Chapter II Literature Review

2.1 Introduction:

Diabetes mellitus is a metabolic disorder in the endocrine system & metabolic disorder causing hyperglycemia. Diabetes affects about 5% of the global population (Chakraborty R et al 2002) and management of diabetes without any side effects is still a challenge to the medical system (Kameswara Rao B et al 2003).

In India, the prevalence rate of diabetes is estimated to be 1-5%. Diabetes is becoming the third "killer" of the health of mankind along with cancer, cardiovascular and cerebrovascular diseases because of its high prevalence, morbidity and mortality (Venkatesh S et al 2003). The cause of diabetes is a mystery, although both genetic and environmental factors such as obesity and lack of exercise appear to play a role (Venkatesh S et al 2003). Ethnic and racial differences have been found in heterogeneous populations within the same area. As a rule, incidence is highest in Scandinavian countries, intermediate in the US, Spain, and Israel, and lowest in Asian and most Latin American countries. Most researchers believe that, in the presence of a genetic predisposition, something in the environment triggers the development of diabetes (Venkatesh S et al 2003). With a long course and serious complications often resulting in high death rate, the treatment of this disorder takes three main forms:

1. Diet and exercise
2. Insulin replacement therapy and
3. The use of oral hypoglycemic agents.

2.2. History of Diabetes Mellitus:

The first clinical description and complication of diabetes mellitus by Egyptian physicians 3,500 years ago who had been striving to diagnose and treat diabetes. Egyptian medicine influenced the medical practices of neighboring cultures; including the culture of ancient Greece (MacCracken J. et al 1997). it is fact that Vedas are oldest literature of civilization. Athervaveda the latest among the Vedas is very close to medical science. It shows rudiment of knowledge about diabetes (Akashpati Gairola et al 1970). In Kausik Sutra of Athervaveda there is reference of prameha. in vedik literature two terms were denoted i.e. asrava and prameha. Asrava means "to flow". Sayana and Kesava commentors of Vedic works included the Mutratisara i.e. excessive urination, which

throws light on existence of reference to state of prameha in Vedic period (Akashpati Gairola et al 1970). .

Table-01: Top 10 countries for estimated numbers of adults with diabetes, 2010 and 2030 (IDF 2009; A Ramachandran et al 2010)

Rank	Country	2010	Country	2030
1.	India	50.8	India	87.0
2.	China	43.2	China	62.6
3.	U. S.	26.8	U. S.	36.0
4.	Russian Federation	9.6	Pakistan	13.8
5.	Brazil	7.6	Brazil	12.7
6.	Germany	7.5	Indonesia	12.0
7.	Pakistan	7.1	Mexico	11.9
8.	Japan	7.1	Bangladesh	10.4
9.	Indonesia	7.0	Russian Federation	10.3
10.	Mexico	6.8	Egypt	8.6

Kautilya, the father of diplomacy of 321-296 B. C. mentioned a method to produce diabetes in his book Arthasastra in the chapter dealing with means to injury the enemies. According to him burnt Krakalasa (Chameleon) and lizard combined with intestine of frog and honey, if administered to subject cause diabetes, this evidently points existence of diabetogenic techniques in ancient time (Akashpati Gairola et al 1970). After vedic period of book Charaka Samhita written in 600 B. C. explains various factors of etiology including heridatory, pathogenesis, symptomatology, complications and method of treatment in details. Regarding etiology of diabetes Charaka Samhita describes over neutrition and lack of exercise as main causative factors. Inherited diabetes is considered as incurable by Charaka (UPV Dhyay et al 1987). Bhela Samhita which is comtemtorary to Charaka Samhita described two types of diabetes mellitus i.e. Prakrti Prabhav (Congenital) and Swarakta (Acquired) (Shukla G. D. 1959)

Around 250 BC Apollonius of Memphis probably coined the name "diabetes" from Ionian Greek. The literal translation, is "to go through" or siphon, reflecting the ear-

ly understanding of a disease that drained patients of more fluid than they could consume (MacCracken J et al 1997).

Aretaeus of CapPVDocia who provided a detailed and accurate account and first used the term "diabetes" in the 2nd century AD and he concluded that it was due to a fault in the kidney, a generic description for conditions that are instantly recognizable today (Pickup JC et al 2003).

He described diabetes with the following words:

"Diabetes is a dreadful affliction, not very frequent among men, being a melting down of the flesh and limbs into urine. The patients never stop making water and the flow is incessant, like the opening of aqueducts. Life is short, unpleasant and painful, thirst unquenchable, drinking excessive, and disproportionate to the large quantity of urine, for yet more urine is passed. The patients are affected by nausea, restlessness and burning thirst, and within a short time they expire (Savona Ventura C 2002).

2.3. Discovery of Insulin:

The discovery of insulin was a seminal event in both the study of diabetes and the care of diabetic patients. The development of procedures for purifying and modifying insulin took an additional 30 years. In his masterful rendition of these developments, Michael Bliss recounts the remarkable story surrounding the discovery of insulin and notes that the discovery of insulin at the University of Toronto in 1921-22 was one of the most dramatic events in the history of the treatment of disease (Joshi SR et al 2007; Shah SN et al 2007).

It was only in 1893 that Edouard Laguesse (1861 – 1927) suggested that these clumps of cells in pancreas might constitute the endocrine tissue of the pancreas, which he named the Islets of Langerhans (named after Paul Langerhans) (Pickup JC et al 2003).

In Germany, Oskar Minkowski and Joseph von Mering observed that total pancreatectomy in experimental animals leads to the development of severe diabetes mellitus and begun the speculation that a mysterious substance produced by the pancreas is responsible for metabolic control (Shah SN et al 2007; Joshi SR et al 2007). Islets of Langerhans this theme was continued further by the Belgian physician, Jean de Meyer (Lasker SP. et al 2010). By the first decade of the Twentieth Century it was widely hy-

pothesized that an “internal solution” of the pancreas controls carbohydrate metabolism. Even so there was so much impressionistic evidence supporting the existence of pancreatic internal secretion emanating from the islet cells that in 1907, J de Meyer who in 1909 isolated glucose lowering hormone from the pancreas proposed it be named “insulin” (Latin, insula= Island, as it was produced by islet cells) In 1916, Sharpey Schafer in Britain independently suggested the same name (Shah SN et al 2007;. Lasker SP et al 2010). As soon as the link between the pancreas and diabetes was recognized, researchers focused on treating diabetes with pancreatic extracts. Several workers including Georg Ludwig Zülzer (Germany), Nicolas Paulesco (Romania), EL Scott and Isael Kleinter (North America) were actively trying to search for the active principal of insulin. In the early 1900s, Zülzer experimented with pancreatic compounds and made an injection called "acomatrol" into a dying diabetic patient. The patient improved but later died when Zülzer’s acomatrol supply was exhausted (Pickup JC et al 2003). Later, in 1911, a European pharmaceutical company funded a small laboratory and some workers to help Zulzer, who took out an American patient on his "Pancreas Preparation Suitable for the Treatment of Diabetes”. But Zulzer’s laboratory was turned over to the military during World War 1 (Pickup JC et al 2003).

In the early years of 20th century purification, removal of toxic products and demonstrations consisting of biological activity were the principal problem. Professor John James Rickard Macleod, a physiologist at the University of Toronto, continued to pursue his work on carbohydrate metabolism. In 1921, Frederick G. Banting was hired and Charles Herbert Best, a 21 year old student was recruited for assisting Banting, who proved that it is an active substance of the pancreas that is associated with hypoglycemia in diabetic dogs under Macleod’s patronization. James Bertram Collip, a biochemist, later joined the team, and improved the extraction and purification of insulin. The most important demonstration was that the pancreatic extract enabled the diabetic liver to store glycogen and it could clear ketouria. Discovery of insulin for the treatment of diabetes represents one of the major humanitarian and scientific milestones of this century. It was a momentous advance in medicine (Pickup JC et al 2003).

The first clinical trial of insulin was yielding encouraging results. On 1 January 1922, at the Medical Service of the Toronto General Hospital the insulin extract made by

Banting and Best was first injected into Leonard Thompson, a 14 year old boy who weighed only 64 pounds, dying of diabetes in Toronto Hospital. Leonard was given a 5 - 10 ml injection in each buttock. But it failed to relieve the symptoms. Abscesses developed at the injection sites, and Leonard became even more acutely ill. However, his blood glucose level dropped initially. On 23rd January insulin extract injection refined by Collip reduced Thompson's blood sugar to normal (from 520-120 mg/dL) in about 24 hours and abolished his glycosuria and ketonuria. He began to gain weight and regain strength. Leonard lived a relatively healthy life for 13 more years but died of pneumonia at the age of 27 (Mac Cracken J et al 1997; Elliott P J 1922). Though, insulin was discovered by the team of four, the credit for the discovery of insulin is given to Banting and Macleod who won the Noble Prize in 1923. In an attempt to remedy this injustice Banting publicly acknowledged best's role in the discovery of insulin and Banting shared the prize money equally with Best while Macleod agreed to do the same with Collip (Lasker SP et al 2010). In 1955, Sir Frederick Sanger characterized the amino acid sequence of human insulin, making it the first protein to whose sequence was determined. He was awarded the 1958 Nobel Prize in Chemistry for this work (Das A. K et al 2011).

2.4. Mechanism of Insulin action:

Insulin is a hormone that is central to regulating glucose metabolism in the body to produce energy. All insulin's actions peak after feeding, whereas on starving, the hormone's main role is glucagon production. There are three main sites to consider for insulin's activities, those being the liver, muscle and adipose tissue. Starting from blood glucose levels upon the fed state, insulin has the main role to increase the rate of glucose oxidation (glycolysis) in the liver and muscle while at the same time it converts glucose concentrations to glycogen (the form in which glucose can be stored in the body) (Espinal J. 1989). These processes are supported by an increase in the number of glucose transporters to the cell membrane which have a significant role, as well as by a variety of phosphorylation and dephosphorylation processes for activation and deactivation of enzymes. In order for an enzyme to be phosphorylated, a PO_3^{2-} group is donated to it by adenosine triphosphate, ATP. The resulting increase in the rate of glycolysis leads to pyruvate production which is then converted to Acetyl-CoA, one of the main components in biologi-

cal processes, since it supplies muscles with energy and it is used in lipid synthesis in the liver and adipose tissue (Espinal J. 1989). Finally, it is worth mentioning that since glucose oxidation is stimulated by insulin, the reverse process in the liver, gluconeogenesis is inhibited.

At low carbohydrate concentrations, fatty-acid breakdown in the liver is reduced, whereas synthesis is facilitated since triacylglycerols in the adipose tissue are converted to long chain fatty acids. This is favoured due to the larger availability of the responsible synthesizing enzyme, lipoprotein lipase (Newsholme E. A et al 1992). Finally, secondary actions of insulin include stimulation of protein synthesis (Newsholme E. A et al 1992) as well as increased blood flow, vasodilatation, and hypotension (Sartori C et al 1999). All of the actions of insulin are performed via insulin receptors, known as *tyrosine kinase receptors*. These are 2 subunit receptors and contain both an extracellular domain for insulin to bind as a ligand as well as an intracellular part, insulin protein kinase (Siddle K 1992) where all phosphorylation events take place. As with most polypeptide hormones, upon binding, conformational changes are undertaken on the two subunits and a series of phosphorylation events proceed through, leading to more of the actions hormones perform. Internalization of insulin upon binding keeps it in the cell followed by degradation at the end (Desbuquois B et al 1980). In addition, it is worth mentioning that all of these actions and signaling pathways are accompanied by the formation of second messengers such as cyclic adenosine phosphate, cAMP. Second messengers are molecules that disperse information around tissues. The decrease in the concentration of cAMP is the main cause for insulin's activities and this is because of insulin suppressing the precursor molecule, adenylate cyclase, from which the second messenger is synthesized (Pertseva N. M et al 2003).

2.5. Classification of Diabetes Mellitus:

2.5.1. Modern Classification Systems for Diabetes Mellitus:

A major requirement for orderly epidemiologic and clinical research on DM and indeed for management of the disease is an appropriate classification of the disorder. Further, a hallmark in the process of understanding the etiology of DM and studying its natural history is the ability to identify and differentiate its various forms and place them into

a rational etiopathologic framework. Although there have been a number of sets of nomenclature, classification systems, and diagnostic criteria proposed for DM, no systematic categorization existed until the late 1970s.

In 1979, a classification for DM and other categories of glucose intolerance, based on scientific research on this heterogeneous syndrome, was developed by an international workgroup sponsored by the National Diabetes Data Group (NDDG) of the National Institutes of Health (National Diabetes Data Group, 1979). This group recognized DM as being a syndrome, a collection of disorders that have hyperglycemia and glucose intolerance as their hallmark characteristics, due either to insulin deficiency or to impaired effectiveness of insulin's action, or to a combination of these. The World Health Organization (WHO) Expert Committee on Diabetes in 1980 endorsed the substantive recommendations of the NDDG (WHO recommendations, 1980). These groups' distinguished two major forms of DM in Western countries, which they termed *insulin-dependent DM* (type 1 DM) and *non–insulin-dependent DM* (type 2 DM). The older terms "juvenile-onset," "maturity-onset," and "adult-onset" DM were recommended to be abolished.

The NDDG/WHO classification system incorporated data from research conducted during the previous several decades clearly establishing that DM is an etiologically and clinically heterogeneous group of disorders that share glucose intolerance in common.

The evidence in favor of this heterogeneity was overwhelming and included the following:

a. There are many distinct disorders, most of which are individually rare, in which glucose intolerance is a feature.
b. There are large differences in prevalence of the major forms of DM among various racial or ethnic groups worldwide
c. Glucose intolerance presents with variable clinical features, such as the differences between thin, ketosis prone, insulin-dependent DM and obese, nonketotic, insulin-resistant DM
d. Genetic, immunologic, and clinical studies show that, in Western countries, the forms of DM with their onset primarily in youth or in adulthood are distinct entities; and

e. A type of non–insulin-requiring DM in young people, which is inherited in an autosomal dominant fashion, is clearly different from the classic acute-onset DM of juveniles.

These and other collective evidence were used to divide DM into the two major and distinct types;

1. Insulin-dependent DM (type 1 DM) and
2. Non–insulin-dependent DM (type 2 DM).

These two types had different clinical presentations and genetic and environmental etiologic factors that permitted their discrimination. The classification system also included a category termed *other types of diabetes*, in which the causes could be attributed to known factors such as pancreatic disease, insulin receptor abnormalities, and pancreatic injury from drugs or chemicals. Gestational DM (GDM) comprised a fourth class recognized as a condition of hyperglycemia that can occur in pregnancy and lead to higher rates of complications for the fetus and the mother. All of these types of DM were characterized by either fasting hyperglycemia or levels of plasma glucose above defined limits during an oral glucose tolerance test. In addition, the NDDG and WHO recognized a category termed *impaired glucose tolerance*, in which plasma glucose levels during an oral glucose tolerance test lie above normal but below those defined as DM.

The NDDG/WHO classification highlighted the marked heterogeneity of the diabetic syndrome. Such heterogeneity had important implications not only for clinical management of DM but for biomedical research. For example, it indicated that the distinct disorders grouped together under the rubric of DM differ markedly in pathogenesis, natural history, and responses to therapy and preventive measures. In addition, it demonstrated that different genetic and environmental etiologic factors can result in similar diabetic phenotypes.

The classification scheme was devised primarily to serve as a uniform framework for conducting clinical metabolic and epidemiologic research so that more meaningful and comparable data could be obtained internationally on the scope and impact of the various forms of DM and other categories of glucose intlerance.

The classification was based on current knowledge of DM and also represented some compromises between different points of view. It was based on a mixture of clinical

manifestations (e.g. insulin dependent, non-insulin dependent) and etiopathogenesis (e.g. drug induced, gestational). However, both the NDDG and WHO anticipated that, as knowledge of DM continued to develop with future research advances; it was likely that the classification would be revised. For example, in 1979 to 1980, a definitive etiology was not well established for any of the DM subclasses. Few genetic markers for DM had been discovered, and the understanding of the immunologic basis for many cases of type 1 DM was rudimentry.

2.5.2. The American Diabetes Association Classification System:

In 1996 and 1997, an expert committee of the American Diabetes Association considered the research findings of the last 20 years and proposed some changes to the NDDG/WHO classification scheme (American Diabetes Association Expert Committee, 1997). The new system is shown in Table-02. Changes to diagnostic criteria were also proposed. The main features of the changes in the classification are:

1. Elimination of the terms *insulin-dependent DM* and *non–insulin-dependent DM* and their acronyms, IDDM and NIDDM. However, the terms *type 1* and *type 2 DM* were proposed to be retained.
2. Inclusion under type 1 DM of forms of DM involving pancreatic β-cell destruction, including those cases due to an autoimmune cause and those cases in which an etiology is not known.
3. More precise definition under type 2 DM of the form of DM that is the most prevalent in the United States and is due to insulin resistance with insulin secretary defects.

Table-02 highlights the different etiologic factors that permit discrimination among the types of DM. The heterogeneity within the syndrome of DM implied in Table-02 has important implications for research and clinical management of patients. For example, different and lifestyle factors can result in similar diabetic phenotypes (hyperglycemia and microvascular complications), although the disorders in Table-02 differ markedly in pathogenesis, natural history, and responses to therapy and preventive measures. The exact causes of type 1 and type 2 DM, the subject of intensive research for decades, remain unknown, although both can be accompanied by ketoacidosis, blindness, kidney failure,

premature cardiovascular disease, stroke, amputations, and other diabetic complications. DM associated with other conditions may be strictly secondary to the pathophysiologic consequences of these conditions. GDM may arise from the physiologic stresses of pregnancy or it may be a degree of abnormal glucose tolerance that precedes pregnancy and is discovered during the routine metabolic testing that occurs during pregnancy. Each class in Table-02 may be heterogeneous in etiology and pathogenesis, and further research is needed to define more precisely the different types of DM, determine their etiologies, and devise more appropriate preventive and therapeutic strategies.

2.6. Type 1 Diabetes Mellitus:

This type of DM comprises approximately 5% to 10% of cases in the DM syndrome. It is the most common form of DM among children and adolescents and was formerly termed juvenile-onset DM. In these peop le, the disease is usually characterized by abrupt onset of severe symptoms, dependence on exogenous insulin to sustain life, and proneness to ketosis even in the basal state, all of which are caused by absolute insulin deficiency (insulinopenia). Onset in adult subjects is not uncommon. In community based studies, 15% to 30% of all cases of type 1 DM were diagnosed after 30 years of age (Laakso M et al1985; Scott RS et al1991). Some studies indicate that approximately 7% of all insulin treated patients with onset at 30 years or more of age may have type 1 DM (Melton LJ et al 1983; Harris MI et al 1994). In adults, the rate of β-cell destruction appears to be slower than in children, and residual β-cell function sufficient to prevent ketoacidosis may be present for many years. Some studies suggest that as many as 20% of subjects who present initially with type 2 DM may have slowly progressive type 1 DM (Niskanen LK et al 1995; Willis J, 1998). Type 1 DM results from β-cell destruction that leads to virtually total loss of insulin secretion and absolute insulin deficiency. Two subclasses are discriminated:

1. Autoimmune class and
2. Idiopathic class.

The autoimmune form is a chronic disease with a subclinical prodromal period characterized by cellular mediated autoimmune destruction of the insulin producing β-cells in the pancreatic islets. The rate and extent of β-cell destruction can be variable.

Table 02: Classification of diabetes mellitus:

Type 1 diabetes mellitus: Caused by β-cell destruction, often immune mediated that leads to loss of insulin secretion and absolute insulin deficiency. The etiologic agents that cause the autoimmune process and β-cell destruction are not well established. Also includes cases in which causes of the β-cell destruction are not understood. Comprises approximately 5% to 10% of cases in the diabetes syndrome.
Type 2 diabetes mellitus: Caused by a combination of genetic and nongenetic factors that result in insulin resistance and insulin deficiency. The specific genes are not known but are under intense investigation. Nongenetic factors include increasing age, high caloric intake, overweight, central adiposity, sedentary lifestyle, and low birth weight. Comprises approximately 90% to 95% of cases in the diabetes syndrome.
Other specific types of diabetes mellitus: These types comprise a heterogeneous etiologic group that includes those cases of diabetes in which the causes are established or at least partially known. The causes include known genetic defects affecting β-cell function or insulin action, diseases of the exocrine pancreas, endocrinopathies, drug or chemical induced pancreatic changes, and diseases and conditions in which the incidence of diabetes is substantially elevated but a precise etiology has not been established. Comprises approximately 1% to 2% of cases in the diabetes syndrome.
Gestational diabetes mellitus: Caused by insulin resistance and relative insulin deficiency associated with pregnancy. Occurs in approximately 3% to 5% of all pregnancies.

Markers of the autoimmune destruction include antibodies to the islet cells and to insulin, glutamic acid decarboxylase (GAD), and tyrosine phosphatases IA-2 and IA-2β (Bingley PJ et al 1997). Often as many as 70% of newly diagnosed patients have islet cell antibodies, compared with only 3% of age and sex matched control subjects (Kolb H et al 1988; Bonifacio E et al 1997). Antibody to GAD has been detected as early as 10 years before diagnosis (Tuomilehto J et al 1994) and is present in up to 75% of patients with newly diagnosed type 1 DM (Rowley MJ et al 1992).

Observations have suggested that β-cell autoimmunity may be induced in any person at any time (Knip M et al 1997). Only a proportion of those with signs of islet cell autoimmunity progress to clinical disease, and harmless β-cell autoimmunity reflected by positivity for single autoantibody specificity seem to appear without any relation to genetic type 1 DM susceptibility. Genetic determinants are important risk factors, in particular certain human leukocyte antigen genes in the histocompatibility system located on chromosome 6 (Concannon P et al 1998; Nepom GT et al 1998; Zamani M et al 1998). However, concordance rates for type 1 DM in identical twins are approximately 35% to 50%, well below the rate required if genetic factors were the only determinants of autoimmune type 1 DM (Kaprio J et al 1992).

The etiologic agents that initiate the autoimmune process and β-cell destruction are not established but are the subject of intensive research. Environmental factors may trigger initial β-cell damage and subsequently accelerate the destructive process. The most likely environmental candidates are viral infections. Genetic and environmental etiologies probably are heterogeneous, as evidenced by the wide variability in the occurrence of type 1 DM. The incidence is highest in Scandinavia, with more than 30 cases/year/100,000 people, of medium incidence in Europe and the United States (approximately 10 to 15 cases/ year/100,000), and lowest in Asian groups (0.5 cases/year/ 100,000) and populations living in the tropics (Karvonen M et al 1993). The disease appears to be virtually absent in some populations (e.g., North American Indians and Pacific Islanders). Type 1 DM also includes cases in which causes of the β-cell destruction are not understood but are thought to not be immune mediated. A minority of patients with type 1 DM are in this category, which is strongly inherited but not associated with histo-

compatibility genes. Patients experience episodic ketoacidosis and varying degrees of insulin deficiency; most are of African or Asian origin.

2.7. Type 2 Diabetes Mellitus

This type of DM comprises approximately 90% of the DM syndrome and, in certain groups such as North American Indians and populations in the South Pacific (King H et al 1998) and India and subcontinent (Mohan V.et al 2007). it is virtually the only form of DM It is characterized by insulin resistance in muscle, liver, and adipose tissue that probably begins at a preclinical stage (possibly at the stage of impaired glucose tolerance). Type 2 DM may be unrecognized for years because of lack of symptoms. Eventually, defects in insulin secretion leading to decompensated hyperglycemia precipitate clinical onset of the disease. In contrast to type 1 DM, patients with type 2 DM do not depend on exogenous insulin for prevention of ketonuria and are not prone to ketosis. However, they may require insulin for correction of fasting hyperglycemia if this cannot be achieved with the use of diet or oral agents, and ketosis may develop under special circumstances such as severe stress precipitated by infections or trauma. In the basal state, there may be normal levels of insulin, mild insulinopenia, or above normal levels of insulin associated with insulin resistance. In response to a glucose or meal challenge, a range of insulin levels from low to supranormal has been found in the group of diabetic patients in this subclass. Although diagnosis in most patients with type 2 DM is made in adult years, the disease also occurs in young people who do not require insulin and are not ketotic and hence could not be considered to have type 1 DM (Neufeld ND et al 1998; Jones KL et al 1898; Scott CR et al 1997). In addition, the average age at diagnosis of type 2 DM is much earlier in very high prevalence groups such as North American Indians and Pacific Islanders, and somewhat earlier in medium prevalence groups such as African Americans and Hispanic Americans, compared with the U.S. white population (Harris MI et al 1995). In the multicentre Study in India, 93% of diabetics from urban areas and 81% of diabetics from rural areas were above the age of 30 (Mohan V et al 2007).

