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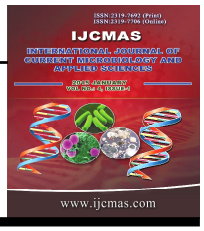

Review Article

A Role of Insulin in different types of Diabetes

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A B S T R A C T

Keywords

Insulin, Hyperglycemia, Diabetes mellitus, Gestational diabetes mellitus

Diabetes mellitus is a group of metabolic diseases characterized by high blood sugar (glucose) levels that result from defects in insulin secretion, or action, or both. Herbal medicines have been highly effective source of medicine throughout the human history. They are widely used today indicating that herbs are a growing part of modern high-tech medicine. The herbal plants fenugreek seed and cinnamon seed which have a role in the management of diabetes mellitus are compiled here are discussed in this review.

Introduction

Diabetes is a complex, chronic disease characterized by an elevation of the level of glucose in the blood. Insulin, a hormone produced by the pancreas, controls the blood glucose level by regulating the production and storage of glucose. In diabetes there may be a decrease in the body's ability to respond to insulin or a decrease in the insulin produced by the pancreas which leads to abnormalities in the metabolism of carbohydrates, proteins and fats. The resulting hyperglycemia may lead to acute metabolic complications including ketoacidosis and in the long term contribute to chronic micro-vascular complications (Smeltzer and Bare, 1992). Diabetes mellitus may be redefined as a complex, chronic disorder characterized by disruption of normal carbohydrates, fat and protein metabolism and the development over time of micro-vascular and macro-vascular complications and neuropathies.

Diabetes –India scenario

India with its dubious distinction of being called "the diabetes capital of the world" is presently estimated to have over 30 million individual affected by this deadly disease. India is ahead of China and USA, which are in second and third places, respectively, in the number. This represents a three hundred percent increase in the number of people with diabetes between 1995 and 2030. A striking example of the rise in prevalence of diabetes in India is the fivefold increase from 2.1% to 12.1% seen from 1970 to 2000

Ahuja *et al.* (1979). Two population based studies, the Chennai Urban population study (CUPS) and Chennai Urban Rural Epidemiological study (CURES) showed a marked increase in the prevalence of diabetes within a short span of five years. The CUPS study conducted in the year 1997 revealed that 12% (crude prevalence rate) of the Chennai population to be affected by diabetes. The national Urban Diabetes Survey (NUDS), carried out in six cities in the year 2001, reported the age-standardized prevalence of diabetes in Chennai according to the study was 13.5%. In addition the study also reported that 14% had impaired glucose tolerance (IGT). The city of Hyderabad showed the highest rates of both diabetes (17%) and IGT (30%). In Chennai, Banglore, Hyderabad and Mumbai, the prevalence of IGT exceeded those of diabetes (Ramachandran *et al.,* 2001). The large population based study, CURES conducted on 26,001 individuals in the year 2001-2002, showed that according to the ADA criteria 19% had diabetes in Chennai and this scaled down to 16% when WHO criteria was used.

Global prevalence of diabetes mellitus

In the year 2000, the global number of individuals with diabetes was estimated to be 171 million (2.8% of the world's population), and this figure has been projected to increase in 2030 to 366 million (6.5%), 298 million of whom will be living in developing countries (WHO, 1998). Type 2 diabetes mellitus has reached epidemic proportions with explosive increase in incidence worldwide over the past few decades. Although type 2 diabetes mellitus is more prevalent in developed countries, the increase in incidence seems to be more pronounced especially in populations that are experiencing rapid westernization (Zimmet *et al.,* 2002). Apart from microvascular complications, cardiovascular disease, with its attendant morbidity and mortality, is on the rise in the developing countries. Current evidence suggests that environmental factors are major determinants of the increasing rates of diabetes (WHO, 1998). Overweight and obesity are increasing dramatically and contribute to the burden of diabetes mellitus and other chronic health conditions. Indeed, the modern environment promotes behaviours that cause obesity.

Classification of diabetes mellitus

Type I diabetes mellitus

Type 1 diabetes is caused by a lack of insulin due to the destruction of insulin-producing beta cells in the pancreas. In type 1 diabetes—an autoimmune disease—the body's immune system attacks and destroys the beta cells. Normally, the immune system protects the body from infection by identifying and destroying bacteria, viruses, and other potentially harmful foreign substances. But in autoimmune diseases, the immune system attacks the body's own cells. In type 1 diabetes, beta cell destruction may take place over several years, but symptoms of the disease usually develop over a short period of time. Type 1 diabetes typically occurs in children and young adults, though it can appear at any age. In the past, type 1 diabetes was called juvenile diabetes or insulin-dependent diabetes mellitus.

Type II diabetes mellitus

Type 2 diabetes—the most common form of diabetes—is caused by a combination of factors, including insulin resistance, a condition in which the body's muscle, fat, and liver cells do not use insulin effectively. Type 2 diabetes develops when the body can no longer produce enough insulin to compensate for the impaired ability to use

insulin. Symptoms of type 2 diabetes may develop gradually and can be subtle; some people with type 2 diabetes remain undiagnosed for years. Type 2 diabetes develops most often in middle-aged and older people who are also overweight or obese. The disease, once rare in youth, is becoming more common in overweight and obese children and adolescents. Scientists think genetic susceptibility and environmental factors are the most likely triggers of type 2 diabetes.

Gestational diabetes mellitus

The onset of gestational diabetes mellitus is during pregnancy, usually in the second or third trimester, as a result of hormones secreted by the placenta, which inhibit the action of insulin. It occurs in about 2–5% of all pregnancies. About 30–40% of patients with gestational diabetes mellitus will develop type II diabetes within 5–10 years (especially if obese). Impaired glucose tolerance and statistical risk groups are examples of gestational diabetes mellitus. Statistical risk groups are individuals at greater risk than the general.

Other types

Other type of diabetes mellitus: This is where diabetes mellitus is associated with other conditions, for example, pancreatic disease, hormonal disorders and drugs such as glucocorticoids and oestrogen-containing preparations. Depending on the ability of the pancreas to produce insulin, the patient may require oral agents or insulin.

LADA

Latent autoimmune diabetes in adults (LADA) may be a slowly developing kind of diabetes. Diagnosis usually occurs after age 30. In LADA, as in type 1 diabetes, the body's immune system destroys the beta cells. At the time of diagnosis, people with LADA may still produce their own insulin, but eventually most will need insulin shots or an insulin pump to control blood glucose levels.

Experimental diabetes

Alloxan and streptozotocin are widely used to induce experimental diabetes in animals. The mechanism of their action in B cells of the pancreas has been intensively investigated and now is quite well understood. The cytotoxic action of both these diabetogenic agents is mediated by reactive oxygen species, however, the source of their generation is different in the case of alloxan and streptozotocin.

Alloxan

Alloxan (2,4,5,6-tetraoxypyrimidine; 5,6-dioxyuracil) was first described by Brugnatelli in 1818. Wöhler and Liebig used the name "alloxan" and described its synthesis by uric acid oxidation. Alloxan exerts its diabetogenic action when it is administered parenterally: intravenously, intraperitoneally or subcutaneously. The dose of alloxan required for inducing diabetes depends on the animal species, route of administration and nutritional status. Human islets are considerably more resistant to alloxan than those of the rat and mouse Eizirik *et al.* (1994). The most frequently used intravenous dose of this drug to induce diabetes in rats is 65 mg/kg b.w. Gruppuso *et al.* (1990). When alloxan is given intraperitoneally or subcutaneously its effective dose must be 2-3times higher. The intraperitoneal dose below 150 mg/kg b.w. may be insufficient for inducing diabetes in the rat Katsumata *et al.* (1992).

