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Diabetes mellitus is a metabolic disorder that either arrives during the early years of growth (Juvenile diabetes) or later in life or is called as maturity onset diabetes. It is observed as the body’s inability to effectively regulate the sugar balance which leads to severe complications such as hyperglycaemia, obesity, neuropathy, nephropathy, retinopathy, cardiopathy, osteoporosis and coma leading to death. Pancreatic damage resulting in the dysfunction of α and β cells causes disordered glucose homeostasis. In diabetic individuals the regulation of glucose levels by insulin is defective, either due to defective insulin production which is called as Insulin Dependent Diabetes Mellitus (IDDM) or due to insulin resistance that is termed as Non Insulin Dependent Diabetes Mellitus (NIDDM).

Diabetes and Brain

Glucose is the only fuel that the neuronal tissue can use for energy under normal circumstances (Sokoloff, 1981). The brain can neither synthesize nor store more than a few minutes worth of glucose; thus a continuous systemic supply is essential for normal cerebral metabolism (Pardridge, 1983). Chronic changes in the antecedent level of glycaemia (either sustained hyperglycaemia or hypoglycaemia) induce alterations in brain glucose metabolism in rodents (McCall et al., 1982; Boyle et al., 1994). Diabetes mellitus is a major global health problem that affects more than 185 million people around the world (Amos et al., 1997; Zimmet, 1999; Zimmet et al., 2001). This disease is an increasingly prevalent metabolic disorder in humans and is characterised by hyperglycaemia (Kumar & Clarke, 2002; Dunne et al., 2004). The number of diabetic patients is expected to reach 300 million by the year 2025. The projected increase in the number of diabetic patients will strain the capabilities of healthcare providers the world over (Adeghate et al., 2006). The pancreatic hormones
have an important role in the regulation of glucose metabolism. The secretion of insulin by β-cells of the endocrine pancreas is regulated by glucose and other circulating nutrients. It is also modulated by several hormones and neurotransmitters, among which serotonin and glutamate plays a prominent role. DM is accompanied by an altered monoamine neurotransmission in the brain, mostly manifested by an increase in content and a decrease in turnover rate (McCall, 1992; Biessels et al., 1994). However, the changes are not generalized, but are regionally specifically distributed, with the appearance dependent on the duration of diabetes (Bitar et al., 1985; Chu et al., 1986; Oliver et al., 1989; Lacković et al., 1990; Ramakrishnan & Namasiyam, 1995). The specific regional character of these changes, particularly in the hypothalamus and the striatum, suggests functional correlations with the distinctive behavioural disorders like increased feeding. Speculation that an adaptation in the CNS might exist in patients with diabetes, depending upon antecedent glycaemia, appeared nearly a decade ago (Cryer & Gerich, 1985; Cryer, 2003). Hormonal defects with unawareness of symptoms can be induced in patients with diabetes (Hepburn et al., 1991; Dagogo et al., 1993) and non-diabetics (Davis & Shamoon, 1991; Heller & Cryer, 1991; Veneman et al., 1993). Sleep, submissive and avoidance behaviour, depression, decreased sexual and aggressive behaviour, spontaneous locomotor activity, and cognitive dysfunctions that are observed in diabetes (Leedom & Mechan, 1989; Lustman et al., 1992; Biessels et al., 1994).

β-cells ability to proliferate in response to rising blood glucose concentrations is remarkably well preserved during severe, chronic beta-cell autoimmunity. Control of the destructive immune response after disease manifestation allows spontaneous regeneration of sufficient β-cell mass to restore normal glucose homeostasis (Pechhold et al., 2009b). Declining glucose levels in the brain stimulate the autonomic nervous system, causing epinephrine and nor epinephrine to be released from the adrenal medulla. Nor epinephrine and acetylcholine from the sympathetic nervous
system is also involved in glucose control. Symptoms occur as these hormones and neurotransmitters simultaneously stimulate α-cells in the pancreas to release glucagon, which consequently induces new glucose production in the liver (Cryer, 1999, 2002a, b, 2003). In this homeostatic mechanism, rising blood glucose levels shut down the neoglucogenesis activities of autonomic nervous system (Towler et al., 1993; Cryer, 1997; McAulay et al., 2001, Charles & Goh, 2005).

Diabetes, Oxidative Stress and Antioxidants

Oxidative stress is produced during normal metabolic process in the body as well as induced by a variety of environmental factors and chemicals. Oxidative stress has been shown to have a significant effect in the cause of diabetes as well as diabetes related complications in human beings (Wilson, 1998). Oxidative stress plays a central role in the pathogenesis of metabolic diseases like diabetes mellitus and its complications (like peripheral neuropathy) as well as in neurodegenerative disorders like Alzheimer's disease (AD) and Parkinson's disease (PD) (Biörkhem et al., 2009; Eggers, 2009). Oxidative stress in diabetes has shown to co-exist with a reduction in the antioxidant status. (Trevisan et al., 2001; Waden et al., 2009). Oxidative stress has shown to produce glycation of proteins, inactivation of enzymes, alterations in structural functions of collagen basement membrane (Boynes, 1991). Oxidative stress may have significant effect in the glucose transport protein (GLUT) or in insulin receptor (Jacqueline et al., 1997). Scavengers of oxidative stress may have an effect in reducing the increased serum glucose level in diabetes and may alleviate the diabetes as well as reduce its secondary complications. Aegle marmelose extract, which is being used in the traditional medicine to reduce the serum glucose level has significant antioxidant activity in vitro (Sabu & Kuttan, 2000). Alloxan, which is an accepted model for the induction of diabetes, has been shown to damage islet cells of pancreas by the liberation of oxygen radicals (Halliwell & Gutteridge, 1985).
Increasing evidence in both experimental and clinical studies suggests that oxidative stress plays a major role in the pathogenesis 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. (Kawamura et al., 1992; Morgan et al., 2002). Abnormally high levels of free radicals and the simultaneous decline of antioxidant defense mechanisms leads to damage of cellular organelles and enzymes, increased lipid peroxidation and development of insulin resistance. Plasma oxidized LDL, a commonly used marker for oxidative stress, is involved in the development of diabetes- and obesity-related traits (Wen-Chi Hsueh et al., 2009). 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, nitrite concentration, nonenzymatic glycosylated proteins, and hyperglycemia in diabetes, and their consequences (Yan & Harding, 1997; Maritim et al., 2003). The insulin signaling cascade constitutes a complex signaling network, adequately activated, in the induction of diverse biological functions. Insulin resistance is the reduced capacity of insulin to induce its biological actions in its target organs. Oxidants are commonly generated by various potential inducers of insulin resistance (Bashan et al., 2009). Even in a healthy population, variations in insulin sensitivity are related to lipid hydroperoxyde levels, reduced catalase and Vitamin E levels (Facchini et al., 2000). ROS oxidize various types of biomolecules, finally leading to cellular lesions by damaging DNA or stimulating apoptosis for cell death (Kaneto et al., 1994).

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related complications in human beings (Wilson, 1998). Oxidative stress in diabetes has been shown to co-exist with a reduction in the antioxidant status. The exact role of oxidative stress in the etiology of human diabetes is however not known. Oxidative stress has been shown to produce glycation of proteins, inactivation of enzymes, alterations in structural functions of collagen basement membrane (Boynes, 1991). Oxidative stress have significant effect in the glucose transport protein (GLUT) or in insulin receptor (Jacqueline et al., 1997). Scavengers of oxidative stress have an effect in reducing the increased serum glucose level in diabetes and alleviate the diabetes as well as reduce its secondary complications. Aegle marmelose extract, which is being used in the traditional medicine to reduce the serum glucose level has significant antioxidant activity invitro (Sabu & Kuttan, 2000). Alloxan, which is an accepted model for the induction of diabetes, has been shown to damage islet cells of pancreas by the liberation of oxygen radicals (Halliwell & Gutteridge, 1985).

β-Cell function: Physiology and Pathophysiology

Islets of Langerhans are microscopic organelles scattered diffusely throughout the pancreas. Each islet contains approximately 2000 cells, which include four types: α, β, δ, and PP cells. The major secretory products of these cells are glucagon, insulin, somatostatin, and pancreatic polypeptide, respectively. The α-cell secretes glucagon primarily in response to hypoglycemia, but also to amino acids. The β-cell secretes insulin in response to elevated glucose levels. Insulin response to intravenous glucose are time-dependent and referred to as first- and second-phase responses. The δ-cell releases somatostatin in response to glucose. The PP cell releases pancreatic polypeptide in response to hypoglycemia and secretin. The functions of these hormones are distinctly different. Glucagon stimulates glycogenolysis in the liver to increase blood glucose levels. Insulin decreases hepatic glucose production and increases glucose entry into muscle and fat cells. Somatostatin inhibits the secretion of
many hormones, including insulin and glucagon and likely is an intra islet paracrine regulator of α and β cells. The function of pancreatic polypeptide in humans remains unclear (Robertson & Harmon, 2006; Nakatsuji et al., 2009).

