# Genetics and Diabetes

## Background

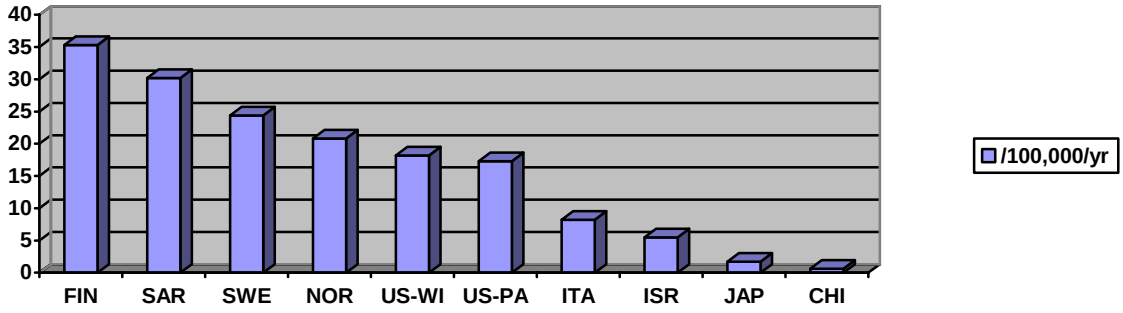
Diabetes mellitus is a heterogeneous group of disorders characterized by persistent hyperglycemia. The two most common forms of diabetes are type 1 diabetes (T1D, previously known as insulin-dependent diabetes or IDDM) and type 2 diabetes (T2D, previously known as non-insulin-dependent diabetes or NIDDM). Both are caused by a combination of genetic and environmental risk factors. However, there are other rare forms of diabetes that are directly inherited. These include maturity onset diabetes in the young (MODY), and diabetes due to mutations in mitochondrial DNA.

All forms of diabetes have very serious effects on health. In addition to the consequences of abnormal metabolism of glucose (e.g., hyperlipidemia, glycosylation of proteins, etc.), there are a number of long-term complications associated with the disease. These include cardiovascular, peripheral vascular, ocular, neurologic and renal abnormalities, which are responsible for morbidity, disability and premature death in young adults. Furthermore, the disease is associated with reproductive complications causing problems for both mothers and their children. Although improved glycemic control may decrease the risk of developing these complications, diabetes remains a very significant cause of social, psychological and financial burdens in populations worldwide.

### *Type 1 Diabetes*

Epidemiology**.** T1D is caused by the autoimmune destruction of the beta cells of the pancreas, and represents approximately 10% of all cases with diabetes. At present, lifelong insulin therapy is the only treatment for the disease. Without exogenous insulin injections, individuals with T1D will not survive. Although the prevalence of T1D is <1% in most populations, the geographic variation in incidence is enormous, ranging from <1/100,000 per year in China to approximately 40/100,000 per year in Finland (Figure 1) (Karvonen et al., 1993). The only chronic childhood disorder more prevalent than T1D is asthma. It has been estimated that approximately 20 million people worldwide, mostly children and young adults, have T1D (Holt, 2004).

**Figure 1. T1D Incidence Rates Worldwide**




FIN = Finland, SAR = Sardinia, SWE = Sweden, NOR = Norway, US-WI = US-Wisconsin, US-PA = US-Pennsylvania, ITA = Italy, ISR = Israel, JAP = Japan, CHI = China

The incidence of T1D is increasing worldwide at a rate of about 3% per year (Onkamo et al., 1999). This trend appears to be most dramatic in the youngest age groups, and is completely unrelated to the current increase in T2D in children. More children with beta cell autoantibodies, a hallmark of T1D,

are being diagnosed with the T1D around the world each year. Although the peak age at onset is at puberty, T1D can also develop in adults. Epidemiologic studies have revealed no significant gender differences in incidence among individuals diagnosed before age 15 (Kyvik et al., 2004). However, after age 25, the male to female incidence ratio is approximately 1.5. There is also a notable seasonal variation in the incidence of T1D in many countries, with lower rates in the warm summer months, and higher rates during the cold winter (Dorman et al., 2003).

Environmental Risk Factors**.** The epidemiological patterns described above suggest that environmental factors contribute to the etiology of the T1D. In particular, the recent temporal increase in T1D incidence points to a changing global environment rather than variation in the gene pool, which require the passage of multiple generations. Twin studies also provide evidence for the importance of environmental risk factors for T1D. T1D concordance rates for monozygous twins are higher than those for dizygous twins (approximately 30% vs. 10%, respectively) (Hirschhorn, 2003). However, most monozygous twin pairs remain discordant. Thus, T1D cannot be completely genetically determined.

Environmental risk factors are thought to act as either 'initiators' or 'accelerators' of beta cell autoimmunity, or 'precipitators' of overt symptoms in individuals who already have evidence of beta cell destruction. They also may function by mechanisms that are directly harmful to the pancreas, or by indirect methods that produce an abnormal immune response to proteins normally present in cells. The T1D environmental risk factors that have received most attention are viruses and infant nutrition.

Enteroviruses, especially Coxsackie virus B (CVB), have been the focus of numerous ecologic and case-control studies (Dahlquist et al., 1998). CVB infections are frequent during childhood and are known to have systemic effects on the pancreas. Recent prospective studies are helping to elucidate the role of viruses to the etiology of T1D. For example, enteroviral infections occurring as early as *in utero* appear to increase a child's subsequent risk of developing the disease (Dahlquist et al., 1995, Hyoty et al., 1995). Other viruses, including mumps (Hyoty et al., 1993), cytomegalovirus (Pak et al., 1988), rotavirus (Honeyman et al., 2000) and rubella, (McIntosh and Menser, 1992) have also been associated with the disease.

Another hypothesis that has been the subject of considerable interest relates to early exposure to cow's milk protein and the subsequent development of T1D. The first epidemiologic observation of such a relationship was by Borch-Johnsen et al., who found that T1D children were breast-fed for shorter periods of time than their non-diabetic siblings or children from the general population (Borsh-Johnsen et al., 1984). The authors postulated that the lack of immunologic protection from insufficient breast-feeding may increase risk for T1D later during childhood. It was also postulated that shorter duration of breast feeding may indirectly reflect early exposure to dietary proteins that stimulate an abnormal immune response in newborns. Most recently it has been hypothesized that the protective effect of breast-feeding may be due, in part, to its role in gut maturation (Kolb and Pozzilli, 1999; Harrison and Honeyman, 1999; Vaarala, 1999). Breast milk contains growth factors, cytokines, and other substances necessary for the maturation of the intestinal mucosa. Breast-feeding also protects against enteric infections during infancy, and promotes proper colonization of the gut. Interestingly, enteroviral infections can also interfere with gut immunoregulation, which may explain the epidemiologic associations between viral infections and T1D.