Streptozotocin

Streptozotocin (STZ,2-deoxy-2-(3-(methyl-

3-nitrosoureido) - D-glucopyranose) is synthesized by *Streptomycetes achromogenes* and is used to induce both insulin-dependent and non-insulin-dependent diabetes mellitus (IDDM and NIDDM, respectively). The range of the STZ dose is not as narrow as in the case of alloxan. The frequently used single intravenous dose in adult rats to induce IDDM is between 40 and 60 mg/kg b.w. Ganda *et al.* (1976), but higher doses are also used. STZ is also efficacious after intraperitoneal administration of a similar or higher dose, but single dose below 40 mg/kg b.w. may be ineffective Katsumata *et al.* (1992). For instance, when 50 mg/kg b.w. STZ are injected intravenously to fed rats, blood glucose (determined 2 weeks after treatment) can reach about 15 mM.

Role of insulin

Release of insulin in diabetes mellitus

Beta cells in the islets of Langerhans release insulin in two phases (Najjar *et al.,* 2001). The first phase release is rapidly triggered in response to increased blood glucose level. The second phase is a sustained, slow release of newly formed vesicles triggered independently of sugar. The description of first phase release as follows:

- Glucose enters the β-cells through the glucose transporter, GLUT2.
- Glucose goes into glycolysis and the respiratory cycle, where multiple, high energy ATP molecules are produced by oxidation, leading to a rise in the ATP:ADP ratio within the cell.
- An increased intracellular ATP: ADP ratio closes the ATP-sensitive SUR1/Kir6.2 potassium channel. This prevents potassium ions (K^+) from leaving the cell by facilitated diffusion, leading to a buildup of potassium ions. As a result, the inside of the cell becomes more positive with respect to the outside, leading to the depolarization of the cell surface membrane.
- On depolarization, voltage-gated calcium ion (Ca^{2+}) channels open which allows calcium ions to move into the cells by facilitated diffusion.
- An increased intracellular calcium ion concentration causes the activation of phospholipase C, which cleaves the membrane phospholipid, phosphatidyl inositol 4,5-bisphosphate into inositol 1,4,5-trisphosphate and diacylglycerol.
- Inositol 1,4,5-trisphosphate (IP3) binds to receptor protein in the plasma membrane of the endoplasmic reticulum (ER). This allows the release of Ca^{2+} ions from the ER via IP3-gated channels, and further raises the intracellular concentration of calcium ions.

Significantly increased amounts of calcium ions in the cells cause the release of previously synthesized insulin, which has been stored in secretary vesicles. This is the primary mechanism for release of insulin. Other substances known to stimulate insulin release include the amino acids arginine and leucine, parasympathetic release of acetylcholine (via phospholipase C), sulphonylurea, cholecystokinin (CCK, via phospholipase C) Nakaki *et al.* (1980) and the gastrointestinally derived incretins glucagon-like peptide-1 (GLP-1) and glucose-dependent insulinotropic peptide (GIP).

Release of insulin is strongly inhibited by the stress hormone, Norepinephrine (noradrealine), which leads to increased blood glucose levels during stress. It appears that release of catecholamines by the

sympathetic nervous system has conflicting influences on insulin release by beta cells, because insulin release is inhibited by α_2^- adrenergic receptors Nakaki *et al.* (1980) and stimulated by β_2^- adrenergic receptors Layden *et al.* (2010). The net effect of norepinephrin from sympathetic nerves and epinephrine from adrenal glands on insulin release is inhibited due to dominance of the α-adrenergic receptors (Sircar *et al.,* 2007).

When the glucose level comes down to the usual physiologic value, insulin release from the β-cells slows or stops. If blood glucose levels drop lower than this, especially to dangerously low levels, release of hyperglycemic hormones (most prominently glucagone from islet of Langerhans α-cells) forces release of glucose into the blood from cellular stores, primarily liver cell stores of glycogen. By increasing blood glucose, the hyperglycemic hormones prevent or correct life-threatening hypoglycemia.

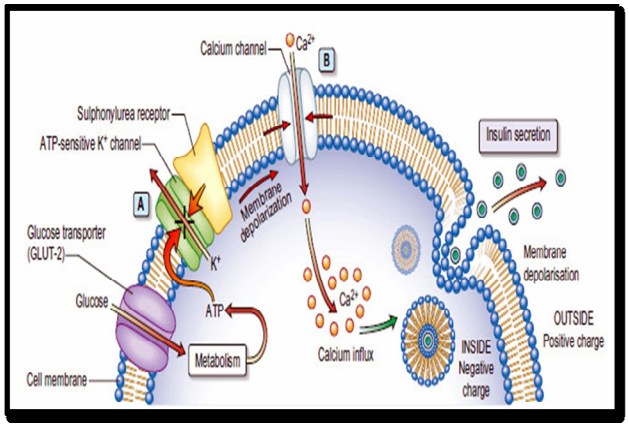

Figure.1 Mechanism of insulin secretion

Phosphorylation of IRS proteins, insulin action, and insulin resistance

Insulin signaling at target tissues is essential for growth and development and for normal homeostasis of glucose, fat, and protein metabolism. Control over this process is therefore tightly regulated. It can be achieved by a negative feedback control mechanism whereby downstream components inhibit upstream elements along the insulin-signaling pathway (autoregulation) or by signals from apparently unrelated pathway that inhibit insulin signaling thus leading to insulin resistance. Phosphorylation of insulin receptor substrate (IRS) proteins on serine residues has emerged as a key step in these control processes under both physiological and pathological conditions. The list of IRS kinases implicated in the development of insulin resistance is growing rapidly, concomitant with the list of potential Ser/Thr phosphorylation sites in IRS proteins. Here, we review a range of conditions that activate IRS kinases to phosphorylate IRS proteins on "hot spot" domains.

How does the insulin receptor work?

The insulin receptor (IR) is a transmembrane receptor that is activated by insulin and belongs to the large class of tyrosine kinase receptors (Ward and Lawrence, 2009). The binding of ligand to the α-chains of the IR induces a structural change within the receptor leading to autophosphorylation of various tyrosine residues within the intracellular TK domain of the β-chain. These changes facilitate the recruitment of specific adapter proteins such as the insulin receptor substrate proteins (IRS) in addition to Src (Kiselyov *et al.,* 2009). Insulin is the major hormone controlling critical energy functions such as glucose and lipid metabolism. Insulin activates the insulin receptor tyrosine kinase (IR), which phosphorylates and recruits different substrate adaptors such as the IRS family of proteins. Tyrosine phosphorylated IRS then displays binding sites for numerous signaling partners. Among them, PI3K has a major role in insulin function, mainly via the activation of the Akt/PKB and the PKCζ cascades. Insulin stimulates glucose uptake

in muscle and adipocytes via translocation of GLUT4 vesicles to the plasma membrane. GLUT4 translocation involves the PI3K/Akt pathway (Altarejos and Montminy, 2011). Whereas interaction of IRS and Shc proteins with the intracellular domain of the insulin receptor constitutes the first step in dispersing the directions of insulin signaling intracellular, their ability to attract multiple signaling intermediates to their own phosphorylated domains further partitions insulin signaling downstream, thus accounting for the multitude of insulin's biological effects. Insulin actions are mediated by the signaling pathway involving IRS proteins, phosphorylation, and activation of phosphatidyl inositol (PI) 3-kinase, Akt (also known as protein kinase B) Shepherd *et al.* (1995). Activation of PI 3-kinase, Akt, and atypical protein kinase C (PKC) via the phosphoinositide-dependent protein kinase appears to be critical in the mechanism of insulin action on GLUT-4 translocation and glucose transport (Bandyopadhyay *et al.*, 1997). In contrast, non-metabolic, proliferative, and mitogenic effects of insulin are mediated largely via the activation of Ras (mostly through Shc and, to a lesser degree, through IRS proteins), Raf, and mitogen-activated protein kinases Erk 1 and Erk 2.