The endocrine pancreas is richly innervated, but the abundance and organization of these innervations are highly variable between species (Kobayashi & Fujita, 1969). Most of the nerve fibers enter the pancreas along the arteries (Woods & Porte, 1974; Miller, 1981). Unmyelinated nerve fibers are found in the neighborhood of all islet cell types at the periphery and within the islet. At some distance from the islets, glial Schwann cells often form a thin sheet around nerve fibers on their travel toward and within the islet. In the vicinity of islet cells, however, it is not rare to see some nerve fibers lacking this glial protection and coming close to or ending blindly 20–30 nm from the endocrine cells (Legg, 1967; Watari, 1968; Kobayashi & Fujita, 1969; Shorr & Bloom, 1970; Fujita & Kobayashi, 1979; Bock, 1986; Radke & Stach (a), 1986; Radke & Stach (b), 1986). Alterations in induced and spontaneous autoimmune diabetes became apparent at diabetes onset, and differed markedly within islets compared with sub-islet-sized endocrine cell clusters and among pancreatic lobes. These changes are adaptive in nature, possibly fueled by worsening glycemia and regenerative processes (Pechhold et al., 2009a). The autonomic innervation of the endocrine pancreas has several origins. The autonomic nervous system uses two interconnected neurons to control effector functions and is divided into two systems, the sympathetic and the parasympathetic nervous systems, according to the location of the preganglionic cell bodies. However, there are indications suggesting that these two systems are not always independent of each other, but display anatomical interactions (Berthoud & Powley, 1993) or share similar neurotransmitters (Sheikh et al., 1988; Verchere et al., 1996; Liu et al., 1998).
The parasympathetic innervation

The preganglionic fibers of the parasympathetic limb originate from perikarya located in the dorsal motor nucleus of the vagus (Ionescu et al., 1983; Luiten et al., 1984; Ahrén et al., 1986; Rinaman & Miselis, 1987; Louis-Sylvestre, 1987; Berthoud et al., 1990; Berthoud & Powley, 1991; Chen et al., 1996) and possibly also in the nucleus ambiguus (Weaver, 1980; Sharkey & Williams, 1983; Sharkey et al., 1984; Luiten et al., 1984; Luiten et al., 1986) which are both under the control of the hypothalamus. They are organized in well separated branches travelling within the vagus nerves (cranial nerve X), and through the hepatic, gastric (Berthoud et al., 1990; Berthoud & Powley, 1991) and possibly celiac branches of the vagus (Kinami et al., 1997). They reach intrapancreatic ganglia that are dispersed in the exocrine tissue. These ganglia send unmyelinated postganglionic fibers toward the islets (Woods & Porte Jr, 1974; Berthoud et al., 1981; Berthoud & Powley, 1990). Preganglionic vagal fibers release ACh that binds to nicotinic receptors on intraganglionic neurons. Postganglionic vagal fibers release several neurotransmitters: ACh, Vasoactive Intestinal Peptide (VIP), gastrin-releasing peptide (GRP), nitric oxide (NO), and pituitary adenylate cyclase-activating polypeptide (PACAP) (Bloom & Edwards, 1981; Bloom et al., 1983; Knuhtsen et al., 1985; Ahrén et al., 1986; Knuhtsen et al., 1987; Ekblad et al., 1994; Sha et al., 1995; Havel et al., 1997; Ahrén et al., 1999; Love & Szebeni, 1999; Wang et al., 1999; Ahren, 2000; Myojin et al., 2000). Cholinergic terminals are found in the neighbourhood of all islet cell types at the periphery and within the islet (Coupland, 1958; Esterhuizen et al., 1968; Stach & Radke, 1982; Radke & Stach, 1986; Van der Zee et al., 1992; Love & Szebeni, 1999).
The sympathetic innervation

The sympathetic innervation of the pancreas originates from preganglionic perikarya located in the thoracic and upper lumbar segments of the spinal cord (Furuzawa et al., 1996). The myelinated axons of these cells traverse the ventral roots to form the white communicating rami of the thoracic and lumbar nerves that reach the paravertebral sympathetic chain (Chusid, 1979). Preganglionic fibers communicate with a nest of ganglion cells within the paravertebral sympathetic chain or pass through the sympathetic chain, travel through the splanchnic nerves, and reach the celiac (Sharkey & Williams, 1983; Fox & Powley, 1986; Brunicardi et al., 1995; Furuzawa et al., 1996; Ahrén, 2000) and mesenteric ganglia (Furuzawa et al., 1996). Ganglia within the paravertebral sympathetic chain, and the celiac and mesenteric ganglia, give off postganglionic fibers that eventually reach the pancreas. The existence of intrapancreatic sympathetic ganglia has also been reported (Liu et al., 1984; Luiten et al., 1986; Luiten et al., 1998). Pancreatic islets are innervated by autonomic fibres. Sympathetic neural cell bodies are located in the superior mesenteric and celiac ganglia, the splanchnic nerve and parasympathetic innervation comes from the vagus nerve (Cabrera-Vásquez et al., 2009). The preganglionic fibers release ACh that acts on nicotinic receptors on intraganglionic neurons, whereas the postganglionic fibers release several neurotransmitters: norepinephrine, galanin, (Ahrén & Taborsky, 1986; Dunning et al., 1988; Ahrén, 2000; Myojin et al., 2000). A rich supply of adrenergic nerves in close proximity of the islet cells has been observed in several mammalian species (Esterhuizen et al., 1968; Ahrén et al., 1981; Stach & Radke, 1982; Radke & Stach, 1986). Neurons found in the CNS and in the sympathetic nervous system serve as links between ganglia and the effected organs (Elseweidy et al., 2009).
**Brain neurotransmitter changes during diabetes**

Neurotransmitters have been reported to show significant alterations during hyperglycaemia resulting in altered functions causing neuronal degeneration. Neuropathic pain and neurons develop hyperexcitability in diabetic rats, attributed to disturbances in neurotransmitters pattern (Elseweidy et al., 2009). Neurotransmitters have been reported to show significant alterations during hyperglycaemia resulting in altered functions causing neuronal degeneration. A significant increase in the catecholamine contents and activity of metabolising enzymes has been reported in experimental diabetes (Gupta et al., 1992). Norepinephrine has been reported to increase in several brain regions during diabetes (Oreland & Shaskan, 1983; Fushimi et al., 1984; Chu et al., 1986; Wesselmann et al., 1988; Chen & Yang, 1991; Tassava et al., 1992), but a significant decrease in NE has been reported in hypothalamus (Ohtani et al., 1997), pons and medulla (Ramakrishna & Namasivayam, 1995). EPI levels were significantly increased in the striatum, hippocampus and hypothalamus of diabetic rats and these changes were reversed to normal by insulin treatment (Ramakrishna & Namasivayam, 1995). Streptozotocin-induced diabetes and acute insulin deficiency were demonstrated to result in increased content of EPI in the supra chiasmatic nucleus. In addition to this, a decreased turnover of dopamine in the ventromedial nucleus in diabetes was found to be reversed by insulin treatment (Oliver et al., 1989). These data indicate that experimental diabetes and acute insulin deficiency result in the rapid onset of detectable alterations in EPI and DA activity in specific hypothalamic nuclei. This can lead to the development of secondary neuroendocrine abnormalities known to occur in the diabetic condition. The DA content was increased in whole brain, (Lackovic et al., 1990; Chen & Yang, 1991) corpus striatum (Chu et al., 1986), cerebral cortex and hypothalamus of diabetic rats (Tassava et al., 1992; Ohtani et al., 1997). The plasma DA content was decreased in diabetic rats (Eswar et al., 2006). Serotonin (5-HT) content is increased in the brain
regions and hypothalamic nuclei (Lackovic et al., 1990; Chen & Yang, 1991), but there are reports suggesting a decrease in brain 5-HT content during diabetes (Sandrini et al., 1997; Sumiyoshi et al., 1997; Jackson & Paulose, 1999). Brain tryptophan was also reduced during diabetes (Jamnicky et al., 1991). Insulin treatment was reported to reverse this reduced tryptophan content to normal (Jamnicky et al., 1993). The cerebellar cortex, like all other motor structures, receives serotoninergic innervation in the form of a plexus of fine varicose fibers that do not face any differentiated postsynaptic element (Ungerstedt, 1971; Chan-Palay, 1975; Bishop and Ho, 1985; Trouillas & Fuxe, 1993). Serotonin is therefore acting in this structure as a paracrine agent, released through volume transmission. In vivo, local ionophoretic applications of serotonin have been shown to modify the spontaneous activity of the Purkinje cells, suggesting that serotonin is able to alter the input–output function of the cortex (Strahlendorf et al., 1988; Darrow et al., 1990; Kerr & Bishop, 1992). It has been reported that application of serotonin potentiates the inhibition of Purkinje cells by exogenous GABA (Strahlendorf et al., 1989, 1991; Kerr & Bishop, 1992) and inhibits their excitation by exogenous non-NMDA glutamatergic agonists in vivo (Hicks et al., 1989; Kerr & Bishop, 1992; Netzeband et al., 1993). Serotonin also modulates the potassium conductance activated by depolarization (Wang et al., 1992) as well as the cationic conductance activated by hyperpolarization (Li et al., 1993) recorded from Purkinje cells in cerebellar slices. Finally, serotonin may affect the efficiency of excitatory transmission at mossy fiber terminals (Maura et al., 1991; Lu & Larson-Prior, 1996) and parallel fiber synapses in the molecular layer (Raiteri et al., 1986; Maura & Raiteri, 1996). In this work we have combined electrophysiological recordings in thin slices of the rat cerebellar cortex with cell reconstruction and immunohistochemical methods to identify a new site of action of serotonin. During exercise, blood glucose fell with placebo but, unexpectedly, rose with exenatide. Plasma adrenaline (epinephrine) and noradrenaline (norepinephrine),
cortisol concentrations increased to a greater extent during exercise after exenatide (Khoo et al., 2010).

**Insulin secretion regulating factors**

**Glucose**

Glucose is an important regulator of various β-cell processes including insulin biosynthesis and release. Glucose, over short intervals stimulates insulin biosynthesis at the level of translation (Permut et al., 1972). Studies have shown that preproinsulin mRNA levels rise 4-10 folds in response to glucose stimulation. Studies of insulin gene expression in primary cultures of rat islets transfected Insulin I gene 5' flanking sequence suggested that metabolic signal from glucose influx is transmitted through the insulin enhancer (German et al., 1990).

Phosphorylation of glucose to glucose-6-phosphate serves as the rate limiting step in glucose oxidation (Schuit, 1996). Glucokinase acts as sensor during this process. The entry of glucose into β-cells is followed by an acceleration of metabolism that generates one or several signals that close ATP-sensitive K⁺ channels in the plasma membrane. The resulting decrease in K⁺ conductance leads to depolarisation of the membrane with subsequent opening of voltage dependent Ca²⁺ channels. The rise in the cytoplasmic free Ca²⁺ eventually leads to the exocytosis of insulin containing granules (Dunne, 1991; Gembal et al., 1992). Glucose induced insulin secretion is also partly dependent upon the activation of typical isoforms of protein kinase C within the β-cell (Harris, 1996). It is suggested that PKC is tonically active and effective in the maintenance of the phosphorylated state of the voltage-gated L-type Ca²⁺ channel, enabling an appropriate function of this channel in the insulin secretory process (Arkhammar, 1994).
**Fatty acids**

Short chain fatty acids and their derivatives are highly active stimulators of insulin release in sheep (Horino et al., 1968). Exogenous saturated long chain fatty acids markedly potentiated glucose-induced insulin release and elevated long chain acyl-CoA esters in the clonal β-cell line, HIT (Prentki et al., 1992). A novel ester of succinic acid 1, 2, 3-tri-(methyl-succinyl) glycerol ester displayed stimulation of insulin release and biosynthetic activity in pancreatic islets of Goto-Kakizaki rats (Laghmich et al., 1997). A monomethyl ester of succinic acid along with D-glucose is required to maintain the β-cell response to D-glucose (Fernandez et al., 1996).

