1. INTRODUCTION

Diabetes mellitus results as a consequence of the body's inability to respond normally to high blood glucose levels. The onset of diabetes is due to several pathological changes, which are a reflection of either the inability of the pancreatic β-cells to secrete sufficient insulin to combat the hyperglycemia or a state of insulin resistance in target tissues. However, the significance of changes in β-cell mass and decreased β-cell proliferation or growth in progression of diabetes has not been given importance till recent years. β-cells, like all other cells of our body are under the regulatory checks and balances enforced by changes in cell cycle progression. However, very little is known regarding the key components of the cell cycle machinery regulating cell cycle control of β-cells. Knowledge of key elements involved in cell cycle regulation of β-cells will improve our understanding of the replication capacity and developmental biology of β-cells. This information is essential for us to design new approaches that can be used to correct β-cell deficiency in diabetes (Rane & Reddy, 2000).

The study of the growth potential of pancreatic β-cells has elicited considerable interest during recent years because of its implications for the better understanding of the pathogenesis of diabetes and potential treatment of this disease. Reduction in the β-cell mass is a critical clinical event in the development of insulin dependent diabetes mellitus which will lead to an insulin insufficiency. The acute onset of disease is preceded by a period of progressive destruction of pancreatic islets without replacement, until the remaining mass is sufficient to respond to hyperglycemia (Kloppel et al., 1985). It would therefore be of interest to examine the regulation of islet cell growth and the factors that prevent or promote replacement of lost islet cells. Glucose homeostasis is tightly controlled by insulin (Ashcroft & Ashcroft, 1992). There is a minute to minute output from the β-cell to meet the changing demands. There is also a long term adaptation of insulin production by changes in total β-cell mass (Kahn, 1998; Taylor, 1999).

The pancreas is a an organ containing two distinct populations of cells, the exocrine cells that secrete enzymes into the digestive tract and the endocrine cells that secrete hormones into the blood stream. The endocrine cells are grouped into the islets of Langerhans, which are compact spheroidal clusters embedded in the exocrine tissue.
There are four principal cell types in the endocrine tissue - \( \alpha \)-cells, which produce glucagon, \( \beta \)-cells produce insulin, \( \delta \)-cells produce somatostatin and the \( pp \) cells which produce pancreatic polypeptides. The endocrine cells are believed to originate from the pancreatic duct, which is the source of endocrine stem cells. The exocrine and the endocrine tissues are capable of regeneration after injury, but the degree of regeneration is variable. The exocrine part respond to regeneration sooner than the endocrine part (Brockenbrough et al., 1988).

A variety of factors are implicated in pancreatic regeneration of which insulin and the glucose are the principal regulators. Growth factors like growth hormone, prolactin, IL-\( \beta \), interferons etc. have a regulatory control over the pancreatic regeneration (Vinik, 1992). Neurotransmitters are one of the important classes of the cell cycle regulators. They are involved in the insulin secretion also. Physiological studies have shown that the insulin secretion from pancreatic \( \beta \)-cells is regulated by the central nervous system through sympathetic and parasympathetic nerves. Catecholamines are found to be principal inhibitors of insulin secretion (Coore & Randle, 1964).

Epinephrine (EPI) and norepinephrine (NE) can regulate insulin secretion in a concentration dependent manner. At a lower concentration they can stimulate insulin secretion while at a higher concentration they can inhibit insulin secretion (Coore & Randle, 1964). It is already found that EPI and NE are increasing during diabetes (Tassava et al., 1992; Fushimi et al., 1984). They are also increasing with age and chances of getting diabetes is more with aging.

As EPI controls insulin secretion, it can regulate the pancreatic cell proliferation also. EPI and NE can mediate their function through same class of receptors called adrenergic receptors. There are mainly two classes of adrenergic receptors, \( \alpha \)- and \( \beta \)-adrenergic receptors. \( \alpha \) has two subtypes - \( \alpha_1 \) and \( \alpha_2 \); and \( \beta \) has - \( \beta_1 \), \( \beta_2 \) and \( \beta_3 \) subtypes. At a higher concentration EPI can bind to the \( \alpha_2 \) adrenergic receptor and can inhibit both the insulin secretion and islet DNA synthesis. At a lower concentration EPI binds and activates \( \beta \)-adrenergic receptors which in turn stimulate insulin secretion and pancreatic regeneration (Sjoholm, 1991).
Capacity of pancreatic β-cells to proliferate, like other cell types, reflects the ability of cells to progress normally through the cell cycle. Defects or anomalies in proteins governing the regulated progression through the cell cycle may impair the capacity of β-cells to proliferate under conditions of increased functional demand on the β-cell mass, as is the case during hyperglycemia in diabetes. Modulation of cell cycle pathways in β-cells can provide alternative approaches to repopulate the β-cells in diabetic patients which will foster development of diabetes therapy.

In the present study, the changes in the brain EPI, adrenergic receptors and the receptor gene expression were investigated during pancreatic regeneration and insulin secretion. The changes in the pancreatic islet EPI and adrenergic receptors were also studied in the pancreatectomised rats. The regulatory function of EPI in association with EGF and glucose were investigated in rat islet cultures. In vitro studies were carried out using antagonists for adrenergic receptor subtypes to see their involvement in the islet DNA synthesis. The mechanism by which the peripheral EPI regulate insulin secretion was also investigated by studying the nuclear binding proteins in the pancreatic islets during pancreatic regeneration and diabetes.
ADRENERGIC REGULATION OF PANCREATIC REGENERATION AND INSULIN SECRETION

BRAIN STEM
- EPI
- α₁-ADRENERGIC RECEPTOR ACTIVITY
- β-ADRENERGIC RECEPTOR ACTIVITY
  - SYMPATHETIC STIMULATION IN ISLETS
  - α₂-ADRENERGIC RECEPTOR ACTIVITY
  - β-ADRENERGIC RECEPTOR ACTIVITY
  - INSULIN
  - REGENERATION

HYPOTHALAMUS
- α₂-ADRENERGIC RECEPTOR ACTIVITY
  - CRF
  - T₃
1.1 OBJECTIVES OF THE PRESENT STUDY

1. To study the DNA synthesis pattern in regenerating pancreas.

2. To study the epinephrine and norepinephrine content in the brain regions as well as in the pancreatic islets during regeneration.

3. To study the kinetic parameters of adrenergic receptor subtypes in brain regions - cerebral cortex, brain stem & hypothalamus and pancreatic islets of sham and pancreatectomised rats.

4. To study the circulating epinephrine and norepinephrine levels in the serum of sham and pancreatectomised young rats.

5. To study the changes in the circulating insulin and T₃ levels of sham and pancreatectomised rats.

6. To study the expression of brain α₂,β- and β-adrenergic receptor subtypes during regeneration by RT-PCR techniques.

7. To establish the role of adrenergic receptors in insulin secretion from pancreatic islets in 1 hr and 24 hr in vitro cultures.

8. To establish the role and mechanism of peripheral epinephrine in regulating insulin release by binding to specific nuclear protein.

9. To study the role of adrenergic receptor subtypes on islet DNA synthesis by using specific antagonists in in vitro in the cultures of pancreatic islets in combination with epidermal growth factor and transforming growth factor.