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GENETIC FACTORS INVOLVED IN THE PATHOGENESIS OF TYPE 2 DIABETES

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Type 2 diabetes (T2D) represents one of the major global health problems of modern societies. Its pathogenesis is complex and it was classically characterized by pancreatic β-cell dysfunction (with diminished insulin secretion) followed by decline of the beta cell mass, peripheral insulin resistance and increased hepatic glucose production, most often associated with obesity. T2D pathogenesis involves both genetic and environmental factors. The common form of polygenic T2D is a complex disease, the genetic risk being influenced by the conjoint effects of variation at an undetermined number of genomic sites. The main methods for mapping the T2D genes were the hypothesis driven candidate gene analysis and the hypothesis free genome-wide scaning studies. The candidate gene approach led to the identification of two T2D genes now considered widely replicated: PPARG and the β-cell potassium channel (Kir6.2) gene, KCNJ11. The genome-wide linkage approach led to the identification of several loci, the most prominent being the TCF7L2 (Transcription Factor 7 Like 2) gene on chromosome 10q25.3. TCF7L2 has been replicated in almost every population examined and, with an OR of about 1.4, represents the strongest T2D gene identified so far. Finally, during the last 5 years, the genome-wide association approach led to the identification of almost 40 T2D genes. The majority of these appear to affect beta cell function. Deciphering the genetic background of T2D will contribute to the prediction of the disease in high risk subjects, with possible benefits for its prevention.

Key words: Type 2 diabetes, genetics, genome wide association scan.

INTRODUCTION

Diabetes mellitus is one of the most common chronic diseases in human populations across the globe, with a current prevalence of 6.5%, representing 285 million adults in 2010[1]. Moreover, the prevalence of diabetes continues to rise in both the Western world and in the developing countries as changing lifestyles lead to reduced physical activity, and increased obesity. Thus, predictions for the next 20 years show that diabetes will become epidemic, reaching a prevalence of 7.7% (439 million adults worldwide) by 2030[1]. More than 80% from these patients represent type 2 diabetes (T2D) cases, the diabetes phenotype formerly known as *non-insulin-dependent diabetes*[2]. Due to its associated morbidities and increased mortality[3], T2D represents one of the major global health problems of modern societies.

T2D has a complex pathogenesis that was classically characterized by pancreatic β-cell dysfunction (with diminised insulin secretion) followed by decline of the beta cell mass, peripheral insulin resistance and increased hepatic glucose production, most often associated with obesity[4]. β-cell dysfunction is characterized initially by diminished (and later by missing) first-phase insulin response after glucose stimulation but also following stimulation with nonglucose secretagogues such as the incretin hormone glucagon-like peptide-1 (GLP-1). The peripheral insulin resistance primarily affects the liver and peripheral tissues (skeletal muscle and adipose

tissue) leading to increased hepatic glucose output and diminished peripheral glucose uptake. Progresses in understanding T2D pathogenesis expanded this "triumvirate" to an "ominous octet"[5], including defects in adipose cells (accelerated lipolysis), gastrointestinal tract (incretin hormones defect), α-cells (increased glucagon production), kidney (increased glucose reabsorption) and brain (insulin resistance).

Similar to other common human complex diseases (such as obesity, hypertension, type 1 diabetes, etc.), T2D pathogenesis involves both genetic and environmental factors. The last include hypercaloric diet, sedentarism, stress and their consequence, obesity, as well as some other conditions such as urbanization and westernized lifestyle. From the genetic point of view, the common form of polygenic T2D is a complex disease, the genetic risk being influenced by the conjoint effects of variation at an undetermined number of genomic sites, some with a predisposing and some with a protective effect[6]. Moreover, the global disease risk is determined by the interaction of the genetic background with the various environmental exposures encountered during each individual's life. In addition T2D is also multigenic, meaning that many different combinations of gene variants may exist among T2D patients, leading to a similar disease phenotype[7].

ARGUMENTS FOR THE IMPORTANCE OF GENETIC FACTORS IN T2D PATHOGENESIS

The evidence for a strong genetic component in the pathogenesis of T2D diabetes and related traits is provided by epidemiologic studies showing marked differences in T2DM prevalence across populations, studies of large families with increased prevalence of T2D (familial aggregation) and by twin studies[8,9].

Ethnic variation of T2D represents strong evidence for the genetic basis of this disease. The maximum prevalence is recorded in Pima Indians from USA and South Sea Island populations (such as Naurus in Polynesia), where it now reaches ~50%[1,10]. A low prevalence (~3%) is recorded in some African populations[1] while the lowest (~1%) is recorded in some isolated rural populations from South America, such as the Mapuche or Aymara tribes in Chile[11].

Additional support derives from the strong familial aggregation of T2D. Thus, offspring have a lifetime risk of developing type 2 diabetes of 35-39% if one parent has T2D and 60–70% if both parents have type 2 diabetes compared with 10% in the general population[12,13,14]. This translates in a sibling relative risk (λs) between 2 and 3.5[13,15]. Family history has been noted to double the risk of T2D, while the presence of obesity plus a positive family history quadruple the diabetes risk. More recently, it was showed that the highest heritability for T2D is present in middle aged people aged 35-60 years and decreases markedly if the upper limit is increased to 75 years[16].

Finally, the study of monozygotic and dizygotic twins can estimate more precisely the relative importance of genetic and nongenetic factors since twins share the same prenatal and postnatal environment and monozygotic twins are virtually genetic identical while dizygotic twins resemble usual siblings according to the phenotype in question[17]. Twin data have shown variable concordance rates of T2D in monozygotic twins. Thus, older studies reported figures of nearly 100%[18]. More recent studies report lower concordance rates, ranging from 30% to 80%, as opposed to 20% to 30% in dizygotic twins[19,20]. Similarly, high degrees of heritability of diabetes-related traits (such as first phase insulin response, basal and insulin stimulated glucose uptake, etc.) were reported in twins[21]. The large differences in estimates for T2D concordance in twins is partially explained by the variable age at T2D onset. Thus, quite often discordant twins may appear concordant after years of evolution, so that the concordance rate will vary according to the timing of the evaluation.

DIFIFICULTIES IN UNRAVELING THE GENETICS OF T2D

Starting with the 1980's, many researchers spent years searching for T2D genes in laboratories worldwide but for two decades their efforts were almost fruitless. Both objective and subjective factors contributed to this failure. Among the objective factors, the most important is T2D own complex pathogenesis. Replication of beta cells, beta cell function, insulin sensitivity, hepatic glucose output, obesity, function of adipocytes etc. are all complex interacting processes that may be controlled by different genes. The inherent

aetiological complexity of multifactorial T2D means that effect sizes expected at any individual gene are likely to be modest and to vary between populations. This seriously complicated their detection and characterization. Moreover, the prolonged poor understanding of T2D pathogenesis rendered difficult the selection of candidates with strong prior odds for disease involvement[22]. In addition, the phenotypic expression of T2D genes is strongly influenced by environmental factors. Thus, some susceptible individuals will never develop diabetes if adopting a healthy lifestyle while some individuals with lower genetic susceptibility will become diabetic in condiditions of westernized lifestyle. All this complicates proper selection of cases and controls for genetic studies.

These objective biological limitations have often been exacerbated by subjective factors such as suboptimal study design and deployment of inadequate sample sizes. However, in the new millennium (and especially after 2006), due to the major advances in molecular biology/genotyping techniques, important progresses in elucidating the genetic background of T2D were made[22,23].

METHODS FOR THE GENETIC ANALYSIS OF T2D

There are many possible methods for mapping the genes involved in the pathogenesis of common chronic diseases, including T2D. These can be classified into two main categories: the hypothesis driven candidate gene studies and the hypothesis free genome-wide studies. The last include both genome wide linkage mapping (GWL) and genome-wide association (GWA) studies[24].

Genome-wide linkage mapping was the method traditionally used to identify disease genes, and has been tremendously successful for mapping genes that underlie monogenic ‘Mendelian’ diseases[25]. Genome-wide linkage analysis has also been carried out for many common diseases but, however, for most of these (including diabetes) it was less successful[26].

Historically, the “first wave” in the discovery of T2D causal genes was represented by the family-based GWL studies and the focused analysis of candidate genes on small scale case/control datasets. These proved effective in identifying genes responsible for rare forms of early-onset non-autoimmune diabetes segregating in Mendelian fashion, including the maturity-onset diabetes of the young (MODY), mitochondrial diabetes, and neonatal diabetes[27]. Several candidate genes identified in these monogenic forms of diabetes proved to be also involved in the genetics of common forms of T2D.

Unfortunately, attempts to apply the GWL technique to the genetic dissection of the common forms of T2D have proved to be largely unrewarding[28]. This is why in the last few years, researchers begun to use more and more for the study of common diseases (including T2D) the technique of genome-wide association study (GWA). GWA is a special type of association study that searches the whole genome (or at least most of it) for causal genetic variants, and it can be attempted even in the absence of convincing evidence regarding the function or location of the candidate causal genes[24]. Practically, it consists in typing thousands or tens of thousands of non-synonymous single nucleotide polymorphisms (nsSNPs) distributed all over the genome in large case/control datasets for different common diseases. The experimental strategy of GWAS is a classic case/control design but uses very large samples (tens of thousands) with the intention to detect minor gene effects. GWAS compares the frequency of allelic variants of SNPs between the cases and the control population. Significant excess of alleles in cases indicates an influential role (pathogenic) and conversely, an excess in controls defines a protective effect.

Due to the high costs of genotyping as well as the huge amount of labor involved, large scale GWA studies were practically impossible until 2006. Since then, some major progresses in human molecular genetics made these studies possible: First, the Human Genome Project was finalized, with a draft sequence in 2001and near-complete sequence in 2003[29,30]. Second, the International HapMap project[31] with its first phase completed in 2005, and now currently in its third phase, provided clear patterns of genome-wide variation and linkage disequilibrium (particularly useful for the methods as GWA that use markers selected on the basis of LD) facilitating the efficient selection of SNPs for GWA as well the analysis of association data. Third, the database of SNPs (dbSNP) includes now most of the ~11 million SNPs with minor allele frequencies of 1% or greater that are estimated to exist in the human genome[32]. Finally, the speed and cost of high throughput genotyping improved dramatically during the last years, analytical tools were developed to assist in the data mining, cleaning,

and interpretation of large databases while large case/control collections for complex diseases became available for investigation[24,33]. As a consequence, it is now possible to analyze up to a million SNPs in a single analysis on case/control datasets comprising thousands of subjects[34,35].