Although the etiology of type 2 DM is unclear, the disease has a strong genetic basis as evidenced by the frequent familial pattern of occurrence, its high prevalence in

certain ethnic groups, and genetic admixture studies. Virtually all race-ethnic groups in the United States are at higher risk than the majority white population (Harris MI et al 1995), probably because of a higher frequency of genes associated with DM in the former groups. The genes causing most cases of type 2 DM remain obscure but are the subject of intense investigation (Cox NJ et al 1999; Hanson RL et al 1998). Although type 2 DM is strongly associated with genetic factors, it is undoubtedly heterogeneous in its etiology because a variety of lifestyle and environmental factors have been identified as being risk factors for the condition (Haffner SM et al 1998). In all probability, the causes of type 2 DM lie in environmental and lifestyle factors superimposed on genetic susceptibility. Prominent among these factors is obesity, and approximately 50% to 90% of all patients with type 2 DM are obese (Harris MI et al 1995). A strong association between upper body obesity (central obesity) and type 2 DM prevalence and incidence has been demonstrated. Intraabdominal fat deposition is the important site conveying enhanced risk for type 2 DM (Bergstrom RW et al 1990). Other risk factors include increasing age, high caloric intake, sedentary lifestyle, and low birth weight. People with impaired glucose tolerance or GDM are also at increased risk, probably because these conditions are preclinical stages of type 2 DM.

2.8. Other Specific Types of Diabetes Mellitus:

This subclass is numerically small, comprising only approximately 1% to 2% of cases in the DM syndrome. However, it is etiologically very heterogeneous because DM can be associated with a variety of other conditions and syndromes. In certain instances, abnormal glucose tolerance is secondary to the condition (e.g., specific endocrine diseases), whereas in others the relationship is apparently causal but not yet explained (e.g., certain genetic syndromes). On this basis, the subclass is defined according to the known or presumed etiologic relationship or the strong association with other conditions.

These conditions are listed in Table-3. The grouping includes cases of DM in which there are known genetic defects in the β-cell (e.g., maturity onset DM of the young, in which several mutations have been identified; point mutations in mitochondrial deoxyribonucleic acid associated with DM and deafness). It also includes cases in which

there are known genetic defects in insulin action (e.g., mutations in the insulin receptor, acanthosis nigricans, leprechaunism, Rabso Mendenhall syndrome).

Diabetes due to diseases of the exocrine pancreas or removal of pancreatic tissue is included in this grouping, including such conditions as pancreatitis, trauma and infections of the pancreas, pancreatectomy, and pancreatic cancer. Cystic fibrosis and hemochromatosis can damage β-cells. Pancreatic fibrosis and calcium stones are included here also. Endocrinopathies form another grouping and act through antagonism of insulin action. These include such conditions as acromegaly, Cushing's syndrome, and glucagonoma.

Certain drugs and chemicals (e.g., thiazide diuretics, corticosteroids) can induce pancreatic injury and destruction, leading to loss of insulin secretion, and these are included as a subgroup in the classification. Another subclass comprises certain diseases such as lupus erythematosus, in which insulin resistance is induced through antiinsulin receptor antibodies. The final subclass comprises genetic syndromes in which there is an increased incidence of DM (e.g., Down's, Turner's, and Wolfram's syndromes), implying an etiologic association. The heterogeneity of the diabetic syndrome is clearly illustrated by the variety of conditions listed in Table-03 with which glucose intolerance is associated.

Table- 03. Other specific types of diabetes mellitus (Derec LeRoith et al 2004)

A. Genetic defects of β-cell function
    1. Chromosome 12, *HNF1*α (formerly *MODY3*)
    2. Chromosome 7, glucokinase (formerly *MODY2*)
    3. Chromosome 20, *HNF4*α (formerly *MODY1*)
    4. Mitochondrial DNA
    5. Others
B. Genetic defects in insulin action
    1. Type A insulin resistance

2. Leprechaunism
3. Rabson Mendenhall syndrome
4. Lipoatrophic diabetes
5. Others

C. Diseases of the exocrine pancreas
1. Pancreatitis
2. Trauma/pancreatectomy
3. Neoplasia
4. Cystic fibrosis
5. Hemochromatosis
6. Fibrocalculous pancreatopathy
7. Others

D. Endocrinopathies
1. Acromegaly
2. Cushing's syndrome
3. Glucagonoma
4. Pheochromocytoma
5. Hyperthyroidism
6. Somatostatinoma
7. Aldosteronoma
8. Others

E. Drug or chemical induced
1. Vacor
2. Pentamidine
3. Nicotinic acid
4. Glucocorticoids
5. Thyroid hormone
6. Diazoxide

7. β-Adrenergic agonists
8. Thiazides
9. Dilantin
10. α-Interferon
11. Others

F. Infections

1. Congenital rubella
2. Cytomegalovirus
3. Others

G. Uncommon forms of immune mediated diabetes

1. "Stiff man" syndrome
2. Antiinsulin receptor antibodies
3. Others

H. Other genetic syndromes sometimes associated with diabetes

1. Down's syndrome
2. Klinefelter's syndrome
3. Turner's syndrome
4. Wolfram's syndrome
5. Friedreich's ataxia
6. Huntington's chorea
7. Lawrence Moon Beidel syndrome
8. Myotonic dystrophy
9. Porphyria.
10. Prader Willi syndrome.
11. Others

2.9. Gestational Diabetes Mellitus:

For many years GDM has been defines as any degree of glucose intolerance with onset of first recognition during pregnancy. Although most cases resolved with delivery, the definition applied whether or not the condition persisted after pregnancy and did exclude the possibility that unrecognized glucose tolerance may have antedated or begun concomitantly with the pregnancy. This definition facilitated a uniform strategy for detection and classification of GDM, but its limitations were recognized for many years. As the ongoing epidemic of obesity and diabetes had led to more type 2 diabetes in woman of childbearing age, the number of pregnant women with undiagnosed type 2 diabetes has increased.

After deliberation in 2008-2009, the international association of diabetes and pregnancy study group (IADPSG), an international consequences group with representative from multiple obstetrical and diabetic organizations, including the American Diabetes Association (ADA), recommended that high risk women found to have diabetes at their prenatal visit, using standard criteria receives a diagnosis overt, not gestational diabetes. Approximately 7% of all pregnancies are complicated by GDM (ADA report; 2012).

Gestational DM is a mild degree of fasting hyperglycemia or glucose intolerance that is detected in approximately 2% to 5% of all pregnancies in the United States. This class is restricted to pregnant women in whom the onset or recognition of DM or impaired glucose tolerance first occurs during pregnancy. Thus, diabetic women who become pregnant are not included in this class.

2.10. Sign and symptoms of diabetes mellitus:

Symptoms are similar in both types of diabetes but they vary in their intensity. The classic signs and symptoms of diabetes are (Kumar PJ et al 2002):

- Polyurea (Excessive urination),
- Polydipsia (Excessive thirst),
- Polyphagia (Excessive hunger or appetite)
- Weight loss,
- Fatigue, lethargy or drowsiness

- Cramps,
- Constipation,
- Blurred vision nearsightedness or other vision problems
- Candidiasis
- Skin problems, such as itchiness or acanthosis nigricans
- Shakiness or trembling
- Mood swings or irritability
- Dizziness or fainting
- Numbness, tingling or pain in the feet, legs or hands

Type 1 diabetes can develop rapidly and often occurs after an illness, but symptoms may be mistaken for the flu or other common conditions. Type 2 diabetes can take many years to develop and sometimes becomes apparent only after long term complications occur, such as sexual dysfunction or leg pain that is due to diabetic neuropathy or claudication (caused by peripheral artery disease).

Some people, especially young people with type 1 diabetes, go undiagnosed until they are brought to a hospital with an emergency condition called diabetic ketoacidosis. Indicators of diabetic ketoacidosis include sweet fruity smelling or wine smelling breath, confusion and heavy labored breathing (Kussmaul breathing). Sometimes patients are diagnosed with diabetes only after suffering other serious complications including insulin shock, hyperosmolar hyperglycemic nonketotic syndrome or diabetic coma. To help prevent such complications, people are advised to undergo periodic screening for diabetes with glucose tests, especially if they have risk factors (Samreen Riaz 2009).

Long standing type 1 DM patients are susceptible to microvascular complications; (Bearse MA Jr et al 2004; Hove MN et al 2004; Seki M et al 2004; Moran A et al 2004; Huang C et al 2002; Shukla N et al 2003) and macrovascular disease (coronary artery, heart, and peripheral vascular diseases) (Svensson M et al 2004; Saely CH et al 2004).

Symptoms in type 2 DM are similar but insidious in onset. Most cases are diagnosed because of complications or incidentally. Type 2 DM caries a high risk of large vessel atherosclerosis commonly associated with hypertension, hyperlipidaemia and obesity. Most patients with type 2 diabetes die from cardiovascular complications and end

stage renal disease. Geographical differences exist in both the magnitude of these problems and their relative contributions to overall morbidity and mortality.

2.11. Prevalence of Diabetes mellitus

2.11.1. Prevalence of diabetes in India and Asian countries:

Phase one results of the Indian Council of Medical Research – India Diabetes (ICMR-INDIAB) Study have provided data from three States and one Union Territory, representing nearly 18.1 per cent of the nation's population. When extrapolated from these four units, the conclusion is 62.4 million people live with diabetes in India, and 77.2 million people are on the threshold, with prediabetes (R. M. Anjana et al 2011*).*

Asia has emerged as the 'diabetes epicenter' in the world (Chan J. C. et al 2009). Asian countries contribute to more than 60% of the world's diabetic population as the prevalence of diabetes is increasing in these countries. Socio-economic growth and industrialization are rapidly occurring in many of these countries. The urban-rural divide in prevalence is narrowing as urbanization is spreading widely, adversely affecting the lifestyle of populations. Asians have a strong ethnic and genetic predisposition for diabetes and have lower thresholds for the environmental risk factors. As a result, they develop diabetes at a younger age and at a lower body mass index and waist circumference when compared with the Western population. The adverse effect of physical inactivity and fatty food are manifested as the increasing rate of over weightiness and obesity, even among children. The health care budgets for the disease management are meager and the health care outcome is far from the optimum. As a result, complications of diabetes are common and the economic burden is very high, especially among the poor strata of the society (Ambady R et al 2012). Unlike in the West, where older populations are most affected, the burden of diabetes in Asian countries is disproportionately high in young to middle aged adults. This could have long lasting adverse effects on a nation's health and economy, especially for developing countries (Ramachandran A et al 2010).

In the past two decades, the prevalence in urban areas has increased remarkably in most countries, the increase being phenomenal in Nepal (Ono K et al 2007) and China (Yang W et al 2010) the national prevalence has increased by two fold or more within a decade in many countries (Janus ED et al 2000; Yang W et al 2010; Tan CE et al 1999;

Lu FH et al 1998; Duc Son LN et al 2004). Rural prevalence has increased considerably in India (Ramachandran A et al 1992) Nepal (Ono K et al 2007) and China (Yang W et al 2010). India and China have large rural populations and hence the increased prevalence of diabetes in rural areas has contributed to the overall national increase in the prevalence of diabetes in these countries.

2.11.2. Global Prevalence of Diabetes:

Over the past three decades, the number of people with diabetes mellitus has more than doubled globally, making it one of the most important public health challenges to all nations (Lei Chen et al 2012).

The global increase in the prevalence of diabetes is due to population growth, aging, urbanization and an increase of obesity and physical inactivity. The primary determinants of the epidemic are the rapid epidemiological transition associated with changes in dietary patterns and decreased physical activity (Ramachandran A et al 2010). The prevalence of diabetes, constituted chiefly by type 2 diabetes (T2DM), is a global public health threat.

In 2010, an estimated 285 million people worldwide had diabetes mellitus, 90% of whom had type 2 diabetes mellitus (T2DM) (Shaw J E et al 2010). In 2011, there were an estimated 366 million people with diabetes (8.3% of the world's population) (IDF 2011), in 2012, there were an estimated 371 million people with diabetes (IDF 2012).

The number of people globally with diabetes mellitus is projected to rise to 439 million by 2030, which represents 7.7% of the total adult population of the world aged 20–79 years (Shaw, J. E. et al 2010).

Table 04: Number of People with Diabetes Top 10 Countries (IDF 2011)			
2011		2030	
Country/ territory	Number of people with diabetes (millions)	Country/ territory	Number of people with diabetes (in Millions)
China	90.0	China	129.7
India	61.3	India	101.2
USA	23.7	USA	29.6
Russian Federation	12.6	Brazil	19.6
Brazil	12.4	Bangladesh	16.8
Japan	10.7	Mexico	16.4
Mexico	10.3	Russian Federation	14.1
Bangladesh	8.4	Egypt	12.4
Egypt	7.3	Indonesia	11.8
Indonesia	7.3	Pakistan	11.4

2.12. Diagnostic Criteria for Diabetes Mellitus:

The clinical diagnosis of diabetes is often prompted by symptoms such as increased thirst and urine volume, recurrent infections, unexplained weight loss and, in severe cases, drowsiness and coma; high levels of glycosuria are usually present.

The diagnosis of diabetes mellitus is based on measuring venous plasma glucose in the fasting state and 2 hours after a 75 gram glucose load (recommended by the WHO) (Mohammad Ashraf Ganie et al 2012).

The American Diabetes Association Expert Committee (American Diabetes Association Expert Committee. Report 1997) recommended modification of the NDDG/WHO diagnostic criteria. Recognizing the difficulties inherent in performing the oral glucose tolerance test, the criteria now essentially exclude the oral glucose tolerance test as a diagnostic method in routine clinical practice. Instead, the criteria rely on fasting hyperglycemia for the diagnosis of DM. The current criteria for diagnosing DM are shown in Table 05.

Table 05: Diagnostic criteria for diabetes mellitus, impaired fasting glucose, and normal fasting glucose

Diabetes	Symptoms of diabetes plus casual plasma glucose(PG) ≥200 mg/dL OR Fasting plasma glucose ≥126 mg/dL, confirmed by repeat testing on a different day OR Plasma glucose ≥200 mg/dL at 2 hours after a 75-g oral glucose challenge, confirmed by repeat testing on a different day. *This method is not recommended for routine clinical use.* *OR*
	Impaired fasting glucose :Fasting plasma glucose 110–126 mg/dL OR
	Normal fasting glucose Fasting plasma glucose <110 mg/dL OR Symptoms of diabetes include such classic symptoms as polyuria, polydipsia, and other acute manifestations of hyperglycemia. Fasting is defined as no caloric intake for at least 8 hours.

DM can be diagnosed by the presence of the classic signs and symptoms of DM and unequivocally elevated blood glucose levels; by elevated fasting plasma glucose; or

by elevated plasma glucose at 2 hours after a 75-g oral glucose challenge. The latter method, however, is not recommended in clinical practice.

ICMR-WHO recommended guidelines for management of Diabetes in 2005 and diagnostic criteria as below (ICMR Guidelines; 2005).

1. Indication of Person with Diabetes:
    - Symptoms of diabetes plus plasma glucose of ≥ 200 mg/dl
    - Fasting plasma glucose ≥ 126 mg/dl
    - 2 hours post 75g glucose ≥ 200 mg/dl

Any of positive tests should be confirmed with another test subsequently.

2. Criteria for diagnosis of Diabetes & Glucose Intolerance

Table 06: ICMR-WHO recommended guidelines for management of Diabetes in 2005 and diagnostic criteria

Normologlycemia	IFG or IGT	Diabetes
FPG <110 mg/dl	FPG ≥ 110 and <126 mg/dl IFG	FPG ≥ 126 mg/dl
2-h PG <140 mg/dl	2-h PG ≥ 140 and <200 mg/dl (IGT)	2-h PG ≥200 mg/dl symptoms of diabetes and casual plasma glucose concentration ≥200 mg/dl

IFG- Impaired Fasting Glucose

IGT- Impaired Glucose tolerance

FPG- Fasting Plasma Glucose

2h PG -2 hr post load Glucose test (oral glucose tolerance test) plasma glucose.

3. Oral Glucose Tolerance Test(OGTT)
    - Person to be tested should be on a normal diet for at least 3 days before the test.
    - The test should be done after an overnight fast of 8-10 hrs and comprises of two blood sample: fasting and 2 hrs after glucose load.
    - Following the collection of the fasting blood sample for analysis of plasma glucose, the individual should be administered 75g of glucose dissolved in 250 ml of water. The glucose load should be drunk within a period of 5 minutes.

- The second and last sample should be collected 2 hrs after the glucose load. The subject should be resting and refrain from smoking in between two sample collections.

4. Criteria for Retesting for Diabetes in Asymptomatic High Risk Individuals
    - Undiagnosed high risk person with normal test retest yearly or at least once in two year.
    - Impaired fasting glucose, impaired glucose tolerance.
5. Testing for Type 2 Diabetes in Children and Adolescents.

    Overweight (Weight > 120% of ideal body weight) plus any of the following high risk factors:
    - Family history of type 2 diabetes in first or second degree relative.
    - Sign of insulin resistance of condition associated with insulin resistance (Acanthosis nigricans, Hypertension, Dyslipidemia or PCOS)

2.13. Glycated haemoglobin (Hba1c) for the diagnosis of diabetes:

HbA1c can be used as a diagnostic test for diabetes providing that stringent quality assurance tests are in place and assays are standardized to criteria aligned to the international reference values, and there are no conditions present which preclude its accurate measurement (Abbreviated Report of a WHO Consultation; 2011).

Glycated haemoglobin (HbA1c) was initially identified as an "unusual" haemoglobin in patients with diabetes over 40 years ago (Rahbar S et al 1969). After that discovery, numerous small studies were conducted correlating it to glucose measurements resulting in the idea that HbA1c could be used as an objective measure of glycaemic control. HbA1c was introduced into clinical use in the 1980s and subsequently has become a cornerstone of clinical practice (Massi Benedetti M et al 2006). HbA1c is a widely used marker of chronic glycemia, reflects average plasma glucose over the previous 8 to 12 weeks (Nathan DM et al 2007*),* the test plays a critical role in the management of the patient with diabetes, since it correlates well with both microvascular and, to a lesser extent, macrovascular complications and is widely used as the standard biomarker for the adequacy of glycemic management. It can be performed at any time of the day and does not require any special preparation such as fasting. These properties have made it the preferred test for assessing glycaemic control in people with diabetes. More recently, there

has been substantial interest in using it as a diagnostic test for diabetes and as a screening test for persons at high risk of diabetes (International Committee report, 2009). Owing in large part to the inconvenience of measuring fasting plasma glucose levels or performing an OGTT, and day-to-day variability in glucose, an alternative to glucose measurements for the diagnosis of diabetes has long been sought. HbA1c has now been recommended by an International Committee and by the ADA as a means to diagnose diabetes (International Committee report, 2009). Although it gives equal or almost equal sensitivity and specificity to a fasting or post load glucose measurement as a predictor of prevalent retinopathy (Abbreviated Report of a WHO Consultation, 2011), it is not available in many parts of the world. Also, many people identified as having diabetes based on HbA1c will not have diabetes by direct glucose measurement and vice versa.

The relationship between HbA1c and prevalent retinopathy is similar to that of plasma glucose, whether glucose and HbA1c are plotted in deciles (McCance DR et al *1994*), The deciles of the three measures FPG, 2-hr PG, and HbA1c at which retinopathy began to increase were the same for each measure within each populations like Pima Indians (McCance DR et al 1994), Egyptians (Engelgau MM et al 1997), the NHANES study in the USA (Report of Expert Committee 1997), in Japanese (Miyazaki M et al 2004) and more recently in the DETECT-2 analysis. The glycemic values above which retinopathy increased were similar among the populations. These analyses helped to inform a new diagnostic cut point of ≥126 mg/dl (7.0 mmol/l) for FPG and confirmed the long standing diagnostic 2-hr PG value of ≥200 mg/dl (11.1 mmol/l).

HbA1c is a widely used marker of chronic glycemia, reflecting average blood glucose levels over a 2- to 3-month period of time. The test plays a critical role in the management of the patient with diabetes, since it correlates well with both microvascular and, to a lesser extent, macrovascular complications and is widely used as the standard biomarker for the adequacy of glycemic management. A recent report from Australia has shown that a model including HbA1c for predicting incident retinopathy is as good as or possibly better than one including fasting plasma glucose (Tapp RJ et al 2008).

The use of HbA1c can avoid the problem of day-to-day variability of glucose values, and importantly it avoids the need for the person to fast and to have preceding dietary preparations. These advantages have implications for early identification and treatment

which have been strongly advocated in recent years. However, HbA1c may be affected by a variety of genetic, haematologic and illness related factors (Gallagher EJ et al 2009). The most common important factors worldwide affecting HbA1c levels are haemoglobinopathies (depending on the assay employed), certain anaemias, and disorders associated with accelerated red cell turnover such as malaria (International Committee report 2009; Robert WL et al 2002).

For patients with a hemoglobinopathy but normal red cell turnover, such as sickle cell trait, an HbA1c assay without interference from abnormal hemoglobins should be used. For conditions with abnormal red cell turnover, such as anemias from hemolysis and iron deficiency, the diagnosis of diabetes must employ glucose criteria exclusively. The established glucose criteria for the diagnosis of diabetes remain valid. These include the FPG and 2-hr PG. Additionally, patients with severe hyperglycemia such as those who present with severe classic hyperglycemic symptoms or hyperglycemic crisis can continue to be diagnosed when random (or casual) plasma glucose of ≥200 mg/dl (11.1 mmol/l) is found. It is likely that in such cases the health care professional would also measure an HbA1c test as part of the initial assessment of the severity of the diabetes and that it would (in most cases) be above the diagnostic cut point for diabetes. However, in rapidly evolving diabetes, such as the development of type 1 diabetes in some children, HbA1c may not be significantly elevated despite frank diabetes. Just as there is less than 100% concordance between the FPG and 2-hr PG tests, there is not full concordance between HbA1c and either glucose based test. Analyses of NHANES data indicate that, assuming universal screening of the undiagnosed, the HbA1c cut point of ≥6.5% identifies one third fewer cases of undiagnosed diabetes than a fasting glucose cut point of ≥126 mg/dl (7.0 mmol/l) (cdc website tbd). However, in practice, a large portion of the population with type 2 diabetes remains unaware of their condition. Thus, it is conceivable that the lower sensitivity of HbA1c at the designated cut point will be offset by the test's greater practicality, and that wider application of a more convenient test (HbA1c) may actually increase the number of diagnoses made. Further research is needed to better characterize those patients whose glycemic status might be categorized differently by two different tests (e.g., FPG and HbA1c), obtained in close temporal approximation. Such discordance may arise from measurement variability, change over time, or because

HbA1c, FPG, and post challenge glucose each measure different physiological processes. In the setting of an elevated HbA1c but "nondiabetic" FPG, the likelihood of greater postprandial glucose levels or increased glycation rates for a given degree of hyperglycemia may be present. In the opposite scenario (high FPG yet HbA1c below the diabetes cut point), augmented hepatic glucose production or reduced glycation rates may be present. As with most diagnostic tests, a test result diagnostic of diabetes should be repeated to rule out laboratory error, unless the diagnosis is clear on clinical grounds, such as a patient with classic symptoms of hyperglycemia or hyperglycemic crisis. It is preferable that the same test be repeated for confirmation, since there will be a greater likelihood of concurrence in this case. For example, if the HbA1c is 7.0% and a repeat result is 6.8%, the diagnosis of diabetes is confirmed. However, there are scenarios in which results of two different tests (e.g., FPG and HbA1c) are available for the same patient. In this situation, if the two different tests are both above the diagnostic thresholds, the diagnosis of diabetes is confirmed.