The role of hygiene in the etiology of T1D is also currently being explored (McKinney et al., 1997; Marshall et al., 2004). It has been hypothesized that delayed exposure to microorganisms due to improvements in standard of living hinders the development of the immune system, such that it is more

likely to respond inappropriately when introduced to such agents at older (compared to younger) ages. This explanation is consistent with recent reports indicating that factors such as day care attendance (McKinney et al. 2000), sharing a bedroom with a sibling, and contact with pets are protective against T1D (Marshall et al., 2004). Further studies are needed to determine if improved hygiene can explain the temporal increase in the incidence of T1D worldwide.

***Type 2 Diabetes***

Epidemiology. T2D is the most common form of the disease, accounting for approximately 90% of all affected individuals. A diagnosis of T2D is made if a fasting plasma glucose concentration is $\geq$ 7.0 mmol/L ($\geq$ 126 mg/dl) or plasma glucose 2 hours after a standard glucose challenge is $\geq$ 11.1 mmol/L ($\geq$ 200 mg/dl) (WHO, 1999) T2D is caused by relative impaired insulin secretion and peripheral insulin resistance. Typically, T2D is managed with diet, exercise, oral hypoglycemic agents and sometimes exogenous insulin. However, it is associated with the same long-term complications as T1D.

The highest rates of T2D are found among Native Americans, particularly the Pima Indians who reside in Arizona in the US, and in natives of the South Pacific islands, such as Nauru (Wild et al., 2004). T2D is also known to be more predominant in Hispanic and African American populations than in Caucasians. In 2000, it is estimated that 171 million people (2.8% of the worlds population) had diabetes and that by 2030 this number will be 366 million (4.4% of the world's population). The vast majority of this increase will occur in men and women aged 45 to 64 years living in developing countries. According to Wild et al.(2004), the 'top' three countries in terms of the number of T2D individuals with diabetes are India (31.7 million in 2000; 79.4 million in 2030), China (20.8 million in 2000; 42.3 million in 2030) and the US (17.7 million in 2000; 30.3 million in 2030). Clearly, T2D has become an epidemic in the 21$^{st}$ century.

In addition to the burden of T2D there is an even larger number of people with raised levels of blood glucose but below the level for diabetes. The World Health Organization defines impaired fasting glucose as a fasting plasma glucose level of $\geq$ 6.1 mmoll$^{-1}$ and less than 7 mmoll$^{-1}$, and impaired glucose tolerance as 2 hour plasma glucose, post glucose challenge, of 7.8 to less than 11.1 mmoll$^{-1}$ (WHO, 1999).

The prevalence of T2D increases with age of population (Wild et al., 2004). In developing countries, the largest number of people with diabetes are in the age group 45 to 64 years, while in developed the largest number is found in those aged 65 years and over. These differences largely reflected differences in population age structure between developed and developing countries. Worldwide rates are similar in men and women, although they are slightly higher in men < 60 years of age and in women > age 65 years.

Of great concern is the recent increase in T2D in children (Bloomgarden, 2004). A report based on the Pima Indians in Arizona noted that between 1967-76 and 1987-96, the prevalence of T2D increased 6-fold in adolescents (Fagot-Campagna et al., 2000). In the US, the incidence of T2D increased from 0.3-1.2/100,000/yr before 1992 to 2.4/100,000/yr in 1994 (Weill et al., 2004). Most T2D children diagnosed during this period were females from minority populations, with a mean age of onset at around puberty. They were also likely to have a positive family history of the disease, particularly maternal diabetes.

Environmental Risk Factors. As early as 1962, Neel hypothesized that T2D represented a 'thrifty genotype', which had a selective advantage (Neel, 1962). He postulated that in primitive times, individuals who were 'metabolically thrifty' and able to store a high proportion of energy as fat when food was plentiful were more likely to survive times of famine. However, in recent years, most populations experience a continuous supply of calorie-dense processed foods, as well as a decrease in physical activity. This likely explains the rise in T2D prevalence worldwide.

The major environmental risk factors for T2D are obesity ($\geq$ 120% ideal body weight or a body mass index $\geq$ 30 k/m$^2$) and a sedentary lifestyle (van Dam, 2003; Shaw and Chisholm, 2003). Thus, the tremendous increase in the rates of T2D in recent years has been attributed, primarily, to the dramatic rise in obesity worldwide (Zimmet et al., 2001). It has been estimated that approximately 80% of all new T2D cases are due to obesity (Lean, 2000). This is true for adults and children. In the Pima Indians, 85% of the T2D children were either overweight or obese (Fagot-Campagna et al., 2000). Another study in the US reported that IGT was detected in 25% of obese children age 4-10 years, and in 21% of obese adolescents (Sinha et al., 2002). Undiagnosed T2D was detected in 4% of the adolescents.

In addition to general obesity, the distribution of body fat, estimated by the ratio of waist-to-hip circumference (WHR), also has an impact on T2D risk. WHR is a reflection of abdominal (central) obesity, which is more strongly associated with T2D than the standard measures of obesity, such as those based on body mass index.

The other major T2D risk factor is physical inactivity. In addition to controlling weight, exercise improves glucose and lipid metabolism, which decreases T2D risk. Physical activity, such as daily walking or cycling for more than 30 minutes, has been shown to significantly reduce the risk of T2D (Hu et al., 2003). Physical activity has also been inversely related to body mass index and IGT. Recently, intervention studies in China (Pan et al., 1997), Finland (Tuomilehto J et al., 2001) and the US (Diabetes Prevention Program Study Group, 2002) have shown that lifestyle interventions targeting diet and exercise decreased the risk of progression from IGT to T2D by approximately 60% . In contrast, oral hypoglycemic medication only reduced the risk of progression by about 30%.

There is also considerable evidence suggesting that the intrauterine environment is an important predictor of T2D risk (Hales and Barker, 2001; Sobngwi et al., 2003), Numerous studies have shown that low birth weight, which is an indicator of fetal malnutrition, is associated with IGT and T2D later in life. However, it is unclear whether low birth weight is causal or related to potential confounding factors that contribute to both poor fetal growth and T2D (Frayling and Hattersley, 2001).

