DISCUSSION

Functional pancreatic β-cell mass is dynamic and although fully differentiated, β-cells are capable of re-entering the cell cycle upon appropriate stimuli. Stimulating regeneration-competent cells in situ is clearly the most desirable way to restore damaged tissue. (Bouckenooghe et al., 2005). A large number of growth factors and growth-stimulating peptides are expressed in or have stimulatory effect in the growing islets (Corbett et al., 1997). The presence of GABA in the cells of the islets of Langerhans is well documented in various species, particularly rats, on the basis of immunohistochemical and biochemical data (Okada et al., 1976; Vincent et al., 1983; Sakaue et al., 1987; Garry et al., 1987, 1988; Gilon et al., 1988; Sorenson et al., 1991; Reetz et al., 1991; Michalik & Erecinska, 1992; Michalik et al., 1993)

DNA synthesis in pancreas after partial pancreatectomy

Partial pancreatectomy was used as a tool to provide the stimulus to regenerate the β-cells of the islets of Langerhans in the rats to study the regeneration. The study was performed by incorporating [3H]thymidine into the replicating DNA as a biochemical index to measure the DNA synthesis during the pancreatic regeneration. An increase in the DNA synthesis was observed after 12 hours after partial pancreatectomy. The rate of [3H]thymidine incorporation was maximum when observed at 72 hours and declined after 7 days. The observation of the peak of the DNA synthesis at 72 hours is concordant with the previous reports (Pearson et al., 1977; Brockenbrough et al., 1988). Enhanced β-cell function and proliferation maintains the normoglycaemic level in rats during pancreatic regeneration (Leahy et al., 1988).
There was an increase in the circulating insulin levels during the regeneration of pancreas. Insulin was reported to increase the cell proliferation of β-cells in vitro (Rabinovitch et al., 1982). Previous studies suggest that the increase in the β-cell proliferation is related to the degree to which insulin biosynthesis and/or release is increased (Chick et al., 1975; King & Chick, 1976). Insulin can stimulate β-cell replication directly possibly through a receptor for multiplication stimulating activity or another insulin-like growth factor (Rabinovitch et al., 1982). There are also reports about the increase in the insulin secretion after the partial pancreatectomy, besides maintaining the normoglycaemic level, it also helps the remaining β-cell mass to regain its original mass and volume by inducing cell division. The signal for islet cell proliferation is related to a long standing demand for increased insulin secretion (Dubuc, 1976). After major pancreatectomy in dogs, insulin treatment enhanced the proliferation of the remnant pancreas and maintains endogenous insulin secretion for a long period, prolonging survival and promoting pancreatic regeneration (Ohashi, 1993).

There was no significant change in the body weight and blood glucose levels in the sham and experimental groups- P 72h and P 7d.

GABA content in brain regions during pancreatic regeneration

GABA is one of the most abundant neurotransmitters in the vertebrate central nervous system and is involved in neuroendocrine processes such as development, reproduction, feeding and stress (Martyniuk et al., 2005). A decrease in GABA content was observed during active pancreatic proliferation in brain stem, cerebellum and hypothalamus. The decreased contents in the brain regions were reversed to basal level when pancreatic DNA synthesis declined to control level. The effect of regeneration in the peripheral tissues to the hypothalamic GABA content was already reported during the regeneration of liver (Biju et al., 2001). This indicates the
A decrease in brain GABA content is important in the DNA synthesis in pancreas. It may be a homeostatic feedback adjustment by the hypothalamus to trigger the sympathetic innervation and thereby DNA synthesis. The pancreas enhance the insulin secretion to compensate the insulin demand in the body during the loss of the cells. Brain GABAergic functional alterations are reported to regulate autonomic nerve function in rats (Martin et al., 1998). GABA has been known to function as an autocrine/paracrine signal molecule in addition to its well-known inhibitory neurotransmitter function. Studies on the developing brain and on primary brain cell cultures showed that neuron formation was facilitated by GABA through GABA_{A} ion channels during postmitotic differentiation, but not earlier during the phases of cell fate commitment (Jelitai et al., 2004). These indicate that a decrease in the brain GABA content is important in the DNA synthesis in pancreas. Brain GABAergic changes are reported to regulate autonomic nerve functions in rats (Martin & Haywood, 1998). So the results show that a reduction in the GABA content in the brain regions may enhance DNA synthesis in pancreas by facilitating the sympathetic tone.

