis
- Lung: Asthma, Hypoxia
- Brain: Trauma, Stroke, Neuro toxins, Alzheimer and Parkins disease
- Multi organ: Radiation, Ageing, Cancer, Inflammatory immune, Diabetes
- Vessel: Vaso spasm, Atherosclerosis
- GI: Iscemic bowel, Liver endotoxin
- Eye: Degenarative retinal damage, Cataracto genesis.

2.7.4 Oxidative stress Vs Diabetes:

Oxidative damages leads to imbalance of free radicals and detoxifying or repair systems in any cell, tissue or organ.
FRs (a loan pair of electrons) induces damage and destroys lipids, proteins, RNA, DNA and this activates diseases and disorders of vital organs [170].

Oxidative stress is a contributing factor to diabetes, cancer, kidney disease, Alzheimer’s disease, Parkinson’s disease, schizophrenia, bipolar disorder, atherosclerosis (hardening of arteries), arthritis, emphysema, and cataracts [129].

Oxidative stress mediates and complicates the hyperglycemia leads to detrimental effect of glucose toxicity which leads to diabetic complications.

When glucose continuously flows through vascular endothelial cells, (since it is the primary vulnerable targets of glucose) hyperglycemic damage the small vessels, arteries and peripheral nerves leads to different pathological changes.

Chronic DMs and oxidative stress may implicate to multiple biochemical pathways and mechanisms of action on the function of vascular, retinal, and renal tissues [178].

Considerably less work has been performed using islet tissue. Diabetic patients are often under oxidative stress and also.

1) Plasma lipid peroxides appear higher than normal in diabetes.
2) Level of F₂ isopretanes and lipid peroxides are also elevated.
3) Erythrocyte GSH levels are also slightly abnormal. Elevated levels of 8-hydroxydeoxy guanosine (8-OH dg)
4) Activated NFKB in blood monocytes have also been reported in diabetes.

2.8 Antioxidants (AO)

2.8.1 Definition:

Antioxidants terminate the free radical generating chain reactions by removing intermediates, inhibiting other oxidation (oxidized themselves). As a result,
AO are often reducing agents such as thiols or polyphenols. They are believed to play a role in preventing the development of chronic diseases such as Diabetic, cancer, heart disease, stroke, Alzheimer's disease, Rheumatoid arthritis, cataracts etc [37].

Antioxidants (AO) are the chemical agents which inhibits the oxidative damage to a target molecule, slows or prevents the oxidation of other molecules, protect cells from damage caused by free radicals [13, 117].

2.8.2 Types antioxidant:

a) In-vivo antioxidant

b) In-vitro antioxidant

c) Natural antioxidants

A) In-vivo antioxidant classification

Enzymatic antioxidants:

These are endogenous antioxidants and are first line of defense against super oxide and hydrogen peroxide radicals mediated injury carried out by using enzymatic antioxidants.

i. Primary antioxidants e.g. SOD, Catalase, Glutathione peroxidase.

ii. Secondary enzymes e.g. - Glutathione reductase [37].

Non-Enzymatic antioxidants:

The second line of defense carried out by non-enzymatic antioxidants.

Natural antioxidants:

Natural antioxidants have been implicated in the prevention of diabetic, cancer and oxidative stress related diseases such as

i. Vitamins: Vitamin A (Retinoid), Vitamin C (Ascorbic acid), vitamin E (Tocopherols), selenium

ii. Carotenoids: β-carotene, Lycopene
iii. Mineral: Zinc, Selenium

iv. Phenolic compounds: vegetables, fruits, cereals, nuts, egg, meat and legumes [28, 62].

