STUDIES ON AUTO ANTIBODIES IN DIABETES MELLITUS

THESIS

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BY

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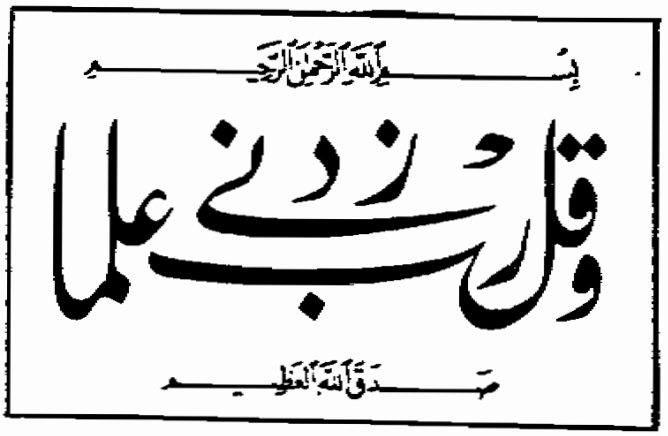

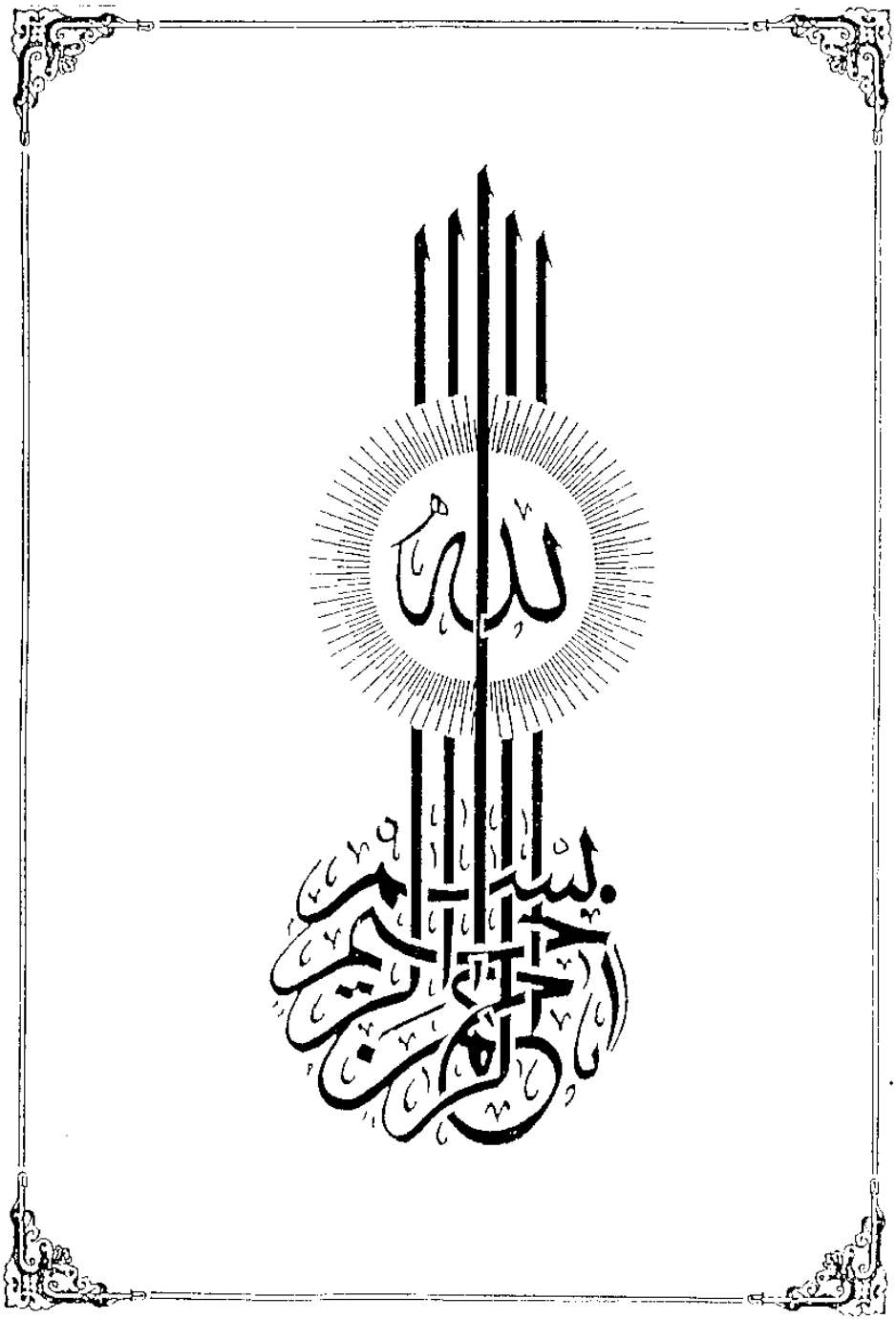

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Though insulin is a protein of low molecular weight it has been shown, both in man and in animal, to be weakly antigenic. Antibodies against parenterally administered insulin have been shown to have a role in insulin resistance and insulin allergy. However, there have been many studies describing the occurrence of antibodies against endogenous insulin and their possible role in the development of diabetes. Such antibodies have been presumed to be autoantibodies.

The evidence suggesting an autoimmune basis for certain types of diabetes is based on several findings. The clinical observations that diabetes coexists with overt autoimmunity and the presence of organ specific humoral autoantibodies are more often than can be accounted for by chance.

The role of autoantibodies in the pathogenesis of diabetic complications is still a matter of discussion, some workers have even denied their existence.

The aim of this work is to determine the incidence and level of circulating insulin autoantibody in newly diagnosed diabetics, and correlate between the level of IAAs and glycosylated haemoglobin, fasting and post prandial serum glucose level.

The ELISA technique is used for estimation of IAA in an attempt to define the sensitivity and specificity of this method in immunodiagnosis and prediction of diabetic subjects.

REVIEW OF LITERATURE

DIABETES MELLITUS

I. Definition:

Diabetes mellitus is generally defined by a particular degree of hyperglycaemia. It is characterized by metabolic abnormalities, by long term complications involving the eyes, kidneys, nerves, and blood vessels; and by a lesion of the basement membranes. Patients fulfilling these criteria are not a homogenous group **(Foster, 1987)**.

II. Classification:

The term diabetes mellitus covers a heterogenous group of disorders with widely varying aetiologies.

A classification of diabetes is given in table 2. The basic categories are those recommended by the National Diabetes Data Group in 1979.