## Role of Genetics in the Development of Diabetes

### *Type 1 Diabetes*

First degree relatives have a higher risk of developing T1D than unrelated individuals from the general population (approximately 6% vs. <1%, respectively) (Dorman and Bunker, 2000). These data suggest that genetic factors are involved with the development of the disease. At present, there is evidence that more than 20 regions of the genome may be involved in genetic susceptibility to T1D. However, none of the candidates identified have a greater influence on T1D risk than that conferred by genes in the HLA region of chromosome 6. This region contains several hundred genes known to be involved in

immune response. Those most strongly associated with the disease are the HLA class II genes (i.e., HLA-DR, DQ, DP).

*IDDM1.* The HLA class II genes, also referred to as *IDDM1*, contribute approximately 40-50% of the heritable risk for T1D (Hirschhorn et al., 2003). When evaluated as haplotypes, DQA1*0501-DQB1*0201 and DQA1*0301-DQB1*0302 are most strongly associated T1D in Caucasian populations. They are in linkage disequilibrium with DRB1*03 and DRB1*04, respectively. Specific DRB1*04 alleles also modify the risk associated with the DQA1*0301-DQB1*0302 haplotype. Other reported high risk haplotypes for T1D include DRB1*07-DQA1*0301-DQB1*0201 among African Americans, DRB1*09-DQA1*0301-DQB1*0303 among Japanese, and DRB1*04-DQA1*0401-DQB1*0302 among Chinese. DRB1*15-DQA1*0602-DQB1*0102 is protective and associated with a reduced risk of T1D in most populations. Recent reports suggest that other genes in the central, class I and extended class I regions may also increase T1D risk independent of HLA class II genes (Nejentsev et al., 1997; Lie et al., 1999).

Individuals with two high risk DRB1-DQA1-DQB1 haplotypes have a significantly higher T1D risk than individuals with no high risk haplotype. The T1D risk among those with only one susceptibility haplotype is also increased, but effect is more modest. Relative risk estimates range from 10 – 45 and 3-7, respectively, for these groups, depending on race (Dorman and Bunker, 2000). In terms of absolute risk, Caucasian individuals with two susceptibility haplotypes have an approximately 6% chance of developing T1D through age 35 years. However, this figure is substantially lower in populations where T1D is rare (i.e., < 1% among Asians). In addition to *IDDM1*, two other genes are now known to influence T1D risk (Anjos and Polychronakos, 2004). These include *INS* and *CTLA-4*.

**Table 1. Several T1D Susceptibility Genes**

| Gene | Locus | Variant | Estimated RR[†] |
|---|---|---|---|
| *HLA-DQB1* | 6p21.3 | *0201 & *0302 | 3 – 45 |
| *INS* | 11p15. 5 | Class I | 1 – 2 |
| *CTLA4* | 2q31-35 | Thr17Ala | 1 – 2 |

[†]RR = relative risk

*INS* (insulin). The *INS* gene, located on chromosome 11p15.5, has been designated as *IDDM2*. Positive associations have been observed with a non-transcribed variable number of tandem repeat (VNTR) in the 5' flanking region (Bennett et al., 1997; Pugliese et al., 1997) . There are two common variants. The shorter class I variant predisposes to T1D (relative increase: 1 – 2), whereas the longer class III variant appears to be dominantly protective. The biological plausibility of these associations may relate to the expression of insulin mRNA in the thymus. Class III variants appear to generate higher levels of insulin mRNA than class I variants. Such differences could contribute to a better immune tolerance for class III positive individuals by increasing the likelihood of negative selection for autoreactive T-cell clones. The effect of *INS* appears to vary by ethnicity, with lesser effects in non-Caucasian populations (Undlien et al. 1994).

*CTLA-4* (cytotoxic T lymphocyte-associated 4). The *CTLA-4* gene is located on chromosome 2q31-35 (Anjos and Polychronakos, 2004), where multiple T1D genes may be located. CTLA-4 variants have been associated with T1D, as well as other autoimmune disease. *CTLA-4* negatively regulates T-cell

function. However, impaired activity, which has been associated with the Thr17Ala variant, may increase T1D risk. Overall, the relative increase in risk for the CTLA-4Ala17 variant has been estimated as ~ 1.5.

***Type 2 Diabetes***

It has long been known that T2D is, in part, inherited. Family studies have revealed that first degree relatives of individuals with T2D are about 3 times more likely to develop the disease than individuals without a positive family history of the disease (Flores et al., 2003; Hansen 2003; Gloyn 2003). It has also been shown that concordance rates for monozygotic twins, which have ranged from 60-90%, are significantly higher than those for dizygotic twins. Thus, it is clear that T2D has a strong genetic component.

One approach that is used to identify disease susceptibility genes is based on the identification of candidate genes (Barroso et al., 2003; Stumvoll, 2004). Candidate genes are selected because they are thought to be involved in pancreatic β cell function, insulin action / glucose metabolism, or other metabolic conditions that increase T2D risk (e.g., energy intake / expenditure, lipid metabolism). To date, more than 50 candidate genes for T2D have been studied in various populations worldwide. However, results for essentially all candidate genes have been conflicting. Possible explanations for the divergent findings include small sample sizes, differences in T2D susceptibility across ethnic groups, variation in environmental exposures, and gene-environmental interactions. Because of current controversy, this review will focus only on a few of the most promising candidate genes. These include *PPARγ*, *ABCC8*, *KCNJ11*, and *CALPN10*.

**Table 2. Several T2D Susceptibility Genes**

| Gene | Locus | Variant | Estimated RR† |
|---|---|---|---|
| *PPARγ* | 3p25 | Pro12Ala | 1 – 3 |
| *ABCC8* | 11p15.1 | Ser1369Ala | 2 – 4 |
| *KCNJ11* | 11p15.1 | Glu23Lys | 1 – 2 |
| *CALPN10* | 2q37.3 | A43G | 1 - 4 |

†RR = relative risk

*PPARγ* (peroxisome proliferator-activated receptor-γ). This gene has been widely studied because it is important in adipocyte and lipid metabolism. In addition, it is a target for the hypoglycemic drugs known as thiazolidinediones. One form of the *PPARγ* gene (Pro) decreases insulin sensitivity and increases T2D risk by several fold. Perhaps more importantly is that this variant is very common in most populations. Approximately 98% of Europeans carry at least one copy of the Pro allele. Thus, it likely contributes to a considerable proportion (~25%) of T2D that occurs, particularly among Caucasians.