On the other hand, when two different tests are available in an individual and the results are discordant, the test whose result is above the diagnostic cut point should be repeated, and the diagnosis is made on the basis of the confirmed test. That is, if a patient meets the diabetes criterion of the HbA1c (two results ≥6.5%) but not the FPG (≥126 mg/dl or 7.0 mmol/l), or vice versa, that person should be considered to have diabetes. Admittedly, in most circumstance the "nondiabetic" test is likely to be in a range very close to the threshold that defines diabetes. Since there is preanalytic and analytic variability of all the tests, it is also possible that when a test whose result was above the diagnostic threshold is repeated, the second value will be below the diagnostic cut point. This is least likely for HbA1c, somewhat more likely for FPG, and most likely for the 2-hr PG. Barring a laboratory error, such patients are likely to have test results near the margins of the threshold for a diagnosis. The healthcare professional might opt to follow the patient closely and repeat the testing in 3–6 months. The decision about which test to use to assess a specific patient for diabetes should be at the discretion of the health care professional taking into account the availability and practicality of testing an individual patient or groups of patients. Perhaps more important than which diagnostic test is used, is that the testing for diabetes be performed when indicated. There is discouraging evidence in-

dicating that many at risk patients still do not receive adequate testing and counseling for this increasingly common disease, or for its frequently accompanying cardiovascular risk factors.

The utility and convenience of HbA1c compared with measures of plasma glucose for the diagnosis of diabetes needs to be balanced against the fact that it is unavailable in many countries, despite being a recognized valuable tool in diabetes management. In addition the HbA1c assay is not currently well enough standardized in many countries for its use to be recommended universally at this time. However, HbA1c assays are now highly standardized so that their results can be uniformly applied both temporally and across populations. International Expert Committee, after an extensive review of both established and emerging epidemiological evidence, recommended the use of the HbA1c test to diagnose diabetes, with a threshold of ≥6.5%, and ADA affirms this decision. The diagnostic HbA1c cut point of 6.5% is associated with an inflection point for retinopathy prevalence, as are the diagnostic thresholds for FPG and 2-hr PG. The diagnostic test should be performed using a method that is certified by the National Glycohemoglobin Standardization Program (NGSP) and standardized or traceable to the Diabetes Control and Complications Trial reference assay. Point of care HbA1c assays are not sufficiently accurate at this time to use for diagnostic purposes.

Table 07: The Current Diagnostic Criteria for Diabetes (ADA report; 2012).

Criteria for the diagnosis of diabetes

- HbA1c ≥6.5%. The test should be performed in a laboratory using a method that is NGSP certified and standardized to the DCCT assay.* **OR**
- FPG ≥126 mg/dl (7.0 mmol/l). Fasting is defined as no caloric intake for at least 8 h.* **OR**
- 2-h plasma glucose ≥200mg/dl (11.1mmol/l) during anOGTT. The test should be performed as described by the World Health Organization, using a glucose load containing the equivalent of 75 g anhydrous glucose dissolved in water.* **OR**
- In a patient with classic symptoms of hyperglycemia or hyperglycemic crisis, a random plasma glucose ≥200 mg/dl (11.1 mmol/l).

*In the absence of unequivocal hyperglycemia, criteria 1–3 should be confirmed by repeat testing.

2.14. Diabetic Complications:

The late complications of diabetes can be broadly classified as (Brajendra Kumar Tripathi et al 2006):

A. Acute complication
B. Chronic complication

2.14.1. Acute complications (Brajendra Kumar Tripathi et al 2006):

These include diabetic ketoacidoses (DKA) and nonketotic hyperosmolar state (NKHS). While the first is seen primarily in individuals with type 1 DM, the latter is prevalent in individuals with type 2 DM. Both disorders are associated with absolute or relative insulin deficiency, volume depletion, and altered mental state. In DKA, insulin deficiency is combined with counter regulatory hormone excess (glucagon, catecholamines, cortisol, and growth hormone). The decreased ratio of insulin to glucagon promotes gluconeogenesis, glycogenolysis, and ketone body formation in the liver and also increases free fatty acid and amino acid delivery from fat and muscle to the liver. Ketosis results from a marked increase in free fatty acid release from adipocytes due to increased lipolysis. In DKA, nausea and vomiting are often present. Lethargy and CNS depression may evolve into coma in severe DKA. Cerebral edema, an extremely serious complication, is seen most frequently in children. NKHS is most commonly seen in elderly individuals with type 2 DM. Its most prominent features include polyuria, orthostatic hypotension, and a variety of neurological symptoms including altered mental state, lethargy, obtundation, seizure, and possibly coma. Insulin defi ciency and inadequate fl uid intake are the underlying causes of NKHS. Insulin defi ciency leads to hyperglycemia, which induces an osmotic diuresis leading to profound intravascular volume depletion.

1. **Diabetic Ketoacidosis**
2. **Lactic Acidosis**
3. **Hypoglycaemia**

2.14.1.1. Diabetic Ketoacidosis (Nicole A S et al 2008)**:**

Diabetic ketoacidosis (DKA) is the result of a relative or absolute deficiency of insulin, and increased levels of the counterregulatory hormones glucagon, cortisol, and

catecholamines. Glucagon seems to be particularly important in the pathogenesis of ketoacidosis. Blood glucose levels rise because there is increased production of new glucose (gluconeogenesis) and failure to store glucose that is absorbed through the gut. In addition, glycogenolysis, as a result of both insulin deficiency and elevations of counter regulatory hormones, contributes to hyperglycemia. With insulin deficiency, there are minimal glycogen stores in the liver or other tissues. Hyperglycemia induces an osmotic diuresis and obligate loss of salt as well as water. Loss of sodium and potassium in adults may amount to up to 20% of total body stores (Nabarrö JDN et al 1957). Without insulin, there is a breakdown of fat with the release of free fatty acids. Free fatty acids are converted to ketones through glucagon dependent hepatic pathways. The released ketoacids are excreted by the kidney as long as there is sufficient hydrogen exchange. With decreasing fluid volume as a result of osmotic diuresis and loss of salt necessary for hydrogen exchange, blood levels of ketoacids rise and acidosis develops. Pulmonary compensation for the metabolic acidosis is not sufficient, leading to increasingly severe acidosis.

Signs & Symptoms DKA

- Fruity breath
- Dehydration: hot/dry skin, dry mucous membranes, rapid weak pulse, and thirst, restless.
- Danger from dehydration and
- coma BG > 250 mg/dl

2.14.1.1.1. Diagnosis:

The diagnosis of DKA is based on biochemical evidence of hyperglycemia (serum glucose levels >200-250 mg/dL), acidosis and ketosis (venous pH <7.25-7.30 and/or serum bicarbonate levels <15 mEq/L), with serum concentrations of ketones (ßhydroxybutyrate plus acetoacetate) >31 mg/dL and/or ketonuria >80 mg/dL. DKA may be characterized as mild (venous pH 7.2-7.3, serum bicarbonate level 10-15 mEq/L), moderate (venous pH 7.1-7.2, serum bicarbonate level 5-10 mEq/L), or severe (venous pH <7.1, serum bicarbonate level <5 mEq/L).

2.14.1.2. Lactic acidosis:

LA consists of elevated lactic acid (lactic academia ≥2.0 mmol/L) with acidosis (pH ≤7.3) and without ketoacidosis. There may be low levels of ketones present (≤1:4 on serum dilution, or beta hydroxybutyrate >0.4 but <0.6 mmol/L). Occasionally a combined LA and DKA may be present. In this situation, the presence of excess lactate may decrease production of acetoacetate, which is measured by dipstick methods for ketones, but beta hydroxybutyrate levels may remain elevated with an increased ratio of beta hydroxybutyrate to acetoacetate. Under the circumstances of combined LA and DKA, LA predominates by laboratory parameters and may mask an associated or underlying DKA (NIH 1995.).

2.14.1.3. Hypoglycaemia:

Hypoglycaemia is a condition that occurs when the blood glucose level has dropped too low, usually below 4mmol/L, although this can vary According to the American Diabetes Association, and for most people hypoglycemia is usually a blood sugar below 70mg/dl. However this can also be individual. It is important to treat hypoglycemia quickly to stop the blood glucose level from falling even lower. It is also commonly referred to as a 'hypo', low blood glucose or insulin reaction.

2.14.1.3.1. Symptoms of Hypoglycemia:

- Weakness, trembling or shaking,
- Sweating
- Light headedness
- Headache
- Lack of concentration/behavior change
- Dizziness
- Tearful/crying
- Irritability
- Numbness around the lips and fingers
- Hunger

Hypoglycemia may range from very mild lowering of glycemia (60-70 mg/dl) with minimal or no symptoms, to severe hypoglycemia with very low levels of glucose (<40 mg/dl) and neurologic impairment. Glucose levels of 40-70 mg/dl usually can be treated with oral carbohydrate and would not require further medical attention. More severe hypoglycemia (glucose levels <40 mg/dl) may require intervention with either intravenous glucose or glucagon, but the patient may be sufficiently responsive to take oral carbohydrate to relieve the hypoglycemia.

Hypoglycemia is one of the most important complications of diabetes treatment. The risk of severe hypoglycemia is higher in elderly patients, those having comorbidities such as vascular disease or renal failure, pregnant women and in children with type 1diabetes. Moreover, in type 2 diabetes, progressive insulin deficiency, longer duration of diabetes, and tight glycemic control increase the risk of hypoglycemia as much as type 1 diabetes (Shafiee et al. 2012)

2.14.2. Chronic complication:

The chronic complications of diabetes mellitus affect many organ systems and are responsible for the majority of morbidity and mortality. Chronic complications can be divided into vascular and nonvascular complications (Brajendra Kumar Tripathi et al 2006). The vascular complications are further subdivided into microvascular (retinopathy, neuropathy, and nephropathy) and macrovascular complications (coronary artery disease, peripheral vascular disease, and cerebrovascular disease). Nonvascular complications include problems such as gastroporesis, sexual dysfunction, and skin changes. As a consequence of its chronic complications, DM is the most common cause of adult blindness, a variety of debilitating neuropathies, and cardiac and cerebral disorders (Tripati B K et al 2006). Treating the complications of diabetes costs more than controlling the disease. Early in the course of diabetes, intracellular hyperglycemia causes abnormalities in blood flow and increased vascular permeability. This reflects decreased activity of vasodilators such as nitric oxide, increased activity of vasoconstrictors such as angiotensin II and endothelin-1, and elaboration of permeability factors such as vascular endothelial growth factor (VEGF) (Tripathi B K et al 2006). In diabetic arteries, endothelial dysfunction seems to involve both insulin resistance specific to the phosphotidylinositol–3-OH kinase pathway and hyperglycemia (Tripathi B K et al 2006).

2.14.2.1. Classifiaction of chronic complications:

1. Microvascular :
    A. Retinopathy
    B. Nephropathy
    C. Neuropathy
2. Macrovascular:
    A. Coronary Heart Disease
    B. Peripheral Vascular Disease
    C. Cerebrovascular Disease
3. Non Vascular Complications
    A. Gastroporesis
    B. Sexual dysfunction
    C. Skin changes

2.14.2.2. Microvascular :

2.14.2.2.1. Retinopathy:

Diabetic retinopathy is the most common cause of new blindness in adults aged 20-74 years (Standards of medical care in diabetes--2010). The pathophysiology of diabetic retinopathy comprises five basic pathologic processes that occur at the level of retinal capillary (Derek LeRoith et al 2004):

a. Formation of microaneurysm
b. Excessive vascular permeability
c. Vasular occlusion
d. Proliferation of new blood vessel and fibrous tissue on retina and
e. Contraction of the fibrovascular proliferation and vitrous

The clinical presentation of diabetes retinopathy follows as orderly progression, with variable contribution from each of these pathogenic processes. the processes that contribute to visual impairment are excessive vascular permeability or vascular occlusion leading to macular oedema and fibrovasular proliferation, which can result in vitreous hemorrhage. such hemorrhage are part of prolifertive stage of diabetic retinopathy and

are common cause of severe vision loss. in addition, the contraction of these fibrovasular proliferation results in distortion or detachment of retina, another cause of vision loss in proliferative stage of diabetes retinopathy (Derel LeRoith et al 2004).

Detection:

Diabetic retinopathy can be detected with hand held opthalmoscopy. However, this method of detection is limited by both the area of the retina that can be observed, the fact that inexperienced opthalmoscopists may miss clinically important retinopathy and the fact that use of monocular vision seriously limits he ability to distinguish background retinopathy (changes within the plane of the retina) from proliferative retinopathy (vessel changes that extend out of the plane of the retina into the vitreous). Similarly, macular edema may be missed by hand held ophthalmoscopy unless it is associated with retinal hard exudates cholesterol/lipid deposits that occur in conjunction with extravasation of crystalloid and protein. To insure adequate screening for diabetic retinopathy, a dilated eye exam is required and should be performed by an experienced ophthalmoscopist most commonly an ophthalmologist. More recently fundus photography has been used as a screening tool with properly obtained photographs then being read by an experienced reader. Stereo photography is required for best results. Technician training is required. Furthermore, this technique is often unsatisfactory in diabetic patients with cataracts (Byron J H et al 2005).

2.14.2.2.2. Nephropathy:

Detection:

Diabetic nephropathy is now most commonly detected by using sensitive measures or urinary albumin excretion commonly called microalbuminuria. The natural history of diabetic nephropathy is generally associated with a progression from microalbuminuria to macroalbuminuria, which is then associated with progressive decline in renal function ultimately resulting in the need for renal replacement therapy. The American Diabetes Association recommends screening for microalbuminuria beginning 5 years after the onset of type 1 diabetes mellitus and at the time of diagnosis in type 2 diabetes

mellitus. This latter recommendation derives from the observation that type 2 diabetes may go unrecognized for many years before it is diagnosed (Byron J H et al 2005).

2.14.2.2.3. Neuropathy:

Detection:

The onset of loss of sensation in the lower extremities is the commonest symptom associated with peripheral neuropathy (Partanen et al 1995; Vinik A et al 1999; Vinik Al et al 2000). However, the onset is often insidious. Careful questioning of patients about loss of sensation or altered sensation to touch and temperature may provide clues to diabetic neuropathy. In addition regular screening with a number of simple techniques has become the standard of care (Meijer JW et al 2002; Armstrong DG et al 1998; Armstrong DG et al 2000). These techniques include testing for lower extremity reflexes, testing for vibration with a tuning fork (preferably 128 hertz), and some measure of touch usually with a pin or monofilament. Monofilament testing has become the gold standard. Patients need to be given instruction in the foot examination with special attention to development of callus formation and loss of skin integrity from either foot ulcers, pressure related blisters or infections such as tinea pedis. Good patient care dictates a foot exam at the time of every routine visit to the health care provider's office. Recognition of loss of sensation and early detection of foot lesions is necessary to help reduce the risk for neurotrophic foot ulcers or peripheral vascular disease both of which contribute the risk for lower extremity amputations in diabetic patients (Meijer JW et al 2002; Armstrong DG et al 1998; Armstrong DG et al 2000).

2.14.2.2.3. Macrovascular:

The macro vascular involves large vessels causing cardiovascular disease, cerebrovascular disease, and peripheral vascular disease.

2.14.2.3.1. Coronary Heart diseases:

Coronary heart disease (CHD) is currently the leading cause of death worldwide and together with diabetes, poses a serious health threat, particularly in the Indian Asian population**.**

2.14.2.3.2. Peripheral Vascular Disease:
Detection:

The prevalence of peripheral vascular disease in both sexes, as measured by al-kle/arm blood pressure ratios, is 22% to 34% those with type 1 DM (Beach KW et al 1979, Orchard TJ et al 1990) and 22% among those with type 2 DM (Beach KW et al 1988) other populations studies of DM and non diabetics have reported PVD prevalence in range of 4% to 6% (Fabsitz RR et al 1999; lamur Welch VL et al 2002; Murabito JM et al 2002). Studies of PVD conducted in excessively diabetic population ranging in size from 70 to 1,018 persons report PVD prevalence ranging from 5.1% to 38.9% (Nilsson SE et al 1975; Klimt CR et al 1979; Melton LJ et al 1980), however in both population based studies clinic populations, PVD is consistently higher in daibetic than in non diabetic patients (Nilsson SE et al 1975; Klimt CR et al 1979; Melton LJ et al 1980). Variability in prevalence estimates of PVD is due to different methods for ascertaining this condition. Use ofRose Questionnaire for intermittent claudication often identifies only advance disease. Thereboy yielding a high false negative rate use of Doppler blood pressure to calculate ABI result in higher PVD prevalence due to its ability ti identify subclinical disease. This was shown in Strong Heart Study, in which only 33% of participants with ABI defined PVD had posterior tibial pulse and only 1.8% had intermittent caludication by the Rose Questionaire. There are few data on progression of PVD. Data from Mayo Clinic, however suggest that progression of PVD did not different between diabetic and nondiabetic individuals (Osmundson PJ et al 1990). On prospective study of PVD showed that 8.2% of people with baseline caludication had amputation or vascular surgery over 5 year's followup (Leng GC et al 1996). Finally, it is important to note that genetic polymorphism may interact with environmental/ behavioral factors such as smoking to increase risk of PVD among people with diabetes, a hypothesis supported by data from Honolulu Asia Aging Study (Resnik HE et al 2000) results of this report, along with other

studies (Palumbo PJ et al 1996) showing the deleterious effects of smoking on PVD, highlight the need to eliminate modifiable CVD risk factors.

2.14.2.3.3. Cerebrovascular Disease (NIH 2013)**:**

Cerebral vascular disease affects blood flow to the brain, leading to strokes and TIAs. It is caused by narrowing, blocking, or hardening of the blood vessels that go to the brain or by high blood pressure.

Stroke: A stroke results when the blood supply to the brain is suddenly cut off, which can occur when a blood vessel in the brain or neck is blocked or bursts. Brain cells are then deprived of oxygen and die. A stroke can result in problems with speech or vision or can cause weakness or paralysis. Most strokes are caused by fatty deposits or blood clots jelly-like clumps of blood cells that narrow or block one of the blood vessels in the brain or neck. A blood clot may stay where it formed or can travel within the body. People with diabetes are at increased risk for strokes caused by blood clots.

A stroke may also be caused by a bleeding blood vessel in the brain. Called an aneurysm, a break in a blood vessel can occur as a result of high blood pressure or a weak spot in a blood vessel wall.

TIAs: TIAs are caused by a temporary blockage of a blood vessel to the brain. This blockage leads to a brief, sudden change in brain function, such as temporary numbness or weakness on one side of the body. Sudden changes in brain function also can lead to loss of balance, confusion, blindness in one or both eyes, double vision, difficulty speaking, or a severe headache. However, most symptoms disappear quickly and permanent damage is unlikely. If symptoms do not resolve in a few minutes, rather than a TIA, the event could be a stroke. The occurrence of a TIA means that a person is at risk for a stroke sometime in the future.

2.15. Treatment of diabetes mellitus:

Glycaemic targets:

The ADA s ‘Standards of Medical Care in Diabetes’ recommends lowering HbA1c to <7.0% (<53 mmol/mol) in most patients to reduce the incidence of microvascular disease (ADA 2011). This can be achieved with mean plasma glucose of ~8.3– 8.9

mmol/l (~150–160 mg/dl); ideally, fasting and pre meal glucose should be maintained at <7.2mmol/l (<130mg/dl) and the postprandial glucose at <10 mmol/l (<180 mg/dl). More stringent HbA1c targets (e.g. 6.0–6.5% (42–48 mmol/mol)) might be considered in selected patients (with short disease duration, long life expectancy, no significant CVD) if this can be achieved without significant hypoglycaemia or other adverse effects of treatment (Ismail-Beigi F et al 2011; Akalin S et al 2009). Conversely, less stringent HbA1c goals—e.g. 7.5–8.0% (58–64 mmol/mol) or even slightly higher—are appropriate for patients with a history of severe hypoglycaemia, limited life expectancy, advanced complications, extensive comorbid conditions and those in whom the target is difficult to attain despite intensive self management education, repeated counseling and effective doses of multiple glucose lowering agents, including insulin (Ismail-Beigi F et al 2011; Lee SJ et al 2011).

The accumulated results from the aforementioned type 2 diabetes cardiovascular trials suggest that not everyone benefits from aggressive glucose management. It follows that it is important to individualize treatment targets (Blonde L 2010; Gerstein HC et al 2008; Turnbull FM et al 2009). As mentioned earlier, the desires and values of the patient should also be considered, since the achievement of any degree of glucose control requires active participation and commitment (Glasgow RE et al 2008; Gandhi GY et al 2008; Ahmed MH et al 2009; May C et al 2009). Indeed, any target could reflect an agreement between patient and clinician. An important related concept is that the ease with which more intensive targets are reached influences treatment decisions; logically, lower targets are attractive if they can be achieved with less complex regimens and no or minimal adverse effects. Importantly, utilizing the percentage of diabetic patients who are achieving an HbA1c <7.0% (<53 mmol/mol) as a quality indicator, as promulgated by various healthcare organizations, is inconsistent with the emphasis on individualization of treatment goals (Inzucchi S E et al 2012).

2.16. Therapeutic options:

Lifestyle Interventions designed to impact an individual's physical activity levels and food intake are critical parts of type 2 diabetes management (Anderson JW et al 2003; Klein S et al 2004). All patients should receive standardized general diabetes edu-

cation (individual or group, preferably using an approved curriculum), with a specific focus on dietary interventions and the importance of increasing physical activity. While encouraging therapeutic lifestyle change is important at diagnosis, periodic counseling should also be integrated into the treatment programme. Weight reduction, achieved through dietary means alone or with adjunctive medical or surgical intervention, improves glycaemic control and other cardiovascular risk factors. Modest weight loss (5–10%) contributes meaningfully to achieving improved glucose control. Accordingly, establishing a goal of weight reduction, or at least weight maintenance, is recommended. Dietary advice must be personalized (Bantle JP et al 2008).

Patients should be encouraged to eat healthy foods that are consistent with the prevailing population wide dietary recommendations and with an individual's preferences and culture. Foods high in fibre (such as vegetables, fruits, wholegrains and legumes), low fat dairy products and fresh fish should be emphasised. High energy foods, including those rich in saturated fats, and sweet desserts and snacks should be eaten less frequently and in lower amounts (Elmer PJ et al 2006; Gordon NF et al 2004; Wing RR 2006). Patients who eventually lose and keep weight off usually do so after numerous cycles of weight loss and relapse. The healthcare team should remain non judgmental but persistent, revisiting and encouraging therapeutic lifestyle changes frequently, if needed. As much physical activity as possible should be promoted, ideally aiming for at least 150 min/week of moderate activity including aerobic, resistance and flexibility training (Boule NG et al 2001).

In older individuals, or those with mobility challenges, so long as tolerated from a cardiovascular standpoint, any increase in activity level is advantageous. At diagnosis, highly motivated patients with HbA1c already near target (e.g. <7.5% (<58 mmol/mol)) could be given the opportunity to engage in lifestyle change for a period of 3–6 months before embarking on pharmacotherapy (usually metformin). Those with moderate hyperglycaemia or in whom lifestyle changes are anticipated to be unsuccessful should be promptly started on an antihyperglycaemic agent (also usually metformin) at diagnosis, which can later be modified or possibly discontinued if lifestyle changes are successful (Inzucchi S E et al 2012). Ultimately, the aims of controlling glycaemia are to avoid acute osmotic symptoms of hyperglycaemia, to avoid instability in blood glucose over

time, and to prevent/delay the development of diabetic complications without adversely affecting quality of life. Information on whether specific agents have this ability is incomplete; an answer to these questions requires long term, large scale clinical trials—not available for most drugs. Effects on surrogate measures for glycaemic control (e.g. HbA1c) generally reflect changes in the probability of developing microvascular disease but not necessarily macrovascular complications. Particularly from a patient standpoint, stability of metabolic control over time may be another specific goal (Inzucchi S E et al 2012).