### 2.8.3 Classification of antioxidants and Natural antioxidants:

**Table: 2.4 Antioxidants and Natural antioxidants [28]:**

<table>
<thead>
<tr>
<th>Primary antioxidants</th>
<th>Secondary antioxidants</th>
</tr>
</thead>
<tbody>
<tr>
<td>They are chain breaking antioxidants which reacts with lipids radicals and converts into stable products.</td>
<td>These are phenolic compounds that perform the function of capturing free radicals and stop the chain reaction.</td>
</tr>
<tr>
<td>1. Antioxidant minerals</td>
<td>1. Butylated hydroxyl anisole (BHA)</td>
</tr>
<tr>
<td>Selenium</td>
<td>2. Butylated hydroxyl toluene (BHT)</td>
</tr>
<tr>
<td>Copper</td>
<td>3. Propyl gallate (PG)</td>
</tr>
<tr>
<td>Zinc and manganese.</td>
<td>4. Tertiary butyl hydroxyl quinine (TBHQ)</td>
</tr>
<tr>
<td>2. Antioxidants vitamins</td>
<td>5. Noedihydroguaretic acid (NDGA)</td>
</tr>
<tr>
<td>Vitamin A</td>
<td></td>
</tr>
<tr>
<td>Vitamin B</td>
<td></td>
</tr>
<tr>
<td>Vitamin E</td>
<td></td>
</tr>
<tr>
<td>3. Flavonoids</td>
<td></td>
</tr>
<tr>
<td>NuHMseg</td>
<td></td>
</tr>
<tr>
<td>Clove</td>
<td></td>
</tr>
<tr>
<td>Black pepper</td>
<td></td>
</tr>
<tr>
<td>Ginger</td>
<td></td>
</tr>
</tbody>
</table>


<table>
<thead>
<tr>
<th>Natural antioxidants</th>
<th>Synthetic antioxidants</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tocopherols</td>
<td>Butylated hydroxyl toluene (BHT)</td>
</tr>
<tr>
<td>Noedihydroguaretic acid (NDGA)</td>
<td>Butylated hydroxyl anisole (BHA)</td>
</tr>
<tr>
<td>Sesamole</td>
<td>Propyl gallate (PG)</td>
</tr>
</tbody>
</table>

2.8.4 Functions of anti oxidants:

- Supports the generation of β cells in pancreas
- Supports kidney functions
- Protects the liver
- Maintain healthy vision
- Reduces obesity
- Helps in systemic circulation
- Maintain good dental health
- Improves reproductive functions
- Possess good anti-aging effects
- Supports the immune system and improves
deference power of body
- Offers protection against digestive disorders
- Supports respiratory system
- Improves quality of sleep [13].
2.8.5 Antioxidant defence:

Diabetes involves superoxide anion and hydroxyl radicals. In addition to these enzymes, GSH-R and glutathione-S-transferase (GST) provide GSH and help to neutralize toxic electrophiles respectively.

SOD, CAT and glutathione peroxidase (GSH-px) can be counteracted the effects of superoxide anion and hydroxyl radicals in addition to that glutathione reductase (GSH-R) and glutathione-S-Transferase (GST) provide glutathione (GSH) help to neutralize toxic electrophiles of FRs.

2.8.6 Source of antioxidants:

(A) **Food source of antioxidants**: Antioxidants are found in varying amount in foods such as vegetables, fruits, grain cereals, eggs, meat, legumes and nuts.

(B) **Synthetic antioxidants** include Butylated Hydroxyl Anisole (BHA), Butylated Hydroxyl Toluene (BHT), Propyl Gallate (PG) and Tertiary Butyl Hydro-Quinone (TBHQ).
(C) Natural anti-oxidants examples: Apple, Blue berrie, Brussel sprout, Carrot, Cauliflower, Olives, Grape, Oranges, Papaya etc.,

2.8.7 Antioxidant enzymes:

The antioxidant enzymes Super Oxide Dismutase (SOD), Catalase (CAT) and Glutathione peroxidase (GPx), Glutathione reductase, Thioredoxin reductase, Hemeoxygenase and Biliverdin reductase serve as primary line of defense in destroying free radicals [13] this also defense against ROS.

---

**Figure: 2-4 Roles of antioxidant enzymes**

A) *Superoxide dismutase*

Superoxide dismutase (SOD) is an enzyme that removes the superoxide \( (\text{O}_2^-) \) radical, repairs cells and reduces the damage done to them by superoxide, the most common free radical in the body. SOD is found in both dermis and epidermis and is key to the production of healthy fibroblasts (skin-building cells).

\[
2 \text{H}_2\text{O}_2 \rightarrow 2 \text{H}_2\text{O} + \text{O}_2
\]
Mechanism of action