**GABA content in pancreas during pancreatic regeneration**

GABA is widely distributed in non-neural tissue including peripheral nervous and endocrine systems (Tillakaratne et al., 1995). GABA was found by immunohistochemistry in glial cells in pancreatic ganglia (Nagamatsu et al., 2001). GABA is stored in microvesicles in pancreatic islet cells (Chessler et al., 2002). GABA released from \(\beta\)-cells functions as an autocrine inhibitor of insulin secretion in pancreatic islets and that the effect is principally due to direct suppression of exocytosis (Braun et al., 2004). A decrease in GABA content was observed during active pancreatic proliferation in the pancreas of the experimental rats when compared with the sham. This effect was reversed to near normal level when the
DNA synthesis was completed during 7 days. Islets possess synaptic-like microvesicles, which have a secretory function and β-cell synaptic-like microvesicles are involved in the secretion of GABA (Thomas-Reetz et al., 1993).

**Brain GABA<sub>A</sub> and GABA<sub>B</sub> receptor alterations in the rats during pancreatic regeneration**

Previous studies in the regeneration of liver have showed significant alterations in the GABA<sub>A</sub> receptor function in brain regions (Biju et al., 2001<sub>a</sub>, 2002<sub>a</sub>). So we have studied the GABA<sub>A</sub> receptor alterations during the regeneration of pancreas of which the endocrine and exocrine secretions have a strong influence from the brain signals. Many gastrointestinal and pancreatic functions are under strong modulatory control by the brain via the vagus nerve (Berthoud et al., 2001). Pancreatic polypeptide when microinjected into the dorsal vagal complex potentiates glucose-stimulated insulin secretion (Krowicki, 1996). Some of the neurons of dorsal motor nucleus of the vagus are presumed to play a role in the brain stem neural control of glycemic homeostasis (Adachi et al., 1995). Targeted pharmacological lesion of the adrenergic innervation of dorsal motor nucleus of the vagus nerve causes hypersecretion by pancreatic β-cells, an effect, which requires an intact vagus nerve (Siaud et al., 1990; 1995). Also, the hypothalamic neurons producing oxytocin that densely project to the dorsal vagal complex are proposed to involve in an inhibitory control of the vagal preganglionic neurons that innervate the pancreas (Siaud et al., 1991). These all suggest the control of brain from hypothalamus and brain stem over pancreas by the vagal innervation. GABA and the hormonal functional studies will elucidate the functional integrity of their control on peripheral tissues including pancreas. A study in our lab in the regeneration of liver has already explained the importance of on the GABAergic receptor function and gene expression (Biju et al., 2001<sub>a</sub>, 2002<sub>a</sub>).
In our present study we analysed the receptor binding parameters and expression of the GABA receptors in sham operated and pancreatectomised rats. Scatchard analysis was performed for determining the $B_{\text{max}}$ and $K_d$ of these receptors (Scatchard, 1949). Receptor mRNA status was analysed by Real Time-PCR technique using specific primers. $\gamma$-receptor binding parameters were analysed using the receptor specific agonist $[^3\text{H}]$bicuculline (Kurioka et al., 1981). $\gamma$-receptor has two affinity sites and the double affinity status of the receptor was confirmed by displacement analysis using bicuculline.

It is well established that the autonomic fibres supplying the pancreas travel via the vagus and splanchnic nerves. These nerves are clearly related to the ventral hypothalamus. The ventro-medial hypothalamic nuclei are considered as the sympathetic centre and the stimulation of this area decreases insulin secretion (Helman et al., 1982). Studies of in vivo pancreatic nerve activity after VMH lesions show increased parasympathetic and decreased sympathetic nerve firing rates (Oommura & Yoshimatsu, 1984). Decreased $\gamma$-receptor binding observed in the hypothalamus reduces the sympathetic nerve stimulation thus reducing the inhibitory effect of EPI on insulin secretion.

Pancreatic $\beta$-cells express glutamate decarboxylase (GAD), which is responsible for the production and release of GABA. Increased cytoplasmic ATP levels can suppress GAD activity in $\beta$-cells, and hence GABA production and release, is compatible with previous findings on ATP suppression of brain GAD activity (Winnock et al., 2002).