The advent of large scale GWA studies represented the "second wave" in the discovery of T2D genes and led to a dramatic progress in understanding the genetic basis of T2D.

MONOGENIC FORMS OF DIABETES

The common forms of polygenic T2D represent the vast majority of diabetes cases in modern societies. However, several monogenic forms of diabetes, representing 1-2% of the total number of cases, were described in the scientific literature[27,36]. While these rare mutations might affect both insulin secretion and insulin action[11], the most frequent and best known are beta-cell genes mutations that alter their insulin secretion capacity[27].

Genetic defects of beta cell function

The candidate gene approach has been remarkably successful in identifying monogenic diabetes genes. Usually these involve proteins from the key rate-limiting steps in insulin secretion and severe mutations will result in β-cell dysfunction. There are five main classes of β-cell dysfunction that encompass most cases of monogenic diabetes[27]: 1) Defective glucose sensing (Glucokinase - *GCK* gene); 2) Abnormal potassium ATP-sensitive (KATP) channels (*KCNJ11* and *ABCC8* genes); 3) Mutated transcription factors (*HNF-4α, HNF-1α, HNF-1β, IPF-PDX1, NEUROD1*, etc); 4) Defective mitochondria (A3243G mutation in (mt)DNA) and 5) Endoplasmic reticulum stress (*EIF2AK3, INS, WFS1* genes). The identification of the etiological genes helped the recognition of novel clinical subgroups.

MODY (*Maturity Onset Diabetes of the Young*) was clinically defined as autosomal dominantly inherited, non–insulindependent, early-onset diabetes. The first cases were described in the 1960s[37], but now there are at least eight genetic subgroups of MODY, most of which have a discrete phenotype[27,38,39]. The term MODY is used to describe a group of clinically heterogeneous, often non-insulin-dependent forms of diabetes with variable age at onset, severity of hyperglycaemia, risk of chronic complications and associated clinical features. The most recent ADA-WHO classification of diabetes[2] includes MODY in the group III - Other Specific Types, Subgroup A – Genetic defects of beta cell function. The main characteristics of MODY diabetes are given in Table 1.

Regarding the effect of MODY genes on susceptibility risk for common, adult onset T2D, some common variants of *HNF1β* gene on chromosome 17q12 showed evidence for robust association with T2D[9,42,43]. Thus, the combined analysis of more than 15,000 samples showed the significant association of the intronic SNP (rs757210) in *HNF1β* with an overall OR of 1.12 and a convincing p value of <10-6[42], results replicated in another large-scale study[44] and independently confirmed in a subsequent GWA study[45]. In the same time, the *GCK, HNF1α* and the *HNF4α* genes have been extensively studied, but no consistent results were obtained regarding an effect on T2D risk[9,43,46].

Table 1

Genetic characteristics of different MODY types (adapted after [11,39,40,41])

Disease	Gene	Frequency	Locus	Protein
MODY1	*HNF-4α* (TCF-14)	~ 4%	20q12-q13.1	Hepatocyte nuclear factor 4 α (Transcription factor 14)
MODY2	*GCK*	~ 22%	7p15-p13	Glucokinase (hexokinase-4)
MODY3	*HNF-1α* (TCF1)	~ 61%	12q24.2	Hepatocyte nuclear factor 1α (Transcription factor 1)
MODY4	*IPF-1* (PDX1)	< 1%	13q12.1	Insulin promoter factor 1, Pancreatic and duodenal homeobox 1
MODY5	*HNF-1β* (TCF-2)	~ 2%	17q12	Hepatocyte nuclear factor 1β (Transcription factor 2)
MODY6	*NEUROD-1*	< 1%	2q32	Neurogenic differentiation 1
MODY7	*KLF11*	< 1%	2p25	Kruppel-like factor 11
MODY8	*CEL*	< 1%	9q37	Carboxyl Ester Lipase
MODY9	*ABCC8*	< 1%	11p15.1	ATP-Binding Cassette, Subfamily C, Member 8
MODYX	Unknown	~ 11%	Unknown	Unknown

Diabetes diagnosed in the first few months of life was defined clinically as neonatal diabetes mellitus[27,36]. Depending on whether diabetes resolves later in life, two phenotypes were described: Permanent neonatal diabetes (PNDM) or transient neonatal diabetes (TNDM). Molecular genetics advances have identified several genetic subgroups of neonatal diabetes or genetic syndromes that include neonatal diabetes. In addition, several genetic syndromes, such as Wolcott-Rallison, Maternally Inherited Diabetes and Deafness (also known as mitochondrial diabetes) and Wolfram Syndrome, are characterized by monogenic transmission and clinically by a cluster of clinical features including diabetes. The main genetic and clinical characteristics of these syndromes are given in Table 2.

It should be noted that severe homozygous mutations in the MODY4 and MODY6 genes are associated with pancreatic agenesis and permanent neonatal diabetes mellitus. Regarding the effect on common T2D risk, as we shall detail below, some common variants in the *KCNJ11* and *WFS1* genes proved to be significantly involved in the predisposition of this disease phenotype.

Genetic defects of insulin action

The most recent ADA-WHO classification of diabetes[2] includes very rare cases of diabetes that result from genetically determined defects of insulin action, with associated severe insulin resistance. The metabolic abnormalities may range from hyperinsulinemia and modest hyperglycemia to severe diabetes.

In the 1970s were described for the first time[11] patients with severe insulin resistance, extreme hypeinsulinemia and *Acanthosis Nigricans*. These were later shown to be induced by mutations of the Insulin Receptor (IR) gene on chromosome 19p13 and are currently classified as Type A syndrome, Leprechaunism and Rabson-Mendenhall syndrome. A second group of monogenic defects of insulin action associate insulin resistance with lipodystrophy. In this group are included the congenital generalized lipodystrophy (also known Berardinelli-Seip Congenital Lipodystrophy - BSCL) and the familial partial lipodystrophy (FPL), also known as Dunnigan-Köberling syndrome. The main genetic and clinical characteristics of these syndromes are given in Table 3. It should be noted that from all these genes, only the *PPAR*γ gene on chromosome 3p25 showed conclusive evidence of association with the common forms of T2D.

Table 2

Neonatal diabetes and other rare monogenic syndromes including diabetes (adapted after [27])

Disease	Gene	Locus	Detailed name	Clinical features
TNDM	*PLAGL1* (ZAC)	6q24	Pleomorphic adenoma gene-like 1	Intrauterine growth retardation,acute onset diabetes, insulin treatment, remission between 3-6 months, usually relapses later in life
	KCNJ11	11p15.1	Potassium channel (subfamily J, member 11)	
	ABCC8 (SUR1)	11p15.1	Sulfonylurea Receptor	
PNDM	*KCNJ11*	11p15.1	Potassium channel (subfamily J, member 11)	Intrauterine growth retardation (IUGR), acute onset diabetes, insulin treatment, KCNJ11 and ABCC8 types can be treated successfully with high dose sulphonylurea theraphy, usually with better results than insulin
	ABCC8 (SUR1)	11p15.1	Sulfonylurea Receptor	
	INS	11p15.5	Insulin gene	
	GCK (homozygous)	7p15-p13	Glucokinase	
Mitochondrial diabetes	*mtDNA*	mt3243A>G	Mitochondrial DNA	Maternally inherited, usually diagnosed later in life, almost all carriers develop diabetes, 75% deafness, increased risk for stroke, epilepsy, renal and cardiac disease
Wolfram Syndrome	*WFS1*	4p16.1	Wolframin	Childhood onset; associates optic atrophy, deafness, diabetes insipidus, gonadal atrophy, neurological and psychiatric disease. Median age at death is 30 years.
Wolcott-Ralisson Syndrome	*EIF2AK3* (PERK)	2p12	Pancreatic EIF2 alpha kinase	Childhood onset, associates epiphyseal dysplasia, renal and hepatic dysfunction and mental retardation. Most cases do not survive beyond 15 years.

Table 3

Monogenic syndromes of severe insulin resistance associating diabetes (adapted after [11])

Disease	Gene	Locus	Protein	Clinical features
Type A Syndrome	INSR	19p13.2	Insulin receptor	Normal or accelerated growth in young women, hyperandrogenism, hirsutism, absence of obesity. By definition, it cannot occur in males
Rabson Mendenhall Syndrome	INSR	19p13.2	Insulin receptor	Childhood onset, dysplastic nails, abnormal dentition, accelerated growth, precocious pseudopuberty and pineal hyperplasia.
Leprechaunism	INSR	19p13.2	Insulin receptor	Very rare; the most severe insulin resistance syndrome. Autosomal recessive transmission with mutations in both copies of IR gene. IUGR and postnatal growth retardation. Thick lips, large ears, globular eyes, dwarfism, hirsutism and acanthosis nigricans, breast enlargement, abdominal distension and lipoatrophy.
Berardinelli-Seip Syndrome (BSCL-1)	AGPAT2	9q34.3	Lysophosphati-dic acid acyltransferase β	Autosomal recessive, more frequent in African origin subjects. Sometimes delayed onset and only partial lipodistrophy. Less frequent intellectual impairment and premature death.
Berardinelli-Seip Syndrome (BSCL-2)	Seipin	11q13	Seipin	Autosomal recessive. Generalized lipodistrophy, ↑ TAG, intelectual impairment, NAFLD which can progress to cirrhosis, hypertrophic cardiomyopathy, cardiac failure, premature death.
Dunnigan-Köberling Syndrome (FPL1/FPL2)	LMNA	1q21.2	Lamin A/C	Autosomal dominant, loss of subcutaneous fat from the limbs and trunk and buttocks. Fat accumulation on neck, shoulders. Lean, muscular limbs, thick nails, premature teeth eruption. Type IV hyperlipoproteinemia.
Dunnigan-Köberling Syndrome (FPL3)	PPARG	3p25	PPARγ	
Alström Syndrome	ALMS-1	2p13	Alström syndrome gene	Atypical pigmentary retinal degeneration, blindness, sensorineural deafness, infantile obesity, acanthosis nigricans, hypogonadism, skeletal anomalies, CKD, alopecia, hypothyroidism, GH deficiency and central diabetes insipidus.