This classification defines the differences in type 1 or Insulin dependant diabetes(IDD) and type 2 or non-insulin dependent diabetes (NIDD). Insulin dependance in this classification is not equivalent to insulin therapy. Many patients classified as non-insulin-dependant require insulin, and are not at risk for ketoacidosis. Some patients with apparent NIDD may become fully IDD and prone to keto-acidosis **(Foster, 1987)**.

a) Type I diabetics: Type 1 diabetics have essentially no insulin synthesis. Type 1 a has a major association with HLA

Table (1): Defintion of diabetes.

Definitions	IDDM	NIDDM	Type 1	Type 2
Clinical	Diabetic state requiring daily insulin injections to prevent a catabolic cascade culminating in diabetic ketoacidosis	Diabetic state that usually does not require daily insulin injections to prevent a catabolic cascade culminating in ketoacidosis	A diabetic state that may begin with a sudden onset of IDDM or evolve gradually through a variable period of NIDDM	A state of NIDDM generally associated with obesity in which weight reduction and oral hypoglycemic drugs may be effective
Pathogenetic	Diabetic state characterized by virtual absence of B-cells irrespective of etiology	A spectrum of diabetic states in which B-cells are present in subnormal,N. or increased quantities but are probably low in number relative to α-cells	Diabetic state in which destruction of B-cells presumably is caused by environment (viral?) or immunologic factors in genetically vulnerable indi-viduals usually expressing HLA DR3 and/or DR4 and islet cell antibodies	Diabetic state in which insullin resistance. usually obesity related, is prominent but accom-panied by a dysfunctional β-cell
Pathophysiological	Diabetic state in which insulin secretion is never sufficient to restrain excessive secretion of glucagon or to counter its enhancement of hepatic glucose and ketone production	Diabetic state in which insulin secretion is usually sufficient to oppose the ketogenic actions of glucagon but not to prevent hyperglcemia	Diabetic state in which insulin secretion may be reduced (NIDDM) or absent (IDDM) and in which coexisting absolute or relative hyperglucagonemia is readily corrected by exogenous insulin.	Diabetes in which insulin secretion may be high, low, or normal, but is insufficient to overcome insulin resistance in peripheral tissues

(Williams endoc., 1985)

antigens on chromosome 6 and with presence of islet cell antibody in the circulation.