**GABA$_\alpha$ and GABA$_\beta$ receptor alterations in the brain stem**

Brain stem region has direct connection with pancreas through the vagal innervation. Autonomic regulation of GABA is reported to mediate through GABA$_\alpha$.
and GABA₉ receptors (Sved, 1990; Coldman, 1998). We studied the brain stem GABAergic receptor subtypes functional regulation to elucidate its role during pancreatic regeneration. Previous reports have referred [³H]GABA as a high affinity GABA₉ receptor agonist (Paulose & Dakshinamurti, 1984). The decreased Bₘₐₓ in partial pancreatectomised rats denotes the decreased receptor density. The displacement analysis of the [³H]GABA against GABA indicates a shift in affinity towards the low-affinity. This suggests an altered receptor function during the pancreatic regeneration.

Bicuculline has a higher affinity for rapidly dissociating low-affinity GABA₉ sites. So we have used [³H]bicuculline to study the status of GABA₉ low-affinity receptors. The decreased Bₘₐₓ of [³H]bicuculline binding indicates a reduction in receptor density. The high-affinity sites of this receptor shifted to low-affinity in P 72 hrs rats denotes a decreased functioning of the receptor. Since GABA has a sympato-inhibitory effect, these changes may be responsible for the increased sympathetic activity observed.

[³H]baclofen were used to study the GABA₉ receptor. In P 72 hrs rats the receptor number of GABA₉ receptor increased. GABA₉ binding shift towards high-affinity in P 72 hrs treated rats indicates increased functioning of this receptor. GABA₉ receptor activation in central nervous system is reported to stimulate the sympathetic nervous system (Nonogaki et al., 1994, Takenaka et al., 1996).

Brain GABA₉ and GABA₉ receptor systems differentially regulate the sympathetic neural activity (Takenaka et al., 1995). It is clear from our results that the brain stem GABA₉ and GABA₉ receptors functions were in the opposite manner. GABA₉ receptor demonstrated to have an inhibitory effect on sympathetic stimulation while GABA₉ receptor activates sympathetic stimulation. The changes in brain stem GABAergic function favoured the pancreatic cell proliferation mediated through sympathetic regulation.
GABA$_A$ and GABA$_B$ receptor alterations in hypothalamus

Hypothalamus is the centre of autonomic nervous system reinforcement. Lateral lesions of hypothalamus caused an increase in DNA synthesis during liver regeneration. Sympathectomy and vagotomy blocked this effect (Kiba et al., 1994, 1995). Hypothalamic GABA$_B$ergic innervation is reported have a stimulatory effect on sympathetic nervous system. The receptor number decreased in P 72 hrs rats. This indicates a decreased high-affinity GABA$_A$ receptor activity in P 72 hrs group. The affinity change was confirmed by displacement analysis with GABA against $[^3H]GABA$ in P 72 hrs rats where we have found a shift in the high-affinity towards low-affinity.

The low-affinity GABA$_A$ receptor binding parameters as determined by $[^3H]bicuculline$ against bicuculline indicate a decrease in number and affinity of the receptor in P 72 hrs rats. Displacement analysis showed a significant shift in the high-affinity site to low-affinity site in P 72 hrs group.

GABA$_B$ receptor density increased in the P 72 hrs group. The affinity change in P 72 hrs group was confirmed by displacement analysis where we have observed a shift in affinity towards high-affinity.

The results showed that hypothalamic high-affinity and low-affinity GABA$_A$ receptor activity decreased during pancreatic regeneration. It is already reported that intrahypothalamic administration of GABA$_A$ receptor antagonist bicuculline methiodide decreased the sympathetic innervation and blood pressure in a dose dependent manner (Tellioglu et al., 1996). So the decreased GABA$_A$ receptor activity may facilitate sympathetic innervation. GABA$_B$ receptor activity was increased in P 72 hrs rats. This kind of differential functioning of GABA$_A$ and GABA$_B$ receptor system and its importance in sympathetic innervation is already reported (Takenaka et al, 1995).
**GABA\(_A\) and GABA\(_B\) receptor alterations in cerebellum**

GABA has been considered as a post-synaptic inhibitory neurotransmitter in the central nervous system particularly in the cerebellum (DeFeudis, 1977). The high affinity GABA\(_A\) receptor number decreased in P 72 hrs rats. Slowly dissociating low-affinity GABA\(_A\) receptor also decreased in p 72 hrs rats. Displacement analysis of the receptor with \(^{3}\text{H}\)bicuculline against bicuculline showed a shift in the low-affinity to very low-affinity.

GABA\(_B\) receptor affinity was increased in P 72 hrs rats. The displacement data are in accordance with the affinity change obtained from Scatchard plot.

