Cell Metabolism
Perspective

Rare and Common Genetic Events in Type 2 Diabetes: What Should Biologists Know?

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Type 2 diabetes (T2D) had long been referred to as the "geneticist's nightmare." Genome-wide association studies have fully confirmed the polygenic nature of T2D, demonstrating the role of many genes in T2D risk. The increasingly busier picture of T2D genetics is quite difficult to understand for the diabetes research community, which can create misunderstandings with geneticists, and can eventually limit both basic research and translational outcomes of these genetic discoveries. The present review wishes to lift the fog around genetics of T2D with the hope that it will foster integrated diabetes modeling approaches from genetic defects to personalized medicine.

Introduction: Is Type 2 Diabetes a Genetic Disorder?

According to the World Health Organization (WHO), approximately 350 million people worldwide have diabetes, and this disorder is likely to be the seventh leading cause of death in 2030. Diabetes is an economic burden on healthcare systems, especially in developing countries (World Health Organization, 2013).

Type 2 diabetes (T2D) emerged in the early 70s and was separated from type 1 diabetes (T1D) thanks to the identification of the role of the major histocompatibility complex in the auto-immune process central to T1D pathogenesis (Bottazzo et al., 1974). T2D, which currently accounts for ~90% of all diabetic patients, was called non-insulin-dependent diabetes or adult-onset diabetes in the US (American Diabetes Association, 2013), and fat diabetes (*diabète gras*) in France, as overweight and unhealthy lifestyles were considered at that time its only triggers (Pirart, 1959). In the 80s, the scientific community started to speculate about the strong familial clustering of T2D (Zimmet, 1982), but also about the extreme difficulty underlying its genetic basis. T2D had long been referred to as the geneticist's nightmare (Neel, 1976). In 1985, a WHO study group suggested that T2D might be an autosomal dominant disorder involving only one dominant gene and characterized by a variable penetrance dependent on both obesity and aging (Vftter, 1986). The first evidence that T2D may be genetically driven was only brought in 1992, through familial linkage analysis of French pedigrees with early-onset, non-auto-immune, non-obese diabetes that was also called maturity-onset diabetes of the young (MODY) (Froguel et al., 1992). Mutations in *GCK* (encoding glucokinase) were shown to cause a relatively benign form of MODY. Incidentally, it was the first time that the direct causative effect of relative insulin deficiency was demonstrated in T2D, when insulin resistance was believed to be the only trigger. The subsequent identification of 28 distinct genes harboring mutations causing monogenic diabetes has brought groundbreaking insights on diabetes physiology, especially on insulin secretion, but they only affect less than 5% of diabetic patients (Shields et al., 2010) (Figure 1).

Common T2D is actually a complex multifactorial polygenic disease, which results from many different genetic events that can interact together and with environmental factors. In other words, none of these numerous associated genetic variants are penetrant enough to lead to T2D alone. Twin or family studies showed that the estimates of the heritability of T2D range from 30 to 70%, depending on the age of diabetes onset and the glycemic status of cases (Almgren et al., 2011; Poulsen et al., 1999). These studies suggested a greater role of genetics in the first phases of development of T2D and a greater contribution of the environment to T2D risk in older patients, which was observed in other complex polygenic disorders like myocardial infarction (Rosamond et al., 2008).

Several recent studies, including genome-wide association studies (GWAS; see below), have fully confirmed the polygenic nature of T2D and demonstrated the modest, but true, role of genetic regions in T2D risk. The tangled picture of T2D genetics is difficult to understand for the diabetes research community, which creates misunderstandings with geneticists, and eventually limits both basic research and translational outcomes of these genetic discoveries. The present review aims to brighten the genetics of T2D so as to encourage integrated T2D modeling approaches from genetic defects to personalized medicine.

The Contribution of Frequent Genetic Variants to Common T2D Risk

Family Linkage Studies: No Major Gene in Common T2D

In contrast to GWAS mainly based on the comparison of unrelated cases and controls (see below), linkage studies looked for co-segregation between genetic markers and disease in multiplex pedigrees. Parametric analyses were very powerful when the mode of inheritance was well known, while non-parametric statistics were more liberal in their a priori mode of transmission assumptions. The hypothesis behind familial linkage in

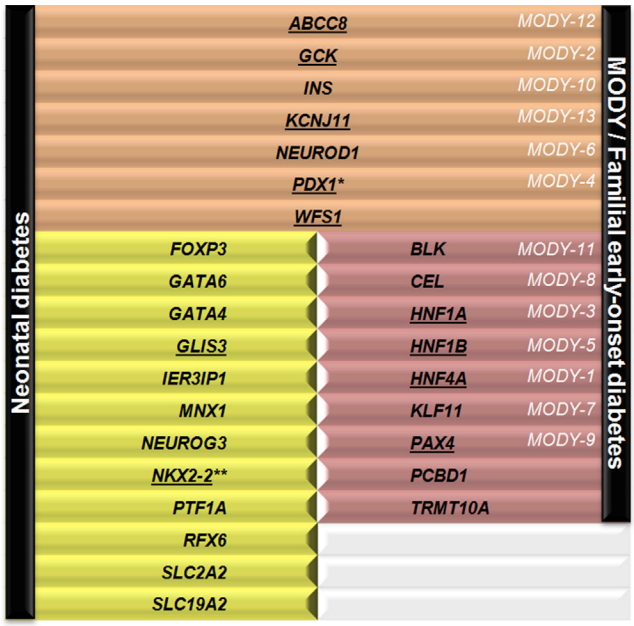

Figure 1. List of Genes Known to Harbor Mutations Causing Monogenic Forms of Diabetes: Neonatal Diabetes and/or MODY/Familial Early-Onset Diabetes