**Amino acids**

Amino acids act as potent stimulators of insulin release. L-Tryptophan, which is the precursor of 5-Hydroxytryptamine (5-HT) act as a stimulator of insulin release (Bird et al., 1980). L-Arginine also stimulates insulin release from pancreatic β-cells. Several in vitro studies have suggested the production of nitric oxides from islet nitric oxide system have a negative regulation of the L-arginine induced secretion of insulin in mice.

**Substrates derived from nutrients**

Substrates like pyruvate (Lisa, 1994), citrate, ATP (Tahani et al., 1979), NADH and NADPH involve in the indirect reflux stimulation triggered by food intake or local islet stimulation through the production of metabolites. The NADH acts as an intracellular regulator of insulin secretion. Heterotrimeric GTP-binding protein G\(\beta\)i is involved in regulating glucose induced insulin release (Konrad et al., 1995). GTP analogues are also important regulators of insulin secretion (Lucia et al., 1987). Glucose induced insulin secretion is accompanied by an increase in the islet content of cAMP (Rabinovitch et al., 1976).
Glucagon

Glucagon is the hormone secreted by pancreatic α-cells. It has been shown that glucagon has a striking stimulatory effect on insulin release in the absence of glucose (Sevi, 1966). Cellular JNK2- and JNK1/JNK2-deficiency divert glucose from oxidation to glycogenesis due to increased glycogen synthase (GS) activity and induction of Pdk4 (Vijayvargia et al., 2010). The presence of specific glucagon receptors on isolated rat pancreatic β-cells as well as a subpopulation of α- and δ-cells shows the relevance of glucagon on regulation of insulin secretion. Intra-islet glucagon appears to be a paracrine regulator of cAMP in vitro (Schuit, 1996). Glucagon stimulates insulin release by elevating cAMP. cAMP through activation of protein kinase A, increases Ca\(^{2+}\) influx through voltage dependent L-type Ca\(^{2+}\) channels, thereby elevating Ca\(^{2+}\) and accelerating exocytosis (Carina, 1993). Protein phosphorylation by Ca\(^{2+}\)/Calmodulin and cAMP dependent protein kinase play a positive role in insulin granule movement which results in potentiation of insulin release from the pancreatic β-cell (Hisatomi, 1996).

Somatostatin

This hormone is secreted by the pancreatic δ-cells of the islets of Langerhans. Somatostatin inhibits insulin release. Its action is dependent on the activation of G-proteins but not associated with the inhibition of the voltage dependent Ca\(^{2+}\) currents or adenylate cyclase activity (Renstrom et al., 1996). Reports from our lab showed that Long-term low dose somatotropin and insulin treatment in regulating cholinergic and glutamergic receptors subtypes in ageing rats and rejuvenation of brain function (Savitha et al., 2010).
**Pancreastatin**

Pancreastatin is known to be produced in islet β-cells and to inhibit insulin secretion. Pancreastatin is a modulator of the early changes in insulin secretion after increase of glucose concentration within the physiological range (Ahren et al., 1996). It is reported to increase Ca\(^{2+}\) in insulin secreting RINm5F cells independent of extracellular calcium (Sanchez et al., 1992).

**Amylin**

Amylin is a 37-amino acid peptide hormone co-secreted with insulin from pancreatic β-cells. Amylin appears to control plasma glucose via several mechanisms that reduce the rate of glucose appearance in the plasma. Amylin limits nutrient inflow into the gut and nutrient flux from the gut to blood. It is predicted to modulate the flux of glucose from liver to blood by its ability to suppress glucagon secretion. Amylin is absolutely or relatively deficient in type I - diabetes and in insulin requiring type II - diabetes (Young, 1997). It inhibits insulin secretion via an autocrine effect within pancreatic islets. Amylin fibril formation in the pancreas cause islet cell dysfunction and cell death in type II - diabetes mellitus (Alfredo et al., 1994).

**Adrenomedullin**

Adrenomedullin is a novel hypotensive adrenal polypeptide isolated from a human pheochromocytoma and is structurally related to calcitonin gene related peptide and amylin. It has been suggested that besides being an adrenal hypotensive peptide, adrenomedullin is a gut hormone with potential insulinoic function (Mulder et al., 1996).
Galanin

Galanin is a 29 amino acid neuropeptide localised in the intrinsic nervous system of the entire gastrointestinal tract and the pancreas of man and several animal species (Scheurink et al., 1992). It inhibits insulin secretion in rat, mouse and also in isolated human islets and pig. In isolated rat and mouse islets galanin inhibits insulin secretion by increasing the K⁺ permeability and interfering with activation of adenylate cyclase and the activity of protein kinase C and cAMP. Among other functions, galanin inhibits insulin release (Ahren et al., 1991), probably via activation of G proteins (Renstrom, 1996) by the mediation of activated galanin receptors.

Macrophage migration inhibitory factor

Macrophage migration inhibitory factor (MIF), originally identified as cytokines and are secreted by T lymphocytes. It was found recently to be both a pituitary hormone and a mediator released by immune cells in response to glucocorticoid stimulation. Recently it has been demonstrated that insulin secreting β-cells of the islets of Langerhans express MIF and its production is regulated by glucose in a time and concentration dependent manner. MIF and insulin were both present within the secretory granules of the pancreatic β-cells and once released, MIF appears to regulate insulin release in an autocrine fashion. MIF is therefore a glucose dependent islet cell product that regulates insulin secretion in a positive manner and play an important role in carbohydrate metabolism (Waeber et al., 1997).

Nerve growth factor

Nerve growth factor (NGF) is a neurotropic growth factor that promotes neurite outgrowth during development. This growth factor is capable of modulating β-cell plasticity because it promotes neurite-like outgrowth in fetal and adult pancreatic β-cells from primary cultures (Vidaltaamayo et al., 1996) and in RINm5F
and insulinoma cells (Polak et al., 1993). In adult rat β-cells, \textit{in vitro} NGF stimulates glucose induced insulin secretion. The presence of the high affinity receptor for NGF has been described in insulinoma cell lines as well as in foetal and adult β-cells. The adult β-cells synthesise and secrete NGF in response to increasing extra cellular glucose concentration (Vidal Tamayo et al., 1996). The effect of NGF on insulin secretion is partly mediated by an increase in calcium current through calcium channels (Rosenbaum et al., 2001).

**Neuropeptides**

Immunocytochemistry has revealed the presence of three neuropeptides in the nerve terminals of pancreatic ganglia and islets of different species: Vasoactive intestinal peptide (VIP), gastrin releasing peptide (GRP) and pituitary adenylate cyclase activating polypeptide (PACAP).

**Gastrin releasing peptide**

Gastrin releasing peptide (GRP) consists of a 27 amino acid residue. It is localised to pancreatic nerves, including islet nerve terminals of several species. GRP released from the pancreas after vagal nerve activation stimulates insulin secretion (Knuhtsen et al., 1987; Sundler & Bottcher, 1991). In islets, activation by GRP receptors is coupled to PLC and phospholipase D (Wahl et al., 1992; Gregersen & Ahren, 1996).

**Vasoactive intestinal peptide**

Vasoactive intestinal peptide (VIP) stimulates insulin secretion in a glucose dependent manner accompanied by increased action of adenylate cyclase with increased formation of cAMP (Klinteberg et al., 1996). VIP increases activity of sympathetic system, including release of catecholamines from the adrenal medulla and
lead to the release of the pancreatic glucagon and inhibition of insulin release, by the activation of adrenergic receptors (Jarrhult & Holst, 1978).

**Pituitary adenylate cyclase activating polypeptide**

Pituitary adenylate cyclase activating polypeptide (PACAP) is localised to the parasympathetic nerves and released by the activation of the vagus nerve (Ahren, 2000). It exists in two forms consisting of 27 and 38 amino acids and show 68% homology (Arimura & Shioda, 1995). PACAP stimulates insulin secretion in a glucose dependent manner accompanied by increased action of adenylate cyclase with increased formation of cAMP (Klinteberg et al., 1996).

**Serotonin and serotonin transporter**

Serotonin content is increased in the brain regions and hypothalamic nuclei (Chen & Yang, 1991); (Lackovic et al., 1990), but there are reports suggesting a decrease in brain 5-HT content during diabetes (Sandrini et al., 1997; Sumiyoshi et al., 1997; Jackson & Paulose, 1999). Ohtani et al., (1997) have reported a significant decrease in extracellular concentrations of NE, 5-HT and their metabolites in the ventro medial hypothalamus (VHM). The ratio of 5-HIAA/5-HT was increased. A similar observation was reported by (Ding et al., 1992) with a decrease in 5-HT in cortex (19%) and 5-HT turnover (5-HIAA/5-HT) that was increased by 48%. Chu et al., (1986) has reported lower 5-HT levels in both hypothalamus and brainstem but not in corpus striatum. Insulin treatment brought about an increase in the cerebral concentration of 5-HIAA and accelerated the cerebral 5-HT turnover (Juszkiewicz, 1985). The 5-HIAA concentration was reported to be approximately twice as high as the controls regardless of duration of treatment. Brain tryptophan, the precursor of 5-HT, was also reduced in brain regions during diabetes (Jamnicky et al., 1991). Insulin treatment was reported to reverse this reduced tryptophan content to normal
(Jamnicky et al., 1993). There was a significant increase in 5-HIAA observed at 2-6 hours after insulin administration (Kwok & Juorio, 1987).

Insulin partly reversed the changes observed in the STZ-treated rats. There was a decrease in the muscarinic receptor number and axonal transport of receptor-bound opiate in STZ induced hyperglycaemia suggesting that impaired axonal transport of receptors partly involved in the neurological disturbance which is seen in diabetic patients (Laduron & Janssen, 1986). It has long been recognized that 5-hydroxytryptamine (serotonin; 5-HT) and its biosynthetic precursor tryptophan, play an important role in regulating immune functions through non-5-HT receptor interactions involving circulating tryptophan and kynurenine levels (Mossner & Lesch, 1998; Schrocksnadel et al., 2006; Muller & Schwarz, 2007). Individual serotonin receptors, however, are expressed in many immune-related tissues and interactions at specific receptors are also known to modulate aspects of the immune response and inflammation (Stefulj et al., 2000; Kubera, et al., 2005; Yu, et al., 2008).