*ABCC8* (ATP binding cassette, subfamily C, member 8). This gene encodes the high-affinity sulfonylurea receptor (SUR1) subunit that is coupled to the Kir6.2 subunit (encoded by *KCNJ11,* also known as the potassium channel, inwardly rectifying subfamily J, member 11). Both genes are part of the ATP-sensitive potassium channel, which plays a key role in regulating the release of hormones, such as insulin and glucagon, in the beta cell. Mutations in either gene can affect the potassium

channel's activity and insulin secretion, ultimately leading to the development of T2D. Interestingly, *ABCC8* and *KCNJ11* are only 4.5 kb apart, and not far from the *INS* gene. Variant forms of *KCNJ11* (Lys) and *ABCC8* (Ala) genes have been associated with T2D, as well as other diabetes-related traits. Because of the close proximity of these genes, current studies are evaluating whether they work in concert with each other, or rather have an independent effect on T2D susceptibility.

Since *PPARγ, ABCC8* and *KCNJ11* are the targets of drugs used routinely in the treatment of T2D, there are pharmacogenetic implications for maintaining good glycemic control. Response to hypoglycemic therapy may actually be related one's genotype. Thus, genetic testing may not only help determine who is at high risk for developing T2D, but may also be useful in guiding treatment regimens for T2D.

*CAPN10* (calpain 10). *CAPN10* encodes an intracellular calcium-dependent cysteine protease that is ubiquitously expressed (Cox et al., 2004). A haplotype that was initially linked to T2D included an intronic A to G mutation at position 43, which appears to be involved in *CAPN10* transcription. Two amino acid polymorphisms (Thr504Ala and Phe200Thr) have also been associated with T2D risk. However, it has been suggested that the coding and noncoding polymorphisms do not independently influence T2D risk, but instead contribute to an earlier age at diagnosis. Physiological studies suggest that variations in calpain 10 activity effects insulin secretion, and therefore, susceptibility to T2D. Studies from different ethnic groups indicate that the contribution of this locus to increased T2D risk may be much larger in Mexican-American than Caucasian populations.

***Maturity-Onset Diabetes of the Young***

An uncommon form of T2D (accounting for <5% of all T2D cases) that generally occurs before age 25 years is MODY. MODY is characterized by a slow onset of symptoms, the absence of obesity, no ketosis, and no evidence of beta cell autoimmunity. It is most often managed without the need for exogenous insulin. MODY displays an autosomal dominant pattern inheritance, generally spanning three generations (Stride and Hattersley, 2002). Because of advances in molecular genetics, it is now known that there are at least six forms of MODY, each of which caused by a mutation in a different gene that is directly involved with beta cell function (Winter, 2003). Table 3 lists the MODY genes that have been identified to date. Because ~15% of MODY patients do not carry mutations in one of these genes, it is anticipated that other genes that cause MODY will be discovered in the near future (Demenais et al., 2003; Frayling et al., 2003; Kim et al., 2004).

**Table 3. MODY Genes**

| Type | Gene | Locus | # Mutations | % MODY |
|---|---|---|---|---|
| MODY1 | *HNF4A* | 20q12-q13.1 | 12 | ~5% |
| MODY2 | *GCK* | 7p15-p13 | ~200 | ~15% |
| MODY3 | *HNF1A* | 12q24.2 | >100 | ~65% |
| MODY4 | *IPF1* | 13q12.1 | Few | |
| MODY5 | *HNF1B* | 17cen-q21.3 | Few | <3% |
| MODY6 | *NEUROD1* | 2q32 | Few | |

*GCK* (glucokinase). The *GCK* gene is currently the only MODY gene that does not regulate the expression of other genes. Rather, the *GCK* gene plays a key role in glucose metabolism and insulin secretion. Thus, the clinical course of MODY2 patients differs from the prognosis associated with other types of MODY. MODY2 patients have a mild fasting hyperglycemia that is present from birth, and generally stable throughout life. There may be a mild deterioration of normoglycemia with age, but patients with

MODY2 mutations are usually asymptomatic. Most are detected during routine medical screening. Women with MODY2 mutations are often diagnosed during pregnancy. However, the outcome of the pregnancy can be influenced by whether the mother and / or fetus carry the mutation. When both mother and fetus are MODY2 positive, there is generally no effect on birth weight. However, MODY2 negative fetuses are carried by MODY2 positive mothers are typically large for gestational age due to maternal hyperglycemia. In contrast, if the fetus, but not the mother, carries the MODY2 mutation, their birth weight will be reduced by approximately 500g due to reduced fetal insulin secretion, which inhibits growth.

*HNF4A* (hepatocyte nuclear factor 4-α). Mutations in promoter and coding regions of the *HNF4A* gene cause MODY1. *HNF4A* is expressed in many tissues, including the liver and pancreas. It regulates hepatic gene expression, and influences the expression of other MODY genes such as *HNF1A,* which causes MODY3. In the beta cell of the pancreas, it directly activates insulin gene expression. Mutations in the *HNF4A* gene also have been associated with T2D (Silander et al., 2004).

*HNF1A* (hepatocyte nucleara factor 1-α). MODY3, the most frequent cause of the disease, results from mutations in the *HNF1A* gene. *HNF1A* is expressed in the liver and pancreas. It can also influence *HNF4A* expression, indicating a connection between MODY1 and MODY3. This suggests that the MODY transcription factors form a regulatory network that maintains glucose homeostasis. In addition to causing MODY3, *HNF1A* mutations have been associated with T1D (Moller et al., 1998; Lehto et al., 1999) and T2D (Pearson et al., 2004).

*IPF1* (insulin promoter factor-1). MODY4, which is a rare form of the disease, is due to mutations in the *IPF1* gene. Homozygosity for such mutations has been associated with newborn pancreatic agenesis and neonatal diabetes. Therefore, infants who carry MODY4 mutations tend to be small for gestational age. Individuals with MODY4 may also develop T2D (Cockburn et al., 2004). *IPF1* regulates expression of glucokinase, insulin and other genes involved in glucose metabolism.