**Pancreatic GABA\(_A\) and GABA\(_B\) receptor alterations in the rats during pancreatic regeneration**

Pancreatic β-cells rank among the few nonneuronal cell types that express GAD and contain its product, GABA (Okada et al., 1976; Reetz et al., 1991). Pancreatic β-cells release GABA in amounts that correspond to the cellular GABA production (Smismans et al., 1997). Gastropancreatic neuroendocrine cells synthesize large amounts of GABA. Results of electrophysiiological studies proved the presence of GABA\(_A\) receptor in the pancreas depolarizing response of GABA (Sha et al., 2001). Studies showed that expression of GABA\(_A\) receptors is abundant in gastropancreatic neuroendocrine cells (von Blankenfeld et al., 1995).

Inhibitory effects of GABA in the endocrine pancreas are consistent with its well-known suppressive actions as a neurotransmitter in the nervous system (Kuffler & Edwards, 1958). Electrophysiologica l measurements in guinea pig islet cells have indicated the presence of a GABA-sensitive chloride channel (Rorsman et al., 1989).

In electrophysiologica l studies GABA pressure microejection depolarized membrane potential. Electrically evoked fast excitatory postsynaptic potentials were
Significantly inhibited after GABA application through GABA_A receptors and suggest that endogenous GABA released from ganglionic glial cells acts on pancreatic ganglion neurons through GABA_A receptors (Sha et al., 2001).

**GABAergic inhibition of insulin synthesis and secretion from pancreatic β-cells in vitro**

A sophisticated interplay between glucose and a plethora of additional factors including other nutrients, neurotransmitters, islet generated factors and systemic growth factors regulate the signal-transduction in the pancreatic β-cell and thereby the insulin secretory process. The coupling of glucose metabolism to electrical activity remains central in all models of β-cell stimulus-secretion coupling.

The resting membrane potential of the β-cell is set by the K_ATP channel (Ashcroft & Rorsman, 1990). Incubation of the pancreatic β-cells with stimulatory glucose concentrations leads to the activation of a cascade of reactions, which ends in the exocytosis of stored insulin. This complex of processes starts with the uptake of glucose by the β-cell high-K_m/low affinity glucose transporter GLUT2 and proceeds with the conversion of glucose into glucose-6-phosphate by the β-cell isoform of glucokinase (Randel, 1993; Matschinsky, 1996). Elevation in the ATP/ADP ratio leads to closure of the K_ATP, which in turn results in depolarization of the plasma membrane. The subsequent opening of voltage-gated L-type Ca^{2+} channels leads to an increase in the cytoplasmic free Ca^{2+} concentration, [Ca^{2+}]_i, which promotes insulin secretion (Berggren & Larsson, 1994).

A reduction in cellular and medium GABA levels is more sensitive than insulin as a marker for the presence of dead β-cells in isolated preparations (Wang et al., 2005). Upon glucose stimulation, GABA and insulin are released from β-cells (Anhert-Hilger et al., 1996). GABA is reported to inhibit glucagon secretion from α-cells (Rorsman et al., 1989) as well as to inhibit insulin secretion from β-cells (Gu et
al., 1993) via GABA\textsubscript{A} and GABA\textsubscript{B} receptor mediated mechanisms, respectively. Most immunocytochemical studies have shown that GABA is found selectively in β cells (Vincent \textit{et al.}, 1983; Garry \textit{et al.}, 1986; Garry \textit{et al.}, 1988; Reetz \textit{et al.}, 1991). Earlier studies, using exogenous GABA, have reported a wide variety of effects on β cells function, including a general inhibition of both phases of insulin secretion, a stimulation of insulin secretion, or no effect at all (Michalik \textit{et al.}, 1992; Satin \textit{et al.}, 1998).

Isolated pancreatic islets were incubated for one hour with different concentrations of GABA, bicuculline, muscimol and baclofen separately in presence of 4mM and 20mM glucose, which would represent normal and diabetic conditions respectively. GABA has inhibited the insulin secretion in a dose dependent manner in presence of both glucose concentrations. This effect was not antagonized in presence on bicuculline, the GABA\textsubscript{A} receptor antagonist. The GABA agonist muscimol also inhibited the insulin secretion except in the lowest muscimol concentration of 10^{-9}M in presence of 4mM glucose. In presence of 20mM glucose, at 10^{-7} and 10^{-4}M muscimol enhanced insulin secretion was found but other concentrations either inhibited or did not significantly change the secretion status. Baclofen at 10^{-5}M concentration enhanced insulin secretion, but inhibited at 10^{-9}, 10^{-7}M and 10^{-4} molar concentrations and remained unafflicting at 10^{-8}M and 10^{-6}M baclofen concentrations. In presence of 20mM glucose, insulin secretion was significantly inhibited at all concentrations except 10^{-9}M baclofen, which did not change the insulin levels \textit{in vitro}. Pancreas perfusion experiments suggest that GABA generated by GAD65 may function as a negative regulator of first-phase insulin secretion in response to glucose by affecting a step proximal to or at the K\textsubscript{ATP} channel (Yuguang \textit{et al.}, 2000).