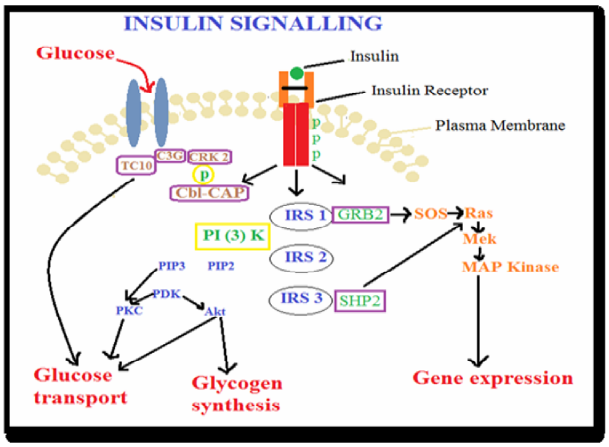

Figure.2 Mechanism of insulin action

IRS kinases and insulin resistance

Insulin resistance is defined as the failure of ordinary levels of insulin to trigger its downstream metabolic actions and is closely associated with obesity and the development of type 2 diabetes (Kahn *et al.*, 2006). It is becoming clear that obesity promotes a state of chronic low-grade inflammation and insulin resistance (Shoelson *et al.*, 2006; Hotamisligil *et al.*, 1993). This is attributed to the release from the adipose tissue of FFA, glycerol, hormones (e.g., leptin, adiponectin, endothelin-1), proinflammatory cytokines (e.g., TNFα, IL-1β, IL-6), and additional products of macrophages that populate adipose tissue in obesity (Shoelson *et al.*, 2006; Fain *et al.*, 2004). It has been shown already a hundred year ago that high doses of salicylates lower glucose levels in diabetic patients implicating the involvement of inflammation in type 2 diabetes. Additionally, increased release of FFAs decreases insulin-mediated glucose transport in skeletal muscle and impairs suppression of glucose production by the liver, a characteristic of insulin resistance (Anderwald and Roden, 2004; Boden and Shulman, 2002).

Many inducers of insulin resistance activate IRS kinases that negatively regulate insulin signaling and action. The list of IRS kinases implicated in the development of insulin resistance is growing rapidly, concomitant with the list of potential Ser/Thr phosphorylation sites in IRS proteins (Draznin, 2006). Recent studies have focused on IRS-1 as a major target for IRS kinases (Draznin, 2006). However, it is now becoming evident that IRS-2 serves as a target as well (Gurevitch *et al.*, 2008; Sharfi and Eldar-Finkelman, 2008).

IRS kinases can be divided into two groups. One includes kinases that are mediators of insulin signaling. These kinases negatively

regulate IRS proteins upon prolonged insulin stimulation [e.g., mTOR/S6K1 (Um *et al.,* 2004), MAPK (De Fea and Roth, 1997) and PKCζ (Lee *et al.* 2008)]. The other group consist of kinases that are activated along unrelated pathways to inhibit insulin action [e.g., glycogen synthase kinase (GSK)-3β (Eldar-Finkelman and Krebs, 1997; Liberman and Eldar-Finkelman, 2005), IKKβ (Gao *et al.,* 2002)]. Of note, several IRS kinases (e.g., S6K1, PKC) are activated both in response to insulin and as inducers of insulin resistance (Morino *et al.,* 2006; Sampson and Cooper 2006; Um *et al.,* 2004).

Glucose transporters

GLUTs are integral membrane proteins that contain 12 membrane-spanning helices with both the amino and carboxyl termini exposed on the cytoplasmic side of the plasma membrane. GLUT proteins transport glucose and related hexoses according to a model of alternate conformation Oka *et al.* (1990) They predicted that the transporter exposes a single substrate binding site toward either the outside or the inside of the cell. Binding of glucose to one site provokes a conformational change associated with transport, and release glucose to the other side of the membrane. The inner and outer glucose- binding sites are, it seems, located in transmembrane segments (Hruz and Mueckler, 1999).

Different types of GLUTs

GLUT1: GLUT1 is widely distributed in fetal tissues. In the adults, it is expressed at high in erythrocytes and also in the endothelial cells of barrier tissues such as the blood-brain. However, it is responsible for the low-level of basal glucose uptake required to sustain respiration in the cells.

GLUT2: GLUT2 is a bidirectional transporter, allowing glucose to flow in 2 directions. It is expressed by renal tubular cells, small intestinal epithelial cells, liver cells and pancreatic beta cells. It is also present in the basolateral membrane of the small intestine epithelium. Bidirectionality is required in liver cells to uptake glucose for glycolysis, and release of glucose during gluconeogenesis. In pancreatic beta cells, free flowing glucose is required so that the intracellular environment of these cells can accurately gauge the serum glucose levels. All three monosaccharides (glucose, galactose and fructose) are transported from the intestinal mucosal cell into the portal circulation by GLUT2.

GLUT3: GLUT3 is expressed mostly in neurons (where it is believed to be the main glucose transporter isoform), and in the placenta.

GLUT4: GLUT4 found in adipose tissue and striated muscle (skeletal muscle and cardiac muscle). GLUT4 has been widely studied due to its role as the main insulin-sensitive member of this family and thus its role in diabetes. Its expression is highest in the insulin sensitive tissues including brown and white adipose tissue and skeletal and cardiac muscle. It has a K_m for glucose of approximately 5-6 mM, similar to GLUT1 and can also transport dehydroascorbic acid and glucosamine.

Under insulin stimulation, GLUT4 undergoes a rapid translocation from the intracellu-lar location to the cell surface, resulting in a dramatic increase in cellular glucose transport activity. GLUT4 is one of 13 sugar transporter proteins (GLUT1-GLUT-12 and HMIT) encoded in human genome that catalyzes hexose transport across cell membrane via an ATP-independent facilitative diffusion mechanism Hruz and Mueckler (1999). These sugar transporters display differences in their kinetics and respective substrate

specificities such that such that GLUT-5 and perhaps GLUT-11 are likely fructose transporter.

A central role for GLUT4 in whole body metabolism is strongly supported by a variety of genetically engineered mouse models where expression of transporter is either enhanced or ablated in muscle or adipose tissue or both. The whole body GLUT4 mouse itself may be less informative due to upregulation of compensatory mechanism that may promote survival of these animals. However heterozygous GLUT4 mice that display decreased GLUT4 protein in muscle and adipose tissue show the expected insulin-resistance and propensity towards diabetes that is consistent with a major role of GLUT4 in glucose disposal. Interestingly, over expression of GLUT4 in skeletal muscle of such GLUT4 animals via crosses with transgenic mice normalizes insulin sensitivity and glucose tolerance. Transgenic mice expressing high levels of GLUT4 in adipose tissues or in skeletal muscles in turn are both highly insulin sensitive and glucose tolerant. Conversely conditional depletion of GLUT4 in either adipose tissue or skeletal muscle causes insulin resistance and a roughly equivalent incidence of diabetes animals. This was particularly surprising in former case since adipose tissue accounts for only small fraction of total body glucose disposal James *et al.* (1989).These tissue specific depletion of GLUT4 have profound metabolic effects on other tissues. For e.g. mice with muscle specific GLUT4 deficiency display decrease insulin responsiveness in adipose tissue and liver, while those with adipose specific GLUT4 depletion exhibit muscle and liver insulin resistance Abel *et al.* (2001). In the muscle specific GLUT4 knockout mice, this at least partially mediated via elevated blood glucose levels that occur in the conditional knockout animals, secondarily impairs insulin signaling Kim *et al.* (2005). Remarkably over expression of GLUT4 in the adipose tissue of muscle-specific GLUT4 deficient mice overcomes the glucose intolerance and diabetes. More recently, it has been reported that a retinol binding protein (RBP4) is released into the serum from GLUT4 deficient adipose tissue, and that RBP4 may contribute to the insulin resistance of obese and diabetes individual.
Even though cell surface GLUT4 is highly dependent on insulin *in vitro,* muscle-specific (MIRKO) or adipose-specific (FIRCO) insulin receptor knockout mice produce surprisingly mild metabolic phenotypes compared to the conditional GLUT4knockout mice.

