

INTRODUCTION

Type 1 Diabetes Mellitus (T1DM) is a chronic autoimmune disease that involves failure in immune regulation. It is characterized by destruction of the insulin-producing cells by auto-reactive T cells (*Atkinson and Brusko, 2007*).

The increasing incidence of type 1 diabetes in many countries challenges health systems because the disease is presently incurable with no known method of prevention (*James et al., 2014*). Around 490,100 children live with the disease worldwide, with incidence estimated to be increasing in children under 15 years by 2.8% per year (*Catanzariti et al., 2009*). This trend is particularly worrying because type 1 diabetes increases mortality and morbidity population-wide (*James et al., 2014*).

The finding of organ-specific autoantibodies in patients with T1DM discloses the existence of a wide spectrum of immunological diseases. Fifteen to 30% of patients with T1DM have autoimmune thyroiditis, 4-9% have celiac disease, and 0.5% have Addison's disease (*Barker et al., 2005*). Autoimmune hepatitis (AIH) has been reported to occur in patients with T1DM (*Barker, 2006; Al-Hussaini et al., 2014*).

In pediatrics, there are 2 liver disorders in which liver damage most likely stems from an autoimmune attack: 'classical' autoimmune hepatitis (AIH) and the AIH/sclerosing

cholangitis overlap syndrome (also known as autoimmune sclerosing cholangitis, ASC) (*Liberal et al., 2015*).

The pathogenesis of pediatric-onset autoimmune liver disease (AILD) is not fully understood, although there is mounting evidence that genetic susceptibility, molecular mimicry and impaired immunoregulatory networks contribute to the initiation and perpetuation of the autoimmune attack. Liver damage is thought to be mediated primarily by CD4 positive T-cells (*Liberal et al., 2015*).

Onset of AILD is frequently insidious with nonspecific symptoms such as fatigue, jaundice, nausea, abdominal pain, and arthralgias at presentation, but the clinical spectrum is wide, ranging from an asymptomatic presentation to an acute severe disease (*Ichai et al., 2007*).

The diagnosis of AIH is based on a combination of biochemical, immunological and histological features and exclusion of other causes of liver disease (*Tiniakos et al., 2015*). The diagnosis is based on histologic abnormalities, characteristic clinical and laboratory findings, abnormal levels of serum globulins, and the presence of one or more characteristic autoantibodies (*Krawitt, 2006*).

Typical histological features include a chronic hepatitis pattern of injury with portal inflammation and interface activity, predominance of plasma cells in the portal infiltrate,

emperipolesis, and hepatocellular rosette formation. Centrilobular injury with prominent hepatocellular necrosis and mononuclear inflammation is now recognized in the histological spectrum of AIH and may represent an early stage of the disease. Liver histology plays a major role in clinical diagnostic scoring systems and is important to confirm or support the clinical diagnosis of AIH (*Tiniakos et al., 2015*).

The detection of diagnostic autoantibodies such as antinuclear antibodies (ANA), anti-smooth muscle antibodies (SMA), anti liver/kidney microsomal type 1 (anti-LKM1), anti-liver cytosol type 1 (anti-LC1) and anti-soluble liver antigen (anti-SLA) is considered of utmost importance in the diagnostic process, as the International Autoimmune Hepatitis Group outlined in the revised criteria for the diagnosis of AIH (*Hennes et al., 2008; Muratori et al., 2015*).

The pediatric literature about type 1 diabetes– related liver disease is largely limited to small case series or case reports for children presenting with symptomatic hepatic dysfunction during metabolic decompensation and ketosis (*Al-Hussaini et al., 2012*).

AIM OF THE WORK

The aim of this study was to determine the prevalence of autoimmune hepatitis-related autoantibodies in children with T1DM and assess the relation between AIH and glycemic control as well as hepatic affection.

Chapter 1

TYPE 1 DIABETES MELLITUS

Definition and Description

Diabetes Mellitus is a metabolic disorder characterized by the presence of hyperglycemia due to defective insulin secretion, defective insulin action, or both (*International Society of Pediatric and Adolescent Diabetes ISPAD, 2014*). The chronic hyperglycemia of diabetes is associated with relatively specific long term microvascular complications affecting the eyes, kidneys and nerves, as well as an increased risk for cardiovascular disease (CVD) (*American Diabetes Association, 2012*).