Type Ib is characterized by the persistant presence of islet cell antibodies and familial association with other autoimmune endocrinopathies. About 10% of all diabetic persons are classified as type I **(Skillman, 1984).**

b) Type 2 diabetes: This is a heterogenous group of disorders, characterized by hyperglycaemia, presence of insulin with tendency to insulin resistance and familial aggregation.

There are two forms of type 2 diabetes mellitus, obese and non-obese forms. The non-obese type 2 diabetics tend to be relatively insulinopenic. While the obese has hyperinsulinaemia.

In most cases the age of onset is delayed to middle or old age. However, many examples of younger onset are present. Nearly 90% of all diabetic persons are of type 2 **(Alberti et al., 1982).**

Unusual type of diabetes may result from insulin resistance. Insulin resistance results from:

i) Severe reduction in receptor number **(Bottazo et al., 1974).**

ii) Production of insulin receptor antibodies **(Flier et al., 1975).**

iii) Synthesis of insulin with an abnormal sequence of amino-acids which leads to reduced biological activity **(Given et al., 1980).**

iv) Secretion of proinsulin rather than insulin **(Gabbay et al., 1976).**

v) Severe deficiency after the receptor binding step **(Kobayashi et al., 1978).**

c) Other types: Secondary Diabetes.

In a small proportion of cases of diabetes, disordered metabolism is secondary to a specific syndrome. There are at least 40 rare hereditary disorders of which diabetes may be a component **(Podolsky and Viswanathan, 1980).**

Secondary diabetes may occur in disorders that produce insulin antagonist. This category include acromegaly, cushing and pheochromocytoma. Glucagonoma patients mostly have diabetes **(Mallinson et al., 1974).**

Few drugs can actually cause secondary diabetes, such as thiazide diuretics which deplete total body potassium. Phenytoin (Dilantin) has a direct effect on beta-cell insulin release. Also adrenergic blocking agents such as propranolol has the same effect.

Clinical diabetes may be found if more than 90% of the islets are destroyed. Few diseases cause sufficient pancreatic destruction such as acute pancreatitis, chronic pancreatitis and chronic calcific pancreatitis **(Skillman, 1984).**

Table (2): Simplified classification of diabetes mellitus.