In conclusion, monogenic forms of diabetes were excellent genetic models for the study of insulin secretion or insulin resistance and provided a high number of functional candidate genes for the study of common forms of T2D. However, with rare exceptions, none of these genes was demonstrated to be singificantly involved in the pathogenesis of the common forms of T2D.

CANDIDATE GENE STUDIES IN T2D

As we already mentioned, the study of T2D genetics was based both on candidate gene studies (comparison of SNPs between diabetes patients and controls in selected genes thought to encode proteins relevant for the pathogenesis of T2D) and genome-wide studies, including genome wide linkage mapping (GWS) and genome-wide association (GWA) studies. T2D candidate genes group both genes related to pancreatic beta cell function and insulin resistance genes (Table 4).

Initial studies focused on a small number of candidate genes such as insulin, insulin receptor, insulin receptor substrate (IRS1 and 2), GLUT4 glucose transporter, glucokinase, etc. These early studies, which focused almost entirely on coding variation, were largely viewed as nonreproducible and relatively uninformative for common forms of T2D. In retrospect, it is obvious that most such studies were seriously underpowered or focused on inappropriate candidates[47]. However, by combining the data of several studies, the candidate gene approach finally identified two T2D genes now considered widely replicated: *PPARG* and the β-cell potassium channel (Kir6.2) gene, *KCNJ11*[28]. Interestingly, both these genes encode proteins that are targets for T2D drugs: the insulin sensitizing thiazolidindiones for PPARγ and sulphonylureas for the partner protein of Kir6.2 (in fact encoded by the *ABCC8* gene).

PPARG gene on chromosome 3 was an attractive candidate gene because it encodes the molecular target for thiazolidinediones. Already in 1997, Yen et al. described an association between the the proline-to-alanine change at position 12 of *PPARG* (Pro12Ala or rs1801282) in and the risk of T2D[48]. Pro12Ala SNP was a plausible candidate

Table 4

T2D Candidate genes

Beta cell function	**Insulin Resistance**			
	CH Metabolism	**Lipid Metabolism**	**Insulin Action**	**Obesity**
HLA	GLUT1	LPL	Insulin	Leptin
Insulin	GLUT2	HSL	IR	Leptin Receptor
K_{ATP} – Kir6.2	GLUT4	ApoAI, Apo AII	IRS 1, 2	β_3 - AR
HNF4α	Hexokinază	ApoB	PI3K	UCP1
Glucokinase	Glicogen Sinthase	ApoE	PKB	UCP2 and UCP3
HNF1α	Fospho-fructokinase	PPARα	ENPP1	TNFα
IPF-1	Adenosine Desaminase	PPARγ		PPARγ
HNF1β	GLP-1	Mitochondrial DNA		PPARGC1A
Wolframin	Mitochondrial DNA			DRD2
				Adiponectin
				Resistin

Kir – Potassium Inward Rectifier; HNF – Hepatocyte Nuclear Factor; GLUT – Glucose Transporter; GLP-1 – Glucagon Like Peptide-1; LPL – Lipoprotein Lipase; HSL – Hormone Sensitive Lipase; IR – Insulin Receptor; IRS – Insulin Receptor Substrate; PI3K – Phosfatidil Inositol 3 Kinase; PKB – Protein Kinase B; β_3 – AR – β_3 Adrenergic Receptor; UCP – Uncoupling protein; DRD2 – Dopamin Receptor; PPARGC1A - PPARγ-coactivator 1A.

since it led to a coding change of amino-acid, with presumably functional significance (this being in contrast to synonymous coding SNPs, which preserve the same amino acid through the degeneracy of the genetic code, or to noncoding SNPs that lie in intergenic or untranslated regions). It was subsequently showed that individuals homozygous for the common 12Pro allele are more insulin resistant[49]. Finally, a large metaanalysis[50] showed that carriers of the 12Pro allele have 20% increased risk of T2D in comparison with carriers of the 12 Ala gene variant.

KCNJ11 gene on chromosome 11p15.1 was the second confirmed T2D candidate gene. Remarkably, this gene was reported to be involved in the pathogenesis of neonatal diabetes. A missense Glu23Lys mutation was described (E23K or rs5210) in which Glutamate is changed into Lysine at codon 23 of *KCNJ11*. Already in 1998 this SNP was reported to be associated with T2D in an initial metaanalysis[51]. Subsequently, this report was confirmed by a large-scale association study[52], with and Odds Ratio of ~1.20 and a convincing overall p value (P < 10-5). Furthermore, the risk allele was also associated with impaired insulin secretion[46]. In retrospect, the P12A variant of *PPARG* and E23K variant in *KCNJ11* have only modest effects on disease risk (OR ~1.2), far too small to offer clinically useful predictive tools.

Another large group of T2D candidate genes included those already known to be involved in monogenic forms of diabetes, mainly MODY. Following these studies, a conclusive association of an intronic MODY5 - *HNFB* gene SNP (rs757210) with T2D was established[46]. Thus, the combined analysis of more than 15,000 samples[42] showed an overall OR of 1.12 with a p value of < 10-6. Similar results were provided by another large-scale study[44]. Of the other MODY genes, suggestive evidence, but not quite reaching genome-wide statistical significance was obtained for two promoter variants in MODY1 gene HNF4α and for the A98V and I27L missense SPNs in the MODY 3 gene *HNF1A*[46]. From the other monogenic forms of diabetes indeuced by defects of beta cell genes, the analysis of the Wolfram syndrome gene *WFS1* on chromosome 4p16.1 in a case-control study comprising about 24,000 samples identified two SNPs that were robustly (p < 10-7) but modestly (OR~1.11) associated with T2D[53].

Finally, another T2D candidate gene that has been studied extensively during the last decade is the *ENPP1* gene on chromosome 6q22-q23 that encodes the protein *Ectonucleotide Pyrophosphatase Phosphodiesterase 1* (also known as *Plasma cell differentiation antigen/glycoprotein* - PC-1). Pizzuti et al. described a K121Q SNP of ENPP1 gene that was strongly associated with insulin resistant, healthy, nonobese, nondiabetic Caucasians in Sicily[54]. Although subsequent larger studies reported a positive association with type 2 diabetes, other well-powered studies failed to replicate this association[46]. However, a recent metaanalysis of about 42,000 samples[55] confirmed the association of this *ENPP1* gene variant with T2D in European populations under a recessive model (OR 1.38, p < 0.005).

More recently, several other T2D associations were identified using the candidate gene approach. These include *SREBF1* (Sterol Regulatory Element

Binding Transcription Factor 1)[56], *PPARGC1A* (PPARγ-coactivator 1A, PGC-1α)[57], *FOXO1* (Forkhead Box O1)[58] and SHBG (sex hormone-binding globulin)[59], but all have to be replicated in other populations in order to become genuine T2D genes.

Finally, a Finnish-USA group[60], using SNP- and gene-based analysis methods, replicated previously reported SNP T2D associations in *PPARG*, *KCNJ11*, and *SLC2A2* genes, it identified significant SNPs in genes with previously reported associations (*ENPP1* rs2021966 and *NRF1* rs1882095) and implicated novel genes, including *RAPGEF1* (rs4740283) and *TP53* (rs1042522).

T2D GENETIC LOCI IDENTIFIED BY GENOME WIDE LINKAGE SCANNING (GWLS)

Linkage analysis is a genetic method that tests the co-segregation of a microsatellite marker with a disease locus by genotyping several hundred microsatellite markers spanned all over the genome on multiplex families with two or more affected siblings. In the last 30 years, several genome-wide linkage scans were performed for T2D in different populations, including Caucasians (European and USA), Mexican-Americans, African-Americans, Asians, etc[11,61]. Results from these studies were usually non-conclusive, and quite often not replicated in other populations. However, a few regions were consistently reported to provide suggestive evidence for linkage with T2D – Table 5 – and we shall discuss below the characteristics of the most important.

Calpain 10 gene. The first report of a positive result emerging from GWL scanning in T2D families was represented by *CAPN-10*, the gene encoding Calpain 10. Already in 1996, a peak of linkage with T2D was described on chromosome 2, the most promising region being located at 2q37.3[62]. Subsequently, by positional cloning in a Mexican American population, the causal gene was identified to be *CAPN 10* wich was later designated as the *NIDDM-1* locus[63,64]. *CAPN-10* encodes a cytoplasmic cysteine protease, member of the calpain family and is ubiquitously expressed in both adult and fetal tissues[65].

The initial studies by Horikawa et al. reported 3 SNPs in the *CAPN-10* gene associated with T2D: SNP -43 (G/A) in intron 3, SNP -19 (3/2) in intron 6 and SNP –63 (C/T) in intron 13 and described several T2D associated haplotypes. Although the haplotypes described originally were not replicated in all populations, selected SNPs in the promoter region do display evidence of association in several metaanalysis[66,67]. The reasons for this heterogeneity remain largely unexplained[11]. The exact mechanism of CAPN-10 gene involvement in T2D pathogenesis remains unknown but functional evidence accumulated in recent studies suggests a potential role in both insulin resistance and insulin secretion[68].

Adiponectin gene. In 2000, a GWL study on French families identified a peak of linkage on chromosome 3q27-qter[69]. Adiponectin (*ADIPOQ*) gene was an excellent candidate since it was shown to seggregate with T2D and MetS on chromosome 3q27 in both French and Japanese populations[70]. Subsequently, strong linkage was found in Hispanic Americans by the *Insulin Resistance and Atherosclerosis Study Family Study*[71] but not reconfirmed in Pima Indians and African Americans[11].