Diabetes mellitus may present with characteristic symptoms such as thirst, polyuria, blurring of vision, and weight loss. In its most severe forms, ketoacidosis or a non-ketotic hyperosmolar state may develop and lead to stupor, coma and, in absence of effective treatment, death. Often symptoms are not severe, or may be absent, and consequently hyperglycemia sufficient to cause pathological and functional changes may be present for a long time before the diagnosis is made (*World Health Organization, 1999*).

Several pathogenic processes are involved in the development of diabetes. These include processes which destroy the beta cells of the pancreas with consequent insulin deficiency, and others that result in resistance to insulin action.

The abnormalities of carbohydrate, fat and protein metabolism are due to deficient action of insulin on target tissues resulting from insensitivity or lack of insulin (*World Health Organization, 1999*).

The long-term effects of diabetes mellitus include progressive development of the specific complications of retinopathy with potential blindness, nephropathy that may lead to renal failure, and/or neuropathy with risk of foot ulcers, amputation, Charcot joints, and features of autonomic dysfunction, including sexual dysfunction. People with diabetes are at increased risk of cardiovascular, peripheral vascular and cerebrovascular disease (*McCance et al., 1994*).

Global burden

Diabetes in all its forms imposes unacceptably high human, social and economic costs on countries at all income levels:

- There are 320.5 million people of working age (20-64 years) with diabetes and 94.2 million people aged 65-79 with diabetes by 2040 this will rise to 441.3 million of working age (20-64 years) and 200.5 million people aged 65-79 (Table 1).
- The number of people with type 2 diabetes is increasing in every country.
- 80% of people with diabetes live in low- and middle-income countries.

- The greatest number of people with diabetes is between 40 and 59 years of age.
- 193 million people with diabetes are undiagnosed.
- Approximately 5.0 million people aged between 20 and 79 years died from diabetes in 2015, equivalent to one death every six seconds.
- For the first time, the estimated number of children living with type 1 diabetes (542,000) exceeds half a million.
- The incidence of type 1 diabetes among children is increasing in many countries, particularly in children under the age of 15 years.

(IDF Diabetes Atlas, 2015)

Table (1): Top 10 countries/ territories for number of people with diabetes (20-79 years), 2015 and 2040

Rank	Country/territory	Number of people with diabetes	Rank	Country/territory	Number of people with diabetes
1	China	109.6 million (9.6-133.4)	1	China	150.7 million (138.0-179.4)
2	India	69.2 million (56.2-84.8)	2	India	123.5 million (99.1-150.3)
3	United States of America	29.3 million (27.6-30.9)	3	United States of America	35.1 million (33.0-37.2)
4	Brazil	14.3 million (12.9-15.8)	4	Brazil	23.3 million (21.0-25.9)
5	Russian Federation	12.1 million (6.2-17.0)	5	Mexico	20.6 million (11.4-24.7)
6	Mexico	11.5 million (6.2-13.7)	6	Indonesia	16.2 million (14.3-17.7)
7	Indonesia	10.0 million (8.7-10.9)	7	Egypt	15.1 million (7.3-17.3)
8	Egypt	7.8 million (3.8-9.0)	8	Pakistan	14.4 million (10.6-20.4)
9	Japan	7.2 million (6.1-9.6)	9	Bangladesh	13.6 million (10.7-24.6)
10	Bangladesh	7.1 million (5.3-12.0)	10	Russian Federation	12.4 million (6.4-17.1)

(IDF Diabetes Atlas, 2015)

Incidence and Prevalence in Egypt

The incidence of type 1 diabetes mellitus (T1DM) is increasing in all population at a rate of approximately 3% per year and the onset of the condition is occurring at younger age. Its incidence varies dramatically between populations and even within the same population, much of this variation is due to genetic defect. Among Eastern Mediterranean and Middle Eastern countries, the largest contribution to the total number of estimated childhood T1DM cases comes from Egypt which accounts for about a quarter of the region's total. The incidence varies between 1/100 000 per year (Pakistan) and 8/100 000 per year (Egypt) in children under the age of 15 years (*Soltész et al., 2006*).