Within the CNS, serotonin and serotonin receptors have been strongly associated with normal function. Certain neuropsychiatric disorders that include depression, bipolar disorder, OCD, anorexia and schizophrenia have been linked to dysregulation of CNS serotonin (Lucki, 1998; Geyer & Vollenweider, 2008). Indeed, therapeutics for these disorders often include inhibition of the serotonin transporter (SERT) with selective serotonin reuptake inhibitor (SSRI) medications, or blockade of specific serotonin receptor subtypes. SSRIs also show an efficacy in treating aspects of cardiovascular disease associated with depression (Halaris, 2009), and have been demonstrated in animal models to have an anti-inflammatory effect (Abdel-Salam et al., 2004, Holmes et al., 2010). The mechanisms underlying the protective effect of antidepressants are not precisely known, but are predicted by some researchers to involve activation of the pituitary-adrenocortical system via increased central serotonin levels (Bianchi et al., 1994), by modulation of cytokine levels in peripheral tissues (Xia et al., 1996;
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Kubera et al., 2004), and by suppression of platelet activation (Serebruany et al., 2003). Furthermore, acute SSRI administration has been shown to have a vasodilatory effect on the coronary artery that is cardioprotective (Van Melle et al., 2004). Interestingly, TNF-α, as well as certain other cytokines, have been shown to influence both expression and transport activity of the serotonin transporter. In neuronally derived cells and choriocarcinoma cells, TNF-α, INF-γ, and IL1β increases function (Ramamoorthy et al., 1995; Mossner et al., 1998; Zhu et al., 2006), whereas in B lymphocytes, IL4 decreases function (Mossner et al., 2001), and in intestinal epithelial derived Caco-2 cells, TNF- α has been found to decrease both expression and transport activity of SERT (Foley et al., 2007). The nature of the influence of cytokines on SERT function (e.g., facilitation or repression) likely depends on the cytokine and tissue modulation of synaptic serotonin levels in various brain regions by inflammatory cytokines would certainly be anticipated to have some effect on neuronal function relevant to psychiatric disorders like depression. In summary, there appears to be a strong link between proper functioning and regulation of the serotonin system and factors underlying cardiovascular disease and neuropsychiatric disorders (Uçeyler et al., 2010). We hypothesize that a particular aspect of the serotonin system, the 5-HT2A receptor, is a common and contributing factor underlying aspects of normal cardiovascular and CNS function. The dysfunction of this receptor results in certain characteristics of cardiovascular and neuropsychiatric disorders. There are seven families of serotonin receptors comprised of fourteen distinct subtypes (Nichols & Nichols, 2008). With the exception of the 5-HT3 receptor, which is a ligand-gated ion channel, all are seven transmembrane-spanning G-protein coupled receptors. Of all the serotonin receptors, the 5- HT2A receptor has been the one most closely linked to complex behaviours and neuropsychiatric disorders (Binder et al., 2009). The 5-HT2A receptor is highly expressed within the frontal cortex, with lower expression levels throughout the brain (Nichols & Nichols, 2008). There has been extensive
research performed to establish the role of 5-HT$_{2A}$ receptors within the brain, where they have been shown to participate in processes such as cognition and working memory (Williams et al., 2004). This mediate the primary effects of hallucinogenic drugs (Nichols, 2004) and has been implicated in mechanisms underlying schizophrenia (Vollenweider et al., 1998; Aghajanian & Marek, 2000). Furthermore, abnormal expression of 5-HT$_{2A}$ receptors has also been linked to depression. For example, some studies have shown that receptor protein expression is increased in certain cortical areas of patients with major depression (Bhagwagar et al., 2006; Shelton et al., 2009), as well as suicide victims (Pandey et al., 2002; Oquendo et al., 2006). 5-HT$_{2A}$ receptor expression decreases, however, have been found in brain limbic regions of patients with major depressive disorder (Mintun et al., 2004). CNS receptor dysfunction results in or contributes to the development of neuropsychiatric disorders including depression, bipolar disease and psychosis. This dysfunction either come from alterations in regulation due to promoter polymorphisms or other regulatory mechanisms influencing expression, or polymorphisms or mutations affecting the protein itself that could influence responsiveness and downstream signal transduction pathways. Polymorphisms in the promoter region of the human HTR$_{2A}$ locus have been shown to alter receptor expression levels (Myers et al., 2007), and these same polymorphisms have been linked to response to antipsychotics and certain SSRIs (Choi, et al., 2005; Benmessaoud, et al., 2008), and in some studies positively associated with various CNS conditions including major depression, bipolar disorder, and schizophrenia (Chee et al., 2001; Choi, et al., 2004; Penas-Lled et al., 2007; S’aiz et al., 2007). Polymorphisms within the coding regions of the HTR$_{2A}$ locus have been found in some studies to be positively associated with neuropsychiatric disorders. There is significant opportunity for future research to investigate how 5-HT$_{2A}$ receptor function mediates certain aspects of both neuropsychiatric and metabolic effects of atypical antipsychotics. If they did, then perhaps long-term therapy with these new
highly selective receptor antagonists would produce metabolic and cardiovascular disorders. The widespread expression and importance of the 5-HT$_2A$ receptor, the knockout animal appears overly normal. There are, however, certain behavioural effects associated with loss of this receptor (Weisstaub et al., 2006; Salomon et al., 2007). Interestingly, some observed behaviours are opposite to the effects of receptor antagonists (Popa et al., 2005), indicating that caution should be exercised in the interpretation of knockout studies using this model. Nevertheless, studies utilizing this mouse in models of cardiovascular related diseases will likely be of value. A better understanding of the relationship between 5-HT$_{2A}$ receptor function and its roles in both the CNS and cardiovascular system should lead to development of improved therapeutics to treat diseases affecting each of these systems either separately or together.

**Serotonin receptors in diabetes**

5-HT receptors comprise a complex family. On the basis of their pharmacology, signal transduction mechanisms and molecular structure, more than a dozen types of 5-HT receptors have been identified (Hoyer et al., 1994). Most of these receptors are coupled to various G proteins with the exception of the 5-HT3 receptor, which is a ligand gated cation channel (Derkach et al., 1989; Maricq et al., 1991; Jackson & Yakel 1995). Multiple 5-HT receptor subtypes are expressed in the cerebral cortex (Mengod et al., 1997). In cerebral cortex, 5-HT3 receptors are only expressed in inhibitory neurons (Morales & Bloom 1997) whereas 5-HT2A receptors are heavily expressed in pyramidal cells and to a lesser extent in inhibitory neurons (Willins et al., 1997; Hamada et al., 1998; Jakab & Goldman-Rakic 1998). Since the 1960s, many experiments using *in vivo* microiontophoretic methods have characterized how 5-HT affects neuronal behaviour. The predominant effect of 5-HT on cerebral cortical pyramidal neurons is an inhibition of spontaneous spiking. (Phillis 1984; Reader &
Intracellular studies in rat cortical slices suggested that 5-HT induces depolarization and action potential firing in pyramidal cells (Davies et al., 1987; Araneda & Andrade 1991; Tanaka & North 1993). Furthermore, Aghajanian & Marek (1997) reported that 5-HT enhances spontaneous excitatory postsynaptic currents (sEPSCs) without significantly changing spontaneous inhibitory postsynaptic currents (sIPSCs) in frontal pyramidal neurons. These in vitro results suggest that 5-HT is mainly excitatory in cortical neuronal circuitry. 5-HT and a-methyl-5-HT had no effect on sEPSCs in layer I neurons. Even though sampling bias might have contributed to this observation, the fact that activation of 5-HT$_2$A receptors induced robust enhancement of sEPSCs in all pyramidal neurons tested suggests that this differential modulation of sEPSCs in the two cell types was real. 5-HT$_2$A receptor expression is high in pyramidal neuron proximal apical dendrites and low in distal parts (Willins et al., 1997; Jakab & Goldman-Rakic, 1998). It is possible that activation of dendritic 5-HT$_2$A receptors induce dendritic transmitter release and/or release of retrograde messenger(s).

**Hyperglycaemia induced by 5-HT$_2$A Receptor Stimulation**

Administration of a selective 5-HT$_2$A receptor agonist DOI produced a rapid increase in blood glucose level. Administration of DOI is also accompanied by an increase in EPI concentration (Glennon, 1987). Pre-treatment of the animals with 5-HT$_2$A receptor antagonists i.e., ketanserin are able to reverse the increase in sympathetic nerve discharge produced by DOI (Hoyer 1988c; McCall & Hornis, 1988; Chaouloff et al., 1990b). These findings show that the central 5-HT$_2$A receptor stimulates sympathetic nerve discharge which in turn increases EPI release from adrenal medulla similar to the 5-HT$_{1A}$ receptor activation.
Administration of the 5-HT<sub>2A</sub> receptor agonist, α-methyl-5-HT, also elicit hyperglycaemic effects which are blocked by 5-HT<sub>2A</sub> antagonist ketaserin (Chaouloff et al., 1990b). α-methyl-5-HT was able to suppress food intake by food-deprived rats and also inhibited 2-deoxy-D-glucose induced hyperphagia in rats. α-methyl-5-HT induced hyperphagia was antagonised by ketanserin (Sugimoto et al., 1996; Yamada et al., 1997) Intraperitonal administration of 5-HT brought about hyperglycaemia mediated through a dose dependent increase in plasma EPI level. 5-HT induced hyperglycaemia was abolished by pre-treatment with ketanserin and also adrenodemedullation (Yamada et al., 1995). This suggests that the hyperglycaemic effects of 5-HT are closely related to the decrease of EPI from the adrenal gland, mediated by 5-HT<sub>2A</sub> receptors. 5-HT<sub>2A</sub> receptors are partly involved in the pharmacological effects of induction of hyperglycaemia induced by the 5-HT<sub>4</sub> receptor agonist 5-methoxytryptamine.