Table 5

T2D loci identified identified in the past by genome wide linkage scaning. (adapted after 11)

Locus	Candidate Gene	Name
1q21-q23	APOA2	Apolipoprotein A-I (APOA2)
1q21-q23	INSRR, IRR	Insulin receptor-related receptor
1q21	PKLR, PK1	Pyruvate kinase, liver and RBC type
1q21.2	LMNA	Lamin A/C
2q37.3	CAPN-10	Calpain-10
3q26.1-q26.3	SLC2A2, GLUT2	Solute carrier family-2 (facilitated glucose transporter)
3q27	ADIPOQ	Adiponectin (gelatin-binding protein, 28kD)
4q28-q31	FABP2	Fatty Acid-Binding Protein 2
8p23	PPP1R3B	Protein Phosphatase 1, Glycogen-Targeting Subunit
10q25.3	TCF7L2	Transcription factor 7-like 2
12q24.31	HNF1α	Hepatocyte nuclear factor 1 α
20q	HNF4α	Hepatocyte nuclear factor 4 α

Adiponectin (ApN or ADPN or ADIPOQ or APM1) is a 30 kDa protein structurally similar to complement 1q, secreted by adipocytes. ApN plays important roles in insulin action, energy homeostasis, inflammation etc. ApN levels are decreased in insulin-resistant patients with obesity, T2D or MetS and correlate well with insulin sensitivity. Moreover, epidemiological studies have shown that ApN is a powerful risk marker for diabetes in predisposed individuals[11]. All these data strongly suggested *ADIPOQ* as an excellent functional T2D candidate gene.

More than 10 SNPs were described in the *ADIPOQ* gene with possible correlation to the plasma ApN levels, MetS and risk to develop T2D, the best studied being SNP-45 in exon 2 and SNP 276 in intron 2. For SNP -45T/G, the G allele would be pathogenic in most studies and associated to high risk of T2D and decreased insulin sensitivity[72]. However other studies suggested that the T allele would be pathogenic while some found no association between SNP45 and T2D[73]. In the French population, two other SNPs (C–11377G and G-11391A) from the promoter region were reported to be associated with hypoadiponectemia and T2D[73]. Discrepancies between these studies may be explained by statistical errors, ethnic differences or biases in inclusion criteria.

TCF7L2 gene. As early as 2003, a modest peak of linkage was described on chromosome 10 by researchers at deCODE genetics consortium on Icelandic T2D families[74]. Subsequently, the thorough exploration of this linkage signal led to the identification of a T2D associated gene[75], namely the gene encoding the Transcription Factor 7-Like 2 (*TCF7L2*) located at 10q25.3. TCF7L2 (also known as TCF-4) is a transcriptional factor involved in *Wnt* signaling, being able to bind β-catenin. This pathway of signaling is involved in embryogenesis, including adipocyte and pancreatic tissue formation. Gene activation is followed by expression of proglucagon and glucagon-like peptides 1 and 2.

The initial association on Icelandic families was quickly reconfirmed by deCODE investigators on samples from the USA and Denmark. Detailed resequencing and fine mapping pointed out to an intronic SNP (rs7903146) as the main source of the association signal in European populations[75]. The effect size in this initial report appeared substantial (each additional copy of the risk allele was associated with an OR of ~1.5) and the strength of the association was very significant, with an overall p value of ~10–18. This means that predisposing variant within the *TCF7L2* gene is approximately 1.5 times more common in T2D patients than in controls, corresponding to an approximately 50% increase in risk of T2D per copy carried.

Further studies confirmed *TCF7L2* as the locus that confers the strongest effect on T2D diabetes risk, so that some authors even stated that this could be "*the biggest story in diabetes genetics since the discovery of HLA's in T1D*"[76]. In fact, the association of *TCF7L2* with T2D has been replicated in almost every population examined, with an OR of about 1.4 and a p value of ~ 10-80[77,78]. As expected, *TCF7L2* has no effect on T1D risk, however, an interesting recent report shows a potential effect on the risk for the latent autoimmune diabetes in adults (LADA) phenotype[79,80]. As for the molecular mechanism of *TCF7L2* involvement in T2D pathogenesis, it seems that the risk allele leads to impaired insulin secretion by altering three different mechanisms: glucose-stimulated insulin secretion, incretin-stimulated insulin secretion and proinsulin-to-insulin conversion[81,82].

T2D GENETIC LOCI IDENTIFIED BY GENOME WIDE ASSOCIATION SCANNING (GWAS)

T2D has been the focus of more GWA studies than any other disorder studied to date. In fact, extraordinary progresses in deciphering T2D genetics using whole genome GWAS were recorded during the last 5 years. Thus, GWA studies identified in only a couple of years (2007–2009) more genuine T2D association genes than the candidate gene approach during decades. These progresses were possible not only due to the advent of the high trhoughput genotyping technology (as described in the *Methods for the genetic analysis of T2D section*) but also through common efforts of many groups in obtaining sufficient biological material from large case/sontrol datasets. These international collaborations substantially contributed to the application of GWAS and include the *Diabetes Genetic Initiative* (DGI), *Wellcome Trust Case Control Consortium (WTCCC)/UK Type 2 diabetes Genetics Consortium, Finland-United States Investigation of NIDDM Genetics* (FUSION), *deCODE* genetics in Island and *McGill/Imperial College study* containing mainly French families[11].

Table 6

Major GWAS performed in T2D (adapted after [11,28]).

GWA Collection	Ref.	Ethnic Groups	Sample size (GWA/Repl.)	T2D associated loci
MacGill/Imperial College	[84]	French	1363 / 5511	*HHEX* and *SLC30A8*
Fusion	[85]	Finnish	2335 / 2473	*CDKAL1, CDKN2A,* and *IGF2BP2*
WTCCC	[33,86,87]	British/Irish	4862 / 9103	*CDKAL1, CDKN2A,* and *IGF2BP2; FTO* with T2D and obesity
DeCode	[88]	Icelandic	6674 / 14138	*CDKAL1* with T2D and insulin secretion
DGI	[89]	Finish, Swedish	2931 / 10850	*CDKAL1, CDKN2A,* and *IGF2BP2*
Framingham	[90,91,92]	American Caucasians	1087 /1390	*MS4A7*; Confirm *TCF7L2* and *SLC30A8*
WTCC, Fusion, DGI (DIAGRAM)	[93]	European GWA meta-analysis	10,128 / 79,792	*NOTCH2, JAZF1, ADAMTS9, TSPAN8, THADA* and *CDC123/CAMK1D*
Japanese Genome Scan	[94]	Japanese, Korean, Chinese	1691 / 18239	*KCNQ1* in East Asians
Japanese Genome Scan	[95]	Japanese, Singaporean	1752 / 19489	*KCNQ1* in East Asians
McGill	[96]	French, Danish	1376 / 27033	*IRS1*
MAGIC (ENGAGE, Fusion, DGI, DeCode)	[97]	European	36610 / 82689	*MTNR1B*
DGI	[98]	Swedish, Finnish	2931 / 18831	*MTNR1B*
MAGIC	[99]	European	46186 / 127677	*ADCY5, PROX1, GCK, GCKR,* and *DGKB*
MAGIC, DIAGRAM	[100]	European	5643 / 84605	*RBMS1*
DIAGRAM+	[83]	European	47117 / 94337	*DUSP9, BCL11A, WFS1, ZBED3, KLF14, TP53INP1, TLE4, CENTD2, HMGA2, HNF1A, PRC1* and *ZFAND6*
East Asia	[101]	East Asians	18817 / 35873	8 new T2D loci in East Asians
-	[102]	African - American	1994 / 6449	10 T2D loci in African Americans

HHEX - Homeobox Hematopoietically Expressed; *SLC30A8* - Solute Carrier family 30 Member 8; *CDKAL1* - CDK5 Regulatory Subunit Associated Protein 1-like 1; *CDKN2A/B* - Cyclin-Dependent Kinase Inhibitor 2A/B; *IGF2BP2* - IGF-II mRNA-Binding Protein 2; *FTO* – Fat Tissue and Obesity associated gene ; *MS4A7* - Membrane-Spanning 4-domains, subfamily A, Member 7; *NOTCH2* - Drosophila Homolog 2 of NOTCH; *JAZF1* - Juxtaposed with Another Zinc Finger gene 1; *ADAMTS9* - A Disintegrin-like And Metalloproteinase with ThromboSpondin type 1 motif 9; *TSPAN8* - Tetraspanin 8; *THADA* - Thyroid ADenoma-Associated gene; *CDC123/CAMK1D* - Cell Division Cycle protein 123 homolog/calcium-calmodulin-dependent protein kinase 1D; *KCNQ1*- Potassium Channel, Voltage-Gated, KQT-like subfamily, member 1; *IRS1* – Insulin Receptor Substrate 1; *MTNR1B* - Melatonin Receptor 1B; *ADCY5* - Adenylate Cyclase 5; *PROX1* - Prospero-Related homeobox 1; *GCK* – Glucokinase; *GCKR* – Glucokinase Regulatory Protein; *DGKB* - Diacylglycerol Kinase Beta, 90-kD; *RBMS1* - RNA-Binding Motif Protein, Single Strand-Interacting 1; *DUSP9* - DUal-Specificity Phosphatase 9; *BCL11A* - B-Cell CLL/Lymphoma 11A; *ZBED3* - Zinc finger, BED-type containing 3; *KLF14* - Kruppel-Like Factor 14; *TLE4* - Transducin-Like Enhancer of split 4; *TP53INP1* - Tumor Protein p53-Inducible Nuclear Protein 1; *CENTD2* - Centaurin Delta-2; *HMGA2* - High Mobility Group At-Hook 2 ; *PRC1* - Protein Regulating Cytokinesis 1; *ZFAND6* - Zinc Finger AN1 Domain-containing protein 6;

Later, combination of these datasets in larger studies led to “Mega-Consortia” in T2D genetics including tens of thousands of cases/controls. Thus, WTCCC, FUSION and DGI combined their data to form the *Diabetes Genetics Replication and Metaanalysis* (DIAGRAM) consortium. MAGIC (*Meta-Analysis of Glucose and Insulin-related traits Consortium*) represents an international collaborative effort to combine data from multiple GWAS to identify additional loci that affect glycemic and metabolic traits. Finally, most T2D genetics cohorts have now combined to form the DIAGRAM+ Consortium, which achieved an effective sample size of over 22,000 subjects of European origin. In a recent DIAGRAM+ report[83], ~2.5 million autosomal SNPs were examined for T2D association and provided 12 new signals that reached genome-wide significance ($p < 5 \times 10{-}8$).

