INTRODUCTION

Diabetes mellitus, as a disease has been known to man for a very long time. Its manifestations such as passing of frequent and large quantities of urine were mentioned in the Ebers Papyrus in Egypt about 1500 B.C. and the presence of honey urine was noted by Sushruta in India around 600 B.C. The disease brings major metabolic changes in the body which not only affect the carbohydrate metabolism but lipid and protein metabolism also. Uncontrolled diabetes often results in a number of disabilities including retinopathy, nephropathy and neuropathy. These eventually cause blindness, kidney failure, coronary thrombosis and gangrene of lower extremities. The disease also lowers immunological status which, at least in part, is responsible for delayed healing and recurrent infection in these patients. Diabetes mellitus affects both sexes equally and individuals at all ages. However, the symptoms, cure and treatment varies with the type of the diabetes. The frequency of the disease is fairly high and according to the current estimate as many as 30 million people around the World suffer from this disease. The National Diabetes Data Group (NDDG) of National Institute of Health (NIH) U.S.A. (1979) have classified diabetes mellitus into three major sub-classes which have been accepted by the
i) **Type-1** : Insulin-dependent diabetes mellitus (IDDM)

It is usually characterized clinically by abrupt onset of symptoms like insulinopenia, dependence on injected insulin to sustain life, and proneness to ketosis. Classically, this type of disease occurs in juveniles and it was formerly termed juvenile onset diabetes (JOD). However, it can occur at any age. It addition to the ketosis-prone stage, this type of diabetes can also be recognized in a pre-ketosis prone stage. IDDM appears to be heterogeneous in term of genetic and environmental factors that precipitate the disease. Genetic determinants as expressed by increased or decreased frequency of certain histocompatibility antigens (HLA) on chromosome number 6 are thought to be important in most of the patients. Abnormal immune responses and autoimmunity also play an etiologic role while islet cell antibodies are frequently present at diagnosis of this type of diabetes.

ii) **Type-II** : Noninsulin-dependent diabetes mellitus (NIDDM)

This type of diabetes is frequently present with minimal or no symptoms of the metabolic disorders. The patients are not dependent on insulin for prevention of ketonurea and are not prone to ketosis. However, they may require insulin for correction of symptomatic or persistent fasting hyperglycemia. Such patients may develop ketosis
under special circumstances, such as severe stress precipitated by infections or trauma. There may be normal levels of insulin, mild insulinopenia, or above normal levels of insulin associated with insulin resistance. The onset of disease of this type generally occurs after the age of 40, but it can occur in young persons who do not require insulin and are not ketotic. NIDDM also has a genetic basis, which appears to be stronger than in IDDM and the autosomal dominant inheritance has been well established. Environmental factors superimposed on genetic susceptibility are undoubtedly involved in onset of NIDDM types. Characteristic aggregation of HLA alleles and islet cell antibodies have not been found. The type was formerly referred to as maturity onset type diabetes of young (MODY).

iii) Other types of diabetes

In this sub-class, diabetes forms a part of certain other conditions and syndromes that often have many clinical features not generally associated with diabetic state. This sub-class has been divided according to the known or suspected etiologic relationships. For example, diabetes may be secondary to:

a) Pancreatic diseases
b) Hormonal abnormalities
c) Drugs or chemicals
d) Insulin receptor abnormalities
e) Certain genetic disorders.
Diabetes mellitus is not a killer disease like ailments of heart and cancer but it deteriorates the quality of life both in terms of physical and mental parameters. Because of its hereditary basis, the disease has some social implications.

Treatment of the disease has been mentioned in ancient documents but there was no scientific basis of any of these treatments till Banting and Best (1922) showed that manifestations of diabetes mellitus can be cured by injection of insulin, a hormone, produced by B-cells of islets of Langerhans of pancreas. A great deal of information is available on the physiological role of B cells and insulin. Similarly, it is well established that disfunctioning of B cells results in lower concentration of insulin in blood and the manifestations of this disease. A number of factors are responsible for the disfunctioning of B cells and insulin and some of these are:

a) Decrease in insulin production by B cells.
b) Ageing of B cells
c) Insulin degradation by enzyme
d) Production of autoantibodies against:
   1. Insulin
   2. Insulin receptors
   3. Islets cells.
e) Invasion of B cells by viruses like Encephalomyocarditis virus (EMC), Reovirus, Coxsackie virus and mumps virus.
f) Selective damage of B cells by chemicals such as alloxan, streptozotocin and fruzemids.
In recent years it has been hypothesised that diabetes may be an autoimmune disease. A number of workers have shown the presence of islet cell antibodies (ICA) in the sera of type 1 diabetics. However, the nature of the antigen(s) responsible for the production of these antibodies has not been identified. Also very little information is available on the substances and/or physiological processes which bring about modification of antigen(s), so that the immune system fails to recognize these antigens as self.

This work, therefore, was undertaken to fill certain gaps in the knowledge regarding the immune basis of diabetes mellitus. The main objectives of this work were to:

1. Establish the presence of autoantibodies against pancreas in the blood of type 1 diabetics.
2. Isolate and purify the pancreatic antigen(s) which react with sera of type 1 diabetics and are responsible for the production of autoantibodies.
3. Localise the antigen(s) responsible for the production of antibodies in the pancreas.
4. Determine, if the purified antigens are related to each other.

To meet these objectives the following experiments were undertaken:

1. The sera obtained from IDDM and newly diagnosed diabetics was tested against cell free homogenate of human pancreas for the presence of specific antibodies against pancreas.
2. The pancreatic homogenate was subjected to differential centrifugation to determine the subcellular components which reacted against the sera of type-1 diabetics.

3. The pancreatic homogenate was subjected to solvent extraction and fractionation by various techniques including \((\text{NH}_4)_2\text{SO}_4\) precipitation, molecular sieve and ion exchange chromatography to isolate and purify the specific antigens against which antibodies were present in sera of type 1 diabetics.

4. The homogeneity of various fractions was tested by acrylamide disc electrophoresis.

5. The molecular weights of the purified antigens were determined by Sephacryl S-200 chromatography.

6. The immune homogeneity of the purified antigens was determined by raising the specific antisera in rabbits and the relatedness of various antigens was tested by immunodiffusion, immunoelectrophoresis and counterimmunoelectrophoresis.

7. The origin of the purified antigens was determined immunocytochemically, by immunoperoxidase technique, using rabbit antisera raised against these purified antigens.