An Egyptian study of incidence and prevalence of T1DM in children and adolescents in four Egyptian Governorates (Fayoum, Minofeya, North Sainai and Sues) was held by *Salem et al. (2007)*, showing a prevalence rate of 0.7/1000 and an incidence rate of 4.01/100.000.

Classification

The American diabetes association (ADA) classification of DM encompasses 4 groups: Type 1, Type 2, other specific types of diabetes, and gestational diabetes. Type 1 DM is further subclassified into Type 1A which is associated with the presence of islet cell autoantibodies, and Type 1B characterized by the absence of such antibodies (*Sperling, 2002; Devendra et al., 2004*). A classification of diabetes is presented in Table 2.

Table (2): Etiological classification of diabetes mellitus

I. Type I-DM: (B-cell destruction, usually leading to absolute insulin deficiency) A. Immune mediated. B. Idiopathic.	II. Type II-DM: (may range from predominantly insulin resistance with relative insulin deficiency to a predominantly secretory defect with insulin resistance).
III. other specific types	
A. Genetic defects of β-cell function: 1. MODY3 (Chromosome 12, HNF1 α) 2. MODY1 (Chromosome 20 HNF4 α) 3. MODY2 (Chromosome 7 glucokinase) 4. Other very rare forms of MODY (e.g. MODY4: Chromosome 13, insulin Promoter factor-1, MODY6: Chromosome 9, carboxyl ester lipase) 5. Transient neonatal diabetes 6. Permanent neonatal diabetes 7. Mitochondrial DNA 8. Others	B. Genetic defect in insulin action 1.Type A insulin resistance 2.Leprechaunism 3.Rabson-Mendenhall syndrome 4.Others
B. Diseases of exocrine pancreas 1.Pancreatitis 2.Truama/ pancreatectomy 3.Neoplasia 4.Cystic fibrosis 5.Haemochromatosis 6.Fibrocalculous pancreatopathy 7.Others	D. Endocrinopathies 1.Acromegaly 2.Cushing's syndrome 3.Glucagonoma 4.Pheochromocytoma 5.Hyperthyroidism 6.Somatostatinoma 7.Aldosteronoma 8.Others
E. Drugs or chemical- induced 1.Vacor 2.Pentamidine 3.Nicotinic acid 4.Glucocorticoids 5.Thyroid hormone 6.Diazoxide 7.B-adrenegic agonist 8.Thiazides 9.Dilantin 10. γ -interferon 11.Others	F. Infections 1.Congenital rubella 2.cytomegalo virus 3.Others
G. Uncommon forms of immune-mediated Diabetes 1."Stiff-man" syndrome 2.Anti-insulin receptor antibodies 4.Others	H. Other genetic syndromes sometimes Associated with diabetes 1.Down syndrome 2.Klinefelter syndrome 3.Turner syndrome 4.Wolfram syndrome 5.Friedreich's ataxia 6.Huntington's chorea 7.Laurence-Moon-Biedle syndrome 8.Myotonic dystrophy 9.Porphyrria 10.Prader-Willi syndrome 11.Others
IV. Gestational diabetes.	

(ISPAD, 2014)

MODY: Maturity onset diabetes of the young, **HNF-4 α :** Hepatocyte Nuclear Factor, **NeuroD1:** Neurogenic differentiation.

Although the vast majority of children and young people with diabetes in the UK have type 1 diabetes, an increasing number are diagnosed with type 2 diabetes, recently reported at around 2% (*Shepherd and Cropper, 2013*). Other causes of childhood diabetes include neonatal diabetes, maturity-onset diabetes of the young (MODY), secondary diabetes and syndromic diabetes (*Royal College of Pediatrics and Child Health, 2009*). However, the correct classification of pediatric diabetes can be challenging (table 4); there may be confusion between the presenting features of different types of diabetes, which can lead to erroneous diagnosis (*Ehtisham et al., 2004*).

Monogenic diabetes results from mutations in single genes that regulate beta-cell function, but is commonly misdiagnosed as type 1 or type 2 diabetes (*Lambert et al., 2003; Shepherd, 2008*), and only a minority of estimated cases have been confirmed by genetic testing in the UK (*Shields et al., 2010*). Individuals with monogenic diabetes do not need to be insulin resistant or obese to develop diabetes (*Ehtisham et al., 2004*).