2.17. Oral Antidiabetic Agents:

Type 2 diabetes mellitus is a progressive and complex disorder that is difficult to treat effectively in the long term. The majorities of patients are overweight or obese at diagnosis and will be unable to achieve or sustain near normoglycaemia without oral antidiabetic agents; a sizeable proportion of patients will eventually require insulin therapy to maintain long term glycaemic control, either as monotherapy or in conjunction with oral antidiabetic therapy. The frequent need for escalating therapy is held to reflect progressive loss of islet βcell function, usually in the presence of obesity related insulin resistance. Today's clinicians are presented with an extensive range of oral antidiabetic drugs for type 2 diabetes (Andrew J et al 2005).

The main classes are heterogeneous in their modes of action, safety profiles and tolerability. These main classes include agents that stimulate insulin secretion (sulphonylureas and rapid acting secretagogues), reduce hepatic glucose production (biguanides), delay digestion and absorption of intestinal carbohydrate (α-glucosidase inhibitors) or improve insulin action (thiazolidinediones) (Andrew J et al 2005).

2.18. Classification of antidiabetic agents

1. Insulin Secretagogues
2. Insulin Sensitisers.
3. Dipeptidyl peptidase-4 (DPP-4) inhibitors
4. α-Glucosidase Inhibitors

2.19. Insulin Secretagogues:

1. Sulfonylurea drugs
2. Meglitinide analogues

2.19.1. Sulphonylureas:

Sulphonylureas have been extensively used for the treatment of type 2 diabetes for nearly 50 years. They lower blood glucose concentrations primarily by stimulating insulin secretion from the β cells of the pancreatic islets. By the 1960s several sulphonylureas were available, including tolbutamide, acetohexamide, tolazamide and chlorpropamide, offering a range of pharmacokinetic options (Andrew J Krentz et al 2005).

A succession of more potent so called second generation sulphonylureas emerged in the 1970s and 1980s, for example glibenclamide (glyburide), gliclazide and glipizide. The latest, glimepiride, was introduced in the late 1990s (Evans AJ et al 1999). Glimepiride is a once daily drug for which claims have been made that it might offer advantages over other sulphonylureas with respect to the risks of weight gain and hypoglycaemia. Compared with older sulphonylureas, glimepiride is relatively expensive and clinical outcome data are not available, as they are for the agents used in the UKPDS. The clinical relevance of theoretical but much debated effects of glimepiride on ischaemic preconditioning – whereby a brief episode of ischaemia protects the myocardium against the detrimental effects of subsequent and more severe interruption of perfusion – remain uncertain. The issues of the importance of ischaemic reconditioning and the possible influence of different sulphonylureas continue to be debated (Ashcroft FM et al 1999).

2.19.1.1. Mechanism of action of Sulphonylureas:

2.19.1.1.1. Pancreatic mechanism:

Sulphonylureas have direct effects on the insulin producing islet β cells. The drugs bind to the β-cell sulphonylurea receptor (SUR)-1, part of a transmembrane complex with adenosine 5′-triphosphate sensitive Kir 6.2 potassium channels (KATP channels) (Ashcroft FM et al 1999; Gribble FM et al 2002). Binding of the sulphonylurea closes these KATP channels; this reduces cellular potassium efflux favouring membrane depolarisation. In turn, depolarisation opens voltage dependent calcium channels, resulting in an

influx of calcium that activates calcium dependent proteins that control the release of insulin. When sulphonylureas interact with SUR1 in the β-cell plasma membrane they cause prompt release of preformed insulin granules adjacent to the plasma membrane – the so called 'first phase' of insulin release (Rorsman P et al 2003). Sulphonylureas also increase the extended ('second phase') of insulin release that begins approximately 10 minutes later as insulin granules are translocated to the membrane from within the β cell (Groop LC 1992). The protracted stimulation of the 'second phase' of insulin release involves the secretion of newly formed insulin granules. The increased release of insulin continues while there is ongoing drug stimulation, provided the β cells are fully functional. Sulphonylureas can cause hypoglycaemia since insulin release is initiated even when glucose concentrations are below the normal threshold for glucose stimulated insulin release (approximately 5 mmol/L) (Ashcroft FM et al 1999).

2.19.1.1.2. Extra Pancreatic Mechanisms (Jack Deruiter 2003):

- The sulfonylureas also reduce serum glucagon levels possibly contributing to its hypoglycemic effects. The precise mechanism by which this occurs remains unclear but may result from indirect (secondary) inhibition due to enhanced release of both somatostatin and insulin.
- Sulfonylureas may also potentiate insulin action at target tissues (drug dependent characteristic).

2.19.1.2. Generations of sulphonylureas:

First generation sulphonylureas:

- Acetohexamide
- Chlorpropamide
- Tolbutamide
- Tolazamide

Second generation sulphonamide:

- Glipizide
- Gliclazide
- Glibenclamide (glyburide)
- Glyclopyramide
- Gliquidone

Third generation sulphonamide:

Glimepiride

2.19.1.3. Mechanism of action of Sulphonylureas

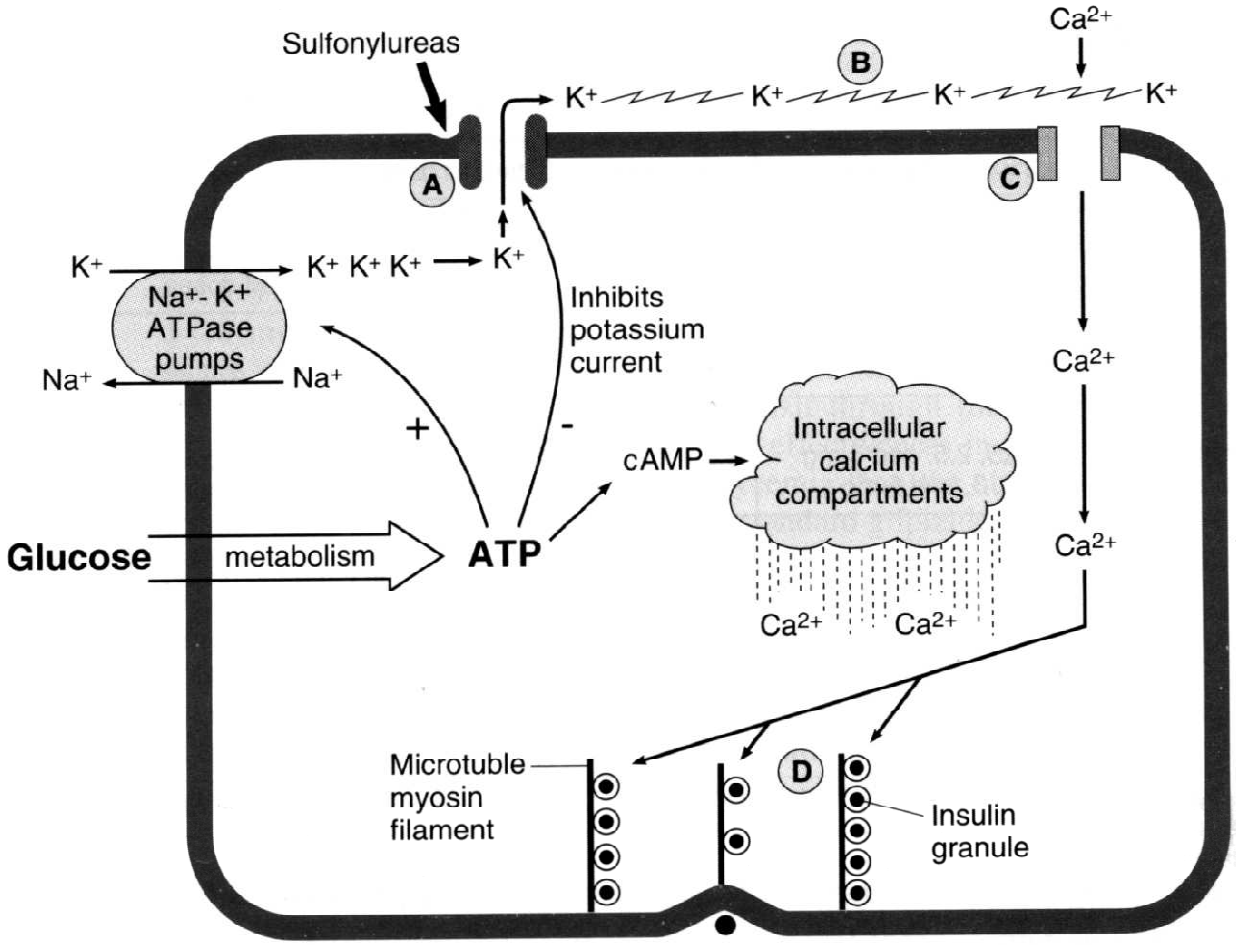

Figure no. 1: Mechanism of action of Sulphonylureas

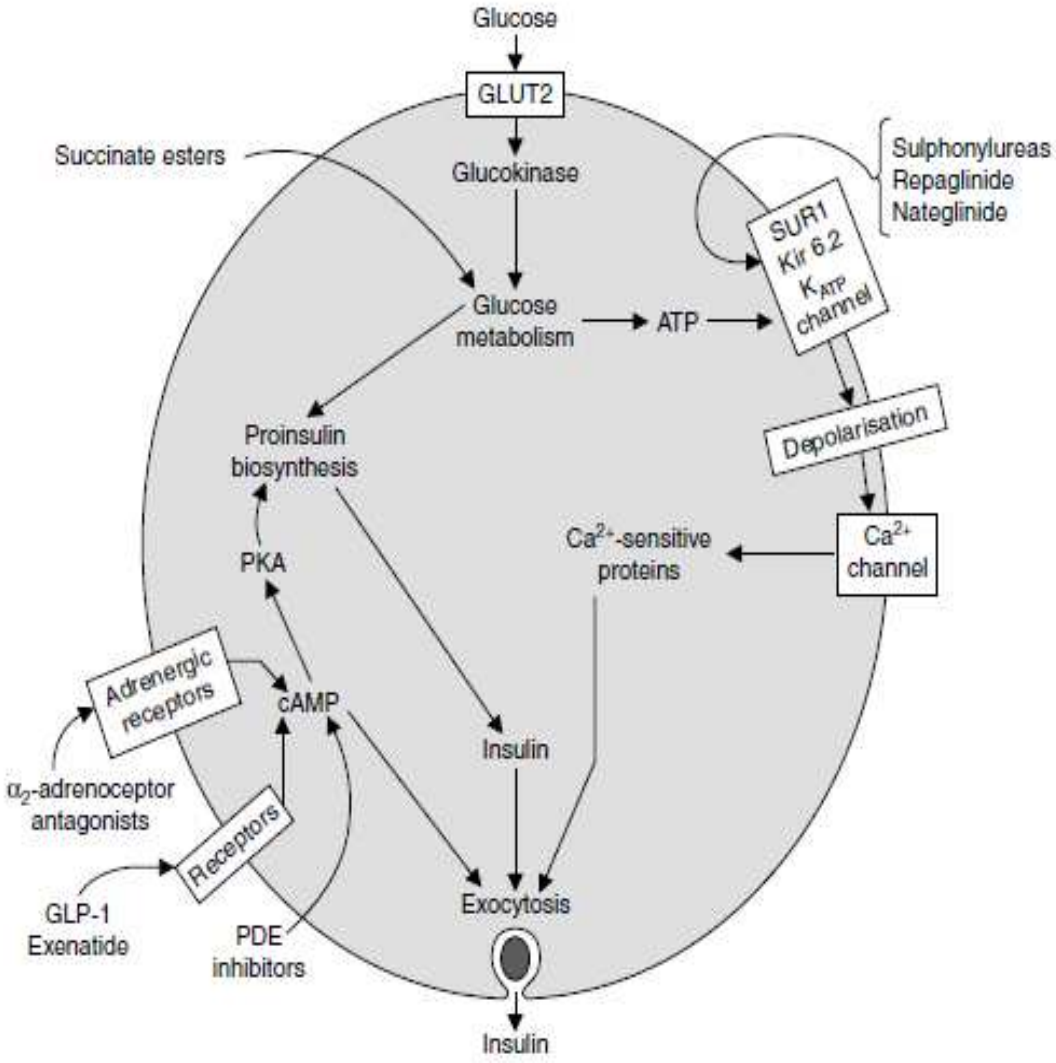

Figure No 2: The insulin releasing effect of sulphonylureas and other agents on the pancreatic islet β cell

Sulphonylureas bind to the sulphonylurea receptor (SUR)-1 located within the plasma membrane. This closes Kir 6.2 potassium channels which reduces potassium efflux, depolarises the cell and opens voltage dependent calcium influx channels. Raised intracellular calcium brings about insulin release. According to the stimulus secretion model, metabolism of glucose generates adenosine 5′-triphosphate (ATP) leading to closure of potassium channels, permitting the normal β cell to link insulin secretion closely to glucose concentration. Sulphonylureas may also enhance nutrient stimulated insulin secretion by other actions on the β cell. Other secretagogues, e.g. repaglinide, nateglinide, also stimulate insulin secretion via the SUR-Kir 6.2 complex. Other agents, e.g. phos-

phodiesterase (PDE) inhibitors, glucagon like peptide (GLP)-1 (7–36 amide), act via cyclic adenosine monophosphate (cAMP) and protein kinase A (PKA) to promote proinsulin synthesis GLUT2 = glucose transporter-2.

2.19.1.4.Pharmacokinetics of first and second generation sulphonylureas:

The principal distinguishing feature between different sulphonylureas relates to their pharmacokinetic characteristics. Duration of action varies from <12 hours for tolbutamide to >24 hours for chlorpropamide because of differences in

(i) Rates of metabolism

(ii) Activity of metabolites

(iii) Rates of elimination (Rendell M et al 2004).

These properties have important implications for the risk of hypoglycaemia associated with various sulphonylureas, an issue that is further complicated by retarded release preparations of some compounds. All sulphonylureas are well absorbed and most reach peak plasma concentration in 2–4 hours. They are metabolised in the liver, although metabolites and their routes of elimination vary considerably between compounds. Since all sulphonylureas are highly bound to plasma proteins they have the potential to interact with other drugs sharing this binding, for example salicylates, sulphonamides and warfarin; displacement from circulating proteins has been implicated in cases of severe sulphonylurea induced hypoglycaemia (Ashcroft FM et al 1999).

2.19.1.5. Indications and Contraindications:

Sulphonylureas remain a popular choice as first line oral therapy for patients with type 2 diabetes who have not achieved or maintained adequate glycaemic control using nonpharmacological measures. Customarily they are preferred for patients who are not overweight since weight gain is usually promoted by their use. Sulphonylureas can be used in combination with agents from other classes of antidiabetic agents, with the exception of other insulin secretagogues. Daytime sulphonylurea treatment may be used in combination with bedtime insulin, and can reduce insulin doses by up to 50%. This is an increasingly accepted practice, albeit one that lacks firm evidence of long term advantages over insulin monotherapy (Yki Ja rvinen H. 2001). As mentioned earlier, continued

gradual loss of β-cell function is to be expected in most patients, this requires escalating insulin doses with increasing duration of diabetes. Sulphonylureas should be introduced at a low dose, usually the lowest recommended by the manufacturer, the blood glucose response being carefully observed over the first few weeks; self monitoring of blood glucose by the patient may be helpful and, as mentioned earlier, is recommended where there are concerns about the potential consequences of hypoglycaemia, for example in the vulnerable elderly patient. In general and intuitively, patients who have achieved less marked degrees of fasting hyperglycaemia after a trial of diet and exercise are more likely to develop hypoglycaemia than those with more marked hyperglycaemia.

The dosage is increased at intervals of 2–4 weeks until the glycaemic target is, hopefully, achieved. If hypoglycemia develops, or if a dosage increment produces no further improvement in glycaemic control, it is advisable to return to the previous dose. Hypoglycaemia, whether actual or, more commonly, perceived as a risk by either the patient or clinician, is the principal limitation to rapid dose escalation of sulphonylureas. The latter point notwithstanding, it should be borne in mind that the maximal blood glucose lowering effect is usually achieved at doses below the maximum recommended by the manufacturer. This is probably a reflection of the fact that maximum stimulation of insulin secretion has already been attained since β-cell function is significantly impaired. Improved β-cell capacity resulting from alleviation of glucose toxicity may contribute to the risk of hypoglycaemia in some patients. Long term glycaemic control should be monitored by periodic measurement of HbA1c (or fructosamine if HbA1c is not available).

2.19.1.6. Efficacy:

The blood glucose lowering efficacy of sulphonylureas has been evaluated in many retrospective and prospective studies, and from decades of collective worldwide clinical experience. When used as monotherapy in patients inadequately controlled by nonpharmacological measures, sulphonylureas can be expected to reduce fasting plasma glucose by an average of 2–4 mmol/L accompanied by a decrease in HbA1c of 1–2% (Krentz AJ et al 2001; Bailey CJ et al 2003, Krentz AJ et al 1994). However, individual responses are variable. Since the hypoglycaemic effect of sulphonylureas is attributable to increased insulin secretion, the effectiveness of these drugs is dependent on adequate

β-cell function. The aforementioned progressive β-cell failure that determines the natural history of type 2 diabetes may require an increased dosage of sulphonylureas if glycaemic control deteriorates. Rapid and uncontrollable deterioration of glycaemic control during sulphonylurea therapy is sometimes termed 'secondary sulphonylurea failure'. This phenomenon, which is something of a misnomer, occurs in approximately 5–10% of patients per annum with suggestions of differences in 'failure' rates between some com pounds (DeFronzo RA 1999, EDPG 1999).The inability to maintain acceptable glycaemic control is common to all sulphonylureas and is held to reflect an advanced stage of β-cell failure, that is, it is a reflection of disease progression rather than a true failure of therapy.

Individuals who have greater degrees of β-cell reserve usually respond well to sulphonylureas; early use of sulphonylureas as first line monotherapy in these patients will produce better blood glucose lowering than late intervention in patients with severely compromised β-cell function. The plasma insulin concentrations achieved during sulphonylurea therapy do not usually extend beyond the range observed in the general non diabetic population (including those with impaired glucose tolerance), and suggestions that sulphonylurea induced hyperinsulinaemia might increase the risk of detrimental insulin induced effects on the cardiovascular system remain unsubstantiated (Krentz Aj 2003).

Sulphonylurea therapy generally has modest effects on blood lipid profiles, although some studies have noted a small decrease in plasma triglyceride levels possibly linked to improved glycaemic control and minor increments in high density lipoprotein (HDL) cholesterol. When a sulphonylurea is used in combination with another antidiabetic agent, the glucose lowering efficacy of the sulphonylurea is approximately additives to the effect of the other agent. Once again, response is crucially dependent on the presence of adequate β-cell function. Early use of such combination therapy is indicated when optimal titration of a single agent does not achieve adequate glycaemic control.

The combination of two different types of agents is more likely to achieve glycaemic targets, albeit for a variable period of time. If combination therapy is started at a stage when hyperglycaemia is already marked (after 'failure' of monotherapy), then β-cell depletion is likely to be advanced. Under these circumstances, oral combination therapy is likely to offer limited benefit and the need for an early move to insulin treatment is

usually clear. Since there are occasional exceptions to this rule, a limited trial of combination oral therapy may be worthwhile. However, the temptation to procrastinate unduly on transferring the patient to insulin treatment should be firmly resisted, not least since some patients derive rapid symptomatic benefit from insulin therapy. Impending metabolic decompensation, with or without ketosis, mandates immediate insulin treat ment; more severe degrees of decompensation, for example obtundation, dehydration, ketosis associated vomiting, necessitates emergency hospitalisation for treatment with intravenous insulin, fluids and electrolytes (Andrew J et al 2005).

2.19.1.7. Adverse Events:

Hypoglycaemia, usually subclinical or minor but occasionally life threatening, is the most common and potentially most serious adverse effect of sulphonylurea therapy (Krentz AJ et al 1994). Patients receiving sulphonylureas should receive instruction on the recognition and prevention of hypoglycaemia and the prompt actions they must take should warning symptoms develop. Severe protracted hypoglycaemia is more likely with longer acting sulphonylureas such as glibenclamide, with tolbutamide holding the lowest place in the hierarchy of risk. Individuals with irregular eating habits or excessive alcohol con sumption are at higher risk of sulphonylurea induced hypoglycaemia.

As mentioned, hypoglycaemia is also more likely to occur in patients with satisfactory glycaemic control, as indicated by an HbA1c concentration within, or just above, the non diabetic reference range. These patients should always be questioned directly about recent symptoms of hypoglycaemia, although their nonspecific nature can raise problems of over diagnosis; self monitoring of capillary blood glucose concentrations during suggestive episodes should help to clarify this issue, although uncertainties may not be completely dispelled. If there is continuing doubt, a temporary reduction in dose is usually indicated.

Estimates of the incidence of mild hyperglycaemia that is, not requiring assistance from another individual are often based on symptoms which have not necessarily been confirmed by contemporaneous self measurement of capillary blood glucose. In the UKPDS, for example, about 20% of sulphonylurea treated patients reported one or more episodes suggestive of hypoglycaemia annually; other studies have suggested similar

rates (Krentz AJ et al 1994). The timing of hypoglycaemia tends to reflect the pharmacokinetics of the sulphonylurea. Thus, glibenclamide has a propensity to cause interprandial benclamide has a propensity to cause interprandial hypoglycaemia whereas chlorpropamide tends to induce hypoglycaemia in the pre-breakfast period. Other adverse events of sulphonylureas include uncommon sensitivity reactions usually cutaneous that are usually transient; erythema multiforme is rare. Fever, jaundice and blood dyscrasias are very rare; some sulphonylureas can reportedly precipitate acute porphyria in predisposed individuals. In its heyday, chlorpropamide was notorious for causing unpleasant facial flushing after consuming small quantities of alcohol; photosensitivity has also been reported. Chlorpropamide could also increase renal sensitivity to antidiuretic hormone, occasionally causing water retention with hyponatraemia.

In contrast, glibenclamide is credited with a mild diuretic action. Weight gain is regarded as a class effect of sulphonylurea therapy, typically amounting to 1–4kg and stabilising after about 6 months. This weight gain, which is always unwelcome, is thought to reflect the anabolic effects of increased plasma insulin concentrations; some studies have suggested that reduced loss of calories as glucose in the urine may account for the majority of the weight gain (Bailey CJ et al 2003; DeFronzo RA 1999).

2.19.2. Meglitinide analogues:

- **Repaglinide**
- **Nateglinide**

Derivatives of meglitinide, such as repaglinide and the phenylalanine derivative nateglinide, are promoted as ‘prandial glucose regulators’; in fact, fasting hyperglycaemia is also improved to a lesser extent, particularly with repaglinide. Clinical experience with these agents remains limited in most countries; these drugs are appreciably more expensive than most sulphonylureas, the atter also having the reassurance of outcome data from the UKPDS (UKPDS Group 1998).

2.19.2.4. Mode of Action of Meglitinide:

Benzamido prandial insulin releasers bind to the SUR1 in the plasma membrane of the ß cell at a site distinct from the sulphonylurea binding site Since the KATP channel

is closed when either the benzamido binding site or the sulphonylurea binding site on the SUR1 is bound with its respective agonist, there is no advantage in giving a prandial insulin releaser in addition to a sulphonylurea. However, drugs are also in development that promote ß- cell proinsulin synthesis and act via signalling pathways distinct from the KATP channel. The short half life of repaglinide results in enhancement of the first phase and early second phase of insulin secretion that is less sustained than that observed with sulphonylureas (Landgraf R 2000; Dornhorst A 2001; Davies M 2002).