MIRKO mice have an enlarged fat mass with increased serum triglyceride and free fatty acids, but otherwise have normal whole-body glucose homeostasis. Insulin stimulated glucose uptake is greatly reduced in MIRkO muscle, but muscle glycogen level is normal, indicating possible compensatory mechanism for glucose import Kim *et al.* (2005). FIRKO mice have severe insulin resistance in adipose tissue but are protected from age and hyperphagia-induced glucose intolerance.

GLUT5: (SLC2A5), a fructose transporter

GLUT7: SLC2A7-(SLC2A7), transporting glucose out of the endoplasmic reticulum

GLUT9: SLC2A9- (SLC2A9)

GLUT11: (SLC2A11)

GLUT6: (SLC2A6)

GLUT8: (SLC2A8)

GLUT10: (SLC2A10)

GLUT12: (SLC2A12)

The H^+ /myoinositol transporter HMIT (SLC2A13)

The function of these new glucose transporter isoforms is still not clearly defined at present.

Complications of Diabetes mellitus

The general categories of chronic diabetic complications include macrovascular disease, microvascular disease and neuropathy.

Macrovascular Complications

Smeltzer and Bare (1992) describe macrovascular diseases as atherosclerotic changes in the larger blood vessels. Different types of macrovascular diseases may result, depending upon the location of the atherosclerotic lesions.

- **Coronary artery disease**

  Atherosclerotic changes in the coronary arteries lead to an increased occurrence of myocardial infarctions in persons with diabetes. It may account for 50-60% of all deaths in patients with diabetes. Sometimes ischemic symptoms may be absent and the patient may not experience early warning signs of decrease coronary blood flow. In such cases the patient may have 'silent' myocardial infarctions which can be detected through the use of electrocardiogram (Smeltzer and Bare, 1992).

- **Cerebral vascular disease**

  These are atherosclerotic changes in cerebral blood vessels or the formation of an embolus elsewhere in the vasculature that then lodges in a cerebral blood vessel which can lead to transient ischemic attacks and strokes. Recovery from stroke may be impaired in diabetic patients whose blood glucose level is elevated at the time of diagnosis. The following are symptoms of cerebro-vascular disease: dizziness, decreased vision, slurred speech and weakness (Smeltzer and Bare, 1992).

- **Microvascular complications**

  These are the diseases of small blood vessels where the basement membrane in the capillaries and arterioles thickens (Smeltzer and Bare, 1992). The retina of the eye and the kidney are affected. This results in diabetic retinopathy and nephropathy. Diabetic retinopathy is the leading cause of blindness in people aged between 20 and 74 years, and one out of every four individuals on dialysis has diabetic nephropathy.

- **Diabetic retinopathy**.

  Diabetic retinopathy is any disease of the retina and a leading cause of blindness. It occurs when diabetes damages the tiny blood vessels inside the retina, the light-sensitive tissue at the back of the eye. A healthy retina is necessary for good vision. If you have diabetic retinopathy, at first you may notice no changes to your vision. But over time, diabetic retinopathy can get worse and cause vision loss. Diabetic retinopathy usually affects both eyes. Retinopathy is classified as background retinopathy, preproliferative retinopathy and proliferative retinopathy.

- **Nephropathy**

  This is the end-stage renal disease which

usually shows no symptoms before this stage, and requires dialysis or transplantation. Nephropathy is indicated by protein in urine which is measured using a dipstick (Smeltzer and Bare, 1992). It occurs in diabetic patients who have had the disease for the past 10-20 years. People with diabetes account for about 25% of patients with end stage renal disease. Once the progression of renal disease takes place it is treated either with dialysis or kidney transplant. Kidney transplantation involves transplanting a kidney from a living donor or human cadaver to a recipient who has end-stage renal disease.

- **Neuropathy**

Diabetic patients often suffer from some degree of neuropathy (Smeltzer and Bare, 1992). We can define neuropathy as a group of diseases that affect all types of nerves, including peripheral, autonomic and spinal nerves. Neuropathy causes a loss of sensation or an alteration in the sensitivity of the feet and legs. Elevated blood glucose levels over a period of years have been implicated in the etiology of neuropathy.

Sensory motor neuropathy affects the distal portion of the nerves, more especially the lower extremities. Symptoms include pricking, tingling and burning sensations at night. The feet may become numb. Decreased sensations of pain and temperature place patients with neuropathy at increased risk of injury and undetected foot infections. This neuropathic pain can resolve on its own within 6 months in some patients, whereas, for others, the pain persists for many years. The patient should be reassured and advised to do the following:

- take enough fluids and high fibre in the diet to prevent constipation
- in the case of diarrhoea, prepare an oral glucose and electrolyte solution and drink it to rehydrate their bodies
- take anti-diarrhoea drugs as prescribed
- express their fears or worries about being embarrassed by lack of control over bowel elimination

- **Infections and skin disorders**

Elevated blood sugar inhibits the action of white blood cells which fight against micro-organisms. This makes the diabetic patient susceptible to infections. Once infection sets in it spreads more rapidly. Foot lesions occur as a result of neuropathy, as the patient does not have sufficient sensory function to be aware of trauma like blisters, tight shoes, etc. The person may tread on a sharp object such as a thorn or nail without feeling any pain. As there is poor circulation, this may result in a high risk of infection. Patients are advised to wear footwear and avoid activities that can traumatize the feet.

Role of Oxidative stress in Diabetic Complications

Increasing evidence in both experimental and clinical studies suggests that oxidative stress plays a major role in the pathogenesis of both types of diabetes mellitus. Free radicals are formed disproportionately in diabetes by glucose oxidation, nonenzymatic glycation of proteins, and the subsequent oxidative degradation of glycated proteins. Abnormally high levels of free radicals and the simultaneous decline of antioxidant defense mechanisms can lead to damage of cellular organelles and enzymes, increased lipid peroxidation, and development of

insulin resistance. Free radicals have been recognized as intermediates of some biological redox reactions essential for the maintenance of life. Biological materials, particularly membranes, contain high concentrations of unsaturated lipids. In the presence of a free radical initiator and oxygen they may be oxidized. This process is known as lipid peroxidation. Pla (1976) implicated, as a general biological degenerative reaction. It has been reported that glucose might be autooxidized generating free radicals (Curcio and Ceriello, 1992). Hydrogen peroxide (H_2O_2) is a product of a variety of cellular reactions such as amino acid oxidation, normal respiration and superoxide dismutation by superoxide dismutase. Although it is not a free radical and is more stable than free radicals, H_2O_2 possesses a serious threat to cells because it can react with O_2 or transition metals to form the serious hydroxyl radical (Robert *et al.*, 1998). These consequences of oxidative stress can promote the development of complications of diabetes mellitus. Changes in oxidative stress biomarkers, including superoxide dismutase, catalase, glutathione reductase, glutathione peroxidase, glutathione levels, vitamins, lipid peroxidation hyperglycemia in diabetes have been reported (Maritim *et al.*, 2003).

Oxidative stress plays a major role in the pathogenesis of diabetes mellitus. Oxidative stress also appears to be the pathogenic factor in underlying diabetic complications. Reactive oxygen species (ROS) are generated by environmental factors, such as ionizing radiations and chemical carcinogens, and also by endogenous processes, including energy metabolism in mitochondria. ROS produced either endogenously or exogenously can attack lipids, proteins and nucleic acids simultaneously in living cells. There are many potential mechanisms whereby excess glucose metabolites traveling along these pathways might promote the development of complications of diabetes mellitus and cause pancreatic β cell damage. However, all these pathways have in common in the formation of ROS, that in excess and over time, causes chronic oxidative stress, which in turn causes defective insulin gene expression and insulin secretion as well as increased apoptosis (Yang *et al.*, 2011).