Guidelines have been developed to highlight when a diagnosis of monogenic diabetes should be considered in children (table 3) and when to suspect that a diagnosis of type 1 or type 2 diabetes is not correct (*Hattersley et al., 2006*). The approaches are separated into:

Those diagnosed less than 6 months of age (indicating neonatal diabetes).

Those who may otherwise have been labeled as having type 1 diabetes (a child with diabetes has an affected parent and evidence of endogenous insulin production outside the honeymoon period).

Those who may otherwise have been labeled as having type 2 diabetes that are not markedly obese, do not have acanthosis nigricans or other evidence of insulin resistance and are from a low-prevalence ethnic group (*Hattersley et al., 2006*).

These guidelines should be used in the clinical setting to identify cases of atypical type 1 diabetes or type 2 diabetes, which warrant further investigation.

Table (3): Criteria to consider monogenic diabetes in children

When to consider monogenic diabetes in children
Diabetes diagnosed below the age of 6 months (neonatal diabetes)
Family history of diabetes with an affected parent
Mild fasting hyperglycemia (5.2-8 mmol/L), especially if young or familial
Diabetes associated with extra-pancreatic features

(*Hattersley et al., 2006*)

Table (4): Characteristic Phenotypes of the commonly encountered diabetes subtypes, illustrating the clinically useful differences between type 1 and type 2 diabetes, and monogenic forms of diabetes

Features associated with diabetes	Type 1 diabetes	Young onset Type 2 diabetes	Monogenic diabetes					
			GCK*	HNF1A#	HNF4A#	HNF1B#	Neonatal diabetes	MIDD#
DKA	Yes	No	No	No ∞	No ∞	No	Yes	Yes/No
Parent affected	2%-4%	Yes	Yes	Yes	Yes	Yes	Variable	Mother
Age of onset	6 months to adulthood	Adolescence and young adulthood	Birth	Teens to young adulthood	Teens to young adulthood	Teens to young adulthood	< 6 months	Young adulthood
Obesity	Population frequency	Increased Frequency	Population frequency	Population frequency	Population frequency	Population frequency	Population frequency	Rare
Glycaemic pattern	Acute General hyperglycemia	Progressive hyperglycemia	Stable, mild fasting glycemia	Post-prandial, 12initially progressing to general hyperglycemia	Post-prandial, 12initially progressing to general hyperglycemia	Post-prandial, 12initially progressing to general hyperglycemia	Acute General hyperglycemia	Variable dysglycaemic pattern either acute or slowly progressive
B cell antibodies±	Yes	No	No	No	No	No	No	No
C-peptide¥	Very low/Absent (< 5 years)	Raised/ Normal	Normal	Low but detectable	Low but detectable	Low but detectable	Absent but detectable once treated with SU	Low but detectable
hsCRP	Normal	High/ High normal	Normal	Very low	Normal	Normal	Normal	Normal
Additional clinical features	Other autoimmune disease (Thyroid, celiac, etc.	Dyslipidaemia, PCOS, Hypertension, Acanthosis nigricans	Absence of microvascular and macrovascular complications	Low renal threshold for glucose in early stages of diabetes	Macrosomia and transient neonatal hypoglycemia	High renal involvement e.g., cysts, etc,	Transient in 50% of cases although may relapse	Deafness, short stature, macular dystrophy

(Carroll & Murphy, 2013)

*= Glucokinase, #= Hepatocyte nuclear factor, ± = Mitochondrial diabetes and deafness, ∞= Excellent responses to sulfonylurea therapy are commonly noted

Type 1 Diabetes Mellitus

Type 1 diabetes is a disorder that arises following the autoimmune destruction of insulin-producing pancreatic b cells (*Atkinson, 2001; Bluestone et al., 2010*). The disease is most often diagnosed in children and adolescents, usually presenting with a classic trio of symptoms (i.e., polydipsia, polyphagia, polyuria) alongside of overt hyperglycemia, positing the immediate need for exogenous insulin replacement-a medicinal introduction to the disorder whose therapeutic practice lasts a lifetime (*Thunandera et al., 2008*).

Classification of type 1 diabetes

Type 1 diabetes may be subdivided in three groups from the etiological point of view: autoimmune, idiopathic and double. The autoimmune group is represented by: type 1A, which is polygenic and it is the most frequent type of this disease, corresponding to approximately 80-90% of all T1DM cases (*Daneman, 2006*).