Theoretical benefits on cardiovascular outcomes from preferentially targeting the postprandial period remain to be confirmed (Dornhorst A 2001; Davies M 2002). It is unclear whether postprandial hyperglycaemia *per se* is detrimental to the vascular endothelium or whether closely associated metabolic disturbances, for example dyslipidaemia, are responsible. Thus, the mechanism of the association between post challenge hyperglycaemia and mortality observed in the multicentre DECODE (Diabetes Epidemiology: Collaborative analysis Of Diagnostic criteria in Europe) study is uncertain (Qiao Q et al 2002).

2.19.2.5. Pharmacokinetics:

Repaglinide is rapidly and almost completely absorbed after oral administration, with peak plasma concentrations achieved in about 1 hour (Landgraf R 2000). The drug is rapidly metabolised in the liver to inactive metabolites, which are mainly excreted in bile. When taken about 15 minutes before a meal repaglinide produces a prompt insulin releasing effect, which is limited to a period of about 3 hours, roughly coinciding with the duration of meal digestion. Nateglinide has a slightly faster onset and shorter duration of action, its binding to target receptors lasting only seconds. A 60mg dose of nateglinide taken 20 minutes before an intravenous glucose tolerance test restored first phase insulin release and lowered glucose concentrations (Dornhorst A 2001; Davies M 2002).

2.19.2.6. Indications and Contraindications:

Repaglinide may be used as monotherapy in patients inadequately controlled by nonpharmacological measures. Suitable candidates for rapid acting insulin releasers include individuals with irregular lifestyles wherein meals are unpredictable or missed. The

lower risk of hypoglycaemia associated with its use makes repaglinide an attractive option for some elderly patients, particularly if other agents are contraindicated. However, the need for multiple daily dosages is a potential disincentive. Repaglinide should ideally be taken about 15–30 minutes before a meal. Starting with a low dose, for example 0.5mg before each main meal the effect on glycaemic control is monitored and the dosage titrated up every 2 weeks to a maximum of 4mg before each main meal; if a meal is not consumed the corresponding dose of repaglinide should be omitted. If glycaemic targets are not met, consider early introduction of combination therapy (e.g. with metformin). Unlike some sulphonylureas and metformin, repaglinide is suitable for patients with moderate renal impairment, although careful upward dose titration and close monitoring is still recommended. In contrast with the US, the UK license for nateglinide currently restricts use to combination therapy with metformin in patients who do not achieve glycaemic targets with the latter drug as monotherapy. (Davies M 2002). In the US, nateglinide may also be used as monotherapy or combined with a thiazolidinedione. Nateglinide should be used with caution in patients with hepatic disease.

2.19.2.7. **Efficacy:**

Repaglinide (0.5–4mg taken about 15–30 minutes before meals) results in dose dependent increases in insulin secretion with reduced postprandial hyperglycaemia; a lesser reduction in fasting hyperglycaemia is also observed. Overall reductions in HbA1c are similar in magnitude to those observed with sulphonylureas, that is 1–2%. Combined with metfmin, nateglinide reduces HbA1c by up to1.5% (Dornhorst A 2001; Davies M 2002).

2.19.2.8. **Adverse Events:**

The overall incidence of hypoglycaemic episodes is lower with repaglinide than with sulphonyureas. Sensitivity reactions, usually transient, can occur. Increased plasma levels of repaglinide have been reported when coadministered with gemfibrozil. A small increase in bodyweight can be expected in patients starting repaglinide as initial monotherapy, but there may be little change in weight among patients switched from a sulpho-

nylurea. Nateglinide appears to have little effect on bodyweight when combined with metformin (Davies M 2002).

2.19.3. Insulin Sensitizer:

Insulin resistance is a prominent metabolic defect in most patients with type 2 diabetes (Reaven GM et al 1988; Krentz AJ 2002; Andrew J. et al 2005) Defective insulin action is not confined to glucose metabolism, subtle defects also being demonstrable in the regulation of other aspects of intermediary metabolism (e.g. lipolysis), using appropriate investigative techniques. Many cross sectional and prospective studies have implicated insulin resistance in the pathogenesis of type 2 diabetes and the related metabolic syndrome of cardiovascular risk. Therefore, defective insulin action at target tissue level is an attractive therapeutic target in type 2 diabetes (Ginsberg HN et al 2000). The biguanides and, in particular, the thiazolidinediones act directly against insulin resistance, and so are regarded as insulin sensitising drugs (Campbell IW et al 2000).

2.19.3.4. Bigunides:

The finding that *Galega officinalis* (goat's rue or French lilac), historically used as a traditional treatment for diabetes in Europe, was rich in guanidine led to the introduction of several glucose lowering guanidine derivatives in the 1920s. These early antidiabetic agents were all but forgotten as insulin became widely available and it was not until the late 1950s that three antidiabetic biguanides were reported: metformin, phenformin and buformin. Phenformin was withdrawn in many countries in the 1970s because of a high incidence of lactic acidosis; buformin received limited use in a few countries, leaving metformin as the main biguanide on a global basis.

1.19.3.1.1. Metformin

Metformin is the only biguanide available in the UK and, since 1995, the US.(Krentz AJ et al 1994; Bailey CJ et al 1996). Extensive clinical experience with metformin has been complemented by favourable results from the UKPDS. Metformin also enjoys the accolade of being among the least expensive of the oral antidiabetic agents.

2.19.3.1.2. Mode of Action of Action Bigunides:

Metformin has a variety of metabolic effects, some of which may confer clinical benefits that extend beyond glucose lowering. However, the molecular mechanisms of metformin have yet to be fully identified. At the cellular level, metformin improves insulin sensitivity to some extent, an action mediated via post receptor signaling pathways for insulin (Kirpichnikov D et al 2002; Cusi K et al 1998). Recent data have suggested that adenosine 5′-monophosphate activated protein kinase (AMPK) is a possible intracellular target of metformin (Zhou G et al 2001). Through phosphorylation of key proteins, AMPK acts as a regulator of glucose and lipid metabolismand cellular energy regulation (Winder WW et al 1999).

Since metformin lowers blood glucose concentrations without causing overt hypoglycaemia it is most appropriately classed as an antihyperglycaemic as distinct from hypoglycaemic agent. The clinical efficacy of metformin in patients with type 2 diabetes requires the presence of insulin. The drug does not stimulate insulin release and a small decrease in fasting insulin concentrations is typically observed in patients with hyperinsulinaemia (DeFronzo RA et al 199). The predominant glucose lowering mechanism of action of metformin is to reduce excessive rates of hepatic glucose production. Metformin reduces gluconeogenesis by increasing hepatic sensitivity to insulin and decreasing the hepatic extraction of certain gluconeogenic substrates (e.g. lactate).

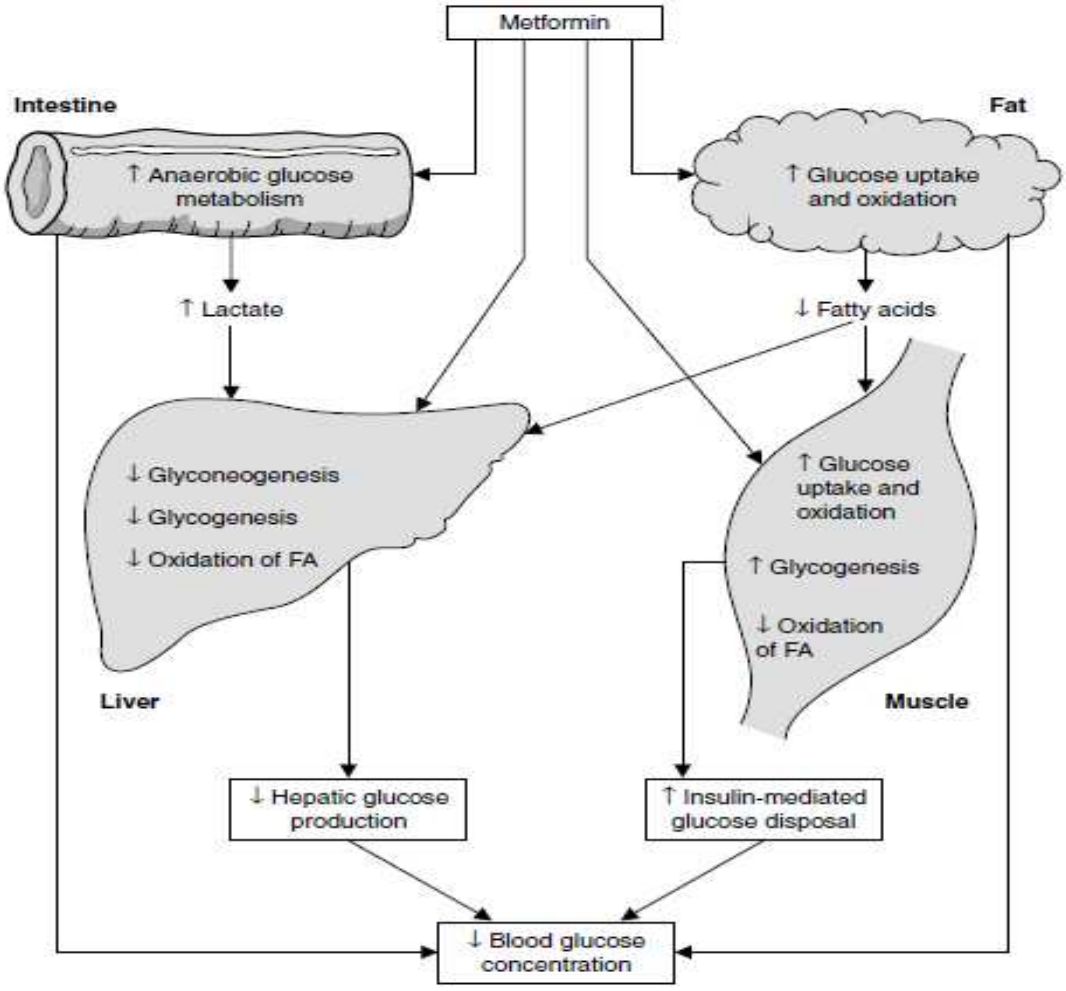

Figure No 03: Mechanism of Action of Metformin

Actions of Metformin: Inhibition of hepatic glucose production is regarded as the principal mechanism through which Metformin lowers blood glucose FA = fatty acids;
↑ indicates increase; ↓ indicates decrease..

Hepatic glycogenolysis is also decreased by metformin. Insulin stimulated glucose uptake in skeletal muscle is enhanced by metformin. This involves an increase in the movement of insulin sensitive glucose transporter molecules to the cell membrane; an increase in the activity of the enzyme glycogen synthase promotes synthesis of glycogen. Metformin also acts in an insulin independent manner to suppress oxidation of fatty acids and to reduce triglyceride levels in patients with hypertriglyceridaemia (Bailey CJ et al 2003). This reduces the energy supply for hepatic gluconeogenesis and has favourable

effects on the glucose fatty acid (Randle) cycle (in which fatty acids are held to compete with glucose as a cellular energy source) (Krentz AJ et al 2002). Glucose metabolism in the splanchnic bed is increased by metformin through insulin independent mechanisms. This may contribute to the blood glucose lowering effect of the drug, and in turn may help to prevent gains in bodyweight. Collectively, the cellular effects of metformin serve to counter insulin resistance and to reduce the putative toxic metabolic effects of hyperglycaemia (glucose toxicity) and fatty acids (lipotoxicity) in type 2 diabetes (Andrew J. Krentz et al 2005).

2.19.3.1.3. Pharmacokinetics*:*

Metformin is a stable hydrophilic biguanide that is quickly absorbed and eliminated unchanged in the urine. It is imperative that metformin is only prescribed to patients with renal function that is sufficient to avoid accumulation of the drug. Renal clearance of metformin is achieved more by tubular secretion than glomerular filtration, the only significant drug interaction being competition with cimetidine, which can increase plasma metformin concentrations. There is little binding of metformin to plasma proteins.

Metformin is not metabolised, and so does not interfere with the metabolism of coadministered drugs. Metformin is widely distributed, high concentrations being retained in the walls of the gastrointestinal tract; this provides a reservoir from which plasma concentrations are maintained. Nevertheless, peak plasma metformin concentrations are short lived: in patients with normal renal function the plasma half life (t/2) for metformin is 2–5 hours, and almost 90% of an absorbed dosage iseliminated within 12 hours (Bailey CJ et al 1996).

2.19.3.1.4. Indications and Contraindications:

Metformin is the therapy of choice for overweight and obese patients with type 2 diabetes (Cusi K, et al 1998). It can be equally effective in normal weight patients. Metformin can also be used in combination with any other class of oral antidiabetic agent or with insulin. The drug is contraindicated in patients with impaired renal function (i.e. serum creatinine>120–130 µmol/L, depending on lean body mass), as a precaution against drug accumulation. Cardiac or respiratory insufficiency, or any other condition predisposing to hypoxia or reduced perfusion (e.g. hypotension, septicaemia) are further contraindications, as well as liver disease, alcohol abuse and a history of metabolic acidosis. Metformin can be used in the elderly, provided that renal insufficiency and other exclusions are not present. A difficulty in practice is that significant renal dysfunction may be present without the aforementioned elevation of serum creatinine. The improvement in insulin sensitivity can cause ovulation to resume in cases of anovulatory polycystic ovary syndrome (PCOS) (an unlicensed application of the drug in the absence of diabetes).

Metformin should be taken with meals or immediately before meals to minimize possible gastrointestinal adverse effects. Treatment should be started with 500 or 850mg once daily, or 500mg twice daily. The dosage is increased slowly – one tablet at a time at intervals of about 2 weeks until the target level of glycaemic control is attained. If the target is not attained and an additional dose produces no greater effect, return to the previous dose and, in the case of monotherapy, consider combination therapy by adding in another agent (e.g. a sulphonylurea, prandial insulin releaser or thiazolidinedione).

The maximal effective dosage appears to be about 2000 mg/day (Lord JM et al 2003) given in divided doses with meals, the absolute maximum being 2550 or 3000 mg/day in different countries. Several single tablet combinations of a sulphonylurea (usually glibenclamide) with a biguanide (metformin or phenformin) have been available in some European countries and elsewhere for more than a decade. A slow release formulation of metformin and a fixed dose combination of metformin with glibenclamide is available in the US Glucovance , Bristol Myers Squibb Company, Princeton, NJ, USA) and elsewhere (although not in the UK). A combined rosiglitazone/metformin (Avandamet, GlaxoSmithKline, Philadelphia, PA, USA) preparation is also available in some parts of the world. During long term treatment with metformin it is advisable to check

(e.g. annually) for the development of contraindications, particularly an elevated serum creatinine concentration (yearly measurement of creatinine clearance posing practical difficulties). Metformin can reduce gastrointestinal absorption of cyanocobalamin (vitamin B12). While anaemia is very rare, an annual haemoglobin measurement is prudent in patients at risk of nutritional deficiencies. It is advised to stop metformin treatment temporarily during use of intravenous radiographic contrast media, surgery and any other intercurrent situationin which the exclusion criteria could be invoked (Howlett HCS et al 2000). Substitution with insulin may be appropriate at such times. Metformin alone is unlikely to cause serious hypoglycaemia, but hypoglycaemia becomes an issue when metformin is used in combination with an insulin releasing agent or insulin.

2.19.3.1.5. Efficacy*:*

The long term blood glucose lowering efficacy of metformin is broadly similar to sulphonylureas. As monotherapy in patients who are not adequately controlled on non-pharmacological therapy, optimal ly titrated metformin therapy typically reduces fasting plasma glucose by 2–4 mmol/L, corresponding to a decrease in HbA1c by approximately 1–2% (Bailey CJ et al 1996). The effect is dependent upon the presence of some endogenous ß-cell function, and is largely independent of bodyweight, age and duration of diabetes. However, given the progressive nature of type 2 diabetes, reassessment of dosage and consideration of additional therapy are required to maintain glycaemic control in the long term (Krentz AJ et al 2001; DeFronzo RA. et al 1999). Metformin has several features that mark it out as a good choice for first line monotherapy. The antihyperglycaemic action of metformin means that it is unlikely to cause severe hypoglycaemia. This may be explained in part because metformin does not stimulate insulin secretion. Indeed, the reduction of basal insulin concentrations, notably in hyperinsulinaemic patients, should itself improve insulin sensitivity by relieving the insulin induced downregulation of insulin receptor number and suppression of post receptor insulin pathways (Chiasson J L et al 2003).

Bodyweight tends to stabilise or decrease slightly during metformin therapy. Small improvements in the blood lipid profile may be observed in hyperlipidaemic patients; plasma concentrations of triglycerides, fatty acids and low density lipoprotein

(LDL) cholesterol tend to fall, whereas cardioprotective High Density Lipoprotein (HDL) cholesterol tends to rise. These effects appear to be independent of the antihyperglycaemic effect, although a lowering of triglyceride and free fatty acids is likely to help improve insulin sensitivity and benefit the glucose fatty acid cycle. In the UKPDS, overweight patients who started oral antidiabetic therapy with metformin showed a statistically significant 39% reduced risk of myocardial infarction compared with conventional treatment (p= 0.01) (UPDSG 1998). No clear relationship is evident between metformin dosage and decreased coronary artery events. This suggests that patients who can only tolerate a low dosage of metformin may benefit from continuing the drug, even when other agents have to be added to optimise glycaemic control. The decrease in myocardial infarction observed with metformin therapy in the UKPDS was not attributable to more effective lowering of HbA1c or major effects on classic cardiovascular risk factors such as plasma lipids. Consequently, other potentially vasoprotective effects of metformin have been invoked. Reported benefits of metformin on non classic cardiovascular risk factors include increased fibrinolysis and a reduced concentration of the antithrombolytic factor plasminogen activator inhibitor-1 (PAI-1) (Kirpichnikov D et al 2002; Howlett HCS et al 2000).

The mechanism of the cardioprotective effects of metformin remains uncertain. Detracting somewhat from this generally favourable view was evidence of an initially greater mortality when metformin was added to a sulphonylurea in a UKPDS substudy, (UPDSG 1998) but longer term follow up has shown the benefits of metformin to be sustained (TOCFDEM 2004). The explanation may have been, at least in part, a spuriously low mortality rate in the comparator sulphonylurea monotherapy group (UPDSG 1998; Hermann LS et al 2002). The small number of events in this sub-study adds to the uncertainty. Sulphonylurea plus metformin is a commonly used combination and it would be reassuring to have definitive safety data. Since each class as monotherapy appears safe from the cardiovascular perspective, alternative explanations have been postulated to explain similar findings seen in observational studies (Hermann LS et al 2002).

One plausible confounder might be greater cardiovascular risk attributable to more severe metabolic derangements in patients treated with the combination. Results from US trials and various large databases of follow up with sulphonylurea plus metfor-

min combination therapy have been reassuring (DeFronzo RA 1999; Hermann LS 2002; Johnson JA et al 2002). Additional well designed compara tive studies of appropriate statistical power would be required to quantify the risk to benefit equation for combination treatment with sulphonylurea plus metformin. However, recent results from the 5 year follow up of UKPDS – with no further attempt to continue in randomized groups – show that the adverse impact of sulphonylurea plus metformin combination seen initially is no longer evident (TOCFDEM 2004).

At this point, the aforementioned benefits observed on mortality and cardiovascular disease in overweight patients initially randomized to metformin monotherapy, while diminished, remained significant. Consistent with the action of metformin on insulin sensitivity, addition of metformin to patients receiving insulin therapy may necessitate a reduction of insulin dosage. Some patients also show an improvement in glycaemic control, although this is not always impressive. Metformin reduces the weight gain associated with insulin therapy and, by decreasing the insulin dosage, there may be a decrease in hypoglycaemic episodes. The regimen has usually involved once daily bedtime long acting (lente) insulin or twice daily insulin suspension isophane with metformin at mealtimes. In the US Diabetes Prevention Program, metformin reduced the incidence of new cases of diabetes in overweight and obese patients with impaired glucose tolerance by 33% overall. This compares with an intensive regimen of diet and exercise, which reduced the risk by 58% (DPPRG 2002). Younger, more obese individuals showed the most response to the preventive effects of metformin.

2.19.3.1.6. Adverse Effects:

Abdominal discomfort and other gastrointestinal adverse effects, including diarrhoea, are encountered fairly commonly during the introduction of metformin. Symptoms may remit if the dose is reduced and reiterated slowly, but about 10% of patients cannot tolerate the drug at any dose. The most serious feared adverse event associated with metformin is lactic acidosis; the occurrence is rare (about0.03 cases per 1000 patient years), but the mortality rate is high (Ashcroft FM ET AL 1999; Ginsberg HN 2000). Since the background incidence of lactic acidosis amongst type 2 diabetic patients has not been established, it is possible that a proportion of cases that have been attributed to the drug

were caused by other factors; this remains an area of controversy. Most cases of lactic acidosis in patients receiving metformin are due to inappropriate prescription of the drug. (Krentz AJ, ET AL 1994; Bailey CJ et al 1996; Hermann LS 2002; Sulkin T et al 1997). The leading contrain dication is renal insufficiency. Metformin increases glycolysis to lactate, particularly in the (Sulkin T et al 1997) splanchnic bed. The situation will be aggravated by any hypoxic condition or impaired liver function. Hyperlactataemia occurs in cardiogenic shock and other illnesses that decrease tissue perfusion, and metformin is often only an incidental factor in these cases (Lalau J D 2000).

In the absence of reliable data to the contrary, metformin treatment should be stopped immediately in all cases of suspected or proven lacticacidosis, regardless of cause. Lactic acidosis is typically characterised by a raised blood lactate concentration(e.g. >5mmol/L), decreased arterial pH and/or bicarbonate concentration with an increased anion gap ((Na+– (Cl– + HCO3–) >15 mmol/L). Presenting symptoms are often nonspecific, but frequently include hyperventilation, malaise and abdominal discomfort. Treatment should be commenced immediately without waiting to determine whether metformin is a cause; carbonate remains the therapy of choice but evidence of its efficacy is scanty. The value of haemodialysis in removing accumulated metformin has been challenged by some authorities, but dialysis may nonetheless be helpful in optimizing fluid and electrolyte balance during treatment with high dose intravenous bicarbonates (Lalau J D 2000).

2.19.4. Thiazolidinediones:

Thiazolidinediones improve whole body insulinsensitivity via multiple actions on gene regulation. These effects result from stimulation of a nuclear receptor peroxisome proliferator activated receptor (PPARγ), for which thiazolidinediones are potent synthetic agonists (Day C. 1999). The antidiabetic activity of thiazolidinediones was described in the early 1980s, troglitazone being the first of the class to become available for clinical use. Troglitazone was introduced in the US in 1997, only to be withdrawn in 2000 because of cases of idiosyncratic hepatotoxicity resulting in fatalities. Roglitazone was available in the UK for only for a few weeks in 1997 before being withdrawn by its distributor as reports of hepatotoxicity accumulated in other countries. To date, two other

thiazolidinediones, rosiglitazone and pioglitazone, have not shown the hepatotoxicity that led to the demise of troglitazone. Rosiglitazone and pioglitazone were introduced in the US in 1999 and in Europe in 2000 (Krentz AJ et al 2000). Combination preparations (e.g.thiazolidinedione plus metformin) are also available.