**Glutamate receptors in diabetes**

Diabetes mellitus induces cognitive impairment and defects of long-term potentiation in the hippocampus as indicated by behavioural and electrophysiological analysis. Considered to be an important mechanism of learning and memory in mammals, long-term potentiation is known to require regulation of the glutamate receptor properties. According to many studies, defects of long-term potentiation in the hippocampus of diabetic animals are due to abnormal glutamate receptors. Earlier studies explained that changes in glutamate receptors account for modifications of long-term potentiation in various models of diabetes mellitus. Deficits in long-term potentiation during chronic diabetes arise from dysfunction of glutamate receptors in early stages of the disease (Trudeau et al., 2004). Previous studies demonstrated that disruption of glutamate homeostasis occurs in the diabetic retina (Qing & Donald, 2002). Binding properties of brain glutamate receptors of STZ induced rats and the possible role of AMPA receptors in cognitive deficits during diabetes is reported by
Gagne et al., (1997). Altered glutamatergic neurotransmission and calcium homeostasis contribute to retinal neural cell dysfunction and apoptosis in diabetic retinopathy. Elevated glucose is reported to change the expression of ionotropic glutamate receptor subunits and impairs calcium homeostasis in retinal neural cells (Ana et al., 2006). It is suggested that enzymes of the glutamate system respond differently towards diabetes or deprivation of food and diabetes affect the glutamate uptake system in glial cells (Galanopoulos et al., 1988). Recent studies suggest that glutamate plays a pivotal role in the processing of sensory information in the spinal cord of patients with diabetic neuropathy. Abnormal expression of multiple glutamate receptors is involved in the development of diabetic neuropathy (Tomiyama et al., 2005).

Studies reported that neurons impaired of energy metabolism are highly sensitive to excitotoxicity (Simon et al., 1984; Wieloch, 1985; Monyer et al., 1989; Cebers et al., 1998). Pathophysiological mechanisms responsible for neuronal cell death in hypoglycaemia include the involvement of glutamate excitotoxicity.

**Inositol 1, 4, 5-trisphosphate (IP3) and activation of calcium release**

Cytosolic Ca\(^{2+}\) is a focal point of many signal transduction pathways and modulates a diverse array of cellular activities ranging from fertilization to cell death (Berridge et al., 2000). In most cell types, the major internal [Ca\(^{2+}\)]\(_{\text{ER/SR}}\) stores are the endoplasmic reticulum/sarcoplasmic reticulum (ER/SR). One mechanism for mobilizing such stores involves the phosphoinositide pathway. The binding of many hormones to specific receptors on the plasma membrane leads to the activation of an enzyme, phosphoinositidase C that catalyses the hydrolysis of phospholipids to produce the intracellular messenger. Although derived from a lipid, IP3 is water soluble and diffuses into the cell interior where it encounters IP3 receptors (IP3Rs) on the ER/SR. The binding of IP3 changes the conformation of IP3Rs such that an
integral channel is opened, thus allowing the \([\text{Ca}^{2+}]\) stored at high concentrations in the ER/SR to enter the cytoplasm. A critical feature of IP3Rs is that their opening is regulated by the cytosolic \([\text{Ca}^{2+}]\) concentration. This sensitivity to cytosolic \([\text{Ca}^{2+}]\) allows them to act as \([\text{Ca}^{2+}]\)-induced calcium release (CICR) channels that promote the rapid amplification of smaller trigger events.

**Alterations of glucose transport during diabetes**

In diabetes mellitus apart from raised blood glucose levels, disturbances in the metabolism of a number of other biomolecules such as glycogen, lipids, proteins and glycoproteins have also been reported (Randle *et al.*, 1963; Williamson *et al.*, 1968). Treatment with insulin generally rectifies these disturbances in diabetic state as it increases the peripheral utilisation of glucose by influencing key enzymes of glucose metabolic pathways (Exton *et al.*, 1966; Lenzen *et al.*, 1990). The liver plays a major role in insulin-regulated glucose homoeostasis through the balance between glucose utilization and glucose production, both processes being tightly coordinated (Nevado *et al.*, 2006). More recently, it has been shown that glucose uptake and release required a family of membrane facilitated-diffusion glucose transporters which are expressed in a tissue-specific manner. In muscle and fat, GLUT-4 is the main isoform of glucose transporters (Burat *et al.*, 1991). In adipose tissue the concentrations of GLUT-4 protein and mRNA are markedly decreased after 2-3 weeks of diabetes, and they are restored by insulin therapy (Berger *et al.*, 1989; Garvey *et al.*, 1989), whereas in skeletal muscle the concentrations of GLUT-4 protein and mRNA are marginally altered (Garvey *et al.*, 1989; Bourey *et al.*, 1990). In liver, GLUT-2 is the main isoform of glucose transporters (Thorens *et al.*, 1988). Much less information is available concerning the expression of GLUT-2 in liver of diabetic rats. Vitamin D3 functional regulation through dopaminergic, cholinergic and insulin receptors and
glucose transport mechanism through GLUT3 in the cerebellum of diabetic rats which play a major role in neuroprotection in diabetes which has clinical application.

**Electrophysiological changes during diabetes**

Neuroelectrophysiological recordings represent a non-invasive and reproducible method of detecting central and peripheral nervous system alterations in diabetes mellitus (Morano *et al.*, 1996). Diabetes mellitus is associated with chronic complications such as nephropathy, angiopathy, retinopathy and peripheral neuropathy. In diabetic patients, hyperglycaemia may precipitate seizures, and in experimental diabetes, indications for an increased neuronal excitability have been found (Anderson *et al.*, 2006). Neurophysiological alterations have also been described in animal models of diabetes, in particular in rats. In the peripheral nervous system (PNS) of diabetic rats the time course of neurophysiological changes is well established. Deficits in both motor and sensory nerve conduction velocity (MNCV and SNCV, respectively) can be detected within weeks after the onset of diabetes and increase up to 2–3 months after diabetes onset, remaining relatively stable thereafter (Moore *et al.*, 1980; Cameron *et al.*, 1986; Brismar *et al.*, 1987; Kappelle *et al.*, 1993). Studies of MNCV and SNCV in diabetic rats have made important contributions to the elucidation of the pathogenesis of the effects of diabetes on the PNS, as well as in the development of putative pharmacotherapy. Neurophysiological alterations have also been reported in the CNS of diabetic rats. Less is known about the underlying mechanisms of alterations in the CNS in diabetic rats. Cerebral metabolic (Knudsen *et al.*, 1989; Kumar and Menon, 1993) and vascular (Duckrow *et al.*, 1987; Jakobsen *et al.*, 1990) disturbances have been demonstrated within weeks after diabetes induction. However, the severity of these disturbances appears to be limited compared with the PNS (Biessels *et al.*, 1994), possibly leading to a less hostile neuronal microenvironment.
Neurotransmitters alterations in diabetes

Diabetes mellitus is a metabolic disorder that either arrives during the early years of growth (Juvenile diabetes) or later in life called as maturity onset diabetes. It is observed as the body’s inability to effectively regulate the sugar balance which leads to severe complications such as hyperglycaemia, obesity, neuropathy, nephropathy, retinopathy, cardiopathy, osteoporosis and coma leading to death. Pancreatic damage resulting in the dysfunction of α and β cells causes disordered glucose homeostasis. In diabetic individuals the regulation of glucose levels by insulin is defective, either due to defective insulin production which is called as Insulin Dependent Diabetes Mellitus (IDDM) or due to insulin resistance that is termed as Non Insulin Dependent Diabetes Mellitus (NIDDM).

Diabetes mellitus has been reported to cause degenerative changes in neurons of the central nervous system (Garris, 1990; Lackovic et al., 1990; Bhattacharya & Saraswathi, 1991). Our previous studies demonstrated adrenergic, serotonergic and dopamine D₂ receptor function alterations in the brain of diabetic rats (Abraham & Paulose, 1999; Padayatti & Paulose, 1999; Paulose et al., 1999; Eswar et al., 2007). The concentration of 5-HT, DA and NE increased in the brain regions of diabetic rats and accumulation of these monoamines is produced by inhibition of monoamine oxidase activity (Salkovic & Lackovic, 1992). Norepinephrine has been reported to increase in several brain regions during diabetes. Ohtani et al., (1997) have reported a significant decrease in extracellular concentrations of NE, 5HT and their metabolites in the ventro medial hypothalamus (VMH). Epinephrine (EPI) levels were significantly increased in the striatum, hippocampus and hypothalamus of diabetic rats and these changes were reversed to normal by insulin treatment (Ramakrishna & Namasivayam, 1995). Diabetes is reported to cause a high level of degeneration in neurons in different regions of the brain. Streptozotocin -induced diabetes and acute
deficiency of insulin is reported to result in increased concentrations of EPI in the supra chiasmatic nucleus. It is also reported that β-adrenergic receptor populations were decreased in diabetes (Garris, 1995). 5-HT content in the brain is reported to be decreased during diabetes (Chu et al., 1986; Sumiyoshi et al., 1997 Jackson & Paulose, 1999). Garris, (1995) reported chronically elevated levels of NE in the brain regions of amygdala, hypothalamus and medulla of diabetic mice. This was proposed to be associated with the expression of the gene causing diabetes mellitus. Hyperglycaemia is reported to alter the noradrenergic and cholinergic nerve components (Akria et al., 1994) with decrease in the Na⁺ - K⁺ ATPase activity in different brain regions (Gurcharan & Sukwinder, 1994). NE, DA and 5-HIAA are reported to be increased in the heart and adrenal gland in STZ rats. In the heart the initial changes in short-term diabetes included an increase in NE concentration but did not persist in the long term diabetic animals. In the adrenal gland there was an initial reduction followed by a steady increase in the concentration of NE and EPI (Cao & Morrison, 2001). Studies of Gireesh et al., (2008a) showed that there is a decrease in total muscarinic and muscarinic M1 receptors during diabetes in the cerebral cortex. A decreased muscarinic M1 receptor gene expression in the hypothalamus, brainstem, and pancreatic islets of diabetic rats was also demonstrated by Gireesh et al., (2008b).

Glucose in brain, supplies energy essential for maintenance of the nervous system. It is reported that 2-D Glucose augmented the turnover of NE, DA and 5-HT under the fasted condition. Insulin perfusion within the medial hypothalamic sites evoked a significant increase in the synthesis and release of DA from the sated rat, but did not alter its turnover. However, in the interval following insulin perfusion, DA and 5-HT turnover were enhanced while the efflux of 5-HT was suppressed.

Severe deterioration in cognitive function and personality in patients with long-standing diabetes as a complication of a consequence of insulin treatment is reported (Gold et al., 1995). The diabetic hippocampus adapt to high circulating
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glucose, with increased susceptibility to reductions in glucose availability. This is accompanied by alterations within the hippocampus, including both ECF glucose and lactate levels during cognitive testing and electrophysiological function.