We are giving in Table 6 the main characteristics/ results of the major T2D GWAS published to date.

The results of the first T2D GWAS were published in 2007 in *Nature*, *Nature Genetics* and *Science* and reported nine genes associated with T2D: *CDKAL1* (CDK5 regulatory subunit associated protein 1-like 1), *SLC30A8* (solute carrier family 30 (zinc transporter), member 8), *HHEX* (homeobox hematopoietically expressed), *LOC387761*, *EXT2* (Exostosin 2), *IGF2BP2* (IGF-II mRNA-binding protein 2), *CDKN2A/B* (cyclin-dependent kinase inhibitor 2A/B), an intragenic region on 11p12, and confirmation of *TCF7L2*, the gene with, by far, the strongest association.

The first GWAS for T2D was conducted in a French cohort comprising 661 T2D cases and 614 nondiabetic controls, genotyped for 392,935 SNPs[84].

Although two associations were not reconfirmed in other follow-up studies (*LOC387761* and *EXT2*), the GWA of Sladek *et al.* identified novel and reproducible association signals at *SLC30A8* and *HHEX* and validated the already described association at *TCF7L2*. *SLC30A8* gene is located on chromosome 8q24.11 and encodes for a zinc transporter that is expressed in the β-cell. The authors described a nonsynonymous SNP (rs13266634) leading to a R325W substitution associated with T2D. The second locus was detected on chromosome 10 which contains 3 genes of interest: *IDE* (Insulin degrading enzyme), a homeodomain protein *HHEX* (a target of Wnt pathway) and KIFF11 (Kinesis Interacting Factor 11). Finally SNP rs1111875 of *HHEX* was reported to be associated with T2D.

Investigators from the Icelandic deCODE consortium and their collaborators[88] confirmed the association of *SLC30A8* and *HHEX* and identified an additional signal in *CDKAL1* on 6p22.3. *CDKAL1* gene encodes a 579-residue, 65-kD protein (sharing considerable amino acid homology with CDK5RAP1, an inhibitor of CDK5 activation) and is expressed in human pancreatic islet and skeletal muscle. On the same day, three other collaborating groups, namely the WTCCC Consortium, FUSION group and the DGI group published their GWAS findings, replicating *SLC30A8* and *HHEX* and independently discovering novel associations at *CDKAL1*, *IGF2BP2*, and *CDKN2A/B*[33,85,89].

Following the results of the first GWASs published in 2007, analysis of larger collections (MAGIC, DIAGRAM) and meta-analysis of already published data led to the identification of numerous other T2D loci. To date, almost T2D 40 loci were confirmed at genome wide significance level. Their complete list is given in Table 7.

FUNCTIONAL SIGNIFICANCE OF THE NEWLY DESCRIBED T2D GENETIC LOCI

The majority of genes identified following the hypothesis-free GWA scanning had an unknown function and have not been previously reported as associated with any diabetic phenotype. To reveal their functional relevance, large cohorts of patients with metabolic disturbances (various degrees of glucose intolerance up to overt T2D, obesity, etc.) were thoroughly phenotyped and subsequently genotyped for these genes. Such efforts led to the description of the principal effect of several GWA identified genes. One of the most interesting finding of these studies was that the vast majority of the T2D genes affect β-cell function and only a minority influence insulin sensitivity or the degree of adiposity. This led some researchers to question even the role of insulin resistance in the pathogenesis of T2D[104] and ask rethorically "*Where are the insulin resistance genes?*"[105].

Genes influencing insulin secretion. At least 20 gene regions have been reported to be associated with impaired insulin secretion[21,82], including *KCNJ11, CAPN10, HNF1B, TCF7L2, WFS1, SGK1, SLC30A8, HHEX, CDKAL1, IGF2BP2, CDKN2A/B, FOXO1, JAZF1, KCNQ1, CDC123/CAMK1D, THADA, ADAMTS9, TSPAN8/LGR5* and *MTNR1B*. The products of these genes may influence different mechanisms involved in insulin secretion and these include: glucose stimulated insulin secretion, incretin mediated insulin secretion, conversion of proinsulin to mature insulin, etc. Moreover, it was reported that these T2D associated genes influence to different extents the rate and speed of progression from IGF to clinically overt T2D[106]. We are giving in Table 8 an overview of the main associations reported so far.

Genes influencing insulin sensitivity. These include *PPARG, CAPN10, ENPP1, ADIPOQ, SREBF1, PPARGC1A, SHBG, ADAMTS9, IRS1* and *TCF7L2*[21,82]. The functional relevance of *PPARG, ENPP1, ADIPOQ* and *CAPN10* was presented above in the respective paragraphs. *SREBF1* gene (Sterol Regulatory Element-Binding Transcription Factor 1on chromosome 17p11.2 encodes for SREBP-1a and 1c - transcription factors that play important roles in lipid metabolism. The precise molecular mechanism by which this gene alters insulin sensitivity is not known and the same is valid for *ADAMTS9*. *PPARGC1A* gene (PPARγ Coactivator 1) on chromosome 4p15.1 encodes for

Table 7

T2D loci reaching statistical significance identified by GWAS (adapted after 103) Loci are presented in chronologically, according to the year of their discovery and chromosomal location.

Locus	Chr.	SNP	Allele	OR	Ref.
2000					
PPARG	3p25	rs18012824	C/ G	1.14 (1.08–1.20)	[50]
2003					
KCNJ11/ ABCC8	11p15.1	rs5219 / rs757110	T/C G/T	1.15 (1.09-1.21)	[52]
2006					
TCF7L2	10q25.3	rs7903146	T/C	1.37 (1.28-1.47)	[75]
2007					
IGF2BP2	3q28	rs4402960	T/ G	1.17 (1.10–1.25)	[85,86,89]
CDKAL1	6p22.3	rs7754840	C/ G	1.12 (1.08–1.16)	[85,86,89]
SLC30A8	8q24.11	rs13266634	C/ T	1.12 (1.07–1.16)	[84,99]
CDKN2A/B	9p21	rs10811661	T/ C	1.20 (1.14–1.25)	[85,86,89]
HHEX	10q24	rs1111875	C/ T	1.13 (1.08–1.17)	[84]
FTO	16q12.2	rs8050136	A/ C	1.15 (1.09–1.22)	[85,86,87,89]
HNF1B	17q12	rs757210	A/ G	1.12 (1.07–1.18)	[42]
2008					
NOTCH2	1p13-p11	rs10923931	T/ G	1.13 (1.08–1.17)	[93]
THADA	2p21	rs7578597	T/ C	1.15 (1.10–1.20)	[93]
ADAMTS9	3p14.3	rs4607103	C/ T	1.09 (1.06–1.12)	[93]
JAZF1	7p15	rs864745	T/ C	1.10 (1.07–1.13)	[93]
CDC123/CAMK1D	10p13	rs12779790	G/ A	1.11 (1.07–1.14)	[93]
KCNQ1	11p15.5	rs2237892 / rs231362	C/T G/A	1.4 (1.34–1.47) 1.08 (1.06–1.10)	[83,94,95]
TSPAN8/ LGR5	12q14.1	rs7961581	C/ T	1.09 (1.06–1.12)	[93]
2009					
IRS1	2q36.3	rs2943641	C/T	1.19 (1.13–1.25)	[96]
2010					
DUSP9	Xq28	rs5945326	G/ A	1.27 (1.18–1.37)	[83]
PROX1	1q32.3	rs340874	C/ T	1.07 (1.05–1.09)	[99]
BCL11A	2p16.1	rs243021	A/G	1.08 (1.06–1.10)	[83]
GCKR	2p23.3	rs780094	C/T	1.06 (1.04–1.08)	[99]
ADCY5	3q21.1	rs11708067	A/G	1.12 (1.09–1.15)	[99]
WFS1	4p16.1	rs1801214	G/ A	1.13 (1.07–1.18)	[83]
ZBED3	5q13.3	rs4457053	G/ A	1.08 (1.06–1.11)	[83]
DGKB/TMEM195	7p21.2	rs2191349	T/G	1.06 (1.04–1.08)	[99]
GCK	7p15-p13	rs4607517	A/G	1.07 (1.05–1.10)	[99]
KLF14	7q32.3	rs972283	G/ A	1.07 (1.05–1.10)	[83]
TP53INP1	8q22.1	rs896854	T/ C	1.06 (1.04–1.09)	[83]
TLE4/CHCHD9	9q21.31	rs13292136	C/T	1.11 (1.07–1.15)	[83]
CENTD2	11q13.4	rs1552224	A/C	1.14 (1.11–1.17)	[83]
MTNR1B	11q14.3	rs10830963	G/C	1.09 (1.06–1.12)	[99]
HMGA2	12q14.3	rs1531343	C/ G	1.10 (1.07–1.14)	[83]
HNF1A	12q24.31	rs7957197	T/ A	1.07 (1.05–1.10)	[83]
PRC1	15q26.1	rs8042680	A/ C	1.07 (1.05–1.09)	[83]
ZFAND6	15q25.1	rs11634397	G/ A	1.06 (1.04–1.08)	[83]

Table 8

Effects of T2D genes on different mechanisms of insulin secretion (adapted after [82])

Gene	Chr.	SNP	Altered mechanism of insulin secretion
CAPN10	2q37.3	rs3792267, rs3842570 rs5030952	Glucose-stimulated insulin secretion Proinsulin-to-insulin conversion
IGF2BP2	3q28	rs4402960	Glucose-stimulated insulin secretion
WFS1	4p16.1	rs10010131	Incretin-stimulated insulin secretion
CDKAL1	6p22.3	rs7754840	Glucose-stimulated insulin secretion Proinsulin-to-insulin conversion
JAZF1	7p15	rs864745	Glucose-stimulated insulin secretion
SLC30A8	8q24.11	rs13266634	Glucose-stimulated insulin secretion Proinsulin-to-insulin conversion
CDKN2A/2B	9p21	rs10811661	Glucose-stimulated insulin secretion
CDC123/CAMK1D	10p13	rs12779790	Glucose-stimulated insulin secretion
HHEX	10q24	rs7923837	Glucose-stimulated insulin secretion
TCF7L2	10q25.3	rs7903146, rs12255372 rs7901695	Incretin-stimulated insulin secretion Proinsulin-to-insulin conversion Glucose-stimulated insulin secretion
KCNJ11	11p15.1	rs5219	Glucose-stimulated insulin secretion
KCNQ1	11p15.5	rs2237892, rs151290	Incretin-stimulated insulin secretion
MTNR1B	11q14.3	rs10830963, rs10830962, rs4753426	Glucose-stimulated insulin secretion
TSPAN8/ LGR5	12q14.1	rs7961581	Decreased glucose-stimulated insulin secretion

PGC-1α which is a co-activator of nuclear receptors, such as PPARδ. It was shown to be associated directly or indirectly (via ectopic intramyocellular or hepatic fat accumulation) with insulin sensitivity[82]. The G482S (rs8192678) polymorphism of *PPARGC1A* was found to be associated with lower insulin sensitivity, although the exact mechanism remains unknown[107]. More recently, the C allele of rs2943641 in *IRS1* gene on chromosome 2q36.3 was reported to be associated with insulin resistance and hyperinsulinemia in 3 European populations from the McGill/Imperial College GWAS[96]. Finally, it should be mentioned that *CAPN10* and *TCF7L2*, despite their presence on this list, have more important roles in the regulation of insulin secretion.