2.19.4.4.**Mode of action of Action Thiazolidinediones*:***

Stimulation of PPARγ is regarded as the principal mechanism through which thiazolidinediones enhance insulin sensitivity. PPARγ is expressed at highest levels in adipose tissue, and less so in muscle and liver. PPARγ operates in association with the retinoid X receptor. The resulting heterodimer binds to nuclear response elements, thereby modulating transcription of a range of insulin sensitive genes, in the presence of necessary cofactors (Day C. 1999; Rosen ED et al 2001). Many of the genes activated or suppressed by thiazolidinediones are involved in lipid and carbohydrate metabolism. Stimulation of PPARγ by a thiazolidinedione promotes differentiation of preadipocytes with accompanying lipogenesis, effects that promote or enhance the local effects of insulin. Thiazolidinediones increase glucose uptake via glucose transporter-4 in skeletal muscle, and some reports indicate that rates of gluconeogenesis in the liver are reduced.
Stimulation of lipogenesis via PPARγ reduces circulating nonesterified fatty acid (NEFA) concentrations through cellular uptake and triglyceride synthesis. The reduction in plasma NEFA concentrations is associated with increased glucose utilisation and reducing luconeogenesis by reducing operation of the glucose fatty acid cycle; reductions in ectopic lipid deposition in muscle and liver may contribute to the improvements on glucose metabolism.

Thiazolidinediones also reduce the production and activity of the adipocyte derived cytokine tumour necrosis factor (TNF)-a (Day C 1999). The latter has been implicated in the development of impaired insulin action although the precise role of TNFa in in muscle (Fasshauer M et al 2003), human states of insulin resistance remains unclear. Reductions in plasma insulin concentrations and lowering of circulating triglycerides are additional indirect mechanisms that may help to improve whole body insulin sensitivity. Thiazolidinediones, like metformin, are antihyperglycaemic agents and require the presence of sufficient insulin to generate a significant blood glucose-lowering effect.

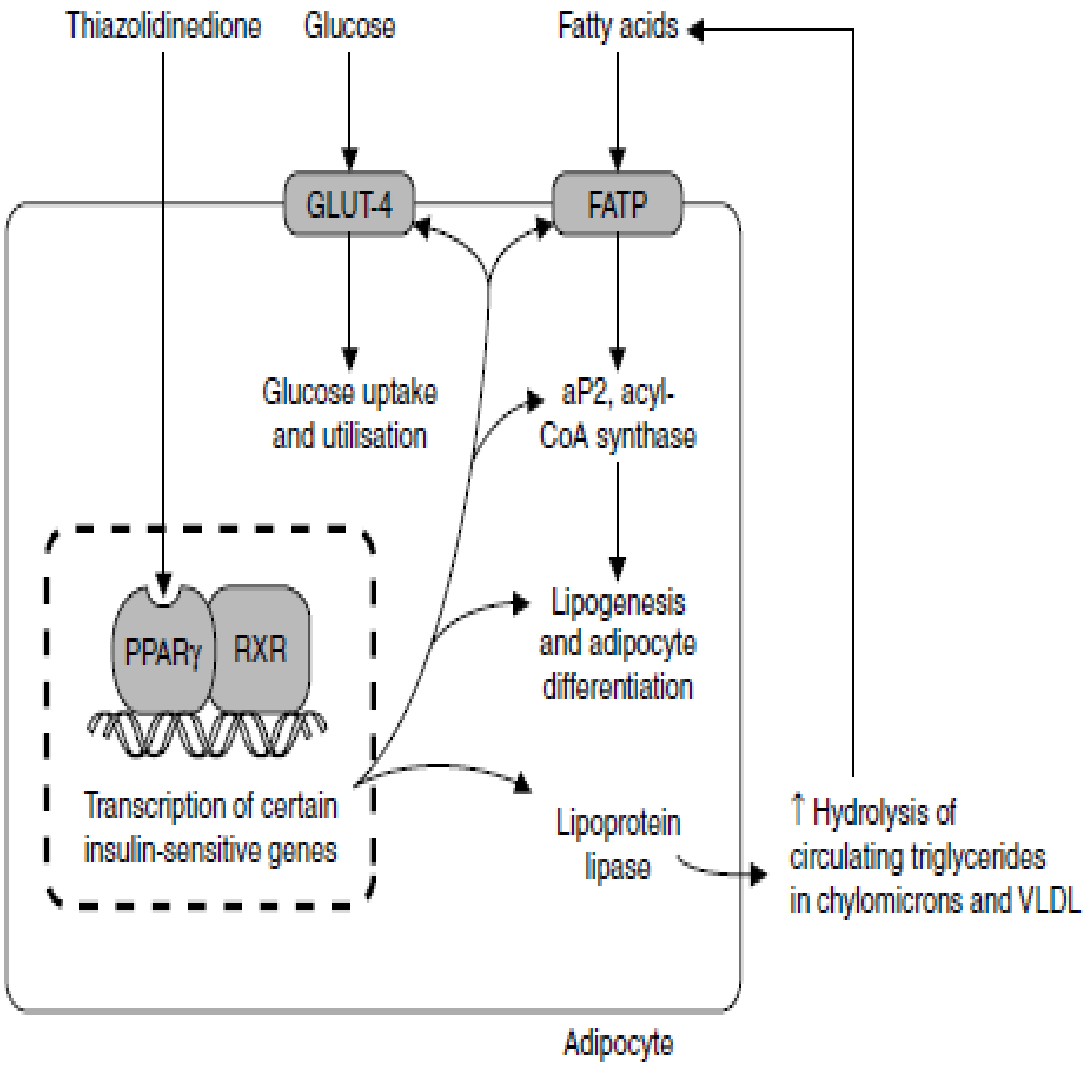

Figure 04: Mechanism of action of a thiazolidinedione on an adipocyte

aP2 = adipocyte fatty acid binding protein; CoA = coenzyme A;

FATP = fatty acid transporter protein; GLUT-4 = glucose transporter-4; PPARγ = peroxi-some proliferator-activated receptor-γ;

RXR = retinoid X receptor; VLDL = very low density lipoproteins;

↑ indicates increase.

2.19.4.5.* Pharmacokinetics*:

Rosiglitazone and pioglitazone are rapidly, and nearly completely absorbed (1–2 hours to peak concentration), although absorption is slightly delayed when taken with

food. Both agents are extensively metabolised by the liver. Rosiglitazone is metabolized mainly to very weakly active metabolites with lesser activity that are excreted predominantly in the urine. The metabolites of pioglitazone are more active and excreted mainly in the bile. Metabolism of rosiglitazone is undertaken mainly by cytochrome P450 (CYP) 2C8, which is not a widely activated isoform of CYP (Baldwin SJ et al 1999).

Thus, rosiglitazone does not interfere with the metabolism of other drugs. Pioglitazone is metabolised in part by CYP3A4 but, to date, no clinically significant reductions in plasma concentrations of other drugs (e.g.oral contraceptives) has been reported. Although both thiazolidinediones are almost completely bound to plasma proteins, their concentrations are low and have not been reported to interfere with other protein bound drugs.

2.19.4.6. **Indications and Contraindications:**

Now a day's rosiglitazone and pioglitazone are available for use as monotherapy in nonobese and obese patients with type 2 diabetes in whom diabetes is not adequately controlled by nonpharmacological measures. They can also be used in combination with various other antidiabetic drugs and in combination with insulin. In Europe, rosiglitazone and pioglitazone can be used as monotherapy if the patient is contraindicated for or intolerant of metformin. Thiazolidinediones can be used in combination with metformin or a sulphonylurea. In Europe, combination with insulin remains a contraindication to thiazolidinediones (Bailey CJ et al 2003).

Substituting a thiazolidinedione for either a sulphonylurea or metformin in patients with inadequate glycaemic control is generally of limited value and risks a temporary deterioration in glycaemic control because of the slow onset of action of thiazolidinediones. Having been disappointed with this experience, some UK diabetologists have elected to use thiazolidinediones in combination with both a sulphonylurea and Metformin (Bailey CJ et al 2003). The former strategy has met with variable success: some patients respond well, others show little response, requiring transfer to insulin.

The combination of thiazolidinedione plus insulin canimprove glycaemic control while reducing insulin dosages in obese patients, although peripheral oedema has been reported (Buch HN et al 2002). Rosiglitazone and pioglitazone can cause fluid retention

with increased plasma volume, a reduced haematocrit and a decrease in haemoglobin concentration. Therefore, the risk of oedema and anaemia should be taken into account, and in Europe, use of thiazolidinediones in patients with any evidence of congestive heart disease or heart failure is contraindicated. The choice of which patients to exclude on the basis of cardiac status varies between the product labelling sheets in the US and Europe. Consensus guidelines from the American Heart Association and the American Diabetes Association have recently been published (Nesto RW et al 2003).

Patients treated with a combination of insulin plus thiazolidinedione appear to be at highest risk of oedema, although the absolute rate of cardiac failure is low despite the fact the diabetes is a major risk factor for this complication (Nesto RW et al 2003). The guidelines urge a cautious approach and careful clinical monitoring, especially for patients likely to be at higher risk of cardiac failure. The haemogloblin concentration should be checked before starting a thiazolidinedione, bearing in mind that reductions of up to 1 g/dL in haemoglobin concentration may occur during therapy. No adverse effects on blood pressure have been noted with the thiazolidinediones, even with the increase in plasma volume; on the contrary, there is some evidence for a modest blood pressure lowering effect (Parulkar AA et al 2001).

As a precautionary measure, liver function should be assessed by measuring serum alanine aminotransferase test (ALT) before starting therapy and subsequently at 2-monthly intervals (or, in the US, as judged necessary by the prescribing clinician) during the first year of treatment; thereafter, periodic monitoring of liver function is prudent. Pre-existing liver disease, the development of clinical hepatic dysfunction or elevated ALT levels >2.5 times the upper limit for the laboratory serve as contraindications to thiazolidinediones. However, as mentioned earlier, hepatotoxicity has not been a concern with either rosiglitazone or pioglitazone. Isolated cases of nonfatal hepatocellular damage have been reported; however, the issue is clouded by reports suggesting an intrinsically higher risk of liver failure in patients with type 2 diabetes. Nevertheless, precautionary monitoring of liver function remains advisable. When initiating therapy with rosiglitazone or pioglitazone, blood glucose monitoring and titration of drug dosage should be undertaken while bearing in mind that thiazolidinediones exert a slowly generated anti-

hyperglycaemic effect that usually requires 2–3 months to reach maximum effect (Andrew J et al 2005).

According to the EU license, rosiglitazone can be given at a dosage of 4 mg/day in combination with a sulphonylurea, increasing to 8 mg/day (either once daily or in divided doses) in combination with metformin. Pioglitazone can be given as a once daily dosage of 15mg, increasing to 30mg if necessary (maximum 45mg in the US and Europe). The therapeutic response varies markedly between patients and it can be difficult to predict those most likely to respond. If no effect is observed after 3 months it is appropriate to consider the patient as a nonresponder and to stop the treatment (Andrew J et al 2005).

Rosiglitazone and pioglitazone can be used in the elderly, provided there are no contraindications. Both drugs may be used in patients with mild-to-moderate renal impairment, although the potential for oedema is a concern. In women with anovulatory PCOS the improvement in insulin sensitivity may cause ovulation to resume during thiazolidinedione therapy. A combination preparation containing rosiglitazone plus metformin (combining rosiglitazone/metformin in strengths 1mg/500mg, 2mg/500mg, 4mg/500mg, 2mg/1000mg, although not all strengths are available in all countries) (Andrew J et al 2005).

2.19.4.7. **Efficacy*:***

Addition of rosiglitazone or pioglitazone to the treatment schedule of patients whose glycaemic control with a sulphonylurea or metformin is suboptimal has consistently resulted in significant reductions in HbA1c. As judged by the available literature, these agents have similar glucose lowering effects, reducing HbA1c by around 0.5–1.5% (Yki J arvinen H 2004). However, the participants in these clinical trials had known diabetes of several years' duration, the effects of thiazolidinediones being more apparent when ß-cell function is less impaired. While earlier use of thiazolidinediones may be advantageous, the longer term picture requires clarification. Estimates of insulin sensitivity and ß-cell function (based on analysis of fasting glucose and insulin concentrations) have indicated that both defects can be improved by the addition of a thiazolidinedione (Yki J arvinen H 2004).

The effects on plasma lipids and apoproteins have been the subject of debate. Rosiglitazone can cause a small rise in the total cholesterol concentration, which stabilizes within about 3 months. This is accounted for by a rise in both the LDL cholesterol and the HDL-cholesterol, leaving the LDL**:** HDL-cholesterol ratio and the total HDL-cholesterol ratio little changed or slightly raised. Pioglitazone generally appears to have little effect on total cholesterol, and has been shown to reduce triglyceride concentrations in several studies. Both thiazolidinediones reduce the proportion of the smaller, more dense (more atherogenic) LDL particles (Yki J arvinen H 2004). To date, no prospective comparative studies of the two drugs have been reported and the clinical implications of these changes are uncertain (Parulkar AA et al 2001).

Weight gain, similar in magnitude to sulphonylurea therapy (typically 1–4kg) and stabilising over 6–12 months, has been observed during thiazolidinedione therapy. There is some evidence that the distribution of body fat is altered such that visceral adipose depots are little changed or reduced, while subcutaneous depots increase as new small, insulin sensitive adipocytes are formed. There are provisional data to suggest that thiazolidinediones exert arrange of effects on aspects of the metabolic syndrome that might reduce the risk of atherosclerotic cardiovascular disease (Parulkar AA et al 2001; Marten FMAC et al 2002). For example, thiazolidinediones have been reported to downregulate PAI-1 expression. Thiazolidinediones have also been reported to decrease urinary albumin excretion to a greater extent than expected for the improvement in glycaemic control and to reduce circulating markers of chronic low grade inflammation.

Preclinical studies suggesting that treatment of glucose intolerant animals with a thiazolidinedione preserved ß-cell function have yet to be confirmed in human studies. In insulin resistant women with a history of gestational diabetes at high risk of type 2 diabetes troglitazone reduced the incidence of new onset diabetes (Buchanan TA et al 2002). Whether thiazolidinediones will prove more effective than conventional antidiabetic agents in reducing the decline in ß-cell function in patients with established type 2 diabetes remains to be determined, although preliminary data in patients who respond to the drugs have been encouraging. Also of considerable interest are the clinical implications of the aforementioned effects of thiazolidinediones on risk factors for cardiovascular disease. These effects, allied to direct antiatherogenic actions reported in animal stu-

dies, are presently being studied in clinical trials with cardiovascular end points (Bell DSH 2002; Roberts AW et al 2003).

2.19.4.8. **Adverse Effects:**

Rosiglitazone and pioglitazone are generally well tolerated. As noted before caution is advised in heart disease; in the UK this includes a history of cardiac failure, oedema, anaemia and liver function requiring intermittent monitoring in accordance with the package labelling. If contraindications arise during treatment, monitoring should be intensified and, if necessary, treatment discontinued. Hypoglycaemia may occur several weeks after adding a thiazolidinedione to a sulphonylurea; selfmonitoring of blood glucose can be helpful in identifying the point at which the dosage of the sulphonylurea should be reduced (Andrew J et al 2005). Since PPARγ is expressed by many tissues, albeit at a low level, we must await the verdict of time for any unforeseen effects of long term stimulation with thiazolidinediones. For example, PPARγ activation in macrophages can reduce the production of some inflammatory cytokines and might increase transformation of monocytes to macrophages in the vascular wall. Stimulation of PPARγ in colon cells has been variously reported to increase and decrease division and differentiation of these cells in different animals and cell models; (Schoonjans K et al 2000) thus, familial polyposis coli is a contraindication to thiazolidinediones on theoretical grounds.

2.19.5. **Dipeptidyl peptidase-4 (DPP-4) inhibitors:**

Dipeptidyl peptidase-4 inhibitors (DPP-4s), also commonly called gliptins, are a relatively new class of drugs for the treatment of type 2 diabetes. These agents work in a unique way to improve insulin secretion from the β-cells of the pancreas in response to an increase in blood sugar and simultaneously decrease glucagon output from the a-cells of the pancreas, which results in decreased hepatic glucose output.

- Sitagliptin
- Vildagliptina
- Saxagliptin
- Linagliptin
- Alogliptin

2.19.5.4. **Mode of Action:**

A novel approach in treating diabetes mellitus is to utilize the physiological actions of endogenous incretin hormones; glucagon like peptide -1 (GLP-1) and glucose dependant insulinotropic peptide (GIP), the intestinal hormone; released in the response to nutrient ingestion. GIP is secreted by k cells from upper small intestine and GLP-1 is by L cells located in distal intestine when simulated by intraluminal glucose. Incretin effect is the augmentation of glucose stimulated insulin secretion by intestinally derives peptides, which are released in the presence of glucose in the gut. The observation that an oral glucose load was more effective at releasing insulin compared with the same amount of glucose given intravenously led to this theory. Studies prove that both GIP and GLP-1 stimulate release in glucose dependent manner in humans, controlling 50-70% of the prostprandial insulin response and both are necessary for maintenance of normal glucose tolerance.

Incretins stimulate insulin secretion only in presence of hypeglycemia with no response in normal or low glucose levels. Pathophysiology of diabetes mellitus also encompass a recently understood defect in incretin effect with decreased insulinotropic effect of GIP and decreased blood levels of GLP-1 in patients with diabetes mellitus. Although hyperinsulinomia is hallmark of the first year after diagnosis, the first phase insulin response (peak after a glucose load) is impaired or absent early in the disease. This first phase insulin response is caused by incretin secreted from the small intestine after glucose load. GIP and GLP -1 are rapidly inactivated by dipeptidyl peptidase 4 (DPP4), a member of serine peptidase family. DPP4 is ubiquitous in distribution. Tissues which strongly express DPP4 include the exocrine pancreas, kindey, GIT, bilary tract, thymus, lymph node, uterus, placenta, prostate, adrenal sweat gland, salivary and mammary glands. DPP4 is anchored to the plasma membrane of endotherlial of almost all organs examined and also found in the body fluids such as blood plasma ans cerebrospinalnal fluid. Different pharmacological strategies to enhance incretin effect in management of diabetes includes continuous administration of GLP-1, DPP4 resistant GLP-1 analogues and DPP4 inhibitors. Data from clinical trials for later approach seem to be promising. the serendipity of all presently available oral hypoglycemic compound, whose precise

mechanism of action have not been known at the time of their discovery, understanding the fact that each one of them have some clinically relevant side effects

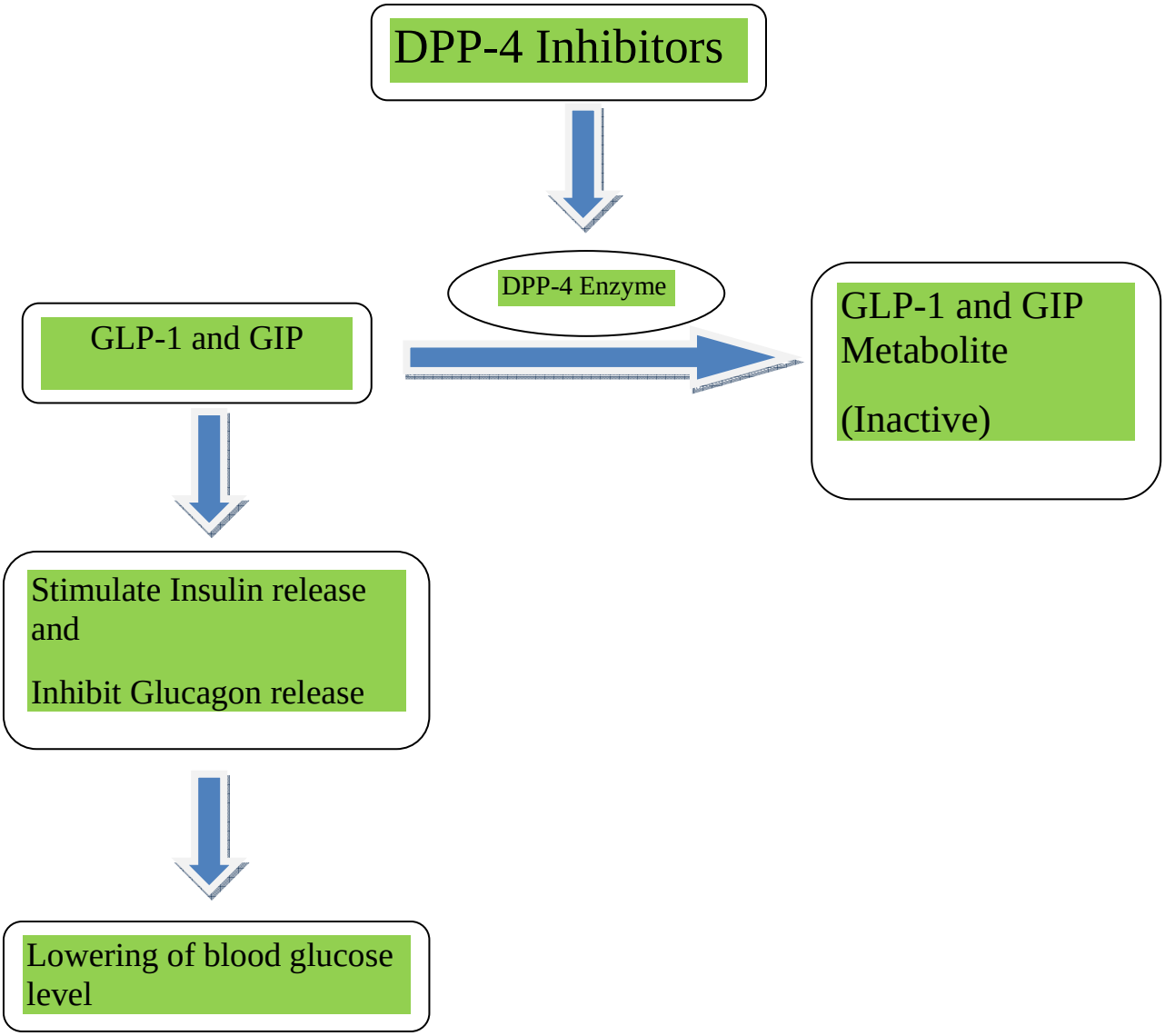

Figure 05: Dipeptidyl peptidase-4 (DPP-4) inhibitors

2.19.5.5. **Pharmacokinetics:**

The DPP-4 inhibitors are all orally available and are rapidly absorbed, with significant inhibition of plasma DPP-4 activity being seen within 5 min of administration. Oral biovailability in humans is generally high (~87% for sitagliptin (Herman GA et al 2007), 85% for vildagliptin (He YL et al 2007) and ~67% for saxagliptin (Fura A et al et al 2009), although somewhat lower for linagliptin (~30%) (Dittberner S et al 2010). Where available, data indicate that the volume of distribution of the various inhibitors in humans is greater than the total body water (~70 l for vildagliptin (EMEA 2010), 198 l for sitagliptin (Villhauer EB et al 2003), 300 l for alogliptin (Covington P et al 2008) and ~2.7 l/kg for saxagliptin (Fura A et al 2003)), suggesting that these compounds distribute widely into the tissues. In the plasma, most of the inhibitors display low, reversible protein binding 38% for sitagliptin (Herman GA et al 2007), 10% for vildagliptin (He H et al 2009; EMEA 2010) and negligible for saxagliptin (EMEA 2010). In contrast, linagliptin binds extensively to plasma proteins in a concentration dependent manner and it has been calculated that at the therapeutic dose (5 mg) most of the drug will be protein bound (primarily to DPP-4) (Fuchs H et al 2009).