*HNF1B* (hepatocyte nucleara factor 1-β). MODY5, another rare form of MODY, has also been linked with MODY1 because *HNF1β* regulates *HNF4α*. However, unlike MODY1, MODY5 is also associated with renal cysts, proteinuria and renal failure.

*NEUROD1* (neurogenic differentiation factor 1). Mutations in *NEUROD1* are responsible for MODY6. MODY6 is also rare. Together, MODY4, MODY5 and MODY6 comprise less then 3% of all MODY cases. *NERUOD1* is expressed in the beta cells of the pancreas, the intestine and the brain. In the pancreas, it contributes to the regulation of the expression of insulin.

To summarize, all MODY genes are expressed in the islet cells of the pancreas, and play a role in the metabolism of glucose, the regulation of insulin or other genes involved in glucose transport, and/or the development of the fetal pancreas. Because MODY phenotypes vary depending which gene is involved (Table 4), genetic testing may also assist in the treatment of the disease.

**Table 4. MODY Phenotypes**

| Type | Disease Onset | Complications | Treatment |
|---|---|---|---|
| MODY1 | Severe | Frequent | Diet, oral agents, insulin |
| MODY2 | Mild | Rare | Diet |

| | | | |
|---|---|---|---|
| MODY3 | Severe | Frequent | Diet, oral agents, insulin |
| MODY4 | Moderate | Little data | Oral agents, insulin |
| MODY5 | Severe | Renal cysts | Oral agents, insulin |
| MODY6 | Severe | Little data | Diet, oral agents, insulin |

## Role of Genetics in the Treatment and Prevention of Diabetes

### *Type 1 Diabetes*

At the present time, there is no way to prevent T1D. Lifelong insulin injections are the only available treatment for the disease. Thus, genetics does not currently play a role in the management or prevention of T1D.

Although a cure for T1D is currently unavailable, several large multi-national investigations have been designed to evaluate a variety of primary and secondary disease interventions (Devendra et al., 2004). The tested interventions have included prophylactic nasal insulin (Diabetes Prediction and Prevention Project (DIPP) in Finland), oral and injected insulin (Diabetes Prevention Trial – 1 (DPT-1) in the US), as well as high doses of nicotinamide (European Nicotinamide Diabetes Intervention Trial - ENDIT), and the avoidance of cow's milk exposure during the first six months of life (Trial to Reduce in Genetically At-Risk (TRIGR) in Finland, US and other countries). These investigations focus on 'prediabetic' individuals identified from families with at least one child with type 1 diabetes. DIPP and TRIGR use HLA-DQB1 screening and recruit only individuals at increased genetic risk. The remaining trials recruit relatives with evidence of beta cell autoimmunity as a pre-clinical marker for disease. To date, none of these interventions have prevented or delayed the onset of T1D (Diabetes Prevention Trial-Type 1 Study Group, 2002; NIDDK, 2003; The ENDIT Group, 2003; Paronen, et al., 2000). However, with the formation of *Type 1 Diabetes TrialNet (www.trialnet.com)*, a collaborative network of clinical centers and experts in diabetes and immunology, new intervention strategies are currently being planned. It is ultimately hoped that through genetic testing, individuals at high risk for T1D could be identified prior to the onset of the disease – at a time when primary prevention strategies could be safely administered. It is most likely that such predictive genetic testing would be offered to families with an affected individual before it was made available to the general population.

### *Type 2 Diabetes*

Unlike T1D, T2D can generally be prevented by maintaining an age-appropriate body weight and engaging in physical activity. Although public health messages that emphasize a nutritious diet and regular physical activity are now commonplace, they have not been effective in terms of disease prevention. Given the recent obesity epidemic, it is obvious that current intervention strategies are being ignored by a majority of individuals in the general population.

Leaders of the Human Genome Project have predicted that genetic tests will become available for many common disorders during the first decade of the 21st century, permitting persons "to learn their individual susceptibilities and to take steps to reduce those risks" by applying interventions based on "medical surveillance, lifestyle modifications, diet or drug therapy" (Collins and McKusick, 2001). In

fact, several companies are now offering genetic susceptibility testing, which can be ordered online by any individual, for conditions such as cardiovascular disease and obesity (Khoury et al., 2004).

Although many scientists and health professionals share this optimistic perspective regarding genetics and disease prevention, others are more pessimistic for a variety of reasons. First, the predictive value of most genetic tests is low (Haga et al., 2003); and risk estimates do not account for well-known environmental determinants of disease. Secondly, it is unclear whether knowledge of one's genetic risk increases motivation to engage in disease interventions. Thirdly, genetic testing presents educational and information-dissemination challenges that were outlined in detail by the Secretary's Advisory Committee on Genetics, Health and Society (Holtzman and Watson, 1998). These include being able to communicate the validity and utility of proposed genetic tests, as well as the potential risks and benefits of being tested, to individuals who may have little knowledge of human genetics. Fourthly, most health care professionals are currently unqualified to interpret the results of genetic tests; and there are no standards for the use of molecular diagnostics in clinical practice. Fifthly, genetic testing may lead to significant distress, the magnitude of which is likely to vary as a function of actual test results, coping skills and resources, risk perception, optimism, health beliefs and pre-existing depression or anxiety.

These factors directly relate to other concerns such as insurance and employment discrimination, confidentiality and stigmatization based on knowing that one is at high genetic risk. In the near future, genetic testing for T2D and other chronic diseases will most certainly become available. Although it is unclear whether this will actually contribute to the prevention of T2D, it may be beneficial in terms of disease management. Many of the current T2D susceptibility genes of interest are drug targets. Evidence for the role of pharmacogenetics in diabetes is already apparent in treatment approaches for MODY.

***Maturity-Onset Diabetes of the Young***

The most common causes of MODY are related to mutations in MODY3, MODY2 and MODY1 genes. Although individuals who carry MODY2 mutations have a very mild form of the disease, those who carry MODY1 and MODY3 variants have a much more severe expression that is associated with long-term complications. In addition, there has been a link between MODY3 and MODY5 because of their interaction in terms of gene expression. However, it is now becoming clear that the metabolic phenotype of individuals with these two forms of MODY is actually quite different (Pearson et al., 2004). To date, little has been known about MODY5 other than its association with renal cysts. However, it now appears that MODY5 is more strongly associated with hyperinsulinemia and dyslipidemia (and more closely related to insulin resistance and T2D) than MODY3. Thus, knowledge about the underlying MODY defect is likely to lead to better management and an improved prognosis for individuals with the disease.