Antioxidant mechanism

Superoxide is converted spontaneously or by manganese superoxide dismutase (MnSOD) in mitochondria or copper-zinc superoxide dismutase (Cu/ZnSOD) in cytosol to hydrogen peroxide (H_2O_2). H_2O_2 can be removed by either catalase or peroxidase. Catalase is mainly expressed in peroxisomes and catalyzes direct decomposition of H_2O_2 to O_2 and water. In contrast, glutathione peroxidase removes H_2O_2 peroxide by coupling the oxidation of glutathione (GSH) (Chang and Chuang, 2010). Antioxidant treatment or over-expression of glutathione peroxidase or catalase can reverse these effects. Beta-cells express low levels of anti-oxidative enzyme including catalase and glutathione peroxidase.

Increased Intracellular AGE Formation

AGEs are formed by the nonenzymatic reaction of glucose and other glycating compounds derived both from glucose and from increased fatty acid oxidation in arterial endothelial cells with proteins (Wautier and Schmidt, 2004). In diabetes, AGEs are found in increased amounts in extracellular matrix (Stitt *et al.*, 1997). Intracellular production of AGE precursors can damage cells by 3 general mechanisms. Firstly, intracellular proteins modified by AGEs have altered function. Secondly,

extracellular matrix components modified by AGE precursors interact abnormally with other matrix components and with matrix receptors (integrins) that are expressed on the surface of cells. Finally, plasma proteins modified by AGE precursors bind to AGE receptors on cells such as macrophages, vascular endothelial cells, and vascular smooth muscle cells. Receptor for AGE (RAGE) binding induces the production of ROS, which in turn activates the pleiotropic transcription factor nuclear factor (NF)-κB, causing multiple pathological changes in gene expression. Methylglyoxal modification of mSin3A results in increased recruitment of *O*-linked *N*-acetylglucosamine (*O*-GlcNAc) transferase.

High glucose induces a decrease in transactivation by the transcription factor hypoxia-inducible factor (HIF)-1α, which mediates hypoxia-stimulated chemokine and vascular endothelial growth factor (VEGF) production by hypoxic tissue, as well as chemokine receptor and endothelial nitric oxide synthase (eNOS) expression in endothelial precursor cells in the bone marrow.

Decreasing superoxide in diabetic mice by either transgenic expression of manganese superoxide dismutase (MnSOD) or by administration of a superoxide dismutase (SOD) mimetic corrected postischemic defects in neovascularization, oxygen delivery, and chemokine expression and normalized tissue survival. Decreased HIF-1α functional activity was specifically caused by impaired HIF-1α binding to the coactivator p300. Hyperglycemia-induced covalent modification of p300 by the dicarbonyl metabolite methylglyoxal is responsible for this decreased association. In diabetic mouse models of impaired angiogenesis and wound healing, decreasing mitochondria ROS formation normalizes both ischemia-induced new vessel formation and wound healing (Schatteman *et al.*, 2000).

Increased Protein Kinase C Activation

PKCs are a family of at least 11 isoforms that are widely distributed in mammalian tissues. The enzyme phosphorylates various target proteins. The activity of the classic isoforms is dependent on both Ca^{2+} ions and phosphatidyl serine and is greatly enhanced by diacylglycerol (DAG) (Geraldes and King, 2010). Persistent and excessive activation of several PKC isoforms operates as a third common pathway mediating tissue injury induced by diabetes-induced ROS. This results primarily from enhanced *de novo* synthesis of DAG from glucose via triose phosphate, whose availability is increased because increased ROS inhibit activity of the glycolytic enzyme GAPDH, raising intracellular levels of the DAG precursor triose phosphate in insulin-resistant arterial endothelial cells and heart, may play an equally important role in diabetic atherosclerosis and cardiomyopathy. Over activity of PKC has been implicated in the decreased NO production in smooth muscle cells (Ganz and Seftel, 2000) and has been shown to inhibit insulin-stimulated expression of eNOS in cultured endothelial cells (Kuboki *et al.*, 2000). Activation of PKC by high glucose also induces expression of the permeability-enhancing factor VEGF in vascular smooth muscle cells (Williams *et al.*, 1997).

Origin of reactive oxygen species

In mitochondria, glucose is oxidized to CO_2 and water. Reducing equivalents generated from oxidation of glucose pass electrons through the mitochondrial electron transfer chain (ETC), building up a proton gradient

to drive the phosphorylation of adenosine 5'-diphosphate (ADP) to adenosine 5'-triphosphate (ATP), a process called "oxidative phosphorylation". The electrons ultimately react with O_2 to form H_2O, a fully reduced form of O_2. In this process, O_2 serves as a terminal oxidant (receiver of electrons) Du *et al.* (2000). This process is highly energy-efficient and is superior to fermentation or respiratory pathways that rely on other terminal oxidants King and Loeken (2004). However, the utilization of O_2 as a terminal substrate in oxidative phsophorylation is risky. When electron pass through mitochondrial ETC, a fraction of them (0.1–0.5%) escape the ETC and combines with O_2 prematurely, resulting in the generation of partially reduced product-superoxide (O_2^-). Superoxides and other partially reduced form of oxygenare reactive oxygen species (ROS) that can react with biomolecules and interfere with biological processes.

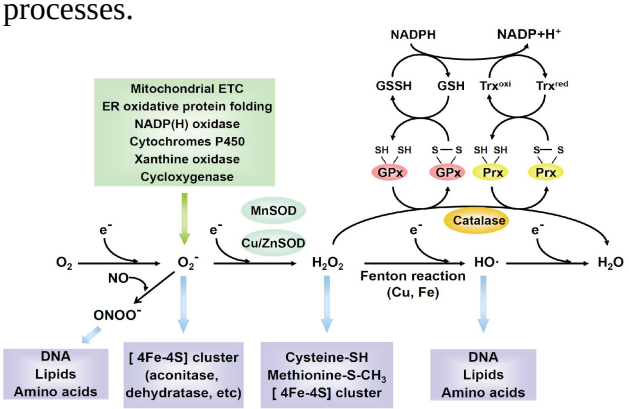

Figure.3 The cellular origins of reactive oxygen species

Medications

Chemical medication

- **Sulphonylureas**

They remain the mainstay of the treatment of type II diabetes in older people. They stimulate the release of insulin in the pancreas. Patients using sulphonylureas need counseling regarding the taking of regular meals and the signs and symptoms of hypoglycaemia, because the drug causes hypoglycaemia. If possible it should not be given to the elderly because they do not eat enough due to many problems associated with their age. Sulphonylureas causes an increase in weight and it should therefore be used by thin or normal weight patients.

- **Metformin**

It acts by reducing glucose production by the liver. Overweight patients can use this since there is less risk of gaining weight. It also lowers cholesterol concentrations, and does not cause hypoglycaemia.

- **Biguanides**

The term biguanide refers to a group of oral type 2 diabetes drugs that work by preventing the production of glucose in the liver, improving the body's sensitivity towards insulin reducing the amount of sugar absorbed by the intestines. The only available biguanide medication is metformin, which is commonly used as a first-line treatment for type 2 diabetes. Metformin is usually prescribed as a single treatment (monotherpay), but it can also be combined with other medication in a single tablet - for example, metformin + pioglitazone (Competact), metformin + vildagliptin (Eucreas) and metformin + sitagliptin (Janumet). It's also sometimes prescribed in combination with insulin for people with type 1 diabetes.

- **α-Glucosidase inhibitors**

The drug delays the absorption of glucose from a carbohydrate-containing meal. It

reduces the fluctuation in daily blood glucose levels and can be used by non-insulin dependent diabetic patients. A patient, whose blood glucose remains high in spite of the diet and the oral anti-diabetic drugs, may be given insulin injections.