During the 24 hour secretion, results obtained were much similar to that obtained in the 1 hour insulin secretion study. GABA significantly inhibited the
Insulin secretion at normal and hyperglycaemic conditions. Addition of bicuculline would not alter this secretion status. Muscimol gave an enhanced secretion of insulin in the presence of 4 mM glucose at $10^{-7}$, $10^{-5}$ and $10^{-4}$ M muscimol concentrations, but remained unvoiced at other concentrations. But, in presence of 20 mM glucose muscimol significantly inhibited the insulin secretion. PK 11195 [1-(2-chlorophenyl)-(N-methyl-N-(1-methylpropyl)-3-isoquinoline-carboxamide], a potent and selective ligand for peripheral benzodiazepine binding sites, was shown to inhibit insulin release from rat pancreatic islets (Pujalte et al., 2000). Baclofen also showed an inhibitory effect on insulin secretion in most of the concentrations in normal as well as hyperglycaemic conditions in vitro.

GABA released from β-cells is reported as an autocrine inhibitor of insulin secretion in pancreatic islets acting through GABA_A receptors and this effect is presumed to be due to direct suppression of exocytosis (Braun et al., 2004). L-glutamine is metabolized preferentially to GABA and L-aspartate, which accumulate in islets, thus preventing its complete oxidation in the Krebs cycle, which accounts for its failure to stimulate insulin secretion (Fernandez-Pascual et al., 2004). When analyzing in terms of GABA release in presence of high glucose, a recent investigation showed inhibition (40%) of GABA release from reaggregated rat β-cells after a 2 hrs culture period in high when compared with low glucose conditions. A threefold increase in insulin secretion was observed in parallel (Winnock et al., 2001). In agreement with this finding, a second study demonstrated a twofold increase in GABA release from rat islets after 30 min when islets were transferred from high to low glucose conditions (Hayashi et al., 2003). A concurrent decrease in insulin secretion can be confirmed from these studies. When compared with the earlier studies GABA can be said to certainly inhibit insulin secretion through its both receptors GABA_A and GABA_B.
**Effect of GABA, muscimol and baclofen on pancreatic DNA synthesis**

Many different types of glutamate and GABA receptor subunits show differential expression that some have prominent expression in the embryonic and/or postnatal brain, whereas others are mainly present in the adult brain (Lujan et al., 2005). We have studied the effect of GABA, the GABA_A receptor agonist muscimol and GABA_B receptor agonist baclofen on pancreatic cell proliferation in in vitro cultures. Addition of EGF caused a marked increase in DNA synthesis from basal level. EGF is a known mitogen for cultured vascular smooth muscle cells (Huang et al., 1992). There are several reports on effects of growth factors in the normal β-cell growth. EGF was shown to stimulate [³H]-thymidine incorporation in islets (Sieradzski et al., 1987). Furthermore, a recent report showed that EGF was an important factor for pancreas precursor cell proliferation in vitro (Corentin et al., 2001). The islet cell migration and differentiation were impaired in the mice lacking EGF receptors (Miettinen et al., 2000). These findings suggest EGF is important in the growth and differentiation of islet cells. The addition of GABA (1ng) did not elicit any significant change in pancreatic DNA synthesis. There was significant increase in DNA synthesis when islets were incubated with 20mM glucose, without EGF but no change was reported in presence of 4mM glucose, which is normal physiological level. The addition of EGF to the cultures containing 10⁻⁶M GABA in presence of both the glucose concentrations obtained marked increase in DNA synthesis.