Given the autosomal dominant inheritance of all forms of MODY, individuals with a diabetic parent may also wish to have genetic testing. Early diagnosis MODY may also help reduce the likelihood of long-term complications. In addition, psychological and family adjustments to diabetes may also be improved when the specific form of the disease is known.

Approximately one-third of individuals with MODY3 and MODY1 are each treated by diet, oral agents and insulin. Some individuals with MODY3 have been previously classified as having T1D because of the severity of the disease (Moller et al., 1998; Lehto et al., 1999). It is now known that individuals with MODY3 mutations are extremely sensitive to the hypoglycemic effects of sulfonylureas. Thus, these oral agents are likely to be the treatment of choice of individuals with MODY3. Recently, there

have been a number of reports of MODY3 individuals being able to change treatment regimens from insulin injections to oral sulphonylurea agents, with considerable improvement in glycemic control (Shepherd, 2003a; Shepherd and Hattersley, 2004)). This is frequently associated with a positive impact on lifestyle and self image, as well as fear and anxiety about the possibility of stopping insulin. Some individuals, particularly those who have long-term complications, have become angry because they were previously misdiagnosed and/or treated inappropriately. These reactions have implications for health professionals who need to be knowledgeable about the potential psychological consequences of changing treatment regimens (Shepherd, 2003b).

## Future Role of Genetics in Diabetes

Within the next decade, the genes that increase risk of developing all forms of diabetes will likely be known. It is, therefore, important that scientists, health professionals, and members of population at large consider how to maximize the advantages, and minimize the disadvantages of predictive genetic testing for diabetes.

In September 2004, the Office of Genomics and Disease Prevention at the Centers for Disease Control in the US held a meeting entitled "Public Health Assessment of Genetic Tests for Screening and Prevention". One of the objectives of this session was to discuss issues related to the evaluation and utilization of genetic tests. Emphasis was placed on three major barriers: 1) the lack of available population data regarding the contribution of genetic variants to disease susceptibility, 2) the lack of an evidence-based process for the integration of genomics into practice and, 3) the lack of readiness of the health care and public health systems to utilize genetic testing for disease prevention. At the end of the meeting it was apparent that we, as a society, are a long way from the practice of 'genomic medicine'.

With regards to diabetes, addressing the first barrier is most critical at the present time. This barrier pertains to the lack of consistent results across populations with regards to the genetic determinants of the disease. Failure to replicate study results may be due to a variety of factors, the most important of which may be that different gene-environment interactions operate different populations to increase risk of developing diabetes. Thus, considerably more epidemiologic research will be needed before we know the actual risk associated with particular genetic variants. This also likely means that we will not be able to apply a 'one size fits all' model when it comes to the genetic testing for any of the forms of diabetes.

To fulfill the promise of the Human Genome Project, several issues that warrant careful consideration. First, multidisciplinary teams will be required to translate genetic discoveries from the laboratory to the community. This is, perhaps, best exemplified by the development of new initiatives such as the NIH Roadmap in the US. Scientists will no longer be able to work in isolation, without input of individuals from other professions, if they are to maximize the impact of their research in terms of improving health. In particular, issues such as quality assurance, health risks and benefits, and economics need to be addressed. This will require expertise from persons who have typically worked outside the profession of science. Finally, the ethical, legal and social issues associated with widespread availability and use of predictive genetic tests must been addressed. These include confidentiality, discrimination, diversity, informed consent, keeping up with genetic discoveries and uncertainty. Ideally, consideration of such issues will lead to the development of practice guidelines for diabetes, which will hopefully serve as a model for genetic testing for other complex diseases.

*Acknowledgement*: We acknowledge the contribution of Dr Janice S. Dorman, Ph.D., University of Pittsburgh, School of Nursing, 350 Victoria Building, Pittsburgh, PA, USA, to this paper.

## References

1. Alberti, K.G.M.M., Zimmett, P. *Definition, diagnosis and classification of diabetes mellitus and its complications part 1: diagnosis and classification of diabetes mellitus provisional report of a WHO consultation.* Diabet Med, 1998. **15**: 539-553.
2. Anjos, S., Polychronakos, C. *Mechanisms of genetic susceptibility to type 1 diabetes: beyond HLA.* Mol Genet Metab, 2004. **81**: 187-195.
3. Barroso, I., Luan, J., Middelberg, R.P.S., et al. *Candidate gene association study in type 2 diabetes indicates a role for genes involved in B-Cell function as well as insulin action.* PLoS Biol, 2003. **1**: 41-55.
4. Bennett, S.T., Wilson, A.J., Esposito, L. *Insulin VNTR allele-specific effect in type 1 diabetes depends on identity of untransmitted paternal allele.* Nat Genet, 1997. **17**: 350-352.
5. Bloomgarden, Z.T. *Type 2 diabetes in the young: the evolving epidemic.* Diabetes Care, 2004. **27**: 998-1010.
6. Borch-Johnsen, K., Joner, G., Mandrup-Poulsen, T., et al. *Relation between breast-feeding and incidence rates of insulin-dependent diabetes mellitus. A hypothesis.* Lancet, 1984. **2**: 1083-1086.
7. Cockburn, B.N., Bermano, G., Boodram, L.-L.G., et al. *Insulin promoter factor-1 mutation and diabetes in Trinidad: identification of a novel diabetes-associated mutation (E224K) in an Indo-Trinidadian family.* J Clin Endocrinol Metab, 2004. **89**: 971-978.
8. Collins, F.S., McKusick, V.A. *Implications of the Human Genome Project for medical science.* JAMA, 2001. **285**: 540-544.
9. Cox, N.J., Hayes, M.G., Roe, C.A., et al. *Linkage of calpain 10 to type 2 diabetes: the biological rationale.* Diabetes, 2004. **53**: S19-S25.
10. Dahlquist, G. *The aetiology of type 1 diabetes: an epidemiological perspective.* Acta Paediatr Suppl, 1998. **425**: 5-10.
11. Dahlquist, G., Frisk, G., Ivarsson, S.A., et al. *Indications that maternal coxsackie B Virus infection during pregnancy is a risk factor for childhood-onset IDDM.* Diabetologia, 1995. **38**: 1371-1373.
12. Demanais, F., Kanninen, T., Lindgren, C.M., et al. *A meta-analysis of four European genome screens (GIFT Consortium) shows evidence for a novel region on chromosome 17p11.2-q22 linked to type 2 diabetes.* Hum Mol Genet, 2003. **12**: 1865-1873.
13. Devendra, D., Liu, E., Eisenbarth, G.S. *Type 1 diabetes: recent developments.* BMJ, 2004. **328**: 750-754.
14. Dorman, J.S., Bunker, C.H. *HLA-DQ locus of the human leukocyte antigen complex and type 1 diabetes mellitus: a HuGE review.* Epidemiol Rev, 2000. **22**: 218-227.
15. Dorman, J.S., LaPorte, R.E., Songer, T.J. *Epidemiology of Type 1 Diabetes*, in *Type 1 Diabetes: Etiology and Treatment*, Mark A. Sperling, Editor. 2003, Humana Press: Totowa, NJ. p. 3-22.
16. Fagot-Campagna, A., Pettitt, D.J., Engelgau, M.M., et al. *Type 2 diabetes among North American children and adolescents: an epidemiologic review and a public health perspective.* J Pediatr, 2000. **136**: 664-672.
17. Flores, J.C., Hirschhorn, J., Altshuler, D. *The inherited basis of diabetes mellitus: implications for the genetic analysis of complex traits.* Annu Rev Genomics Hum Genet, 2003. **4**: 257-291.
18. Frayling, T.M., Hattersley, A.T. *The role of genetic susceptibility in the association of low birth weight with type 2 diabetes.* Br Med Bull, 2001. **60**: 89-101.