- **Thiazilidinediones (Tzds)**

This is a new drug Commonly called glitazones or actos (e.g. pioglitazone), thiazolidinediones lower blood glucose by increasing the sensitivity of your body's cells to insulin, so more glucose is taken into cells for the same amount of insulin in the bloodstream. They are not usually used alone, but are an option to take in addition to metformin or a sulfonylurea. Actos is effective as monotherapy and reduces the risk of microvascular complications by 2.6%.

- **Amylin analogues**

Amylin analogues, or agonists, are injectable drugs used in the treatment of both type 1 diabetes and type 2 diabetes. These compounds are administered before meals, and work similarly to the hormone amylin. Amylin has a number of benefits in terms of weight loss and reducing blood glucose levels.

- **Humulin**

Humulin is synthesized in a laboratory strain of Escherichia coli bacteria which has been genetically altered with recombinant DNA to produce biosynthetic human insulin. Humulin R consists of zinc-insulin crystals dissolved in a clear fluid. The synthesized insulin is then combined with other compounds or types of insulin which affect its shelf life and absorption. For example, Humulin N is combined with protamine to extend the time-activity profile of Humulin R for an extended period. Humulin by itself is short-acting insulin. HumulinR, like many other form of injectable insulin, is intended for subcutaneous injection, so it should not be used intramuscularly, since its dispersion in the rest of the body would take more time.

Types of humulin:

Humulin R [REGULAR human insulin injection (rDNA origin)] is a short-acting insulin that has a relatively short duration of activity as compared with other insulins.

Humulin R Regular U-500 (Concentrated) insulin human injection, USP (rDNA Origin) is a stronger concentration (500 units/mL) of Humulin R.

Humulin N [human NPH insulin injection (rDNA origin)] is an intermediate-acting insulin with a slower onset of action and a longer duration of activity than Humulin R.

Humulin 70/30 [70% human insulin isophane suspension, 30% human insulin injection (rDNA origin)] is a mixture insulin. It is an intermediate-acting insulin combined with the onset of action of Humulin

Humulin 50/50 [50% human insulin isophane suspension, 50% human insulin injection (rDNA origin)] is a mixture insulin. It is an intermediate-acting insulin combined with the onset of action of Humulin R

Herbal medication

Because of these limitations there is continued need for new and more effective therapies. Dietary measures and traditional

plant therapies prescribed by ayurvedic and other indigenous systems of medicine were used commonly in India and other countries. A multitude of herbs spices and other plant materials such as *fenugreek seed, Momordica charantia, Eugenia jambolana, Aloevera* and *Aloe barbadensis, Allium sativum,* holy basil, amla, and cinnamon have been described for the treatment of diabetes throughout the world. The medicinal plants might provide a useful source of new oral hypoglycemic compound for development of pharmaceuticals entities or as a dietary adjunct to existing therapies. Even WHO has also recommended the evaluation of safe modern drugs.

Apart from their nutritive value, various herbal preparations serve as prophylactic or therapeutic agents in various disorders like inflammation, would healing, cancer, diabetic neuropathy, and osteoporosis. Thus it has been experimentally proved, *in vitro* and *in vivo* that it is effective to treat certain allergies, anemia, cancer, hepatotoxicity, viral, cardiovascular disease, hyperglycemia, hyperlipidemia, immunodeficiency and inflammatory processes, among others. Several of these activities are attributed to some of it components including fatty acids, omega-3 or omega-6, β-carotene, alpha-tocopherol, phycocyanin, phenol compounds and recently isolated complex Ca-Spirulan.

Trigonella foenumgraecum (fenugreek)

Fenugreek is a legume that grows in India, North Africa and Mediterranean regions. Its seeds are rich in alkaloid trigonelline, nicotinic acid and coumari, 4-hydroxyleucine, a novel amino acid increased glucose stimulated insulin release by isolated islet cells in both rats and humans. Animal and human studies have reported that the legume lowers blood glucose and lipid levels, as well as increases HDL cholesterol levels (Ribes *et al.,* 1984). Oral administration of 2 and 8 g/kg of plant extract produced dose dependent decrease in the blood glucose levels in both normal as well as diabetic rats. Administration of fenugreek seeds also improved glucose metabolism and normalized creatinine kinase activity in heart, skeletal muscle and liver of diabetic rats. It also reduced hepatic and renal glucose-6-phosphatase and fructose -1,6-biphosphatase activity. This plant also shows antioxidant activity.

Figure.4 Seed of fenugreek

Momordica charantia (bitter gourd)

Momordicacharantia is commonly used as an antidiabetic and antihyperglycemic agent in India as well as other Asian countries. Extracts of fruit pulp, seed, leaves and whole plant was shown to have hypoglycemic effect in various animal models. *Polypeptide p,* isolated from fruit, seeds and tissues of *M. charantia* showed significant hypoglycemic effect when administered subcutaneously to langurs and humans. Ethanolic extracts of *M. charantia* (200 mg/kg) showed an antihyperglycemic and also hypoglycemic effect in normal and STZ diabetic rats (Khanna *et al.,* 1981). This may be because of inhibition of glucose-6-phosphatase besides fructose-1, 6-biphosphatase in the liver and stimulation of hepatic glucose-6-phosphate dehydrogenase activities.

Eugenia jambolana **(Indian gooseberry, jamun)**

In India decoction of kernels of *Eugenia jambolana* is used as household remedy for diabetes. This also forms a major constituent of many herbal formulations for diabetes. Antihyperglycemic effect of aqueous and alcoholic extract as well as lyophilized powder shows reduction in blood glucose level. This varies with different level of diabetes. The extract of jamun pulp showed the hypoglycemic activity in streptozotocin induced diabetic mice within 30 min of administration while the seed of the same fruit required 24 h. The oral administration of the extract resulted in increase in serum insulin levels in diabetic rats (Acherekar *et al.,* 1997). Insulin secretion was found to be stimulated on incubation of plant extract with isolated islets of Langerhans from normal as well as diabetic animals. These extracts also inhibited insulinase activity from liver and kidney.

Aloe vera

Aloe, a popular houseplant, has a long history as a multipurpose folk remedy. The plant can be separated into two basic products: gel and latex. *Aloe vera* gel is the leaf pulp or mucilage, aloe latex, commonly referred to as "aloe juice," is a bitter yellow exudate from the pericyclic tubules just beneath the outer skin of the leaves. Extracts of aloe gum effectively increases glucose tolerance in both normal and diabetic rats (Al-Awadi, 1987).

Allium sativum **(garlic)**

This is a perennial herb cultivated throughout India. Allicin, a sulfur-containing compound is responsible for its pungent odour and it has been shown to have significant hypoglycemic activity (Sheela and Augusti, 1992). This effect is thought to be due to increased hepatic metabolism, increased insulin release from pancreatic beta cells and/or insulin sparing effect (Bever and Zahnd, 1979). S-allylcysteinsulfoxide (SACS), the precursor of allicin and garlic oil, is a sulfur containing amino acid, which controlled lipid peroxidation better than glibenclamide and insulin. It also improved diabetic conditions. SACS also stimulated in vitro insulin secretion from beta cells isolated from normal rats (Augusti and Sheela, 1992).

Ocimum sanctum L. **(Holy basil)**

It is commonly known as Tulsi. Since ancient times, this plant is known for its medicinal properties. The aqueous extract of leaves of *Ocimum sanctum* showed the significant reduction in blood sugar level in both normal and alloxan-induced diabetic rats (Significant reduction in fasting blood glucose, uronic acid, total amino acid, total cholesterol, triglyceride and total lipid indicated the hypoglycemic and hypolipidemic effects of tulsi in diabetic rats (Oral administration of plant extract (200 mg/kg) for 30 days led to decrease in the plasma glucose level by approximately 9.06 and 26.4% on 15 and 30 days of the experiment, respectively. Renal glycogen content increased 10 fold while skeletal muscle and hepatic glycogen levels decreased by 68 and 75% respectively in diabetic rats as compared to control. This plant also showed antiasthemitic, antistress, antibacterial, antifungal, antiviral, antitumor, gastric antiulcer activity, antioxidant, antimutagenic and immunostimulant activities.