Exposure to stress is known to precipitate or exacerbate many neuropsychiatric disorders such as depression, Parkinson's disease, schizophrenia, and others (Schwab & Zieper, 1965; Mazure, 1995). All these disorders involve a working memory deficit caused by prefrontal cortical (PFC) dysfunction (Mattes, 1980; Weinberger et al., 1986; Deutch, 1993; Fibiger, 1995). This increase of DA from the dendrites of dopamine neurons is due to an alteration in GABA regulation of the dopamine neurons. As with noradrenergic systems, single or repeated exposures to stress potentiates the capacity of a subsequent stressor to increase DA function in the forebrain without altering basal DA turnover, suggesting that the receptors have been hyper-sensitized (Basso et al., 1999). DA neurons are vulnerable to metabolic stress (Callahan et al., 1998). The maintenance of normal energy metabolism in T1DM during hypoglycaemia effect glucose sensing in the brain and contribute to hypoglycaemia-associated autonomic failure (Bischof et al., 2006).

The projected increase in the number of diabetic patients will strain the capabilities of healthcare providers the world over (Adeghate et al., 2006). The pancreatic hormones have an important role in the regulation of glucose metabolism. The secretion of insulin by β-cells of the endocrine pancreas is regulated by glucose and other circulating nutrients. The progression of diabetes is associated with an impaired ability of the neurons in the CNS to release neurotransmitters (Broderick & Jacoby, 1989). Neurotransmitters show significant alterations during hyperglycaemia and causes degenerative changes in neurons of the central nervous system (Garris, 1990; Lackovic et al., 1990; Bhardwaj, et al., 1999). Studies on STZ-induced diabetic rat models have shown similar results which exhibits morphological, behavioural and electrophysiological alterations on diabetes (Jakobsen et al., 1987; Biessels et al.,
Learning and memory deficits are associated with Type I and Type II diabetes mellitus (Gispen & Biessels, 2000) and brain morphological abnormalities have been found in diabetic patients, mainly in the cortical area (Dejgaard et al., 1991). STZ-induced diabetes results in structural alterations of mAChRs in the brain (Latifpour et al., 1991) which in turn alters cholinergic nerve components (Akria et al., 1994) with decrease in the Na⁺, K⁺-ATPase activity (Gurcharan & Sukwinder, 1994). Studies of Latifpour and McNeill, (1984) on long-term STZ-induced diabetes reported large reduction in muscarinic receptor densities as compared with their age-matched controls. Diabetes is intimately related at a molecular level and hence diabetes is able to provide the link between disease treatment and the prevention of age-related diseases. If specific molecular pathways controlling the rate of ageing are modulated genetically, then perhaps they are modulated pharmacologically (Geesaman, 2006). These insights ultimately have an important impact on the discovery and development of drugs to both treat and prevent a wide range of diseases.

**Factors affecting insulin regulation from pancreatic β-cells**

D-Glucose is the major physiological stimulus for insulin secretion. The mechanism of glucose induced insulin release is not completely understood. Phosphorylation of glucose to glucose-6-phosphate serves as the rate limiting step in glucose oxidation (Schuit, 1996). Glucokinase acts as a glucose sensor during this process. An increased ATP/ADP ratio is believed to close K⁺-ATP channel at the plasma membrane, resulting in decreased K⁺ efflux and subsequent depolarisation of the β-cell (Dunne, 1991). Depolarisation activates voltage-dependent Ca²⁺ channels, causing an influx of extracellular Ca²⁺ (Liu et al., 1998). Although intracellular Ca²⁺ activates protein kinases such as Ca²⁺ and calmodulin dependent protein kinase (Breen & Aschercroft, 1997), it remains unclear how increase in intracellular Ca²⁺ leads to
intracellular Ca$^{2+}$ stores appear to regulate a novel plasma membrane current [Ca$^{2+}$] release activated non-selective cation current], whose activity controls glucose activated secretion. Glucose induced insulin secretion is also partly dependent upon the activation of typical isoforms of PKC within the β-cell (Harris et al., 1996). It is suggested that PKC is tonically active and effective in the maintenance of the phosphorylated state of the voltage-gated L-type Ca$^{2+}$ channel, enabling an appropriate function of this channel in the insulin secretory process (Arkhammar et al., 1994). Glucose is an important regulator of various β-cell processes including insulin biosynthesis and release. Glucose, over short intervals stimulates insulin biosynthesis at the level of translation (Permut & Kipnis, 1972). Studies have shown that preproinsulin mRNA levels raise 4-10 folds in response to glucose stimulation. Studies of insulin gene expression in primary cultures of rat islets transfected Insulin I gene 5'-flanking sequence suggested that metabolic signal from glucose influx is transmitted through the insulin enhancer (German et al., 1990).

**ROLE OF NEUROTRANSMITTERS IN INSULIN REGULATION**

**Epinephrine and Norepinephrine**

Epinephrine and norepinephrine has an antagonistic effect on insulin secretion and glucose uptake (Renstrom et al., 1996; Porte, 1967). They also inhibit insulin stimulated glycolysis through inactivation of glycogen synthase and activation of phosphorylase with consequent accumulation of glucose-6-phosphate. In addition, it has been reported that epinephrine enhances glycolysis through an increased activation of phosphofructokinase. EPI and NE at low concentrations bind and activate β-adrenergic receptors which in turn stimulate the insulin secretion from pancreatic islets and at high concentration they can bind to α$_{2A}$ receptors and inhibit insulin secretion (Lacey et al., 1993). Previous studies had shown that in diabetic
condition α2A receptors are more activated which brought out the insulin inhibition and in turn hyperglycaemia (Lacey et al., 1993). Rat islet cell membrane is equipped with α2A-adrenoceptors (Filipponi et al., 1986) which are linked to adenylyl cyclase inhibiting insulin secretion.

NE and EPI, the flight and fright hormones are released in all stress conditions and are the main regulators of glucose turnover in strenuous exercise (Simartirkis et al., 1990). In severe insulin induced hypoglycaemia, a 15 to 40-fold increase of epinephrine plays a pivotal role in increasing glucose production independently of glucagon (Gauthier et al., 1980). In humans, adrenaline stimulates lipolysis, ketogenesis, thermogenesis and glycolysis and raises plasma glucose concentrations by stimulating both glycogenolysis and gluconeogenesis. It is already known that, when used in high doses in vivo or in vitro, EPI reduces the insulin response to stimulators (Malaisse, 1972). In vitro studies with yohimbine showed that the insulin secretion from the pancreatic islets increased significantly suggesting that when the α2-adrenergic receptors are blocked, it enhances islet cell proliferation and insulin secretion. Our previous studies demonstrated the role of α and β-adrenergic receptors in the insulin secretion (Ani et al., 2006a; b; c). We also reported the effect of NE in DA mediated insulin secretion (Eswar et al., 2006).

Acetylcholine

Acetylcholine is the neurotransmitter of the parasympathetic system. Cholinergic receptors are classified as ionotropic nicotinic receptor and metabotropic muscarinic receptor. Acetylcholine increases insulin secretion through muscarinic receptors in pancreatic islet cells (Tassava et al., 1992; Greenberg & Pokol, 1994). Muscarinic receptors are classified as M1, M2, M3, M4 and M5. They are G protein coupled receptors. They are characterized by having seven hydrophobic transmembrane-spanning regions that interacts with G-proteins and other effector
molecules to mediate the physiological and neurochemical effects. Expression studies have revealed the presence of M₁ and M₃ receptors in the pancreas. Acetylcholine is reported to be involved in the activation of glucose transport in the chromaffin cells. The cholinergic activation affecting this process is coupled with calmodulin and protein kinase C (Serck-Hanssen et al., 2002). It is reported that the role of acetylcholine in insulin secretion is mediated through M₁ and M₃ receptors (Renuka et al., 2004; 2005; 2006).

**γ-Aminobutyric acid**

Gamma aminobutyric acid (GABA) is the major inhibitory neurotransmitter in central nervous system. GABA is reported to be present in the endocrine pancreas at concentrations comparable with those found in central nervous system. The highest concentration of GABA within the pancreatic islet is confined to β-cells (Sorenson et al., 1991). Glutamate decarboxylase, the primary enzyme that is involved in the synthesis of GABA, has been identified as an early target antigen of the T-lymphocyte mediated destruction of pancreatic β-cells causing insulin-dependent diabetes mellitus (Baekkeskov et al., 1990). The brain GABAergic mechanisms also play an important role in glucose homeostasis. Recent studies reported the regulatory role of GABA during pancreatic regeneration (Balaram et al., 2007; 2008). Also, we reported the role of GABA in hepatocyte proliferation (Biju et al., 2002). GABA through its receptors has been demonstrated to attenuate the glucagon and somatostatin secretion from pancreatic α-cells and δ-cells respectively (Gaskins et al., 1995). GABA which is present in the cytoplasm and in synaptic-like microvesicles is co-released with insulin from β-cells in response to glucose (Reetz et al., 1991). GABA inhibits islet α and δ-cell hormonal secretion in a paracrine manner. GABA release is decreased in diabetes resulting in the enhancement of glucagon secretion from α-cells leading to hyperglycaemia. GABA is involved in the maintenance of glucose homeostasis and
inhibition of central $\text{GABA}_A$ receptors increasing the plasma glucose concentration (Lang, 1995). Thus, any impairment in the GABAergic mechanism in central nervous system and/or pancreatic islets is important in the pathogenesis of metabolic stress.

**Serotonin**

Brain serotonin content decreased during diabetes (Jackson & Paulose, 1999). This decrease is reported to be due to a decrease in uptake of tryptophan through the blood brain barrier (BBB) (Fernstrom & Wurtman, 1971; Fernstrom & Wurtman, 1972; Madras et al., 1974) and a decrease in rate of 5-HT synthesis (Carndall et al., 1981). The turnover rate of 5-HT to 5-HIAA in diabetic rats was also reported to be lower (Kwok & Juorio, 1987; Sandrini et al., 1997). A decrease in brain 5-HT lead to an up regulation of 5-HT$_{2A}$ receptors of cerebral cortex and brain stem which in turn inhibit insulin secretion due to increased sympathetic activity (Jackson & Paulose, 1999). Our earlier studies reported the role of serotonin in cell proliferation (Sudha & Paulose, 1998). The functional regulation of brain 5-HT during pancreatic regeneration is also reported (Mohanan et al., 2005a, b; 2006).