Muscimol is the specific agonist of the GABA_A receptor. We used muscimol to study the GABA_A receptor mediated effect on DNA synthesis of islets kept in primary culture. The addition of 10⁻⁶M muscimol also did not elicit any marked difference in DNA replication in cultures. But the DNA synthesis was enhanced in the presence of EGF and with the addition of 4mM and 20mM glucose along with EGF. Also, it was found that there was a dose dependent inhibition of DNA
In the presence of different concentrations of muscimol when incubated with and without EGF. GABA<sub>A</sub> receptors are peripheral benzodiazepine receptor ligands, which are reported to inhibit the proliferation of various tumors. Peripheral benzodiazepine receptor ligands inhibited the proliferation of hepatocellular carcinoma cell lines by inducing apoptosis and cell cycle arrest (Sutter et al., 2004).

Peripheral benzodiazepine receptor is involved in numerous biological functions, including steroid biosynthesis, mitochondrial oxidative phosphorylation and cell proliferation. Recent studies support the idea that the subcellular localisation of peripheral benzodiazepine receptor defines its function and this receptor could be a possible target for new strategies against cancer (Corsi et al., 2005). Apoptosis of GABAergic interneurons was demonstrated in the molecular layer and white matter of the cerebellar cortex during the first two weeks of development (Yamanaka et al., 2004). Exposure of cultured Schwann cells to (-)-baclofen inhibits their proliferation and reduces the synthesis of specific myelin proteins providing evidence for a physiological role of GABA<sub>B</sub> receptors in the glial cells of the peripheral nervous system (Magnaghi et al., 2004). Brown rice extracts with enhanced levels of GABA have an inhibitory action on leukemia cell proliferation and have a stimulatory action on the cancer cell apoptosis (Oh & Oh., 2004). Receptor functional alterations studied during hepatic proliferation in hepatocyte cultures showed that GABA<sub>A</sub> receptor agonist, muscimol, dose dependently inhibited epidermal growth factor induced DNA synthesis and enhanced the transforming growth factor β<sub>1</sub> mediated DNA synthesis suppression in primary hepatocyte cultures suggesting GABA<sub>A</sub> receptor acts as an inhibitory signal for hepatic cell proliferation (Biju et al., 2001b).

Baclofen is the specific agonist of GABA<sub>B</sub> receptor and we have studied the effect of baclofen in primary pancreatic cultures to learn about the GABA<sub>B</sub> receptor mediated changes during pancreatic DNA synthesis in detail. Baclofen has obtained a ranked increase in the DNA replication in primary pancreatic cultures with and
without EGF. The result was similar and significant when cells were incubated in the presence of 4mM and 20mM glucose also. The dose-dependent increase in GABA_B receptor mediated EGF mitogenicity was abolished by G_i protein inhibitor pertussis toxin. GABA_B receptor is coupled to G_i protein. The stimulation of these G_i protein coupled receptors induces cell proliferation in various tissues (Biesen et al., 1996). Several lines of evidence suggest that activation of receptors that couple to heterotrimeric G-proteins is important in regulating proliferation in regenerating liver cells following a partial hepatectomy. The expression of the stimulating and inhibitory α subunits of G proteins that couple various receptors to their effector targets like adenylyl cyclase is differentially regulated during the early pre-replicative period in the liver (Mahler & Wilce, 1988). ERK activation via endogenous IGF-I receptor and G_i-coupled LPA receptor is sensitive to pertussis toxin treatment suggesting a cross talk between leading mitogenic effect as reported in rat fibroblasts. Such a cross talk between GABA_B receptor α-subunit and for EGF receptor may be responsible for the triggering of pancreatic DNA synthesis. Neurotransmitter receptors like α1 adrenergic and S2 class serotonin receptors act as co-mitogenic signals in EGF mediated DNA synthesis in hepatocyte cultures by the same mechanism (Sudha & Paulose, 1997, Michalopoulos et al., 1997). A similar result was obtained during the primary culture of pancreatic islets, pertussis toxin inhibited potentiation of EGF effect induced by 8-OH DPAT, where an inhibition of ERK2 activation by the 5-HT_{1A} receptor-selective agonist is suggested (Mohanan, 2005b). Studies on functional alterations of receptors during hepatic proliferation in hepatocyte cultures showed that GABA_B receptor enhancement induce hepatic neoplasia. Also, baclofen is seen to act as a potent co-mitogen, triggering DNA synthesis in primary cultures of rat hepatocytes, mediated through the G_i protein coupled GABA_B receptors (Biju et al., 2002b).
Our studies have revealed the significance of \( \text{GABA}_A \) and \( \text{GABA}_B \) receptors functional regulation during pancreatic regeneration and insulin secretion in rats. This will have immense clinical significance in the management of diabetes.