Superoxide Dismutase (SOD) catalyzes the reduction of superoxide anions to hydrogen peroxide. It plays a critical role in the defense of cells against the toxic effects of oxygen radicals. SOD competes with nitric oxide (NO) for superoxide anion, which inactivates NO to form peroxy nitrite. Therefore, by scavenging superoxide anions, SOD promotes the activity of NO. SOD has suppressed apoptosis in cultured rat ovarian follicles, neural apoptosis in neural cell lines and transgenic mice by preventing the conversion of NO to peroxynitrate, an inducer of apoptosis. Covalent conjugation of superoxide dismutase with polyethylene glycol (PEG) has been found to increase the circulatory half-life and provides prolonged protection from partially reduced oxygen species [67, 166].

\[
M^{(n+1)+} + \text{SOD} + O_2^- M^{n+} \rightarrow \text{SOD} + O_2
\]

\[
M^{n+} - \text{SOD} + O_2^- + 2H^+ M^{(n+1)+} \rightarrow - \text{SOD} + H_2O_2
\]

Where M = Cu (n=1)
Mn (n=2), Fe (n=2), Ni (n=2)

In this reaction the oxidation state of the metal cations oscillates between n and n+1.

\[
\text{Superoxide Dismutase}
\]
\[
O_2^\cdot \quad \text{O} \quad O_2
\]

\[
\text{Superoxide Dismutase}
\]
\[
O_2 + 2H^+ \quad \text{H}_2O_2
\]

It is found in both prokaryotic and eukaryotic cells that catalyzes the superoxide radical into oxygen and hydrogen peroxide. The rate of spontaneous decay of superoxide is significantly increased by the action of superoxide dismutases
(SODs) found in many cell types. In eukaryotic cells, copper containing enzyme (Cu-SOD) and zinc containing enzyme (Zn-SOD) are present in cytosol. The second type manganese containing enzyme (Mn-SOD) is present in mitochondrial matrix. In prokaryotic cells, iron containing enzymes (Fe-SOD) and manganese containing enzyme (Mn-SOD) are present.

$$2O_2 \cdot^− + 2H → H_2O_2 + O_2$$

A) Catalase:

Catalase is a haeme containing enzyme that catalyses the dismutation of hydrogen peroxide into water and oxygen. The enzyme is found in all aerobic eukaryotes and is important in the removal of hydrogen peroxide generated in peroxisomes by oxidases involved in β-oxidation of fatty acids, glyoxalate cycle and purine catabolism [3].

$$2 H_2O_2 → 2 H_2O + O_2$$

B) Glutathione reductase

GSH is highly abundant in all cell compartments and is the major soluble antioxidant. GSH/GSSG ratio is a major determinant of oxidative stress. GSH shows its antioxidant effects in several ways. It detoxifies hydrogen peroxide and lipid peroxides via action of GSH-Px. GSH donates its electron to $H_2O_2$ to reduce it into $H_2O$ and $O_2$. GSSG is again reduced into GSH by GSH reductase that uses NADPH as the electron donor. GSH-Pxs are also important for the protection of cell membrane from lipid peroxidation. Reduced glutathione donates protons to membrane lipids and protects them from oxidant attacks.

Mechanism of action

GSH is a cofactor for several detoxifying enzymes, such as GSH-Px and transferase. It has a role in converting vitamin C and E back to their active forms. GSH protects cells against apoptosis by interacting with proapoptotic and anti apoptotic
signalling pathways. It also regulates and activates several transcription factors, such as AP-1, NF-kB and Sp-1.

\[ 2\text{GSH} + \text{H}_2\text{O}_2 \rightarrow \text{GSSG} + 2\text{H}_2\text{O} \]

**C) Peroxiredoxins**

Peroxy redoxins are peroxidases that catalyse the reduction of hydrogen peroxide, organic hydro peroxides, as well as peroxynitrite. They are divided into three classes: typical 2-cysteine peroxiredoxins; atypical 2-cysteine peroxiredoxins; and 1-cysteine peroxiredoxins. These enzymes share the same basic catalytic mechanism, in which a redox-active cysteine (the peroxidatic cysteine) in the active site is oxidized to a sulfonic acid by the peroxide substrate. Over-oxidation of this cysteine residue in peroxiredoxins inactivates these enzymes, but this can be reversed by the action of sulfiredoxin. Peroxiredoxins seem to be important in antioxidant metabolism, as mice lacking peroxiredoxin 1 or 2 have shortened lifespan and suffer from haemolytic anaemia, while plants use peroxiredoxins to remove hydrogen peroxide generated in chloroplasts.