Table 08: Pharmacokinetics of DPh4 Inhibitors:

Inhibitor	**Metabolism**	**Elimination route**
Sitagliptin	Not appreciably metabolized	Renal (~80% unchanged as parent)
Vildagliptin	Hydrolyzed to inactive metabolite (P450 enzyme independent)	Renal (22% as parent, 55% as primary metabolite)
Saxagliptin	Hepatically metabolized to active metabolite (via P450 3A4/5)	Renal (12–29% as parent, 21–52% as metabolite)
Alogliptin	Not appreciably metabolized	Renal (>70% unchanged as parent)
Linagliptin	Not appreciably metabolized	Biliary (>70% unchanged as parent);<6% via kidney

Sitagliptin, alogliptin and linagliptin do not undergo appreciable metabolism *in vivo* in humans; around 80% of the dose is eliminated unchanged as the parent compound. For sitagliptin, the limited metabolism produces six metabolites in trace amounts (each accounting for <1% to 7% of sitagliptin related material in plasma), with *in vitro* studies indicating that the primary enzyme responsible is CYP3A4 with a lesser contribution from CYP2C8 (Vincent SH et al 2007). Three of these metabolites (M1, M2 and M5) are active, but are not expected to contribute to the pharmacodynamic profile of sitagliptin because of the combination of low plasma concentration and low affinity for DPP-4 (Vincent SH et al 2007; EMEA 2010). For alogliptin, the parent molecule accounts for >80% of alogliptin related material in plasma and two minor metabolites have been identified, N-demethylated (active) and N-acetylated (inactive) alogliptin, accounting for less than 1% and approximately 5%, respectively (Karim A et al 2007). In the case of linagliptin, the parent compound made up around 70% of drug related material in plasma, while exposure to the major metabolite (CD1790, identified as *S*-3-hydroxypiperidinyl derivative of linagliptin) was around 18% of that of the parent compound. Formation of CD1790, which is pharmacologically inactive, is dependent upon CYP3A4. In addition, seven minor metabolites (each accounting for 0.3 to <5% of linagliptinrelated material in plasma) were identified (Blech S et al 2010). In contrast, both vildagliptin and saxagliptin undergo extensive metabolism in humans.

The major metabolic pathway for vildagliptin is hydrolysis at its cyano moiety, which occurs in the liver and other tissues via aCYP independent mechanism, to produce a carboxylic acid metabolite (M20.7/LAY151) and four minor metabolites. The parent molecule and the major metabolite, which is pharmacologically inactive, account for the majority of vildagliptin related material in the plasma (approximately 22 and 55%, espectively) (He H et al 2009; EMEA 2010). Saxagliptin is hepatically metabolized by CYP 3A4/5 to produce a major metabolite (5hydroxy saxagliptin; BMS-510849), which is also a competitive, reversible inhibitor of DPP-4 with approximately 50% of the potency of the parent drug. Systemic exposure to saxagliptin related material is accounted for by the parent molecule (22%) and BMS-510849 plus other unidentified minor monohydroxylated metabolites (76%) (EMEA 2010). Generally, the DPP-4 inhibitors are eliminated primarily via the kidney, with the rate of renal clearance exceeding glomerular filtration,

suggesting that active transport is involved. For sitagliptin, around 70% of the dose is excreted as the parent molecule and active transport has been shown to account for around 50% of its clearance (Herman GA et al 2005); the human organic anion transporter (OAT)-3, organic anion transporting polypeptide (OATP)-4C1 and Pgp transporters in the proximal tubule have been indicated to be involved (Chu XY et al 2007). Alogliptin (and its minor metabolites) is renally eliminated, with around 60–70% of the dose appearing in the urine as the parent compound (Covington P et al 2008; Christopher R et al 2008).

Clearance of alogliptin is greater than normal glomerular filtration, but the renal transporters involved have not been identified, although drug interaction studies suggest that Pgp is unlikely to be involved (Covington P et al 2008). Similarly, both saxagliptin and its primary metabolite (BMS-510849) are primarily renally eliminated, accounting for 24 and 36% of the dose, respectively (EMEA 2010). Again, renal clearance of the parent compound is greater than the glomerular filtration rate, indicating the involvement of active renal secretion, but the mechanism is unknown; saxagliptin is reported not to be a substrate for OAT1, OAT3, OATPA, OATPC, OATP8,organiccationtransport (OCT)-1, OCT2, sodium taurocholate co-transporting peptide or peptide transporters (PepT1 and PepT2) (EMEA 2010). In contrast, clearance of BMS-510849 is similar to the glomerular filtration, suggesting that this is a main mechanism involved in its elimination (EMEA 2010). Data for vildagliptin also indicate the kidneys to be the predominant route of elimination, with 22% of the dose appearing in the urine unchanged and 50% appearing as the major metabolite (M20.7); active transport in addition to glomerular filtration was indicated to be involved in the elimination of both compounds (He H et al 2009). Linagliptin is the exception, with <6% of the dose being excreted in the urine (Heise T et al 2009). This may be, at least in part, because of the high degree of protein binding (Fuchs H et al 2009), meaning that the drug escapes glomerular filtration. Rather, linagliptin has a hepatic route of elimination, with 78% appearing in the faeces unchanged. Renal excretion of the primary metabolite (CD1790) is negligible; this undergoes further metabolism and is also eliminated in the feces (Blech S et al 2010).

2.19.5.6. Indications and Contraindications:

In general the DPP-4 inhibitors have not been reported to result in any meaningful activation or inhibition of the CYP enzyme system, suggesting that they are unlikely to be involved in clinically meaningful drug interactions involving these systems. There are data suggesting that there is no great propensity for the DPP-4 inhibitors to be involved in any clinically relevant drug–drug interactions with other commonly prescribed medications (EMEA 2010(a); EMEA 2010(b), EMEA 2010(c)), including metformin (Herman GA et al 2006; He YL et al 2009; Karim A et al 2010; Graefe Mody EU et al 2009), pioglitazone (Serra D et al 2008; Karim A et al 2009; Graefe Mody EU et al 2009), rosiglitazone (Graefe Mody EU et al 2009), glyburide (Serra D et al 2008; Karim A et al 2009; Mistry GC et al 2008) and simvastatin (Ayalasomayajula SP et al 2007; Bergman AJ et al 2009; Graefe Mody U et al 2010), suggesting that these agents can be co-administered with the DPP-4 inhibitors without the need for dose adjustment of either drug. As mentioned, CYP3A4/5 is involved in the conversion of saxagliptin to the active metabolite (BMS-510849), and strong inhibitors of CYP3A4/5, such as ketoconazole, increase the exposure to the parent compound. For this reason, dose reduction by half (2.5 mg qd) is recommended when saxagliptin is co administered with strong CYP3A4/5 inhibitors (OPI 2010). Linagliptin is also a substrate for CYP3A4, and ketoconazole prevents the generation of the metabolite, CD1790. However, because this is of only minor importance in the clearance of linagliptin, inhibition or induction of CYP3A4 by concomitantly administered drugs was not considered likely to alter the overall exposure to linagliptin (Blech S et al 2010). Additionally, linagliptin has been identified as a weak competitive and a poor tomoderate mechanism based inhibitor of CYP3A4, resulting in a decrease in the clearance of other compounds metabolized by this pathway by less than twofold; linagliptin was therefore considered to have only a weak potential for clinically relevant interactions with drugs metabolized by this system (Blech S et al 2010).

2.19.5.7. **Efficacy:**

As might be expected from their similar efficacy in inhibiting DPP-4 activity broadly speaking, the DPP-4 inhibitors all seem to show similar efficacy in lowering HbA1c levels, although it must be stressed that these are observations made in different

studies and so must be interpreted with some caution. At present, data are available only from one direct head-to-head comparison between the inhibitors, in which the efficacy of saxagliptin and sitagliptin as addon therapy in Metformin treated patients was compared (Scheen AJ et al 2010). This showed non inferiority of saxagliptin to sitagliptin in terms of HbA1c lowering (−0.5 vs. −0.6% from a baseline of ∼7.7%; i.e. from 60 to 55 mmol/mol for saxagliptin vs. sitagliptin from 61 to 54 mmol/mol for sitagliptin) at week 18, with similar proportions of subjects (26 vs. 29%) reaching target HbA1c levels of <6.5% (<48 mmol/mol). However, in terms of the reduction in fasting plasma glucose, it did appear that there might be a small difference, with sitagliptin being more efficacious (−0.6 vs. −0.9 mmol/l; difference 0.30±0.115 mmol/l, 95% confidence interval: 0.08–0.53); this could potentially be related to differences in the half life of the compounds. In other direct head-to-head studies, the DPP-4 inhibitors have shown similar efficacy to metformin (Aschner P et al 2010; Schweizer A et al 2009), the sulphonylureas (Ferrannini E et al 2009; Nauck MA et al 2007), the glitazones (Bolli G et al 2008; Scott R et al 2008) and the alpha-glucosidase inhibitors (Pan C et al 2008). In line with other antidiabetic agents (Bloomagarden ZT et al 2007), greater reductions in HbA1c are seen in subjects with higher baseline levels.

Although the described DPP-4 inhibitors are all competitive reversible inhibitors, it can be difficult to compare them using data reported in individual studies, because these are influenced by differences in the assay conditions used to estimate the extent of DPP-4 inhibition. However, one study in which the inhibitors were directly compared under identical experimental conditions reported that all five inhibitors showed similar efficacy (i.e. maximal effect) for inhibition of DPP-4 in vitro, but that there were differences in potency (i.e. amount of compound needed; IC50 = ∼1 nM for linagliptin vs. 19, 62, 50 and 24 nM, for sitagliptin, vildagliptin, saxagliptin and alogliptin, respectively) (Thomas L et al 2008). With regard to half-life, there are also differences between the various inhibitors. Vildagliptin (EMEA 2010(b); He YL et al 2007) and saxagliptin (EMEA 2010(c); Boulton DW et al 2007) are cleared from the plasma relatively quickly, whereas sitagliptin (EMEA 2010(a); Bergman AJ et al 2006), alogliptin (Convington P et al 2008) and linagliptin (Heise T et al 2009) have much longer survival times. These differences are reflected in the therapeutic doses, which range from 5 mg for saxagliptin to

100 mg for sitagliptin, and in the dosing frequency (once daily for most of them, twice daily for vildagliptin. Nevertheless, despite the differences in potency, when used at their therapeutic doses, the effects of the inhibitors, in terms of the extent of DPP-4 inhibition in vivo, are broadly similar. Over 90% inhibition is attained within 15 min of inhibitor administration, with around 70–90% inhibition being sustained at 24 h postdose for vildagliptin, although the extent of plasma DPP-4 inhibition drops to around 50% after 24 h with the 50 mg dose, the twice daily therapeutic dosing regimen maintains plasma DPP-4 inhibition at >80% over the full 24-h period. However, it should be pointed out that plasma DPP-4 activity is assessed ex vivo (i.e. in plasma samples taken after in vivo dosing) and is generally not corrected for the inherent dilution of the sample in the assay. Hence, the true extent of DPP-4 inhibition in vivo is probably higher than the measured values suggest.

Table 09: Efficacy of DPP-4 inhibitors:

Inhibitor	**Compound t 1/2 h**	**1/2 (h) Dosing**	**DPP-4 inhibition∗**
Sitagliptin	8–24	100 mg qd	Max~97%;>80% 24 h postdose
Vildagliptin	11/2–41/2	50 mg bid	Max~95%;>80% 12 h postdose
Saxagliptin	2–4 (parent) 3–7 (metabolite)	5 mg qd	Max~80%;~70% 24 h postdose
Alogliptin	12–21	25 mg qd	Max~90%;~75% 24 h postdose
Linagliptin	10–40	5 mg qd (anticipated)	Max~80%;~70% 24 h postdo

2.19.5.8. **Adverse Effects:**

Vildagliptin and saxagliptin, but not sitagliptin or alogliptin, were reported to be associated with adverse skin toxicology in monkeys. However, this may be a finding which is specific to monkeys, as it has not been observed in other preclinical species, and

importantly, there have been no reports of skin problems in the clinical trials with any of the inhibitors (EMEA 2010(b); EMEA 2010(c); Williams Herman D et al 2010; Ligueros Saylan M et al 2010; Sato K et al 2008). For Saxagliptin, small, reversible, dose dependent reductions in absolute lymphocyte count have been noted in some of the clinical trials, but this has not been reported for the other DPP-4 inhibitors. The effect was more apparent at saxagliptin doses =20 mg (which is greater than the therapeutic dose), but values still remained within normal limits (EMEA 2010(c); Rosenstock J et al 2008).

There was no effect on white blood cell or neutrophil count and no evidence of altered immune function. At present, the clinical significance of this (if any) remains unknown. At the time of initial registration of vildagliptin (in EU), a Meta analysis of the clinical trial data revealed that the 100 mg qd dose was associated with small numerical elevations in liver transaminases compared to placebo or 50 mg bid. For this reason, the recommended therapeutic dose was changed to 50 mgbid, with the recommendation that liver function tests be performed before initiation and at three monthly intervals for the first year of treatment and periodically thereafter (EMEA 2010(b); Press Release 2008). Subsequently, the trend for mild increases (greater than three times the upper limit of normal) in liver enzymes was confirmed in the larger pooled safety analysis, but notably, this was not associated with any increased incidence of actual hepatic adverse events (Ligueros Saylan M et al 2010). Nevertheless, liver function tests are still recommended and vildagliptin is not approved for use in patients with hepatic insufficiency. Despite the above observations, overall, the DPP-4 inhibitors as a class appear to be very well tolerated, and rates of adverse effects have been low, and generally not different to placebo or comparator.

An early meta analysis of incretinbased therapies (in which inhibitor data were available only for sitagliptin and vildagliptin) did, however, suggest that there was an increased risk of some infections (urinary tract infections with both inhibitors and nasopharyngitis more evident with sitagliptin) and headache (more evident with vildagliptin) (Amori RE et al 2007; Richter B et al 2008). Since then, updated safety analyses (each >10 000 patients, exposed for up to 2 years) of the sitagliptin and vildagliptin clinical trials have been published, showing no increased risk for urinary tract or respiratory infections or headache (and indeed, no increased risk of any other adverse effect) with the

DPP-4 inhibitors compared to placebo or comparator (Williams Herman D et al 2010; Ligueros Saylan M et al 2010). Notably, recent debate over potential links between some antidiabetic medications and cancer (Pocock SJ et al 2009) or bone fracture (Bodmer M et al 2009) does not seem to extend to the DPP-4 inhibitors, with no evidence for increased signals being observed in the safety analyses (Williams-Herman D et al 2010; Ligueros Saylan M et al 2010). Cardiovascular safety of new drugs, including antihyperglycaemic agents, has also been the focus of concern, with the FDA requiring pharmaceutical companies to show that new agents to not increase the risk of adverse cardiovascular events. Retrospective analyses of data from the clinical development programmes of sitagliptin, vildagliptin and saxagliptin do not appear to indicate any increased cardiovascular risk with the DPP-4 inhibitors relative to alogliptin on cardiovascular outcomes are underway. There has also been some debate over whether incretin based therapies, including the DPP-4 inhibitors, are associated with elevated risk of pancreatitis (Butler PC et al 2010). This does not seem to be borne out by the pooled safety analyses (Ligueros Saylan M et al 2010; Engel SS et al 2010) or retrospective analyses of large healthcare data bases (Dore DD et al 2009; Herrera V et al 2009). Continued vigilance and longer term reports are still needed to confirm these observations.

2.19.6. Alpha Glucosidase Inhibitors:

Inhibitors of intestinal α-glucosidase enzymes retard the rate of carbohydrate digestion, thereby providing an alternative means to reduce postprandial hyperglycemia (Lebovitz HE 1998).

Acarbose has been to reduce the risk of myocardial infarction and other cardiovascular events in meta analysis (Hanefeld, M. et al 2004). Miglitol has an adiponectin-dependent anti-atherothrombotic effect that may be beneficial for primary prevention of atherothrombosis in patients with type 2 diabetes (Shosaku Nomura et al 2011).

2.19.6.4. Mode of Action of Alpha Glucosidase Inhibitors:

The α-glucosidase inhibitors competitively inhibit the activity of α-glucosidase enzymes in the brush border of enterocytes lining the intestinal villi. High affinity binding prevents these enzymes from cleaving their normal disaccharide and oligosaccharide

substrates into monosaccharides prior to absorption. This defers the completion of carbohydrate digestion until further along the intestinal tract, in turn causing glucose absorption to be delayed. The α-glucosidase inhibitors should be taken with meals containing digestible carbohydrates, not monosaccharides; these drugs generally do not significantly affect the absorption of glucose. Since α-glucosidase inhibitors move glucose absorption more distally along the intestinal tract they alter glucose dependent release of intestinal hormones that enhance nutrient induced insulin secretion. Release of gastric inhibitory polypeptide, which occurs mainly from the jejunal mucosa, may be reduced by α-glucosidase inhibitors, whereas glucagon like peptide-1 (7–36 amide) secretion (mostly from the ileal mucosa) is increased. Overall, α-glucosidase inhibitors reduce postprandial insulin concentrations through the attenuated rise in postprandial glucose levels (Lebovitz HE 1998).

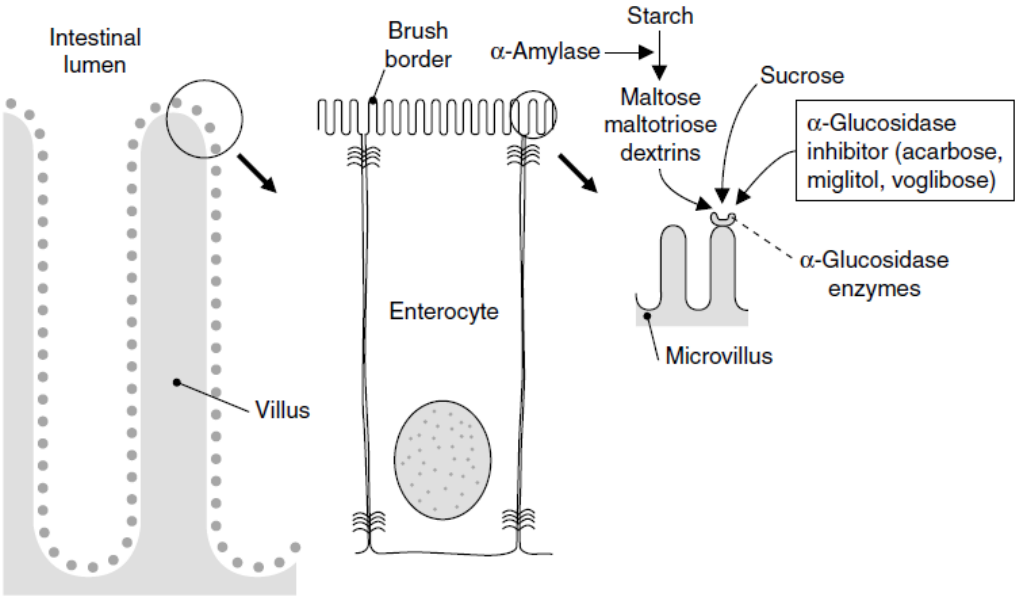

Figure no 06: Mechanism of action of alpha glucosidase inhibitor.

Secondary metabolic effects:

In addition to reducing the postprandial hyperglycemia these drugs also reduce the fasting BSL and insulin resistance. An increase in glucagon like peptide 1 (GLP-1) are thought to contribute to these actions α-glucosidase inhibition has also been associated with a significant improvement in lipid metabolism and may blunt postprandial increases in serum triglyceride levels (Hanefeld M et al 2004).

Effect on body weight:

Clinical studies show that α-glucosidase inhibitors, unlike sulphonylureas and insulin, do not cause weight gain because of decrease in postprandial insulin concentration (Hanefeld M et al 2004).

2.19.6.5. Indications and Contraindications:

An α-glucosidase inhibitor may be used as monotherapy for patients with type 2 diabetes that is inadequately controlled by nonpharmacological measures. Because α-glucosidase inhibitors target postprandial hyperglycaemia, they can be a useful firstline treatment in patients who have a combination of only slightly raised basal glucose concentrations and more marked postprandial hyperglycaemia. A recent multicentre clinical trial (STOP NIDDM (Study TO Prevent NonInsulin Dependent Diabetes Mellitus)) confirmed the utility of acarbose in preventing the transition from impaired glucose tolerance to diabetes (Chiasson J L et al 2002). Acarbose can be used in combination with other antidiabetic agents. When starting therapy with a α-glucosidase inhibitor it is said to be important to ensure that the patient is taking a diet rich in complex carbohydrates, as opposed to simple sugars. Acarbose should be taken with meals, starting with a low dose, for example 50 mg/day, and slowly titrating up over several weeks. Monitoring of glycaemic control, particularly postprandially, may be helpful.

The postprandial action of these agents would not be expected to induce hypoglycaemia, at least when they are used as monotherapy. The maximum dosage of α-glucosidase inhibitors may be limited by gastrointestinal symptoms; this is certainly our experience with acarbose. Intuitively, patients experiencing gastrointestinal adverse effects with metformin may not be the best candidates in whom to add an α-glucosidase inhibitor. A history of chronic intestinal disease serves as a largely theoretical contraindi-

cation to acarbose and other agents in this class. High dosages of acarbose can occasionally increase liver enzyme concentrations, and it is recommended that transaminase concentrations are measured at intervals in patients receiving the maximum dosage (200mg three times daily in the UK, a dosage rarely attained in practice for the aforementioned reasons). If liver enzymes are raised, the dosage of acarbose should be reduced 2.5 to a level at which normal enzyme concentrations are reestablished. Alternative causes of hepatic dysfunction should be considered. Pregnancy and breast feeding are traditionally regarded to be contraindications for all oral antidiabetic drugs, mainly because of a lack of safety data rather than evidence of detrimental effects.

2.19.6.6. Efficacy:

An α-glucosidase inhibitor can reduce peak concentrations of blood glucose and reduce interprandial troughs. Used as monotherapy to patients who comply appropriately with dietary advice, an α- glucosidase inhibitor will typically reduce postprandial glucose concentrations by 1–4 mmol/L. The incremental area under the postprandial plasma glucose curve can be more than halved in some individuals. There seems to be a 'carry over' effect that may produce a reduction in basal glycaemia up to 1 mmol/L. The decrease in HbA1c is usually about 0.5–1.0%, provided that a high dose of the drug is tolerated and dietary compliance is maintained (Holman RR et al 199). There may be a trivial alteration in the gastrointestinal absorption of other oral antidiabetic agents when used in combination therapy. In general the extra benefit to glycaemic control achieved by addition of an α-glucosidase inhibitor to another antidiabetic agent is additive. In the recently published multicentre STOP-NIDDM trial acarbose reduced the risk of progression from impaired glucose tolerance to type 2 diabetes (relative hazard 0.75; 95% CI 0.63, 0.90; p= 0.0015). This study randomised 1429 patients with impaired glucose tolerance to acarbose 100mg three times daily or placebo, of whom data were available for a modified intention-to-treat analysis in 1368 patients. Glucose tolerance was determined using a 75g oral glucose tolerance test. Intriguingly, new cases of hypertension and major cardiac events, including overt and clinically silent myocardial infarction, were also reduced by acarbose therapy (Chiasson J L et al 2003). The latter were not primary endpoints of the study, a limitation acknowledged by the investigators (Chiasson J L et al 2003). The re-

sults of ongoing trials using acarbose and other agents in this class are awaited (Scheen A 2003)

2.19.6.7. Pharmacokinetics:

In a study of 6 healthy men 2% of an oral dose of acarbose was absorbed as active drugs, while approximately 35% of total radioactivity from a C14 labeled oral dose was absorbed. An average of 51% of an oral dose was excreted in the feces as unabsorbed drug related radioactivity within 96 hrs of ingestion (US patent no. 6,387,361B1). Because acarbose acts locally within the gastrointestinal tract, this low systemic bioavailabilty of parent compound is therapeutically desired following oral dose of healthy volunteers with C-14 labeled acarbose, peak plasma concentration of radioactivity were attained 14-24 hrs after dosing, while peak plasma concentration of active drugs were attainted approximately 1hr. the delayed absorption of acarbose related radioactivity reflects the absorption of metabolites that may be formed by either intestinal enzymatic hydrolysis (US patent no. 6,387,361B1).