19. Frayling, T.M., Lindgren, C.M., Chevre, J.C., et al. *A genome-wide scan in families with maturity-onset diabetes of the young: evidence for further genetic heterogeneity.* Diabetes, 2003. **42**: 872-881.
20. Gloyn, A.L. *The search for type 2 diabetes genes.* Ageing Res Rev, 2003. **2**: 111-127.
21. Group, D.C.a.C.T.R. *The effect of intensive treatment of diabetes on the development and progression of long-term complications in insulin-dependent diabetes mellitus.* N Engl J Med, 1993. **329**: 977-986.
22. Group, D.P.T.-T.S. *Effects of insulin in relatives of patients with type 2 diabetes mellitus.* N Engl J Med, 2002. **346**: 1685-1691.
23. Group, T.E.N.D.I.T.E. *Intervening before the onset of type 1 diabetes: baseline data from the European Nicotinamide Diabetes Group.* Diabetologia, 2003. **46**: 339-346.
24. Haga, S.B., Khoury, M.J., Burke, W. *Genomic profiling to promote a healthy lifestyle: not ready for prime time.* Nat Genet, 2003. **34**: 347-350.
25. Hales, C.N., Barker, D.J. *The thrifty phenotype hypothesis.* Br Med Bull, 2001. **60**: 5-20.
26. Hansen, L. *Candidate genes and late-onset type 2 diabetes mellitus. Susceptibility genes or common polymorphisms?* Dan Med Bull, 2003. **50**: 320-346.
27. Harrison, L.C., Honeyman, M.C. *Cow's milk and type 1 diabetes.* Diabetes, 1999. **48**: 1501-1507.
28. Hirschhorn, J.N. *Genetic epidemiology of type 1 diabetes.* Pediatr Diabetes, 2003. **4**: 87-100.
29. Holt, R.I.G. *Diagnosis, epidemiology and pathogenesis of diabetes mellitus: an update for psychiatrists.* Br J Psychiatry, 2004. **184**: s55-s63.
30. Holztman, N.A., Watson, M.S. eds. *Promoting Safe and Effective Genetic Testing in the United States*. Johns Hopkins University Press, 1998.
31. Honeyman, M.C., Coulson, B.S., Stone, N.L. *Association between rotavirrus infection and pancreatic islet autoimmunity i nchildren at risk of developing type 1 diabetes.* Diabetes, 2000. **49**.
32. Hu, F.B. *Sedentary lifestyle and risk of obesity and type 2 diabetes.* Lipids, 2003. **38**: 103-108.
33. Hyoty, H., Hiltunen, M., Knip, M., et al. *A prospective study of the role of coxsackie B and other enterovirus infections in the pathogenesis of IDDM. Childhood Diabetes in Finland (DiMe) Study Group.* Diabetes, 1995. **44**: 652-657.
34. Hyoty, H., Hiltunen, M., Reuranen, A. *Decline of mumps antibodies in type 1 (insulin-dependent) diabetic children with a plateau in the rising incidence of type 1 diabetes after introduction of the mumps-measles-rubella vaccine in Finland.* Diabetologia, 1993. **41**: 40-46.
35. Karovonen, M., Tuomilehto, J., Libman, I., et al. *A review of the recent epidemiological data on the worldwide incidence of type 1 (insulin-dependent) diabetes mellitus. World Health Organization DiaMond Group.* Diabetologia, 1993. **36**: 883-892.
36. Khoury, M.J., Yang, Q., Gwinn, M., et al. *An epidemiologic assessment of genomic profilling for measuring susceptibility to common diseases and targeting interventions.* Genet Med, 2004. **6**: 38-47.
37. Kim, S.-H., Ma, X., Weremowicz, S., et al. *Identification of a locus for maturity-onset diabetes of the young on chromosome 8p23.* Diabetes, 2004. **53**: 1375-1384.
38. Kolb, H., Pozzilli, P. *Cow's milk and type 1 diabetes: the gut immune system deserves attention.* Immunol Today, 1999. **20**: 108-110.
39. Kyvik, K.O., Nystrom, L., Gorus, F., et al. *The epidemiology of type 1 diabetes mellitus is not the same in young adults as in children.* Diabetologia, 2004. **47**: 377-384.
40. Lean, M.E. *Obesity: burdens of illness and strategies for prevention or management.* Drugs Today (Barc), 2000. **36**: 773-784.