**D) Glutathione peroxidases (GPX)**

Glutathione peroxidases catalyze the reaction of hydro peroxides with reduced glutathione to form glutathione disulphide and the reduction product of the hydro peroxide (OH-). GSH, which consists of three amino acids, is an essential component of this system and serves as a cofactor for an enzyme called glutathione transferase.

\[ \text{H}_2\text{O}_2 + \text{GSH} \stackrel{\text{GPX}}{\rightarrow} \text{H}_2\text{O} + \text{GSSG} \]

\[ \text{GSSG} + \text{NAD} (\text{P}) \text{H} \stackrel{\text{GR}}{\rightarrow} \text{GSH} + \text{NAD} (\text{P})^+ \]
2.9 Experimental models for diabetes mellitus

Animal models have been extremely contributing to DMs studies and metabolic disorder with abnormal blood glucose level analysis. It helps the investigators to get more opportunity to control DMs.

Animal models are human-like biological characteristics thus develop diabetes either spontaneously or by using chemical, surgical, genetic or other techniques.

2.9.1 Chemically induced diabetes:

Chemical which induces diabetes is called as diabetogenic agent. Chemical agents which produce diabetes can be classified into three categories that, specifically damage β-cells, cause temporary inhibition of insulin production and / or secretion and diminish the metabolic efficacy of insulin in target tissue [41, 122, 146, 177].

Streptozotocin (STZ) induced diabetes mellitus

1. Streptozotocin (STZ) is chemically 2-deoxy-2-(3-(methyl-3-nitrosoureido)-D-glucopyranose), derived from Streptomyces achromogenes.

2. STZ is used for the induction of T-1 & T-2 DMs (ie., IDDM and NIDDM).

3. 40-60mg/kg b.w, STZ is used as a single intravenous dose in adult rats to induce IDDM. Some times higher doses also used (ip): 100 mg/kg of STZ is used for NIDDM induction.

4. After two hours injections of STZ, acts on β-cells accompanied by characteristic change in blood insulin and glucose concentrations, 6 hr later high level of insulin followed by drop in blood glucose occurs, finally the consistent drop of blood insulin will leads to hyperglycemia by impairing the glucose oxidation and decreases insulin bio-synthesis and secretion.
5. The action of STZ was observed that first protects the β-cell response to glucose followed by cells damage and permanent loss of the β cells (changes of DNA pancreatic β-cells fragmentation).

6. STZ reaches the pancreatic β-cells through GLUT 2. alkylation of DNA in β cells leads to cell death.

7. STZ donates nitric oxide and ROS which destructs pancreatic islet cells and induces DNA damage then activates poly ADP-ribosylation. This process causes cellular NAD+ depletion further reduction of the ATP content and subsequent inhibition of insulin synthesis.

8. Potential problem of STZ: The toxic effects of STZ are not restricted to pancreatic β-cells which also leads to cause renal injury, inflammation and endothelial dysfunction as well [188, 189].

Figure 2-5: Mechanism of action Streptozotocin on β-cells in pancreas
Table: 2.5 Dose and Route of administration (ROA) of STZ in different animals

<table>
<thead>
<tr>
<th>Species</th>
<th>Dose(mg/kg)</th>
<th>Route</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rats</td>
<td>35-65</td>
<td>i.v or i.p</td>
</tr>
<tr>
<td>Mouse</td>
<td>100-200</td>
<td>i.v or i.p</td>
</tr>
<tr>
<td>Hamsters</td>
<td>50</td>
<td>i.p</td>
</tr>
<tr>
<td>Dogs</td>
<td>20-30</td>
<td>i.v</td>
</tr>
<tr>
<td>Pigs</td>
<td>100-150</td>
<td>i.v</td>
</tr>
</tbody>
</table>

Disadvantages of STZ as diabetogenic agent:

1. Mortality of STZ induced diabetic animal is 7%
2. Variability of hyperglycemia is high
3. Hyperglycemia develops by direct cytotoxic action on β cells
4. Guinea pig and rabbits are resistant to its diabetogenic action.