The other subtype of this group is the latent autoimmune diabetes in adults (LADA) (*Fourlanos et al., 2006*), which appears after the age of 35 and is frequently associated with other autoimmune endocrine diseases.

The third subtype includes called "monogenic" T1DM. They correspond to T1DM of the autoimmune polyglandular syndrome type 1A (*Su and Anderson, 2004*) and of IPEX

syndrome (Immune Dysfunction, Polyendocrinopathy, Enteropathy, X-linked) (*Wildin and Freitas, 2005*).

Type 1B, also called idiopathic, has all the clinical features of type 1A, but the autoimmune component is not detected (*American Diabetes association, 2008*). Another 1B subtype is the fulminant diabetes most described in Asian peoples, mainly Japan, China and Korea, characterized by a short clinical history, before to the first acute metabolic decompensation, presents the impairment of beta and alpha cells of the pancreatic islet and no autoimmune etiology (*Imagawa et al., 2000*).

Finally, the denomination of mixed, 1.5 or double (type 1 plus type 2) diabetes has been proposed when we have the type 1A (autoimmunity) plus type 2 (obesity, insulin resistance, dyslipidemia) diabetes characteristics in the same individual (*Libman and Becker, 2003*).

Etiology of type 1 diabetes

The etiology of type 1 diabetes remains poorly understood, but it is likely that an environmental factor triggers an autoimmune process in a predisposed individual (figure 1). Although the genetic susceptibility to type 1 diabetes is inherited, only 12–15% of type 1 diabetes occurs in families. Twin studies have shown that the concordance rate for type 1 diabetes in monozygotic twins is around 20–30% (*Holt, 2004*).

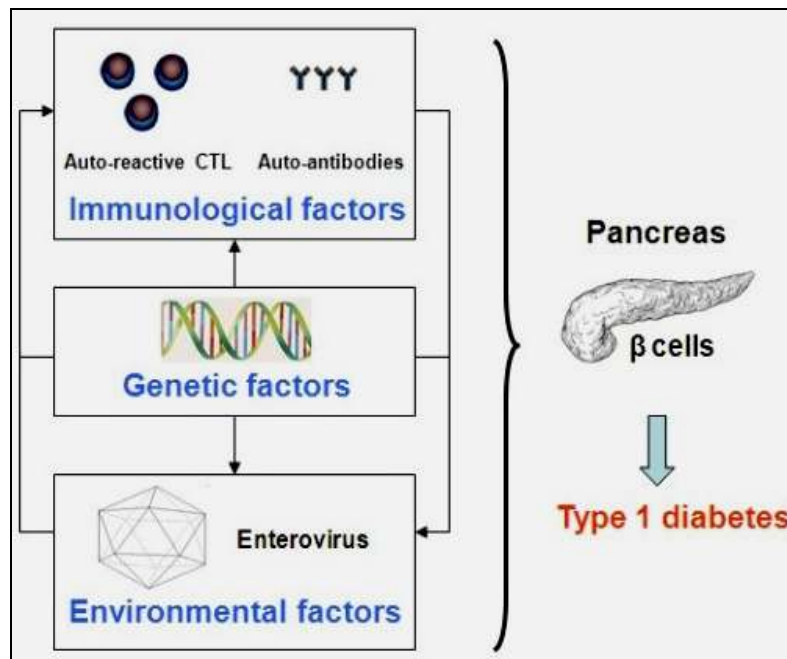


Figure (1): Types 1 diabetes and environmental factors. Type 1 diabetes is the result of interactions between immunological, genetic, and environmental factors, especially viruses mainly represented by enteroviruses (www.discoverymedicine.com/enteroviral-pathogenesis-of-type-1-diabetes).

Studies indicate that genetic factors do not account entirely for the development of type 1 diabetes, and several environmental triggers, including viral infections, nutritional factors, parental age and low birth weight, have been implicated (*Akerblom et al., 2002*).

1. Genetics of type 1 diabetes

Despite being strongly influenced by genetic factors, type 1 DM does not fit any simple pattern of inheritance and is considered a complex, multifactorial disease (*Noble and*