Emblica officinali**s (Amla)**

Amla is a rich natural source of vitamin C. It

is used as anti-diabetic. It contains 0.5% fat, phyllemblin, 5% tannin. It also contains phosphorus, iron & calcium. It contains pectin & 75% moisture**.**

Cinnamom umzeylanicum (Cinnamon)

It consists of dried inner bark of shoots of coppiced trees of *Cinnamom umzeylanicum***,** belonging to the family Lauraceae. Cinnamon bark contains volatile oil, tannins, mucilage, calcium oxalate, starch & mannitol. Cinnamon oil contains cinnamaldehyde, other terpenes like phellandrene, pinene, cymene, caryophyllene. Cinnamon is used in the treatment of type II diabetes mellitus & insulin resistance.

Figure.5 Powder of cinnamon

References

Abel, E.D., Peroni, O, Kim, J.K., Kim, Y.B., Boss, O., Hadro, E., Minnemann, T., Shulman, G.I., Kahn, B.B. 2001. Adipose selective targeting of the GLUT4 gene impairs insulin action in muscle and liver. *Nature*, 409: 729–733.

Acherekar, S., Kaklij, G.S., Kelkar, S.M. 1991. Hypoglycemic activity of Eugenia Jambolana and ficusbangalensis: mechanism of action. *In vivo*, 5: 153–147.

Ahuja, M.M.S. 1979. Epidemiology studies on diabetes mellitus in India. *Nature*, 409: 729–733.

Al-Awadi, F.M., Gumma, K.A. 1987. Studies on the activity of individual plants of an antidiabetic plant mixture. *Acta Diabetol.*, 24: 37–41.

Altarejos, J.Y., Montminy, M. 2011. CREB and the CRTC co-activators: sensors for hormonal and metabolic signals. *Nat. Rev. Mol. Cell Biol.*, 12: 141–151.

Anderwald, C., Roden, M. 2004. Adipotoxicity and the insulin resistance syndrome. *Pediatar. Endocrinol. Rev.*, 1: 310–319.

Augusti, K.T., Sheela, C.G. 1992. Antidiabetic effects of S-allyl cysteine sulphoxide isolated from garlic Allium sativum Linn. *Indian J. Exp. Biol.*, 30: 523–526.

Bandyopadhyay, G.K., Standaert, M.L., Zhao, L., Yu B., Avignon, A., Galloway, L., Karnam, P., Moscat J., Farese, R.V. 1997. Activation of protein kinase (α, β, and ξ) by insulin in 3T3–L1 cells: transfection studies suggest a role for PKC-zeta in glucose transport. *J. Biol. Chem.*, 272: 2551–2558.

Bever, B.O., Zahnd, G.R. 1979. Plants with oral hypoglycemic action. *Quart. J. Crude Drug Res.*, 17: 139–146.

Boden, G., Shulman, G.I. 2002. Free fatty acids in obesity and type 2 diabetes: defining their role in the development of insulin resistance and beta-cell dysfunction. *Eur. J. Clin. Invest.*, 3: 14–23.

Chang, Y., Chuang, L. 2010. Review article the role of oxidative stress in the pathogenesis of type 2 diabetes: from molecular mechanism to clinical implication. *Am. J. Transl. Res.*, 2: 316–331.

Curcio, F., Ceriello, A. 1992. Decreased cultured endothelial cell proliferation in high glucose medium is reversed by antioxidants. *In Vitro Cell. Deve. Biology.*, 45: 787–790.

De Fea, K., Roth, R.A. 1997. Modulation of insulin receptor substrate-1 tyrosine phosphorylation and function by

mitogen activated protein kinase. *J. Biol. Chem.*, 272: 31400–31406.

Draznin, B. 2006. Molecular mechanisms of insulin resistance: serine phosphorylation of insulin receptor substrate-1 and increased expression of p85alpha: the two sides of a coin. *Diabet.*, 55: 2392–2397.

Du, X.L., Edelstein, D., Rossetti, L., Fantus, I.G., Goldberg, H., Ziyadeh, F., Wu J., Brownlee, M. 2000. Hyperglycemia-induced mitochondrial superoxide overproduction activates the hexosamine pathway and induces plasminogen activator inhibitor-1 expression by increasing Sp1 glycosylation. *Proc. Natl. Acad. Sci. USA.*, 55:12222–12226.

Eizirik, D.L., Pipeleers, D.G., Ling, Z., Welsh, N., Hellerstrom, C., Andersson, A. 1994. Major species differences between humans and rodents in the susceptibility to pancreatic beta-cell injury. *Proc. Natl. Acad. USA.*, 91: 9253–9256.

Eldar-Finkelman, H., Krebs, E.G. 1997. Phosphorylation of insulin receptor substrate 1 by glycogen synthase kinase 3 impaires insulin action. *Proc. Natl. Acad. Sci. USA.*, 94: 9660–9664.

Fain, J.N., Madan, A.K., Hiler, M.L., Cheema, P., Bahouth, S.W. 2004. Comparison of the release of adipokines by adipose tissue, adipose tissue matrix, and adipocytes from visceral and subcutaneous abdominal adipose tissues of obese humans. *Endocrinology.*, 145: 2273–2282.

Ganda, O.P., Rossi, A.A., Like, **A.A**. 1976. Studies on streptozotocin diabetes. *Diabetes.*, 25: 595–603.

Ganz, M.B., Seftel, A. 2000. Glucose-induced changes in protein kinase C and nitric oxide are prevented by vitamin E. *Am. J. Physiol. Endocrinol. Metab.*, 278: E146–E152.

Gao, z., Hwang, D., Bataille, F., Lefevre, M., York, D., Quon, M.J., Ye, J. 2002. Serine Phosphorylation of insulin receptor substrate 1 by inhibitor kappa B kinase complex. *J. Biol. Chem.*, 277: 48115–48121.

Geraldes, P., King, G.L. 2010. Activation of protein kinase C isoforms and its impact on diabetic complications. *Circ. Res.*, 106: 1319–31.

Gruppuso, P.A., Boylan, J.M., Posner, B.I., Faure, R., Brautigan, D.L. 1990. Hepatic protein phosphotyrosine phosphatase. Dephosphorylation of insulin receptor and epidermal growth factor receptors in normal and alloxan diabetic rats. *J. Clin. Invest.*, 85: 1754–1760.

Gurevitch, D., Boura-Halfon, S., Issac, R., Alberstkein, M., Rnen, D., Lewis, E., Zick, Y. 2008. Phosphorylation of selected serines at the PBT domain of IRS-2 regulates beta cell growth, survival, and insulin secretion. In: Keystone Symposia on Islet and beta-cell Development and Transplantation.

Hotamisligil, G.S., Shargill, N.S., Spiegelman, B.M. 1993.Adipose expression of tumor necrosis factor-alpha: direct role in obesity-linked insulin resistance. *Science*, 259: 87–91.

Hruz, P., Mueckler, M. 1999.Cysteine-scanning mutagenesis of transmembrane segment 7 of the GLUT1 glucose transporter. *J. Biol. Chem.*, 274: 36176–80.

James, D.E., Strube, M., Mueckler, M., 1989. Molecular cloning and characterization of an insulin-regulatable glucose transporter. *Nature*, 338: 83–7.

Kahn, S.E., Hull, R.L., Utzschneider, K.M. 2006. Mechanisms linking obesity to insulin resistance and type 2 diabetes. *Nature*, 444: 840–846.

