CONCLUSION

Our results demonstrate the functional alterations of the GABA_A and GABA_B receptors and the gene expression during the regeneration of pancreas following partial pancreatectomy. The role of these receptors in insulin secretion and pancreatic DNA synthesis using the specific agonists and antagonists also are studied in vitro. The alterations of GABA_A and GABA_B receptor function and gene expression in the brain stem, cerebellum and hypothalamus play an important role in the sympathetic regulation of insulin secretion during pancreatic regeneration. Previous studies have given much information linking functional interaction between GABA and the peripheral nervous system. The involvement of specific receptor subtypes functional regulation during pancreatic regeneration has not given emphasis and research in this area seems to be scarce. We have observed a decreased GABA content, down regulation of GABA_A receptors and an up regulation of GABA_B receptors in the cerebral cortex, brain stem and hypothalamus. Real Time-PCR analysis confirmed the receptor data in the brain regions. These alterations in the GABA_A and GABA_B receptors of the brain are suggested to govern the regenerative response and growth regulation of the pancreas through sympathetic innervation. In addition, receptor binding studies and Real Time-PCR analysis revealed that during pancreatic regeneration GABA_A receptors were down regulated and GABA_B receptors were up regulated in pancreatic islets. This suggests an inhibitory role for GABA_A receptors in islet cell proliferation i.e., the down regulation of this receptor facilitates proliferation. Insulin secretion study during 1 hour showed GABA has inhibited the insulin secretion in a dose dependent manner in normal and hyperglycaemic conditions. Bicuculline did not antagonize this effect. GABA_A agonist, muscimol inhibited glucose stimulated insulin secretion from pancreatic islets except in the lowest concentration of 10^{-9}M in presence of 4mM glucose.
Musclinol enhanced insulin secretion at $10^{-7}$ and $10^{-4}$M muscimol in presence of 20mM glucose- 4mM glucose represents normal and 20mM represents hyperglycaemic conditions. GABA<sub>B</sub> agonist, baclofen also inhibited glucose induced insulin secretion and enhanced at the concentration of $10^{-5}$M at 4mM glucose and at $10^{-9}$M baclofen in presence of 20mM glucose. This shows a differential control of the GABA<sub>A</sub> and GABA<sub>B</sub> receptors over insulin release from the pancreatic islets. During 24 hours in vitro insulin secretion study it showed that low concentration of GABA has inhibited glucose stimulated insulin secretion from pancreatic islets. Muscimol, the GABA<sub>A</sub> agonist, inhibited the insulin secretion but, gave an enhanced secretion of insulin in presence of 4mM glucose at $10^{-7}$, $10^{-5}$ and $10^{-4}$M muscimol. But in presence of 20mM glucose muscimol significantly inhibited the insulin secretion. GABA<sub>B</sub> agonist, baclofen also inhibited glucose induced insulin secretion in presence of both 4mM and 20mM glucose. This shows the inhibitory role of GABA and its specific receptor subtypes over insulin synthesis from pancreatic β-islets. In vitro DNA synthesis studies showed that activation of GABA<sub>A</sub> receptor by adding muscimol, a specific agonist, inhibited islet DNA synthesis. Also, the addition of baclofen, a specific agonist of GABA<sub>B</sub> receptor resulted in the stimulation of DNA synthesis.

Thus, we conclude that brain and pancreatic GABA<sub>A</sub> and GABA<sub>B</sub> receptor gene expression differentially regulates pancreatic insulin secretion and islet cell proliferation during pancreatic regeneration. This will have immense clinical significance in therapeutic applications in the management of Diabetes mellitus.