Genes influencing adiposity. By far the most important (and in fact the only one replicated) is the FTO gene on chromosome 16q12.2. As part of the WTCCC T2D GWAS, the SNPs rs9939609 and rs8050136 of *FTO* gene were described to be strongly associated with T2D[87]. Carriers of the pathogenic allele of rs9939609 (who represent almost 16% of the population) weighed 3 kg more than the rest of the population. The association was quickly replicated by other groups[108] but was abolished after adjustment for BMI[82,109], leading to the conclusion that *FTO* is directly linked to BMI and indirectly to T2D. In addition, *FTO* gene variants were reported to influence insulin sensitivity but these effects can be fully explained by the correlation between BMI and the traits of insulin resistance[109].

FTO gene in humans is expressed in almost all tissues including pancreatic islets, adipose tissue and brain and, at the cellular level, has a nuclear localization. The product of the gene is a nuclear demethylase, probably playing a role in activating genes silenced by DNA methylation[110]. How *FTO* gene is involved in the pathogenesis of obesity or T2D remains largely unknown. There are data suggesting that the risk alleles in *FTO* influence energy intake or the intake of energy-dense foods, and not energy expenditure[110] while one study reported a relationship with cortical insulin resistance[111].

In addition, a series of about 15 other “obesity” genes were identified by GWA studies[28,112]. The most significant association is for the *MC4R* (melanocortin-4 receptor) gene, but this gene was not associated with T2D[113].

CONCLUSIONS

Major progresses have been made in deciphering the genetics of T2D, especially following the breakthrough represented by the publication of the major GWA studies. Almost 40 diabetes risk genes were identified. Except a few functional candidate genes, most of the newly described genes were not suspected to be associated with diabetes. The majority of the loci identified by GWAS appear to affect insulin secretion, but the precise molecular mechanisms

are still incompletely established. Unraveling these mechanisms will help to understand the pathophysiology of T2D possibly leading to the identification of functional targets for diabetes prevention or for the pharmacological treatment of this disease. In addition, deciphering the genetic background of T2D will contribute in estimating the risk for developing type 2 diabetes in high risk subjects, with positive consequences for disease prevention.

REFERENCES

1. Shaw JE, Sicree RA, Zimmet PZ., Global estimates of the prevalence of diabetes for 2010 and 2030. *Diabetes Res Clin Pract* **2010**, 87:4-14.
2. American Diabetes Association, Diagnosis and classification of diabetes mellitus. *Diabetes Care* **2012,** 35[Suppl 1]:S64-S71.
3. Roglic G, Unwin N., Mortality attributable to diabetes: estimates for the year 2010. *Diabetes Res Clin Pract* **2010**, 87:15-19.
4. DeFronzo RA., The triumvirate: β-cell, muscle, liver. A collision responsible for NIDDM. *Diabetes* **1988**, 37:667-687.
5. DeFronzo RA., From the triumvirate to the ominous octet: A new paradigm for the treatment of type 2 diabetes mellitus. *Diabetes* **2009**, 58:773-795.
6. McCarthy MI., Progress in defining the molecular basis of type 2 diabetes mellitus through susceptibility-gene identification. *Hum Mol Gen* **2004**, 13[Review Issue 1]:R33–R41.
7. Froguel P, Velho G., Genetic determinants of type 2 diabetes. *Recent Prog Horm Res* **2001**, 56:91-105.
8. Barroso I., Genetics of Type 2 diabetes. *Diabet Med* **2005***,* 22:517-535.
9. Elbein SC., Genetics factors contributing to type 2 diabetes across ethnicities. *J Diabetes Sci Technol* **2009**, 3:685-689.
10. Knowler WC, Pettitt DJ, Saad MF, Bennett PH., Diabetes mellitus in the Pima Indians: incidence, risk factors and pathogenesis. *Diabetes Metab Rev* **1990***,* 6:1-27.
11. Grigorescu F, Attaoua R, Ait El Mkadem S, Radian Ş., Susceptibility genes for insulin resistance and type 2 diabetes. In Cheța D (ed). *Genetics of diabetes. The Truth Unveiled.* Ed Acad Rom, Bucureşti & S. Karger AG, Basel, **2010**, pp. 131-192.
12. Köbberling J, Tillil H., Empirical risk figures for first degree relatives of non-insulin dependent diabetics. In Köbberling J & Tattersall R (eds). *The Genetics of Diabetes Mellitus*.: Academic Press, London, **1982,** pp 201–209.
13. Rich SS., Mapping genes in diabetes. Genetic epidemiological perspective. *Diabetes* **1990,** 39:1315-1319.
14. Meigs JB, Cupples LA, Wilson PW., Parental transmission of type 2 diabetes: the Framingham Offspring Study. *Diabetes* **2000**, 49:2201-2217.
15. Lyssenko V, Almgren P, Anevski D, Orho-Melander M, Sjögren M, Saloranta C et al., Genetic prediction of future type 2 diabetes. *PLoS Med* **2005**, 2:e345.
16. Almgren P, Lehtovirta M, Isomaa B, Sarelin L, Taskinen MR, Lyssenko V, et al., Heritability and familiality of type 2 diabetes and related quantitative traits in the Botnia study. *Diabetologia* **2011**, 54:2811-2819.
17. Vaag A, Poulsen P., Twins in metabolic and diabetes research: what do they tell us? Curr Opin *Clin Nutr Metab Care* **2007**, 10:591–596.
18. Barnett AH, Eff C, Leslie RD, Pyke DA., Diabetes in identical twins. A study of 200 pairs. *Diabetologia* **1981**, 20:87–93.
19. Kaprio J, Tuomilehto J, Koskenvuo M, Romanov K, Reunanen A, Eriksson J *et al.*, Concordance for type 1 (insulin-dependent) and type 2 (non-insulin-dependent) diabetes mellitus in a population-based cohort of twins in Finland. *Diabetologia* **1992**, 35:1060–1067.
20. Poulsen P, Kyvik KO, Vaag A, Beck-Nielsen H., Heritability of type II (non-insulin-dependent) diabetes mellitus and abnormal glucose tolerance -- a population-based twin study. *Diabetologia* **1999**, 42:139-145.
21. Grarup N, Sparsø T, Hansen T., Physiologic characterization of type 2 diabetes-related loci. *Curr Diab Rep* **2010**, 10:485-497.
22. McCarthy MI, Smedley D, Hide W., New methods for finding disease susceptibility genes: Impact and potential. *Genome Biol* **2003**, 4:119.1–119.8.
23. Risch N., Searching for genetic determinants in the new millennium. *Nature* **2000**, 405:847–856.
24. Hirschorn JN, Daly MJ., Genome-wide association studies for common diseases and complex traits. *Nature Rev Genet* **2005**, 6:95–108.
25. Jimenez-Sanchez G, Childs B, Valle D., Human disease genes. *Nature* **2001**, 409:853–855.
26. Altmüller J, Palmer LJ, Fischer G, Scherb H, Wjst M., Genomewide scans of complex human diseases: true linkage is hard to find. *Am J Hum Genet* **2001**, 69: 936–950.
27. Waterfield T, Gloyn AL., Monogenic β-cell dysfunction in children: clinical phenotypes, genetic etiology and mutational pathways. *Pediatr Health* **2008**, 2:517-532.
28. McCarthy MI., Genomics, type 2 diabetes, and obesity. *NEJM* **2010**, 363:2339-2350.
29. Lander ES, Linton LM, Birren B, Nusbaum C, Zody MC, Baldwin J *et al.*, Initial sequencing and analysis of the human genome. *Nature* **2001**, 409: 860–921.
30. Venter JC, Adams MD, Myers EW, Li PW, Mural RJ, Sutton GG *et al.*, The sequence of the human genome. *Science* **2001**, 291:1304–1351.
31. The International HapMap consortium., A haplotype map of the human genome. *Nature* **2005**, 437:1299–1320.
32. Kruglyak L, Nickerson DA., Variation is the spice of life. *Nature Genet* **2001**, 27:234–236.
33. Wellcome Trust Case Control Consortium., Genome-wide association study of 14,000 cases of seven common diseases and 3,000 shared controls. *Nature* **2007**, 447:661–678.
34. Hardenbol P, Yu F, Belmont J, Mackenzie J, Bruckner C, Brundage T *et al.*, Highly multiplexed molecular inversion probe genotyping: over 10,000 targeted SNPs genotyped in a single tube assay. *Genome Res,* **2005**, 15:269-275.
35. McCarthy MI, Abecasis GR, Cardon LR, Goldstein DB, Little J, Ioannidis JP, Hirschhorn JN., Genome-wide association studies for complex traits: consensus, uncertainty and challenges. *Nat Rev Genet* **2008**, 5:356-369.