**Glutamate**

Although the role of glutamate as a signaling molecule is well established in the central nervous system, a similar role in the periphery has only recently been suggested. Inagaki et al., (1995) and Weaver et al., (1996) have detected functional glutamate receptors in the pancreatic islets of Langerhans. Pancreas is composed of four major cell types: the insulin-secreting β-cell, the glucagon-secreting α-cell, the pancreatic polypeptide-secreting PP cell and the somatostatin-secreting delta cells. The electrically excitable β-cells are stimulated to secrete insulin in response to changes in serum glucose concentrations. Secretion of insulin and the three other major peptide hormones found in islets is also believed to be affected by other
metabolic and neuronal signals (Boyd, 1992; Ashcroft et al., 1994). Bertrand et al. (1992; 1993) have shown that AMPA receptor agonists can potentiate both insulin and glucagon secretion from a perfused pancreas preparation and that oral or intravenous glutamate can increase insulin secretion and glucose tolerance \textit{in vivo} (Bertrand et al., 1995).

The precise role of a glutamatergic signaling system in islet physiology or pathology is important. Glutamate also subserves communication between islets and the central nervous system. Glucose-stimulated insulin release is \( \text{Ca}^{2+} \)-dependent, perhaps because \( \text{Ca}^{2+} \) couples the process of stimulus recognition to that of insulin discharge (Douglas, 1968; Milner & Hales, 1970; Matthews, 1970; Malaisse, 1973; Malaisse et al., 1974). Although several studies have indicated that glucose alters the state of \( \text{Ca}^{2+} \) in the pancreatic cells, the nature of the changes and the mechanisms by which they occur has to be understood (Hellman et al., 1976).

\textbf{Effect of insulin on glucose uptake and metabolism}

The insulin receptor is a transmembrane receptor that is activated by insulin (Ward & Lawrence, 2009). It belongs to the large class of tyrosine kinase receptors. Two alpha subunits and two beta subunits make up the insulin receptor. The beta subunits pass through the cellular membrane and are linked by disulfide bonds. The alpha and beta subunits are encoded by a single gene (\textit{INSR}). The insulin receptor has also recently been designated CD220 (cluster of differentiation 220). Insulin binds to its receptor which in turn starts many protein activation cascades. These include: translocation of Glut-4 transporter to the plasma membrane and influx of glucose. Tyrosine kinase receptors, including the insulin receptor, mediate their activity by causing the addition of a phosphate group to particular tyrosines on certain proteins within a cell. The "substrate" proteins which are phosphorylated by the Insulin receptor include a protein called "IRS-1" for "insulin receptor substrate 1". IRS-1
binding and phosphorylation eventually leads to an increase in the high affinity glucose transporter (Glut4) molecules on the outer membrane of insulin-responsive tissues, including muscle cells and adipose tissue and therefore to an increase in the uptake of glucose from blood into these tissues. The glucose transporter Glut4 is transported from cellular vesicles to the cell surface, where it then can mediate the transport of glucose into the cell. The main activity of activation of the insulin receptor is inducing glucose uptake. Courses of glucose and insulin mechanism for production, elimination and homeostatic feedback, has been extensive to oral glucose provocations, meal tests and insulin administration (Silber et al., 2009). For this reason "insulin insensitivity", or a decrease in insulin receptor signaling, leads to diabetes mellitus - the cells are unable to take up glucose and the result is hyperglycaemia (an increase in circulating glucose) and all the sequelae which result from diabetes.

Specific membrane transporters facilitate the movement of glucose into cells to reduce plasma glucose concentrations in response to insulin stimulation. The transported glucose is subsequently used as metabolic fuel or stored as the complex macromolecular structure, glycogen. Two major types of glucose transporters are known: Na\(^+\)-dependent and Na\(^+\)-independent transporters. Only the Na\(^+\)-independent transporters possess an insulin responsive isoform. The Na\(^+\)-dependent glucose transporter has been identified in several tissues particularly in the small intestine epithelium and the proximal tubule cells of the kidney, as well as in other kidney tubule cells (Takayama et al., 1988). These transporters are located on the luminal side of intestinal and kidney cells and act to absorb glucose against its concentration gradient by coupling the movement of glucose into these cells with the concomitant movement of Na\(^+\) into the cell. Since Na\(^+\) is moving down its electrochemical gradient this energy can be used to co-transport glucose into the cells. Thus, this
transporter is dependent on the concentrations of extracellular and intracellular sodium which are maintained by a Na+/K+-ATPase ion pump.

The Na+-independent glucose transporter family consists of several isoforms which facilitate the movement of glucose down its concentration gradient across a plasma membrane. Although seven isoforms have been identified (Glut1-7) (Brozinick et al., 2003) only one will be discussed in detail here, Glut4, because it is the transporter that is in highest concentration in insulin-sensitive tissues such as, skeletal muscle, fat and cardiac muscle (Cai & Helke, 2003). Glut4, and to a lesser extent Glut1 enable these cells to increase their glucose uptake, thereby lowering circulating glucose levels. Because the intracellular concentration of glucose is low due to the rapid phosphorylation of glucose to glucose-6-phosphate and its dissimulation to other metabolic products, the presence of active transporters in the plasma membrane favours the movement of glucose into cells.

Insulin enhances glucose uptake by increasing the number of transporters in the plasma membrane of cells. Insulin stimulation of cells mobilize transporters from intracellular compartments to the plasma membrane to facilitate glucose transport. This translocation of receptors to the plasma membrane has been demonstrated to occur within 30 seconds of insulin stimulation (Hill et al., 2001) and as the stimulus dissipates the decrease in the number of plasma membrane receptors declines coincident with a decline in glucose transport (Puro & Agardh, 1984).

The impaired ability of insulin to signal Glut4 translocation from intracellular stores is currently believed to be an important contributory factor to postprandial hyperglycaemia in diabetes (Song et al., 2003). In fact, decreased insulin levels in diabetic animals have been shown to, not only decrease transporter translocation but diminish expression of Glut4 (Brussee et al., 2004). Thus, it appears that insulin serves not only to acutely increase glucose transporter translocation, but also to maintain a basal level of expression of transporters in cells. Thus one mechanism by
which diabetes characterized by either low insulin levels, as in type 1 diabetes, or insulin resistance, as in type 2 diabetes, could cause pathologically high plasma glucose levels is via loss of regulation and expression of transmembrane glucose transporters. Several authors have also proposed that Glut2 on the β-cell membrane is relevant in regulating insulin secretion from islets (Yi et al., 2005).

**Triiodothyronine (T3) regulation in diabetes**

Diabetes mellitus and thyroid diseases are the two common endocrinopathies seen in the adult population. Insulin and thyroid hormones being intimately involved in cellular metabolism and excess/deficit of either of these hormones could result in the functional derangement of the other. In euthyroid individuals with diabetes mellitus, the serum T3 levels, basal TSH levels and TSH response to thyrotropin releasing hormone (TRH) is influenced by the glycemic status (Schlienger et al., 1982). Alterations in serum T3 levels have been described in association with energy deprivation (Vagenakis et al., 1975; Eisenstein et al., 1978) wasting illnesses (Burke & Eastman, 1974) the neonatal period (Larsen, 1972) and the use of such drugs as propylthiouracil (Oppenheimer et al., 1972), dexamethasone (Chopra et al., 1975) and propranolol (Roszkowska et al., 1974; Tevaarwerk & Boyd, 1977; Tevaarwerk et al., 1978). Fasting appears to inhibit 5'-monodeiodination, causing a decrease in the rate of conversion of T4 to T3 and an increase in the reverse T3 Concentration (Monnier et al., 2009). Poorly controlled diabetes, both Type 1 and Type 2, induce a “Low T3 state” characterized by low serum total and free T3 levels, increase in reverse T3 (rT3) but near normal serum T4 and TSH concentrations. Low serum T3 is due to reduced peripheral conversion of thyroxine (T4) to T3 via 5’ monodeiodination reaction. Studies indicate that long term diabetic control determines the plasma T3 levels. TSH responses and low T3 state normalized with improvement in glycaemic status but even with good diabetes control, the normal nocturnal TSH peak is not restored in C-
peptide negative patients i.e., those with totally absent pancreatic β cell function (Coiro et al., 1997). Studies show decreased insulin secretion (Ahren et al., 1985) as well as normal or increased levels of insulin is reported in the peripheral and portal circulation in hyperthyroidism (Dimitriadis et al., 1985). Long term thyrotoxicosis has been shown to cause beta cell dysfunction resulting in reduced pancreatic insulin content, poor insulin response to glucose and decreased rate of insulin secretion (Bech et al., 1996).

In hyperthyroidism, the endogenous glucose production is greatly increased by a variety of mechanisms: (a) an increase in the availability of gluconeogenic precursors in the form of lactate, glutamine and alanine from skeletal muscles and glycerol from adipose tissue, (b) an increase in the concentration of plasma FFA stimulating hepatic gluconeogenesis (Dimitriadis & Raptis, 2001); (c) an increase in glycogenolysis due to inhibition of glycogen synthesis resulting in hepatic glucose output even in fed state (Holness & Sugden, 1987); (d) an up regulation of GLUT-2 glucose transporters protein expression in the hepatocyte plasma membrane. This permits increased glucose efflux to occur without intracellular glucose accumulation which would limit hepatic glucose production (Mokuno et al., 1990); and (e) an increased secretion and exaggerated effects of glucagon and adrenaline on liver cells (Dimitriadis & Raptis, 2001). In skeletal muscle, there is a preferential increase in glucose uptake and lactate formation relative to glucose oxidation and storage in hyperthyroid state. This is due to increase in both basal and insulin stimulated GLUT1 and GLUT-4 transporters (Haber et al., 1995), increased responsiveness of glycogenolysis to beta adrenergic stimulation (Dimitriadis & Raptis, 2001), increased activity of hexokinase and 5-phosphofructokinase and decreased sensitivity of glycogen synthesis to insulin (Dimitriadis et al., 1997). In hypothyroidism, the synthesis and release of insulin is decreased (Ahren et al., 1985). The rate of hepatic glucose output is decreased probably due to reduced gluconeogenesis. A post receptor
defect has been proposed to explain the decrease in insulin stimulated glucose utilization in peripheral tissues (Dimitriadis & Raptis, 2001).