Katsumata, K., Katsumata, K. Jr., Katsumata, Y.1992. Protective effect of diltiazem hydrochloride on the occurrence of alloxan- or streptozotocin-induced diabetes in rats. *Horm, Metab. Res.*, 24: 508–510.

Khanna, P., Jain, S.C., Panagariya, A., Dixit, V.P. 1981. Hypoglycemic activity of polypeptide- p from a plant source. *J. Nat. Prod.*, 44: 648–655.

Kim, J.A., Yeh, D.C., Ver, M., Li, Y., Carranza, A., Conrads, T.P., Veenstra, T.D., Harrington, M.A., Quon, M.J. 2005. Phosphorylation of Ser24 in the pleckstrin homology domain of insulin receptor substrate-1 by Mouse Pelle-like kinase/interleukin-1 receptor associated kinase: cross-talk between inflammatory signaling and insulin signaling that may contribute to insulin resistance. *J. Biol. Chem.*, 280: 23173–23183.

King, G.L., Loeken, M.R. 2004. Hyperglycemia-induced oxidative stress in diabetic complications. *Histochem. Cell Biol.*, 128: 333.

Kiselyov, V.V., Versteyhe, S., Gauguin, L., De Meyts, P. 2009. Harmonic oscillator model of the insulin and IGF1 receptors' allosteric binding and activation. *Mol. Syst. Biol.*, 5: 243–250

Kuboki, K., Jiang, Z.Y., Takahara, N., Ha, S.W., Igarashi, M., Yamauchi, T., Feener, E.P., Herbert, T.P., Rhodes, C.J., King, G.L. 2000. Regulation of endothelial constitutive nitric oxide synthase gene expression in endothelial cells and in vivo: a specific vascular action of insulin. *Circulation*, 101: 676–681.

Layden, B.T., Durai, V., Lowe, W.L. Jr. 2010. G-Protein-Coupled Receptors, Pancreatic Islets, and Diabetes. *Nature Educ.*, 3: 9–13.

Lee, S., Lynn, E.G., Kim, J.A., Quon, M.J. 2008. Protein kinase C-ζ phosphorylates insulin receptor substrate-1, -3, and -4, but not -2: Isoform specific determinants of specificity in insulin signaling. *Endocrinology*, 149: 2451–2458.

Liberman, Z., Eldar-Finkelman, H. 2005. Serine 332 Phosphorylation of insulin receptor substrate-1 by glycogen synthase kinase-3 attenuates insulin signaling. *J. Biol. Chem.*, 280: 4422–4428.

Maritim, A.C., Sanders, R.A., Watkins, J.B. 2003.Oxidative stress in diabetics. *J. Biochem. Mol. Toxicol.*, 17(1): 24–38.

Morino, K., Petersen, K.F., Shulman, G.I. 2006.Molecular mechanisms of insulin resistance in humans and their potential links with mitochondrial dysfunction. *Diabetes*, 2: 9–15.

Najjar, S. 2001. Insulin action: Molecular basis of diabetes. Encyclopedia of life sciences (John wiley& Sons). doi: 10.1038/npg.els.0001402

Nakaki, T., Nakadate, T., Kato, R. 1980. Alpha 2-adrenoceptores modulating insulin release from isolated pancreatic islets. *Naunyn Schmiedebergs Arch. Pharmacol.*, 313: 151–153.

Oka, Y., Asano, T., Shibasaki, Y., Lin, J., Tsukuda, K., Katagiri, H., Akanuma, Y., Takaku, F. 1990. C-terminal truncated glucose transporter is locked into an inward-facing form without transport activity. *Nature.*, 345: 550–553.

Pla, G. 1976.Drugs and lipid peroxidation. *Ann. Rev. Pharmacol.*, 3: 16–125.

Ramachandran, A., Snehalatha, C., Kapur, A. 2001. Diabetes Epidemiology Study Group in India (DESI): high prevalence of diabetes and impaired glucose tolerance in India. National urban Diabetes Surbey. *Diabetologia*, 44: 1094–1101.

Ribes, G., Sauvaire, Y., Baccou, J.C., Valette, G., Chenon, D., Trimble, E.R., Marian, M.M.L**.** 1984. Effects of fenugreek seeds on endocrine pancreatic secretion in dogs. *Ann. Nutr. Metab.*, 28: 37–43.

Robert, K., Daryl, K., Victor, W.1998. Biological Oxidation, In: Biochemistry., 22nd edn. Elsevier, New York, 111 Pp.

Sampson, S.R., Cooper, D.R. 2006. Specific protein kinase C isoforms as transducers and modulators of insulin

signaling. *Mol. Genet. Metab.*, 89: 32–47.

Schatteman, G.C., Hanlon, H.D., Jiao, C., Dodds, S.G., Christy, B.A. 2000.Blood-derived angioblasts accelerate blood-flow restoration in diabetic mice. *J. Clin. Invest.*, 106: 571–578.

Sharfi, H., Eldar-Finkelman H. 2008**.** Sequential phosphorylation of insulin receptor substrate-2 by glycogen synthase kinase-3 and c-Jun NH2-terminal kinase plays a role in hepatic insulin signaling. *Am. J. Physicol. Endocrinol. Metab.*, 294: E307–E315.

Shepherd, P.R., Nave, B.T., Siddle, K. 1995. Insulin stimulation of glycogen synthesis and glycogen synthase activity is blocked by wortmannin and rapamycin in 3T3–L1 adipocytes: evidence for the involvement of phosphoinositide 3-kinase and p70 ribosomal protein-S6 kinase. *Biochem. J.*, 305: 25–28.

Shoelson, S.E., Lee, J., Goldfine, A.B. 2006. Inflammation and insulin resistance. *J. Clin. Invest.*, 116: 1793–1801.

Sircar, S. 2007. Medical physiology. Thieme Publishing Group, Stuttgart. 537–538 Pp.

Smeltzer, S., Bare, B.G. 1992. Overview of study on diabetes. *Med. Surg. Nurs. Suzanne C.*,10: 1022–1025.

Stitt, A.W., Li, Y.M., Gardiner, T.A., Bucala, R., Archer, D.B., Vlassara, H. 1997. Advanced glycation end products (AGEs) co-localize with AGE receptors in the retinal vasculature of diabetic and of AGE-infused rats. *Am. J. Pathol.*, 150: 523–31.

Um, S.H., Frigerio, F., Watanabe, M., Picard, F., Joaquin, M., Sticker, M., Fumagalli, S., Allegrini, P.R., Kozma, S.C., Auwerx, J., Thomas, G. 2004. Absence of S6K1 protects against age- and diet-induced obesity while enhancing insulin sensitivity. *Nature*, 431: 200–205.

Ward, C.W., Lawrence, M.C. 2009. Ligand induced activation of insulin receptor. A multi step process involving structural changes in both the ligand and the receptor. *Bioassay.*, 31: 422–434.

Wautier, J.L., Schmidt, A.M. 2004. Protein glycation: a firm link to endothelial cell dysfunction. *Circ. Res.*, 95: 233–238.

Williams, B., Gallacher, B., Patel, H., Orme, C. 1997. Glucose-induced protein kinase C activation regulates vascular permeability factor mRNA expression and peptide production by human vascular smooth muscle cells in vitro. *Diabetes*, 46: 1497–1503.

World Health Organization Consultation Group. 1998. Diagnosis and classification of diabetes mellitus. Provisional report of a WHO Consultation. *Diabet. Med.*, 15: 539–553.

Yang, H., Jin, X., Kei Lam, C.W., Yan, S.K. 2011. Oxidative stress and diabetes mellitus. *Clin. Chem. Lab. Med.*, 49: 1773–1782.

Zimmet, P., Unwin, N., Shaw, J., Alberti, K.G. 2002. Impaired glucose tolerance and impaired fasting glycaemia: the current status on definition and intervention. *Diabet Med.*, 19: 708–23.