36. Grulich-Henn J, Wagner V, Thon A, Schober E, Marg W, Kapellen TM *et al.*, Entities and frequency of neonatal diabetes: data from the diabetes documentation and quality management system (DPV). *Diabet Med* **2010**, 27:709-712.
37. Fajans SS, Bell GI, Polonsky KS., Molecular mechanisms and clinical pathophysiology of maturity-onset diabetes of the young. *N Eng J Med* **2001**, 345:971-980.
38. Ellard S, Bellanné-Chantelot C, Hattersley AT, European Molecular Genetics Quality Network (EMQN) MODY group., Best practice guidelines for the molecular genetic diagnosis of maturity-onset diabetes of the young. *Diabetologia* **2008**, 51:546-553.
39. Vlad A, Timar R, Ivan D., Maturity onset diabetes of the young. *Rom J Diab Nutr Metab Dis* **2010**, 17:229-237.
40. McCarthy MI, Hattersley AT., Learning from molecular genetics: novel insights arising from the definition of genes for monogenic and type 2 diabetes. *Diabetes* **2008**, 57:2889-2898.
41. Bowman P, Flanagan SE, Edghill EL, Damhuis A, Sheperd MH, Paisey R *et al.*, Heterozygous ABCC8 mutations are a cause of MODY. *Diabetologia* **2012**, 55:123-127.
42. Winckler W, Weedon MN, Graham RR, McCarroll SA, Purcell S, Almgren P *et al.*, Evaluation of common variants in the six known maturity onset diabetes of the young (MODY) genes for association with type 2 diabetes. *Diabetes* **2007**, 56:685–693.
43. Staiger H, Machicao F, Fritsche A, Häring HU., Pathomechanisms of type 2 diabetes genes. *Endocrine Reviews* **2009**, 30:557–585.
44. Bonnycastle LL, Willer CJ, Conneely KN, Jackson AU, Burrill CP, Watanabe RM *et al.*, Common variants in maturity onset diabetes of the young genes contribute to risk of type 2 diabetes in Finns. *Diabetes* **2006**, 55: 2534–2540.
45. Gudmundsson J, Sulem P, Steinthorsdottir V, Bergthorsson JT, Thorleifsson G, Manolescu A et al., Two variants on chromosome 17 confer prostate cancer risk, and the one in TCF2 protects against type 2 diabetes. *Nat Genet* **2007,** 39:977–983.
46. Florez JC., The genetics of type 2 diabetes: A realistic appraisal in 2008. *J Clin Endocrinol Metab* **2008**, 93:4633–4642.
47. Hattersley AT, McCarthy MI., What makes a good genetic association study? *Lancet* **2005**, 366:1315-1323.
48. Yen CJ, Beamer BA, Negri C, Silver K, Brown KA, Yarnall DP *et al.*, Molecular scanning of the human Peroxisome proliferator activated receptor γ (hPPARγ) gene in diabetic Caucasians: identification of a Pro12Ala PPARγ2 missense mutation. *Biochem Biophys Res Commun* **1997**, 241:270–274.
49. Deeb SS, Fajas L, Nemoto M, Pihlajamaki J, Mykkanen L, Kuusisto J *et al.*, A Pro12Ala substitution in PPARγ2 associated with decreased receptor activity, lower body mass index and improved insulin sensitivity. *Nat Genet* **1998**, 20:284–287.
50. Altshuler D, Hirschhorn JN, Klannemark M, Lindgren CM, Vohl MC, Nemesh J *et al.*, The common PPARγ Pro12Ala polymorphism is associated with decreased risk of type 2 diabetes. *Nat Genet* **2000**, 26:76–80.
51. Hani EH, Boutin P, Durand E, Inoue H, Permutt MA, Velho G, Froguel P., Missense mutations in the pancreatic islet beta cell inwardly rectifying K+ channel gene (KIR6.2/BIR): a meta-analysis suggests a role in the polygenic basis of Type II diabetes mellitus in Caucasians. *Diabetologia* **1998**, 41:1511–1515.
52. Gloyn AL, Weedon MN, Owen KR, Turner MJ, Knight BA, Hitman G et al., Large-scale association studies of variants in genes encoding the pancreatic β-cell KATP channel subunits Kir6.2 (*KCNJ11*) and SUR1 (*ABCC8*) confirm that the *KCNJ11* E23K variant is associated with type 2 diabetes. *Diabetes* **2003**, 52:568–572.
53. Sandhu MS, Weedon MN, Fawcett KA, Wasson J, Debenham SL, Daly A *et al.*, Common variants in WFS1 confer risk of type 2 diabetes. *Nat Genet* **2007**, 39:951–953.
54. Pizzuti A, Frittitta L, Argiolas A, Baratta R, Goldfine ID, Bozzali M *et al.*, A polymorphism (K121Q) of the human glycoprotein PC-1 gene coding region is strongly associated with insulin resistance. *Diabetes* **1999**, 48:1881-1884.
55. McAteer JB, Prudente S, Bacci S, Lyon HN, Hirschhorn JN, Trischitta V, Florez JC., The *ENPP1* K121Q polymorphism is associated with type 2 diabetes in European populations: evidence from an updated meta-analysis in 42,042 subjects. *Diabetes* **2008**, 57:1125–1130.
56. Grarup N, Stender-Petersen KL, Andersson EA, Jorgensen T, Borch-Johnsen K, Sandbaek A, et al., Association of variants in the sterol regulatory element-binding factor 1 (SREBF1) gene with type 2 diabetes, glycemia, and insulin resistance: a study of 15,734 Danish subjects. *Diabetes* **2008**, 57:1136–1142.
57. Barroso I, Luan J, Sandhu MS, Franks PW, Crowley V, Schafer AJ, *et al.*, Meta-analysis of the Gly482Ser variant in PPARGC1A in type 2 diabetes and related phenotypes. *Diabetologia* **2006**, 49:501–505.
58. Mussig K, Staiger H, Machicao F, Stancakova A, Kuusisto J, Laakso M, *et al.*, Association of common genetic variation in the FOXO1 gene with beta-cell dysfunction, impaired glucose tolerance, and type 2 diabetes. *J Clin Endocrinol Metab* **2009**, 94:1353–1360.
59. Ding EL, Song Y, Manson JE, Hunter DJ, Lee CC, Rifai N, *et al.*, Sex hormone-binding globulin and risk of type 2 diabetes in women and men. *N Engl J Med* **2009**, 361:1152–1163.
60. Gaulton KJ, Willer CJ, Li Y, Scott LJ, Conneely KN, Jackson AU *et al.*, Comprehensive association study of type 2 diabetes and related quantitative traits with 222 candidate genes. *Diabetes* **2008**, 57:3136-3144.
61. Florez JC, 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.
62. Hanis CL, Boerwinkle E, Chakraborty R, Ellsworth DL, Concannon P, Stirling B *et al.*, A genome-wide search for human non-insulin-dependent (type 2) diabetes genes reveals a major susceptibility locus on chromosome 2. Nat Genet **1996**, 13:161-166.
63. Cox NJ, Frigge M, Nicolae DL, Concannon P, Hanis CL, Bell GI, Kong A., Loci on chromosomes 2 (NIDDM1) and 15 interact to increase susceptibility to diabetes in Mexican Americans. *Nat Genet* **1999**, 21:213-215.
64. Horikawa Y, Oda N, Cox NJ, Li X, Orho-Melander M, Hara M et al., Genetic variation in the gene encoding calpain-10 is associated with type 2 diabetes mellitus. *Nat Genet* **2000**, 26:163-175.
65. Suzuki K, Hata S, Kawabata Y, Sorimachi H., Structure, activation, and biology of calpain. *Diabetes* **2004**, 53:S12-S18.
66. Weedon MN, Schwarz PEH, Horikawa Y, Iwasaki N, Illig T, Holle R *et al.*, Meta-analysis and a large association study confirm a role for calpain-10 variation in type 2 diabetes susceptibility. *Am J Hum Genet* **2003**, 73:1208–1212.