**Calcium imaging**

The Langerhans’ islet is another example of the presence of peripheral glutamatergic systems (Satin & Kinard, 1998). Intracellular free Ca\(^{2+}\) concentration plays a pivotal role in the regulation of various cellular functions as an intracellular messenger system. After stimulation of islets with AMPA or kainate, intracellular Ca\(^{2+}\) increased by way of activation of voltage-gated Ca\(^{2+}\) channels (Inagaki et al., 1995; Weaver et al., 1999), resulting in an elevated level of insulin secretion through increased exocytosis of insulin granules in \(\beta\)-cells (Bertrand et al., 1992). Since the development of digital video imaging of Ca\(^{2+}\) novel findings including Ca\(^{2+}\) oscillations (Berridge & Galione, 1988; Berridge, 1991) and Ca\(^{2+}\) waves (Berridge, 1993) have been described in many different cultured cell types. Ca\(^{2+}\) spots were reported as an elementary Ca\(^{2+}\) influx event through mechanosensitive channels directly coupled with the initial step in mechanotransduction in cultured endothelial (Ohata et al., 2001a, b; Tanaka & Takamatsu, 2001) and cultured lens epithelial cells (Ohata et al., 2001b, c). The Ca\(^{2+}\) spots, which develop sporadically, exhibit a spatiotemporal pattern distinct from Ca\(^{2+}\) sparks, the elementary Ca\(^{2+}\) release events from intracellular stores (Cheng et al., 1993; Nelson et al., 1995).

**Medicinal Plants as antidiabetic agents**

Antidiabetic plants have often been used by practitioners of herbal medicine in treating individuals with non-insulin-dependent diabetes. In such cases patient response must be carefully monitored and significant benefit can be gained from such therapies. While hypoglycemic herbs offer promise in the treatment of diabetes in their combined effect with insulin, treatment is inherently disruptive and extreme
caution must be exercised in order to promote a smooth transition, maintain suitable blood sugar levels and avoid insulin shock. Plants still remain a major source for drug discovery inspite of the development of synthetic molecules. Consequently, the uses of traditional plant extract in the treatment of various diseases have been flourished (Fabricant & Farnsworth, 2001). According to the World Health Organisation (WHO), more than 150 plants are known to be used for the treatment of diabetes mellitus and the study of hypoglycemic plants is then encouraged (Marles & Farnsworth, 1995). The ethnobotanical information reports about 800 plants that possess anti-diabetic potential (Alarcon-Aguilara et al., 1998). Several such herbs have shown anti-diabetic activity when assessed using presently available experimental techniques.

A wide array of plant derived active principles representing numerous chemical compounds has demonstrated activity consistent with their possible use in the treatment of NIDDM (Ivorra et al., 1988; Bailey & Day, 1989; Marles & Farnsworth, 1995). Among these are alkaloids, glycosides, galactomannan gun, polysaccharides, peptidoglycans, hypoglycans, guanidine, steroids, carbohydrates, glycopeptides, terpenoids, amino acids and inorganic ions. Even the discovery of widely used hypoglycemic drug, metformin came from the traditional approach of using Galega officinalis . Thus, plants are a potential source of anti-diabetic drugs but this fact has not gained enough momentum in the scientific community. The reasons may be many including lack of belief among the practitioners of conventional medicine over alternative medicine, alternative forms of medicine are not very well-defined, possibility of quacks practising such medicine providing alluring and magical cures and natural drugs vary tremendously in content, quality and safety (Grover et al., 2002).

In modern medicine, no satisfactory effective therapy is still available to cure the diabetes mellitus. Though insulin therapy is also used for the management of diabetes mellitus but there are several drawbacks like insulin resistance (Piedrola et
al., 2001), anorexia nervosa, brain atrophy and fatty liver (Yaryura-Tobias et al., 2001) after chronic treatment. In recent years, there has been renewed interest in plant medicine (Dubey et al., 1994; Prince et al., 1998; Ladeji et al., 2003) for the treatment against different diseases as herbal drugs are generally out of toxic effect (Geetha et al., 1994; Rao et al., 2003) reported from research work conducted on experimental model animal.

**Aegle marmelose**

Medicinal plants have formed the basis for Indian traditional medicine systems. *Aegle marmelose* Corr. (Rutaceae) commonly called as ‘Koovalam’ in Malayalam and ‘Bael’ in Hindi is indigenous to India. It is a medium sized, armed deciduous tree found in wild, especially in dry forests and is also cultivated throughout Indian subcontinent for its fruit. The fruit are globose with smooth, hard and aromatic rind. The ripe fruit is used for digestive and stomachic complications. Leaves, fruits, stem and roots of *Aegle marmelose* have been used in ethno medicine for several medicinal properties: astringent, antidiarrheal, antidysenteric, demulcent, antipyretic, antiscourbutic, haemostatic, aphrodisiac and as an antidote to snake venom (Kirtikar & Basu, 1935; Nandkarni, 1976). *Aegle marmelose* is also known as herbal medicine for the treatment of diabetes mellitus (Alam et al., 1990; Prakash, 1992). Preliminary report indicates blood glucose lowering activity in green leaves of *Aegle marmelose* (Chakrabarti et al., 1960). Oral administration of aqueous decoction of *Aegle marmelose* root bark (1 ml/100 g) showed hypoglycemic effect, which was maximum (44%) at 3 h in normal fasted rats. In addition, the same extract completely prevented peak rise of blood sugar at 1 h in OGTT (Karunanyake et al., 1984). Ponnachan et al. (1993) have observed that the crude aqueous leaf extract (1 g/kg for 30 days) exhibit hypoglycemic effect in alloxan induced diabetic rats. Aqueous leaf extract reversed the increase in Km values of liver malate dehydrogenase enzyme.
(Seema et al., 1996) and improved histopathological alterations in the pancreatic and kidney tissues of streptozotocin (STZ) induced diabetic rats (Das et al., 1996).

The aqueous extracts of fruits have also been reported to possess hypoglycemic activity (Kamalakkannan & Prince, 2003, 2004). Aqueous seed extract of Aegle marmelose possess antidiabetic and hypolipidemic effects in diabetic rats (Kesari et al., 2006). Aegle marmelose extract effectively reduced the oxidative stress induced by alloxan and produced a reduction in blood sugar (Sabu et al., 2004). Anandharajan et al., (2006) reported that methanolic extracts of Aegle marmelose activate glucose transport in a PI3 kinase-dependent fashion. Aegle marmelose root extract treated animals showed significant inhibitory activity against castor oil induced diarrhea (Mazumder et al., 2006). Aegle marmelose fruit extract exhibits protective effects on the pancreas of streptozotocin induced diabetic rats (Kamalakkannan & Prince, 2005).

Scopoletin (7-hydroxy-6-methoxy coumarin) was isolated from the leaves of Aegle marmelose and evaluated for its potential to regulate hyperthyroidism, lipid peroxidation and hyperglycaemia in levo-thyroxine-induced hyperthyroid rats. Scopoletin (1.0 mg/kg, p.o.) administered daily for 7 days to levo-thyroxine-treated animals decreased the levels of serum thyroid hormones and glucose as well as hepatic glucose-6-phosphatase activity, demonstrating its potential to regulate hyperthyroidism and hyperglycaemia (Panda & Kar, 2006).

The leaves of Aegle marmelose Correa were reported as a source of aegeline (Chatterjee et al., 1959). An examination of the fruits by various workers has revealed the occurrence of a coumarin termed ‘marmelosin’ (Asima & Sudhangsu, 1949). There molecular aspects of Aegle marmelose therefore, on blood glucose and lipids in normal and streptozotocin induced diabetic rats has been investigated. Aegle marmelose on blood sugar levels and markers of oxidative stress, i.e. lipid peroxidation, conjugated diene and hydroperoxide levels in serum and catalase,
glutathione and superoxide dismutase in blood and liver in streptozotocin treated rats (Sabu & Kuttan, 2000). Natural antioxidants strengthen the endogenous antioxidant defences from reactive oxygen species (ROS) and restore the optimal balance by neutralizing the reactive species. They are gaining immense importance by virtue of their critical role in disease prevention. In this context, *Aegle marmelose* can rightly be mentioned as a plant of considerable interest.

**Neurobiology of Pyridoxine**

Pyridoxine is required for the production of the monoamine neurotransmitters serotonin, dopamine, norepinephrine and epinephrine, as it is the precursor to pyridoxal phosphate cofactor for the enzyme aromatic amino acid decarboxylase. This enzyme is responsible for converting the precursors 5-hydroxytryptophan (5-HTP) into serotonin and levodopa (L-DOPA) into dopamine, noradrenaline and adrenaline. It has been implicated in the treatment of depression and anxiety (Dakshinamurti et al., 1990). Imbalance between dopamine and serotonin in the hypothalamus of the pyridoxine-deficient rat leads to severe neuroendocrine consequences. The decrease in pineal serotonin leads to a deficiency of melatonin (Yehuda et al., 1984). The decrease in cerebral and cerebellar GABA content in the pyridoxine-deficient rat is accompanied by a significant increase in the concentration of the excitatory amino acid, glutamic acid. Spontaneous or drug induced seizure activity in the pyridoxine-deficient rat is ascribed to the neurotransmitter imbalance (Dakshinamurti et al., 1984). An overdose of pyridoxine cause a temporary deadening of certain nerves such as the proprioceptory nerves; causing a feeling of disembodiment common with the loss of proprioception (Jones, 1982). Although vitamin B₆ is a water-soluble vitamin and is excreted in the urine, high doses of pyridoxine over long periods of time results in painful neurological symptoms known as sensory neuropathy (Perry et al., 2004).
Pyridoxine has a role in preventing heart disease. Without enough pyridoxine, a compound called homocysteine builds up in the body. Homocysteine damages blood vessel linings, setting the stage for plaque buildup when the body tries to heal the damage. Vitamin B6 prevents this buildup, thereby reducing the risk of heart attack. Pyridoxine lowers blood pressure and blood cholesterol levels and keeps blood platelets from sticking together (Perry et al., 2007). All of these properties work to keep heart disease. Nutritional supplementation with high dose vitamin B$_6$ and magnesium is one of the most popular alternative medicine choices for autism (Angley et al., 2007). Some studies suggest the B6-magnesium combination help attention deficit disorder, citing improvements in hyperactivity, hyperemotivity and aggressiveness (Mousain-Bosc et al., 2006).