	Other names	Aetiological features	Clinical characteristics
e Ia	Insulin-dependent diabetes Juvenile onset diabetes Ketosis-prone diabetes	(i) Association with HLA types (e.g. DR3, DR4) (ii) Generally cytoplasmic and complement-fixing islet-cell-antibody positive at diagnosis but later become negative	In early phase retain some endogenous insulin secretion, and may have honeymoon phase. Later have no endogenous insulin. Develop ketosis when insulin withdrawn or with stress. May present with weight loss. Mostly young.
e Ib	Same as type Ia	(i) Close association with other autoimmune endocrinopathies (ii) Persistent islet-cell antibodies. (iii) Presumed autoimmune aetiology	Same as type Ia
e II n-obese)	Non-insulin-dependent diabetes (NIDDH) Maturity-onset diabetes (MOD) Maturity-onset diabetes of the young (MOD).	(i) Heterogeneous aetiologies (ii) Familial aggregation (iii) Environmental factors ?	Always measurable insulin present. Tendency to insulin resistance. Ketosis not provoked by insulin withdrawal but may become ketoacidotic with severe illness.Onset usually above age of 40.
e II (obese)	As for type II (non-obese)	(i) Related to obesity (ii) Glucose tolerance often normal after weight loss. (iii) Probably different aetiology from type II (non-obese)	Hyperinsulinaemic and insulin resistant. Rarely if ever ketotic.
her types Pancreatic disease	Secondary diabetes J type } type III K type } Z type }	Chronic pancreatitis. Pancreatic calcification. Alcohol may be important (K type). Malnutrition and cassava consumption also implicated (Z type). Pancreatectomy). Haemochromatosis.	Often underweight with history of malnutrition. Mostly restricted to tropical countries non-caucasians. May have severe insulin resistance although pancreatectomized subjects are insulin sensitive.
Hormonal		(i) Corticosteroid excess (exogenous or endogenous) (ii) Glucagonoma (iii) Acromegaly (iv) Thyrotoxicosis (v) Phaeochromocytoma. (vi) ? Hypothalamic lesions	Obvious signs of steroid excess. Associated with skin rash. Diabetes mild. Obvious signs and symptoms of excess of particular hormone.
Drug-induced		(i) Diuretics. (ii) Psychoactive agents. (iii) Catecholaminergic agents. (iv) Analgesics.	
Insulin receptor abnormalities.		(i) Congenital lipodystrophy (ii) Associated with acanthosis nigricans. (iii) Autoimmune insulin receptor antibodies.	
Genetic syndromes		Examples: glycogen-storage disease; ataxia telangiectasia; DIDMOAD syndrome; Huntigton's chorea; Laurence-Moon-Biedl syndrome; Werner's syndrome; Prader-Willi syndrome.	
npaired glucose lerance (IGT)	Asymptomatic diabetes Chemical diabetes Borderline diabetes. Latent diabetes. Subclinical diabetes.	Mild glucose intolerance from any cause.	Increased risk of macrovascular Likely to be obese.
estational diabetes.		Pregnancy. Normal GTT before hand	Diagnosis as for IGT or more severe forms of glucose intolerance.

National Diabetes Data Group, 1979.

III. Etiology:

a) Type I Diabetes Mellitus:

In the etiology of type I diabetes three factors interact to produce insulin deficiency: the genetic, the environmental and the immunological factor. These factors are present in different proportions and in some cases one of them predominates.

i) Genetic factors:

The genetic predisposition is probably permissive and not causal. The susceptibility genes in IDDM resides on the sixth chromosome in view of strong association between diabetes and certain human leukocyte antigens (HLA). HLA is coded by the major histocompatibility region on this chromosome. Four loci designated by the letters A, B, C, and D are recognized with alleles at each site identified by numbers.

Major alleles conferring enhanced risk for IDDM are HLA-DR_3, HLA-DW_3, HLA-DR_4, HLA-DW_4, HLA-B_8 and HLA-B_{15}. However, many persons carrying "high-risk" alleles never develop diabetes. Also diabetes can develop in absence of these "high-risk" allels.

Antigens B_7 and DR_2 (DW_2) have been called "low-risk" alleles. They are found with less frequency in diabetics than in general population **(Foster, 1987).**

ii) Environmental Factors:

An environmental factor is directly responsible for the islet cell damage in few cases. It is either a viral infection or a high dose of diabetogenic chemical agent.

Viruses are the most common environmental agents inducing diabetes in genetically susceptible persons. The onset of type I diabetes frequently coincides with or follows viral infection **(Notkin, 1979).** Neonatal rubella appears to impose a special risk (20%) **(Menser et al., 1978).**

Beta cell destruction and acute and chronic inflammation of the islets were observed in many cases of fatal Coxackie B and cytomegalo virus infection **(Jenson et al., 1980).**

Exposure to other non viral islet toxins enhances host susceptibility to viral damage.

Chemicals may be responsible for diabetes through islet cell damage **(Toniolo et al., 1980).**

iii) Immunological factors:

The immune-directed destruction of beta cells involves both humoral and cell-mediated mechanisms. Many types of antibody have been identified e.g insulin, cytoplasmic and surface. Islet cell surface antibody can fix complement and lyse beta cells. It can also impair insulin release even before beta cell damage.

Cytotoxic T-lymphocytes and antibody-dependant killer T cells participate in and complete the destructive process. By the time overt diabetes appears, most insulin-producing cells has disappeared **(Foster, 1987).**