67. Tsuchiya T, Schwarz PEH, Bosque-Plata LD, Geoffrey Hayes M, Dina C, Froguel P *et al.*, Association of the calpain-10 gene with type 2 diabetes in Europeans: results of pooled and meta-analyses. *Mol Genet Metab* **2006**, 89:174–184.
68. Turner MD, Cassell PG, Hitman GA., Calpain-10: from genome search to function. *Diabetes Metab Res Rev* **2005**, 21:505–514.
69. Vionnet N, Hani El-H, Dupont S, Gallina S, Francke S, Dotte S *et al.*, Genomewide search for type 2 diabetes-susceptibility genes in French whites: evidence for a novel susceptibility locus for early-onset diabetes on chromosome 3q27-qter and independent replication of a type 2-diabetes locus on chromosome 1q21-q24. *Am J Hum Genet* **2000**, 67:1470-1480.
70. Kissebah AH, Sonnenberg GE, Myklebust J, Goldstein M, Broman K, James RG et al., Quantitative trait loci on chromosomes 3 and 17 influence phenotypes of the metabolic syndrome. *Proc Natl Acad Sci USA* **2000**, 97:14478–14483.
71. Guo X, Saad MF, Langefeld CD, Williams AH, Cui J, Taylor KD *et al.*, Genome-wide linkage of plasma adiponectin reveals a major locus on chromosome 3q distinct from the adiponectin structural gene: The IRAS Family Study. *Diabetes* **2006**, 55:1723-1730.
72. Tso AW, Sham PC, Wat NM, Xu A, Cheung BM, Rong R *et al.*, Polymorphisms of the gene encoding adiponectin and glycaemic outcome of Chinese subjects with impaired glucose tolerance: a 5-year follow-up study. *Diabetologia* **2006**, 49:1806-1815.
73. Vasseur F, Helbecque N, Dina C, Lobbens S, Delannoy V, Gaget S *et al.*, Single-nucleotide polymorphism haplotypes in the both proximal promoter and exon 3 of the APM1 gene modulate adipocyte-secreted adiponectin hormone levels and contribute to the genetic risk for type 2 diabetes in French Caucasians. *Hum Mol Genet* **2002**, 11:2607-2614.
74. Reynisdottir I, Thorleifsson G, Benediktsson R, Sigurdsson G, Emilsson V, Einarsdottir AS *et al.*, Localisation of a susceptibility gene for type 2 diabetes to chromosome 5q34–q35.2. *Am J Hum Genet* **2003**, 73:323–335.
75. Grant SFA, Thorleifsson G, Reynisdottir I, Benediktsson R, Manolescu A, Sainz J *et al.*, Variant of transcription factor 7-like 2 (*TCF7L2*) gene confers risk of type 2 diabetes. *Nat Genet* **2006,** 38:320–323.
76. Zeggini E, McCarthy MI., TCF7L2: the biggest story in diabetes genetics since HLA? *Diabetologia* **2007**, 50:1–4.
77. Florez JC., The new type 2 diabetes gene *TCF7L2. Curr Opin Clin Nutr Metab Care* **2007**, 10:391–396.
78. Cauchi S, El Achhab Y, Choquet H, Dina C, Krempler F, Weitgasser R *et al.*, TCF7L2 is reproducibly associated with type 2 diabetes in various ethnic groups: a global meta-analysis. *J Mol Med* **2007**, 85:777–782.
79. Grant SF, Hakonarson H, Schwartz S., Can the genetics of type 1 and type 2 diabetes shed light on the genetics of latent autoimmune diabetes in adults? *Endocr Rev* **2010**, 31:183-193.
80. Lukacs K, Hosszufalusi N, Dinya E, Bakacs M, Madacsy L, Panczel P., The type 2 diabetes-associated variant in TCF7L2 is associated with latent autoimmune diabetes in adult Europeans and the gene effect is modified by obesity: a meta-analysis and an individual study. *Diabetologia* **2012**, 55:689-693.
81. Lyssenko V, Lupi R, Marchetti P, del Guerra S, Orho-Melander M, Almgren P *et al.*, Mechanisms by which common variants in the TCF7L2 gene increase risk of type 2 diabetes. J Clin Invest **2007**, 117:2155–2163.
82. Schäfer SA, Machicao F, Fritsche A, Häring HU, Kantartzis K., New type 2 diabetes risk genes provide new insights in insulin secretion mechanisms. *Diabetes Res Clin Pract* **2011**, 93[Suppl 1]:S9-S24.
83. Voight BF, Scott LJ, Steinthorsdottir V, Morris AP, Dina C, Welch RP *et al.*, Twelve type 2 diabetes susceptibility loci identified through large-scale association analysis. *Nat Genet* **2010**, 42:579-589.
84. Sladek R, Rocheleau G, Rung J, Dina C, Shen L, Serre D *et al.*, A genome-wide association study identifies novel risk loci for type 2 diabetes. *Nature* **2007**, 445:881-885.
85. Scott LJ, Mohlke KL, Bonnycastle LL, Willer CJ, Li Y, Duren WL *et al.*, A genome-wide association study of type 2 diabetes in Finns detects multiple susceptibility variants. *Science* **2007**, 316:1341-1345.
86. Zeggini E, Weedon MN, Lindgren CM, Frayling TM, Elliott KS, Lango H *et al.*, Replication of genome-wide association signals in UK samples reveals risk loci for type 2 diabetes. *Science* **2007**, 316:1336-1341.
87. Frayling TM, Timpson NJ, Weedon MN, Zeggini E, Freathy RM, Lindgren CM *et al.*, A common variant in the FTO gene is associated with body mass index and predisposes to childhood and adult obesity. *Science* **2007**, 316:889-894.
88. Steinthorsdottir V, Thorleifsson G, Reynisdottir I, Benediktsson R, Jonsdottir T, Walters GB *et al.*, A variant in CDKAL1 influences insulin response and risk of type 2 diabetes. *Nat Genet* **2007**, 39: 770-775.
89. Diabetes Genetics Initiative of Broad Institute of Harvard and MIT, Lund University, Novartis Institutes of BioMedical Research. Genome-wide association analysis identifies loci for type 2 diabetes and triglyceride levels. *Science* **2007**, 316:1331-1336.
90. Cupples LA, Arruda HT, Benjamin EJ, D'Agostino RB Sr, Demissie S, DeStefano AL *et al.*, The Framingham Heart Study 100K SNP genome-wide association study resource: overview of 17 phenotype working group reports. *BMC Med Genet* **2007**, 8:S1.
91. Florez JC, Manning AK, Dupuis J, McAteer J, Irenze K, Gianniny L *et al.*, A 100K genome-wide association scan for diabetes and related traits in the Framingham Heart Study: replication and integration with other genome-wide datasets. *Diabetes* **2007**, 56: 3063-3074.
92. Moore AF, Jablonski KA, McAteer JB, Saxena R, Pollin TI, Franks PW *et al.*, Extension of type 2 diabetes genome-wide association scan results in the diabetes prevention program. *Diabetes* **2008**, 57:2503-2510.
93. Zeggini E, Scott LJ, Saxena R, Voight BF, Marchini JL, Hu T *et al.*, Meta-analysis of genome-wide association data and large-scale replication identifies additional susceptibility loci for type 2 diabetes. *Nat Genet* **2008**; 40:638-645.
94. Yasuda K, Miyake K, Horikawa Y, Hara K, Osawa H, Furuta H *et al.*, Variants in KCNQ1 are associated with susceptibility to type 2 diabetes mellitus. *Nat Genet* **2008**, 40:1092-1097.
95. Unoki H, Takahashi A, Kawaguchi T, Hara K, Horikoshi M, Andersen G *et al.*, SNPs in KCNQ1 are associated with susceptibility to type 2 diabetes in East Asian and European populations. *Nat Genet* **2008**, 40: 1098-1102.

96. Rung J, Cauchi S, Albrechtsen A, Shen L, Rocheleau G, Cavalcanti-Proença C, *et al.*, Genetic variant near IRS1 is associated with type 2 diabetes, insulin resistance and hyperinsulinemia. *Nat Genet* **2009**, 41:1110-1115.
97. Prokopenko I, Langenberg C, Florez JC, Saxena R, Soranzo N, Thorleifsson G *et al.*, Variants in MTNR1B influence fasting glucose levels. *Nat Genet* **2009**, 41:77-81.
98. Lyssenko V, Nagorny CL, Erdos MR, Wierup N, Jonsson A, Spégel P, *et al.*, Common variant in MTNR1B associated with increased risk of type 2 diabetes and impaired early insulin secretion. *Nat Genet* **2009**, 41:82-88.
99. Dupuis J, Langenberg C, Prokopenko I, Saxena R, Soranzo N, Jackson AU *et al.*, New genetic loci implicated in fasting glucose homeostasis and their impact on type 2 diabetes risk. *Nat Genet* **2010**, 42:105-116.
100. Qi L, Cornelis MC, Kraft P, Stanya KJ, Linda Kao WH, Pankow JS, *et al.*, Genetic variants at 2q24 are associated with susceptibility to type 2 diabetes. *Hum Mol Genet* **2010**, 19:2706-2715.
101. Cho YS, Chen CH, Hu C, Long J, Ong RT, Sim X *et al.*, Meta-analysis of genome-wide association studies identifies eight new loci for type 2 diabetes in east Asians. *Nat Genet* **2011**, 44:67-72.
102. Palmer ND, McDonough CW, Hicks PJ, Roh BH, Wing MR, An SS, *et al.*, A genome-wide association search for type 2 diabetes genes in African Americans. *PLoS One* **2012**, 7(1):e29202.
103. Billings LK, Florez JC., The genetics of type 2 diabetes: what have we learned from GWAS? *Ann NY Acad Sci* **2010**, 1212:59–77.
104. Ionescu-Tirgoviste C., Insulin resistance - What is myth and what is reality? *Acta Endo (Buc)* **2011**, 7:123-146.
105. Florez JC. Newly identified loci highlight beta cell dysfunction as a key cause of type 2 diabetes: where are the insulin resistance genes? *Diabetologia* **2008**, 51:1100–1101.
106. Walford GA, Green T, Neale B, Isakova T, Rotter JI, Grant SFA *et al.*, Common genetic variants differentially influence the transition from clinically defined states of fasting glucose metabolism. Diabetologia **2012**, 55:331-339.
107. Fanelli M, Filippi E, Sentinelli F, Romeo S, Fallarino M, Buzzetti R, *et al.*, The Gly482Sermissense mutation of the peroxisome proliferator-activated receptor gamma coactivator-1 alpha (PGC-1 alpha) gene associates with reduced insulin sensitivity in normal and glucose-intolerant obese subjects. *Dis Markers* **2005**, 21:175–180.
108. Dina C, Meyre D, Gallina S, Durand E, Korner A, Jacobson P, *et al.*, Variation in FTO contributes to childhood obesity and severe adult obesity. *Nat Genet* **2007**, 39:724–726.
109. Freathy RM, Timpson NJ, Lawlor DA, Pouta A, Ben-Shlomo Y, Ruokonen A *et al.*, Common variation in the FTO gene alters diabetes-related metabolic traits to the extent expected given its effect on BMI. *Diabetes* **2008**, 57:1419-1426.
110. Gerken T, Girard CA, Tung YC, Webby CJ, Saudek V, Hewitson KS, *et al.*, The obesity-associated FTO gene encodes a 2-oxoglutarate-dependent nucleic acid demethylase. *Science* **2007**, 318:1469–1472.
111. Tschritter O, Preissl H, Yokoyama Y, Machicao F, Häring HU, Fritsche A. Variation in the FTO gene locus is associated with cerebrocortical insulin resistance in humans. *Diabetologia* **2007**, 50:2602-2603.
112. Burgdorf KS, Gjesing AP, Grarup N, Justesen JM, Sandholt CH, Witte DR, *et al.*, Association studies of novel obesity-related gene variants with quantitative metabolic phenotypes in a population-based sample of 6,039 Danish individuals. Diabetologia **2012**, 55:105-113.
113. Loos RJ, Lindgren CM, Li S, Wheeler E, Zhao JH, Prokopenko I, *et al.*, Common variants near MC4R are associated with fat mass, weight and risk of obesity. *Nat Genet* **2008**, 40:768–775.