Acarbose is metabolized exclusively within the GIT, principally by intestinal bacteria, but also by digestive enzyme, a fraction of these metabolites was absorbed and subsequently excreted in the urine. at least 13 metabolites have been separated chromatographically from urine specimens. the major metabolite have been identified as 4- methylpyrogallol derivatives (that is sulphate, methyl and glucuronide conjugates) (US patent no. 6,387, 361B1). One metabolites (formed by cleavage of a glucose molecule from acarbose) also has alpha glucosidase inhibitory activity, this metabolite together with parent compound from urine account for less than 2% of the total administration dose (US patent no. 6,387,361B1). The fraction of acarbose that is absorbed as intact drug is almost completely excreted by the kidney. When acarbose was given intravenously 89% of the dose was recovered in the urine as active drug within 48 hrs. In contrast to, less than 2% of an oral dose was recovered in the urine as active drug. this is consistent with low bioavailability of parent drugs. the plasma elimination half life of acarbose activity is approximately 2 hrs in healthy volunteers. Consequently, drugs accumulation dose not occur with three times a day oral dosing (US patent no. 6,387,361B1).

Table 10: Pharmacokinetics of α-Glucosidase Inhibitor (Zargar AH et al 2002; Fuhlendorff J et al 1998; Ambavane V et al 2000)

Drugs	tmax (hrs)	t1/2 (hrs)	Protein Binding (%)	Dose (mg)	Excretion
Acarbose	1	2	-	25-100 mg TDS	Renal < 2 faeces – 51
Miglitol	2 – 3	2	< 4 %	25-100 mg TDS	Renal – 95
Voglibose	1 – 1.5	1.5	-	0.2 – 0.3mg TDS	Renal <2

Table 11: Mean Plecebo Subtracted Change in HbA1c in the Fixed Dose Monotherapy Studies (Acarbose) (US patent no. 6,387,361B1)

Dose of Acrbose	N	Changwe in **HbA1c %**	p- valie
25 mg tid	110	0.44	0.0307
50 mg tid	131	0.77	0.0001
100mg tid	244	0.74	0.0001
200 mg tid	231	0.86	0.0001
300 mg	53	1.00	0.0001

Table 12: Mean Plecebo- Subtracted Change in HbA1c in the Fixed Dose Monotherapy Studies

Study	Treatment	HbA1c %			p-value
		Mean Baseline	Mean change from baseline	Treatment difference	
1	Placebo Plus Diet	8.67	+0.33	----	-----
	Acarbose 100mg tid plus Diet	8.69	-0.45	-0.78	0.0001
2	Placebo Plus SFU	9.56	+0.24	------	-----
	Acarbose 50-300mg tid Plus SFU	9.64	-0.3.	-0.54	0.0096
3	Plecebo Plus Metformin	8.17	+0.08	-----	----
	Acarbose 50-100mg tid Plus Metformin	8.46	-0.57	-0.65	0.0001
4	Plecebo Plus Insulin	8.69	+0.11	---	---
	Acarbose 50-100mg tid Plus Insulin	8.77	-0.58	-0.69	0.0001

Absorption of miglitol is saturable at high doses: a dose of 25 mg is completely absorbed, whereas a dose of 100 mg is only 50% - 70% absorbed. For all doses, peak concentrations are reached in 2-3 hours (U.S. Patent No. 4,639,436). There is no evidence that systemic absorption of miglitol contributes to its therapeutic effect (U.S. Patent No. 4,639,436).

The protein binding of miglitol is negligible (<4.0%). Miglitol has a volume of distribution of 0.18 L/kg, consistent with distribution primarily into the extracellular fluid (U.S. Patent No. 4,639,436).

Miglitol is not metabolized in man or in any animal species studied. No metabolites have been detected in plasma, urine, or feces, indicating a lack of either systemic or presystemic metabolism (U.S. Patent No. 4,639,436).

Miglitol is eliminated by renal excretion as unchanged drug. Thus, following a 25-mg dose, over 95% of the dose is recovered in the urine within 24 hours. At higher doses, the cumulative recovery of drug from urine is somewhat lower due to the incomplete bioavailability. The elimination half life of miglitol from plasma is approximately 2 hours (U.S. Patent No. 4,639,436).

Because miglitol is excreted primarily by the kidneys, accumulation of miglitol is expected in patients with renal impairment. Patients with creatinine clearance <25 mL/min taking 25 mg 3 times daily exhibited a greater than two fold increase in miglitol plasma levels as compared to subjects with creatinine clearance >60 mL/min. Dosage adjustment to correct the increased plasma concentrations is not feasible because miglitol acts locally. Little information is available on the safety of miglitol in patients with creatinine clearance <25 mL/min (U.S. Patent No. 4,639,436).

Miglitol pharmacokinetics were not altered in cirrhotic patients relative to healthy control subjects. Since miglitol is not metabolized, no influence of hepatic function on the kinetics of miglitol is expected (U.S. Patent No. 4,639,436). No significant difference in the pharmacokinetics of miglitol was observed between elderly men and women when body weight was taken into account (U.S. Patent No. 4,639,436). Several pharmacokinetic studies were conducted in Japanese volunteers, with results similar to those observed in Caucasians. A study comparing the pharmacodynamic response to a single 50-mg dose in Black and Caucasian healthy volunteers indicated similar glucose and insulin responses in both populations (U.S. Patent No. 4,639,436).

Voglibose is poorly absorbed after oral doses. Plasma concentrations after oral doses have usually been undetectable. After an 80 mg dose (substantially higher than recommended dose), peak plasma levels of about 20 ng/mL were observed in 1 to 1.5 hours. When Voglibose tablets were repeatedly administered to healthy male adults (6 subjects) in a single dose of 0.2 mg, 3 times a day, for 7 consecutive days, Voglibose was not detected in plasma or urine. Similarly, when Voglibose was administered to healthy male adults (10 subjects) as a single dose of 2 mg, Voglibose was not detected in plasma or urine. After ingestion of Voglibose (and other glucosidase inhibitors), the majority of active unchanged drug remains in the lumen of the gastrointestinal tract to exert its pharma-

cological activity. Voglibose is metabolized by intestinal enzymes and by the microbial flora. Voglibose is excreted in the urine and feces.

Table 13: Alpha Glucosidase Inhibitors Pharmacokinetic Data

Drug	Bioavailability	Half- Life (hr)	Metabolites	Excretion
Acarbose	Less than 2%	2	13; one active with less than 2% of parent activity	Almost completely renal
Miglitol	1005 at 25 mg; 50-70% at 100 mg	2	None	renal> 95% at 25 mg; Renal > 95% at higher doses
Voglibose		1-1.5	Voglibose is metabolized by intestinal enzymes and by the microbial flora.	Voglibose is excreted in the urine and feces.

2.19.6.8. Adverse Effects:

The most common problems with α-glucosidase inhibitors are gastrointestinal adverse effects. In the STOP-NIDDM trial 31% of acarbose treated patients compared with 19% on placebo discontinued treatment early (Chiasson JL et al 2002) If the dosage is too high (relative to the amount of complex carbohydrate in the meal), undigested oligosaccharides pass into the large bowel (Krentz AJ et al 1994) Carbohydrates fermented by the flora of the large bowel cause flatulence, abdominal discomfort and sometimes diarrhoea. This is most likely to occur during the initial titration of the drug and can sometimes be minimised by slow titration and by ensuring dietary compliance with meals rich in complex carbohydrate. In some patients the gastrointestinal symptoms may gradually subside with time, suggesting an adaptive response within the gastrointestinal tract. Hypoglycaemia is only likely to be encountered when an α-glucosidase inhibitor is used in combination with a sulphonylurea or insulin (Krentz AJ et al 1994). No clinically significant drug

interations have been reported. However, agents affecting gut motility can potentially influence the efficacy and gastrointestinal effects of acarbose; cholestyramine may increase the glucose lowering effect of acarbose.

2.19.6.9. **Clinical studies of acarbose** (Adiyodi JR 1999)**:**

1) In a multicentric trial conducted by the department of clinical pharmacology, Chinese university of Hongkong, Prince of Wales hospital, Shatin china, found that Acarbose 100 mg t.i.d.is an effective, safe and well tolerated. There were greater reduction of mean HbA1c (-0.70[-1.0 to -0.39] vs -0.27[-0.54 to 0]; p=0.04), fasting plasma glucose (-0.37[-0.75 to 0.02] vs 0.41mmo/l [0.08 to 0.90] p=0.017), postprandial plasma glucose (-0.77[-1.44to-1.0] vs 0.65mmol/l[-0.07 to 1.36]; p=0.05) in the Acarbose group as compared to placebo.

2) A study conducted by Unidad de Diabetes y nutricion, faculted de medicinia university, patient received Acarbose 150mg/day which was increased to 300mg/day during 3 months found that HbA1C decreased from 8.36±1.33 to 7.71±1.7% ($p<0.001$), fasting blood glucose decreased from 173±48 to 159±59mg/dl ($p<0.03$) and postprandial blood glucose decreased from 254±80 to 241±80 mg/dl. Fifty nine patients had developed gastrointestinal symptoms that were mild in 59% and moderate in 39%.

3) A study conducted by the university of Pittsburgh School of medicine, Division of Endocrinology, PA,USA, in which patient received Acarbose 25mg TDS to 100mg TDS or placebo in which BSL was not controlled with insulin and diet shown that there was reduction in mean HbA1C of 0.69% compared with placebo.

2.19.6.10. Clinical Studies of Miglitol (Stephen N et al 2006; Mitrakou A et al 1998; R quejo F et al 1990; Temelkova K et al 2000):

1) Clinical experience in NIDDM patients on diet only. Miglitol was evaluated in 2 U.S. and 3 non US controlled fixed dose monotherapy studies in which 735 patients treated with Miglitol were evaluated for efficacy analysis.

2) Clinical experience in NIDDM patients receiving Sulphonylureas. Miglitol was studied as adjunctive therapy to background of maximal or near maximal Sulfonylurea

treatment in three large double blind, randomized studies (Two U.S. and one non U.S.) in which 471 patients treated with Miglitol were evaluated for efficacy.

3) An observational study was carried out from October 1999 to August 2000 by H.C. Fehmann. 2654 patients with Type 2 D.M. were treated for 3 month with Miglitol which shows that HbA1C reduced from 8.4% to 7.1%. Pre and postprandial glucose level reduced minus 49 mg/dl and minus 59 mg/dl. The average weight reduced is 1.9 kg. ADR reported only in 47 patients mainly in GI Tract.

4) A double blind randomized placebo controlled trial of Miglitol 25 mg TDS & 50 mg TDS compared with placebo and titrated dose of glyburide was conducted in 30 OPD patient across USA, shows that treatment with Miglitol offers significant reduction in HbA1C as compared to Glyburide.

2.19.6.11. Clinical studies of Voglibose:

1) Maksumoto et al 1998 investigated the effect of Voglibose on daily glycemic excursion, insulin secretion and insulin sensitivity in NIDDM ; shown that HbA1C and plasma glucose were in patients who received Voglibose were comparable to those of patients in control group(5.7 ± 0.9 vs 9.8 ± 1.2 $p<0.05$) (Matsumoto K et al 1998).

2) Saino et al 1998 studied Voglibose (0.6mg TID) in combination with sulfonylurea in multicentric trial and shown that there was significant reduction of fasting plasma glucose, postprandial glucose and HbA1C as compare to baseline (Saino N et al 1998).

3) Fujisawa et al 2005 compared the Voglibose (0.9mg/day) and Acarbose (150mg/day) for postprandial hyperglycemia and found that both drug has overall similar effect on postprandial glucose (Fujisawa T et al 2005).

4) Vicharant et al compared the efficacy and safety of Voglibose with Acarbose in type II diabetes patients and found that Voglibose (0.2mg t.d.s.) and Acarbose (100mg t.d.s.) significantly reduces HbA1C, PPG and postprandial insulin level. At these doses level of Voglibose was associated with less GIT side effects (Vichayanrat A. et al. 2002).

5) Van de Laar FA et al compared the efficacy and safety of Miglitol (50mg t.d.s.) versus Voglibose (0.2mg t.d.s.) in type II diabetes patients, found that there is mean reduction of

HbA1C and PPG which was statistically significant ($p < 0.05$) (Van de Laar FA et al 2005).

2.20. Alpha Glucosidae Inhibitor from Plant Source:

The medicinal plants are widely used by the traditional medical practitioners for curing various diseases in their day to day practice. The medicinal plants are rich in secondary metabolites (which are potential sources of drugs) and essential oils of therapeutic importance. The important advantages claimed for therapeutic uses of medicinal plants in various ailments are their safety besides being economical effective and their easy availability (Atal CK et al 1989; Siddiqui HH 1993; Soumya KV et al 2012). Because of these advantages the medicinal plants have been widely used by the traditional medical practitioners in their day to day practice.

Salacia species (e.g. *Salacia Oblonga, S. prinoides, Salacia Reticulata*), known as 'Ponkoranti' in Ayurvedic medicine, are widely distributed in Sri Lanka, India, China, Vietnam, Malaysia, Indonesia and other Asian countries, where these species have been used for thousands of years in traditional medicines particularly for the treatment of diabetes (He *et al*. 2009; Yuhao *et al*. 2008). Extracts from *Salacia* species (including *Salacia chinensis*, *Salacia reticulata*, and *Salacia oblonga*) are consumed in Japan, Korea, the United States, and India as a food supplement to prevent and manage obesity and diabetes (Li Y *et al*. 2008)

2.21. Salacia Oblonga:

Salacia oblonga is a perennial wild, woody, climbing vine native to India and Sri Lanka. A member of the Hippocrateaceae family, it is commonly known as ''ponkoranti'' due to its golden colored root bark. *Salacia oblonga* has been used for thousands of years in Ayurvedic medicine for the oral treatment of diabetes (Flammang AM et al 2006; Grover et al 2002). Root extracts of various Salacia species have been shown to inhibit the activity of intestinal a-glucosidases enzyme (Matsuda et al 2005). Inhibition of the carbohydrate metabolizing enzymes results in delayed breakdown of oligosaccharides and inhibits glucose absorption into the bloodstream. Augusti et al (Augusti KT et al 1995; Krishnakumar K et al 2000) isolated 2 active terpenoid fractions from the root bark

of this plant and showed its antidiabetic action, these two principles demonstrated about 60% and 76% of the hypoglycemic potency of an equal dose of tolbutamide (250 mg/kg) in albino rats (Augusti KT et al 1995).

Matsuda et al (Matsuda H et al 1999) demonstrated that the aqueous methanolic extract of the roots of *Salacia oblonga* inhibited the increase in serum glucose level in sucrose and maltose loaded rats. The water soluble and ethyl acetate soluble portions from the aqueous methanolic extract showed inhibitory activities on α-glucosidase and aldose reductase, respectively. From the water soluble portion, 2 potent α-glucosidase inhibitors, salacinol and kotalanol, were isolated as well as 9 sugar related components; a new friedelane type triterpene, kotalagenin 16-acetate, was isolated from the ethyl acetate–soluble portion along with known diterpenes and triterpenes. The structure of kotalagenin 16-acetate was elucidated on the basis of physicochemical evidence. Principal components from this natural medicine were examined in terms of inhibitory activity on aldose reductase. The diterpene and triterpene constituents, including the new kotalagenin 16-acetate, were found to be responsible components for the inhibitory activity on aldose reductase.

In another attempt, extract of *Salacia oblonga* constituents Mangiferin (Muruganandan S et al 2005) (a xanthone from the roots) and sulfonium ion derivatives, kotalanol (Mentreddy, Rao S et al 2007) and salacinol (from the roots and stems) (Yoshikawa M 2002; Murakami T et al 1997), have been identified as the antidiabetic principles. Mangiferin also inhibits aldose reductase activity, thereby delaying the onset or progression of diabetic complications (eg: diabetic neuropathy and nephropathy). The polyphenol constituents of *S.oblonga*, the catechins, also contribute to the antidiabetic property of the plant (Nadkarni, Dr.K.M.1976). Therefore, *Salacia oblonga* holds potential as a natural method to mitigate the blood glucose response for people with diabetes. As an ingredient incorporated into foodstuff, *Salacia oblonga* may provide people with diabetes a convenient way to help manage their blood glucose levels (A.M. Flammang et al 2006).

Williams et al (Williams JA et al 2007) evaluated the effect of an herbal extract of *Salacia oblonga on* postprandial glycemia and insulinemia in patients with type 2 diabetes mellitus after ingestion of a high carbohydrate meal. Sixty six patients with diabetes participated in a randomized, double blind, crossover study. In a fasted state, sub-

jects consumed 1 of the following 3 meals: a standard liquid control meal a control meal plus 240 mg *Salacia oblonga* extract, or a control meal plus 480 mg *Salacia oblonga* extract. Both doses of the salacia extract significantly lowered the postprandial positive area under the glucose curve (14% for the 240 mg extract and 22% for the 480 mg extract) and the adjusted peak glucose response (19% for the lower dose and 27% for the higher dose of extract) compared with the control meal. In addition, both doses of the salacia extract significantly decreased the postprandial insulin response, lowering both the positive area under the insulin curve and the adjusted peak insulin response (14% and 9%, respectively, for the 240 mg extract; 19% and 12%, respectively, for the 480 mg extract), compared with the control meal.

Huang et al (Huang TH et al 2008) investigated the effect of the water extract of *Salacia Oblonga* on obesity and diabetes associated cardiac hypertrophy. They discussed the modulation of the cardiac angiotensin II type 1 receptor (AT1) expression in the effect. *Salacia oblonga,* 100 mg/kg, was given orally to male Zucker diabetic fatty rats for 7 weeks. At the end of the study, the hearts and left ventricles were weighed, cardiomyocyte cross sectional areas were measured, and cardiac gene profiles were analyzed. Angiotensin II stimulated embryonic rat heart derived H9c2 cells and neonatal rat cardiac fibroblasts were pretreated with water extract of *Salacia oblonga* and 1 of its prominent components, mangiferin, respectively. Atrial natriuretic peptide, mRNA expression, protein synthesis, and (3H) thymidine incorporation were determined.

Salacia oblonga treated Zucker diabetic fatty rats showed less cardiac hypertrophy, as shown by a decrease in the weights of the hearts and left ventricles and by reduced cardiomyocyte cross sectional areas. *Salacia oblonga* treatment suppressed cardiac overexpression of atrial natriuretic peptide, brain natriuretic peptide, AT1 mRNAs, and AT1 protein. Aqueous extract of *Salacia oblonga* (50-100 μg/mL) and mangiferin (25μmol) suppressed angiotensin II induced ANP mRNA overexpression and protein synthesis in H9c2 cells. They also inhibited angiotensin II stimulated (3H) thymidine incorporation by cardiac fibroblasts. The findings demonstrate that *Salacia Oblonga* decreased cardiac hypertrophy in Zucker diabetic fatty rats, at least in part by inhibiting cardiac AT1 overexpression (Amritpal Singh et al 2010). Heacock P M et al reported that in a double masked randomized crossover study, 1000mg/day *Salacia oblonga* extract on

four separate occasions, lowered postprandial glycemia and significantly reduced the postprandial insulin response. Breath hydrogen excretion increased, suggesting the mechanism similar to alpha glucosidae inhibitors (Heacock P M et al 2005).

In randomized crossover clinical study the ethanol water extract of *Salacia oblonga* reduced the the glycemic response to high starch meal with mild flatulence suggesting alpha glucosidase inhibition as the main mechanism of action (Hertzler S et al 2007). In similar type of study it has lowered acute glycemia and insuinemia in person with type 2 diabetis after a high carbohydrate meal (William J A et al 2007).

Negative results in the bacterial reverse mutation and mouse micronucleus assay demonstrated that *Salacia oblonga* extract is devoid of any significant genotoxic activity, hence use of *Salacia oblonga* is expected to be safe (Flammang AM et al 2006). The no observable adverse effect level for *Salacia oblonga* was determined to be 2500mg/kg/day following daily subcronic oral gavages administration to rats (Flammang AM et al 2007). hot water extract of *Salacia oblonga* did not result in clinical chemistry of histopathologic indication of toxic effect in male Sprague Dawley rats (Wolf BW et al 2003). In traditional Indian medicine the root bark of *S.oblonga* is also used in gonnorhoea, rheumatism and skin diseases (Nadkarni Dr.K.M.1976; Kirtikar KR et al 1984).

2.22. ***Salacia reticulata*:**

Salacia reticulata is a climbing plant in the Hippocrateaceae family Wight ('Kotala himbutu' in Singhalese), a large woody climbing plant in the submontane forests, is distributed in Sri Lanka and the south region of India, that has been used traditionally in Indian Ayurvedic medicine and is said to be effective for the prevention and treatment of diabetes, rheumatism, gonorrhea and skin diseases (Yoshikawa M et al 1998; Yoshikawa M et al 2002).

The aqueous extract from the root bark of Sri Lankan *Salacia reticulata* was reported to show hypoglycemic activity in rats (Karunanayake E H et al 1984) while the aqueous extract was also reported to produce a significant lowering of plasma glucose in streptozotocin induced diabetic rats (Serasinghe S et al 1990). As the constituents of this plant, many triterpenes as well as hydrocarbon, sitosterol, mangiferin, and gutta percha were isolated from the root and stem barks (Yoshikawa M et al 2002). However, pharma-

cologically active components were left uncharacterized In the course of Masayuki Yoshikawa, studies on antidiabetogenic compounds from natural medicines and medicinal foodstuffs (Yoshikawa M et al 2002) he found that the aqueous methanolic extract of Sri Lankan *Salacia reticulata* did not show hypoglycemic effects on oral d-glucose-loaded rats and alloxaninduced diabetic mice, while the extract and the water soluble portion showed potent hypoglycemic effect in oral sucrose loaded rats and α-glucosidase inhibitory effect. Through bioassay guided separation using α-glucosidase inhibitory activities, he isolated potent α-glucosidase inhibitors termed salacinol (Yoshikawa M et al 1997) and kotalanol (Yoshikawa M et al 1998) from the water soluble portion together with several phenolic compounds (Yoshikawa M et al 2001). Furthermore, salacinol and kotalanol were also isolated from Indian *Salacia oblonga* and Thai S. chinensis together with several new triterpenes with aldose reductase inhibitory activity (Matsuda H et al 1999).

Kajimoto et al. (2000) reported that a double blind placebo controlled study performed in Japan resulted in significantly decreased blood sugar levels in humans with mild type II diabetes, receiving *Salacia reticulata* extract as part of their diet, as compared to control. In a sucrose tolerance test on human volunteers, pretreatment with the aqueous extract of *Salacia reticulata* prior to sucrose loading significantly suppressed postprandial hyperglycemia (Shimoda et al 1998; Tanimura et al 2005). Water extract prepared from *Salacia reticulata* leaves can also prevent diabetes and obesity similarly to that of roots and stems (Yoshino et al 2009). Mangiferin, one of the main components in *Salacia* species (Li et al 2004), has been reported to be potent α-glucosidase inhibitors that have been shown to inhibit increases in serum glucose levels (Yoshikawa et al 1997, 1998, 2001). Aqueous extract of *Salacia reticulata* strongly inhibited the activities of α-glucosidase and α-amylase, but not that of β-glucosidase (Shimoda et al 1998). *Salacia oblonga* root extract lowered acute glycemia and insulinemia in patients with type II diabetes after a high carbohydrate meal (Williams et al 2007). *Salacia oblonga* root extract concentration dependently inhibited α-glucosidase activity *in vitro* (Li et al 2004). *S. chinensis* also showed α-glucosidase inhibitory activity (Yoshikawa et al 2003). The intestinal enzymes α-glucosidase and α-amylase break down starches, dextrins, maltose and sucrose into absorbable monosaccharides, thus, it could be suggested that the antidiabetic

property of *Salacia* is partially attributed to intestinal α-glucosidase inhibitory activity (Yuhao et al 2008). Furthermore, inhibition of above enzymes delays glucose absorption into the blood and suppresses postprandial hyperglycemia, resulting in improved glycemic control (Heacock et al 2005). In addition, mangiferin can activate PPAR-α luciferase activity in human embryonic kidney 293 cells and enhances PPAR-α-dependent lipoprotein lipase expression and activity in the THP-1 derived macrophage cell line (Huang et al 2006). This compound could also inhibit aldose reductase activity, thereby delaying the onset or progression of diabetic complications (Yoshikawa et al 2001).