Table: 2.6 Differences between Alloxan and Streptozotocin

<table>
<thead>
<tr>
<th>Streptozotocin</th>
<th>Alloxan</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Maximum blood glucose level during phase of acute hyperglycemia occurs within 120 min.</td>
<td>1. Maximum blood glucose level during phase of acute hyperglycemia occurs within 45 min.</td>
</tr>
<tr>
<td>2. Liver glycogen depletes slowly</td>
<td>2. Liver glycogen depletes faster</td>
</tr>
<tr>
<td>3. Hypoglycemia more severe</td>
<td>3. Hypoglycemia less severe</td>
</tr>
<tr>
<td>4. Mortality rate 8%</td>
<td>4. Mortality rate 37%</td>
</tr>
</tbody>
</table>
### iii) Other diabetogenic agents

**Table: 2.7 Diabetogenic agents**

<table>
<thead>
<tr>
<th>S. No</th>
<th>Compound</th>
<th>Dose</th>
<th>Animal</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Dehydro ascorbic acid</td>
<td>650 mg/kg (for 3 days)</td>
<td>Rats</td>
</tr>
<tr>
<td>2.</td>
<td>Dehydroglucoascorbic acid</td>
<td>3.5-3.9 gm/kg</td>
<td>Rats</td>
</tr>
<tr>
<td>3.</td>
<td>Methyl Alloxan</td>
<td>53 mg/kg</td>
<td>Rats</td>
</tr>
<tr>
<td>4.</td>
<td>Ethyl Alloxan</td>
<td>53-130 mg/kg</td>
<td>Rats</td>
</tr>
<tr>
<td>5.</td>
<td>Oxime and Dithizone</td>
<td>53 mg/kg</td>
<td>Rabbits</td>
</tr>
<tr>
<td>6.</td>
<td>Sodium Diethyldithiocarbonate</td>
<td>0.5-1 gm/kg</td>
<td>Rabbits</td>
</tr>
<tr>
<td>7.</td>
<td>Potassium Xanthate</td>
<td>200-350 mg/kg</td>
<td>Rabbits</td>
</tr>
<tr>
<td>8.</td>
<td>Uric Acid</td>
<td>1 gm/kg</td>
<td>Rabbits</td>
</tr>
<tr>
<td>9.</td>
<td>Alloxan</td>
<td>40-200 mg/kg</td>
<td>Rats</td>
</tr>
</tbody>
</table>

**2.9.2 Surgical and other diabetic induction methods:**

i. Pancreatectomy [23, 61]

ii. Growth hormone induced diabetes

iii. Steroid induced diabetes

iv. Genetically induced diabetes [206]

v. Virus induced diabetes [104, 206].
2.10 Therapeutic role of phytomedicine in the management of diabetes

Depends on the nature of disease, insulin and certain synthetic drugs like Sulphonylureas, Biguanidines and Acarbose (SABC treatment) are widely used in DMs treatment. In recent years, evidence of cases of “IRc” and the occurrences of side effects from prolonged administration of conventional drugs have triggered the search for safe and effective alternatives. Several plant extracts and isolated phyto-chemicals have been examined for anti-diabetic activity with a view to identify alternative treatment strategies for diabetes. It has been observed that certain resistant cases of diabetes that do not respond well to SABCS drugs often respond well to supplementation with natural remedies [19].

Herbal remedy the ancient healing system, from India, has steadily increased in popularity in the western world in recent years. Treatment with specific herbs and minerals to cure various diseases widely developed. The botanicals in the Ayurvedic Meteria Medica have been proven to be safe and effective, through several hundred to thousand years of use. The positive role of traditional medicinal plant in the prevention or control of some metabolic disorders like diabetes, heart disease and certain types of cancer. The great advantages of these medicinal plants are easily available and have no proven side effects.

The incidence of diabetes mellitus is increasing all over the world and is becoming a problem of significant importance in the affluent societies. Oral anti-hyperglycemic drugs have been frequently used for controlling non-insulin dependent diabetes mellitus (T-2) but synthetic medicines have side effects.

Medicinal plants and herbs are of great importance to the health of individuals and communities. Despite the existence of HMs over many centuries, only relatively small numbers of plant species have been studied for their application. However, in the recent past, an increasing research evidence is getting accumulated, which clearly indicate the positive role of traditional medicinal plants in the prevention or control of some metabolic disorders like diabetes. The great advantage of this medicinal plant is that these are easily available and have minimal side effects. The hypoglycemic and
hypolipidemic effect of various traditional medicinal plants like fenugreek, jambu and bitterguard are reported but, there is limited information on the nutritional and anti-nutritional factors of these three medicinal plants which lower the blood glucose and lipid profile [38].