41. Lehto, M., Wipemo, C., Ivarsson, S.A., et al. *High frequency of mutations in MODY and mitochondrial genes in Scandinavian patients with familial early-onset diabetes.* Diabetologia, 1999. **42**: 1131-1137.
42. Lie, B.A., Todd, J.A., Pociot, F., et al. *The predisposition to type 1 diabetes linked to the human leukocyte antigen complex includes at least one non-class II gene.* Am J Hum Genet, 1999. **64**: 793-800.
43. Marshall, A.L., Chetwynd, A., Morris, J., et al. *Type 1 diabetes mellitus in childhood: a matched case control study in Lancashire and Cumbria, UK.* Diabet Med, 2004. **21**: 1035-1040.
44. McIntosh, E.D.G., Menser, M. *A fifty-year follow-up of congenital rubella.* Lancet, 1992. **340**: 414-415.
45. McKinney, P.A., Okasha, M., Parslow, R., et al. *Ante-natal risk factors for childhood diabetes mellitus, a case-control study of medical record data in Yorkshire, UK.* Diabetologia, 1997. **40**: 933-939.
46. Moller, A.M., Dalgaard, L.T., Pociot, F., et al. *Mutations in the hepatocyte nuclear factor-1a gene in Caucasian families originally classified as having type 1 diabetes.* Diabetologia, 1998. **41**: 1528-1531.
47. Neel, J. *Diabetes mellitus: a thrifty genotype rendered detrimental by "progress"?* Am J Hum Genet, 1962. **14**: 353-362.
48. Nejentsev, S., Reijonen, H., Adojaan, B., et al. *The effect of HLA-B allele on the IDDM risk defined by DRB1*04 subtypes and DQB1*0302.* Diabetes, 1997. **46**: 1888-1892.
49. (NIDDK), N.I.o.D.D.K.D. *Oral Insulin Does Not Prevent Type 1 Diabetes.* News Briefs (June 15, 2003): Available at: www.niddk.nih.gov/welcome/releases/6-15-03.htm March 8, 2004.
50. Onkamo, P., Vaananen, S., Karvonen, M., et al. *Worldwide increase in incidence of type 1 diabetes--the analysis of the data on published incidence trends.* Diabetologia, 1999. **42**: 1395-1403.
51. Pak, C.Y., McArthur, R.G., Eun, H.M. *Association of cytomegalovirus infection with autoimmune type 1 diabetes.* Lancet, 1988. **2**: 1-4.
52. Pan, X.R., Li, G.W., Hu, Y.H., et al. *Effects of diet and exercise in preventing NIDDM in people with impaired glucose tolerance. The Da Qing IGT and Diabetes Study.* Diabetes Care, 1997. **20**: 537-544.
53. Paronen, J., Knip, M., Savilahti, E., et al. *Effect of cow's milk exposure and maternal type 1 diabetes on cellular and humoral immunization to dietary insulin in infants at genetic risk for type 1 diabetes.* Diabetes, 2000. **49**: 1657-1665.
54. Pearson, E.R., Badman, M.K., Lockwood, C., et al. *Contrasting diabetes phenotypes associated with hepatocyte nuclear factor-1a and -1B mutations.* Diabetes Care, 2004. **27**: 1102-1107.
55. Pugliese, A., Zeller, M., Ferndandez, J.A. *The insulin gene is transcribed in human thymus and transcription levels correlated with allelic variation at the INS VNTR-IDDM 2 susceptibility locus for type 1 diabetes.* Nat Genet, 1997. **15**: 293-297.
56. Shaw, J., Chisholm, D. *Epidemiology and prevention of type 2 diabetes and the metabolic syndrome.* MJA, 2003. **179**: 379-383.
57. Shepherd, M. *'I'm amazed I've been able to come off injections': Patients' perceptions of genetic testing in diabetes.* Pract Diab Int, 2003. **20**: 338-342.
58. Shepherd, M. *Genetic testing in maturity onset diabetes of the young (MODY)-practical guidelines for professionals.* Pract Diab Int, 2003. **20**: 108-110.
59. Shepherd, M., Hattersley, A.T. *'I don't feel like a diabetic any more': the impact of stopping insulin in patients with maturity onset diabetes of the young following genetic testing.* Clin Med, 2004. **4**: 144-147.
60. Silander, K., Mohlke, K.L., Scott, L.J., et al. *Genetic variation near the hepatocyte nuclear factor-4a gene predicts susceptibility to type 2 diabetes.* Diabetes, 2004. **53**: 1141-1149.

61. Sinha, R., Fisch, G., Teague, B., et al. *Prevalence of impaired glucose tolerance among children and adolescents with marked obesity.* N Engl J Med, 2002. **346**: 802-810.
62. Sobngwi, E., Boudou, P., Mauvais-Jarvis, F., et al. *Effect of a diabetic environment in utero on predisposition to type 2 diabetes.* Lancet, 2003. **361**: 1861-1865.
63. Stride, A., Hattersley, A.T. *Different genes, different diabetes: lessons from maturity-onset diabetes of the young.* Ann Med, 2002. **34**: 207-216.
64. Stumvoll, M. *Control of glycaemia: from molecules to men. Minkowski Lecture 2003.* Diabetologia, 2004. **47**: 770-781.
65. Tuomilehto, J., Lindstrom, J., Eriksson, J.G., et al. *Prevention of type 2 diabetes mellitus by changes in lifestyle among subjects with impaired glucose tolerance.* N Engl J Med, 2001. **344**: 1343-1350.
66. Undlien, D.E., Hamaguchi, K., Kimura, A. *Type 1 diabetes susceptibility associated with polymorphism in the insulin gene region: a study of blacks, Caucasians, and orientals.* Diabetologia, 1994. **37**: 745-749.
67. Vaarala, O. *Gut and the induction of immune tolerance in type 1 diabetes.* Diabetes Metab Res Rev, 1999. **15**: 353-361.
68. van Dam, R.M. *The epidemiology of lifestyle and risk for type 2 diabetes.* Eur J Epidemiol, 2003. **18**: 1115-1125.
69. Weill, J., Vanderbecken, S., Froguel, P. *Understanding the rising incidence of type 2 diabetes in adolescence.* Arch Dis Child, 2004. **89**: 502-504.
70. WHO, 1999. Definition, Diagnosis and Classification of Diabetes Mellitus and its Complications. WHO/NCD/NCS/99.2.
71. Wild, S., Roglic, G., Green, A., et al. *Global prevalence of diabetes.* Diabetes Care, 2004. **27**: 1047-1053.
72. Winter, W.E. *Newly defined genetic diabetes syndromes: maturity onset diabetes of the young.* Rev Endocr Metab Disord, 2003. **4**: 43-51.
73. Zimmett, P., Alberti, K.G.M.M., Shaw, J. *Global and societal implication of the diabetes epidemic.* Nature, 2001. **414**: 782-787.