Underlined genes are also known to harbor common SNPs contributing to common T2D. MODY, maturity-onset diabetes of the young. *This gene has been found to harbor a rare (and not frequent) mutation associated with common T2D. **This gene has been found to harbor common SNPs associated with fasting glucose levels.

complex traits was the presence of major genes that are potent enough to cause disease under certain circumstances. Although the community put a great deal of effort into genome-wide familial linkage studies during 10 years, only a few T2D putative linked regions were identified through this strategy: *CAPN10* (Hanis et al., 1996), *ADIPOQ* (Vionnet et al., 2000), *HNF4A* (Silander et al., 2004), *ENPP1* (Meyre et al., 2005), and *TCF7L2* (Grant et al., 2006). Yet, only the associations of *HNF4A* and *TCF7L2* loci with T2D risk were subsequently replicated by GWAS analyses, casting doubt about the contribution of the others (Figure 2) (Kooner et al., 2011; Sladek et al., 2007). Importantly, the hypothesis of major genes involved in common T2D has not been demonstrated, as even *HNF4A* and *TCF7L2* single nucleotide polymorphisms (SNPs) were unable to explain the observed linkage.

The only big success of the pre-GWAS era was the identification of *TCF7L2* as a T2D-susceptibility gene. A common intronic SNP within *TCF7L2* has been repeatedly demonstrated to confer the strongest effect on T2D risk discovered so far, with an odds ratio (OR) roughly 1.50 (Grant et al., 2006). *TCF7L2* encodes a transcription factor involved in the Wnt signaling pathway, which is expressed in many tissues, including those with a key metabolic role (Nobrega, 2013). Though various cell or animal studies reported compelling evidence for a strong involvement of TCF7L2 into pancreatic beta cell mechanisms including insulin production and processing (Zhou et al., 2014), some reports also raised the possibility that the beta cell dysfunction related to TCF7L2 was indirect and was instead the consequence of disruptions in liver, brain, or gut (Boj et al., 2012; Shao et al., 2013). It remains that, in humans, the T2D risk allele in *TCF7L2* is markedly associated with a pancreatic phenotype evoking a primary islet dysfunction, and not with insulin resistance or with liver abnormalities (Saxena et al., 2010).

Candidate Genes: Too Much Noise for Very Few Advances

In the last 20 years, a plethora of biological candidate genes for T2D were reported to harbor common SNPs nominally associated with T2D risk. However, GWAS (that should be considered as the gold standard for common SNPs analyses) identified association signals that lie within only a few physiologically compelling T2D genes: *GCK* (März et al., 2004; Stone et al., 1996), *HNF1B* (Winckler et al., 2007), *WFS1* (Sandhu et al., 2007), *KCNJ11* (Hani et al., 1998), *PPARG* (Deeb et al., 1998), and *IRS1* (Almind et al., 1993) (Figure 2).

The studies of a putative role of frequent SNPs within *GCK*, *HNF1B*, and *WFS1* for common T2D logically followed the identification of the genetic etiology of monogenic forms of diabetes, including MODY-2, MODY-5, and the Wolfram syndrome, respectively (Figure 1). Interestingly, although homozygous *WFS1* mutations in consanguineous families cause the Wolfram syndrome (Inoue et al., 1998), it has been shown recently that rare heterozygous *WFS1* mutations can also lead to dominantly inherited isolated adult-onset T2D (Figure 1) (Bonnycastle et al., 2013). Therefore, there is a gradient of clinical severity depending on the functional effect and the state of deleterious mutations (i.e., homozygous or heterozygous) in *WFS1*, something that has also been found in other genes involved in monogenic diabetes, including *GCK*, *PDX1*, and *NEUROD1* (Vaxillaire et al., 2012).

The assessment of the susceptibility of *KCNJ11*, *PPARG*, and *IRS1* to T2D was based on their known involvement into pancreatic beta cell function or insulin resistance. *KCNJ11* encodes the pore-forming subunit KIR6.2 of the ATP-dependent potassium channel in pancreatic beta cells, which plays a pivotal role in insulin granule exocytosis (Hani et al., 1998). Rare heterozygous mutations in this gene were subsequently shown to cause monogenic neonatal diabetes mellitus (Gloyn et al., 2004) as well as MODY-13 (Bonnefond et al., 2012a) (Figure 1). *PPARG* encodes the nuclear receptor PPAR-γ, which plays a crucial role in adipocyte differentiation and function and is a well-known molecular target for thiazolidinedione insulin sensitizer to treat T2D (Deeb et al., 1998). Deleterious heterozygous dominant-negative mutations in *PPARG* were shown to cause monogenic severe insulin resistance with lypodystrophy and liver disease progressing to T2D at an early age (Barroso et al., 1999). *IRS1* encodes the insulin receptor substrate-1 that is necessary for insulin action in insulin-sensitive tissues and pancreatic beta cells (Almind et al., 1993). *IRS1* has been the only success story of an obvious candidate gene that was not responsible for monogenic diabetes too. However, the initial T2D-associated coding mutation in *IRS1* was not subsequently confirmed by GWAS, as the GWAS signal was actually located adjacent to the gene (Rung et al., 2009).

GWAS Legacy to T2D Research

Since GWAS were chosen as a Breakthrough of the Year 2007 by *Science*, they have generated many enthusiastic commentaries followed by excessive criticisms. Obviously, GWAS have brought groundbreaking advances in the genetics of common T2D, which was still poorly elucidated in the mid-2000s. This

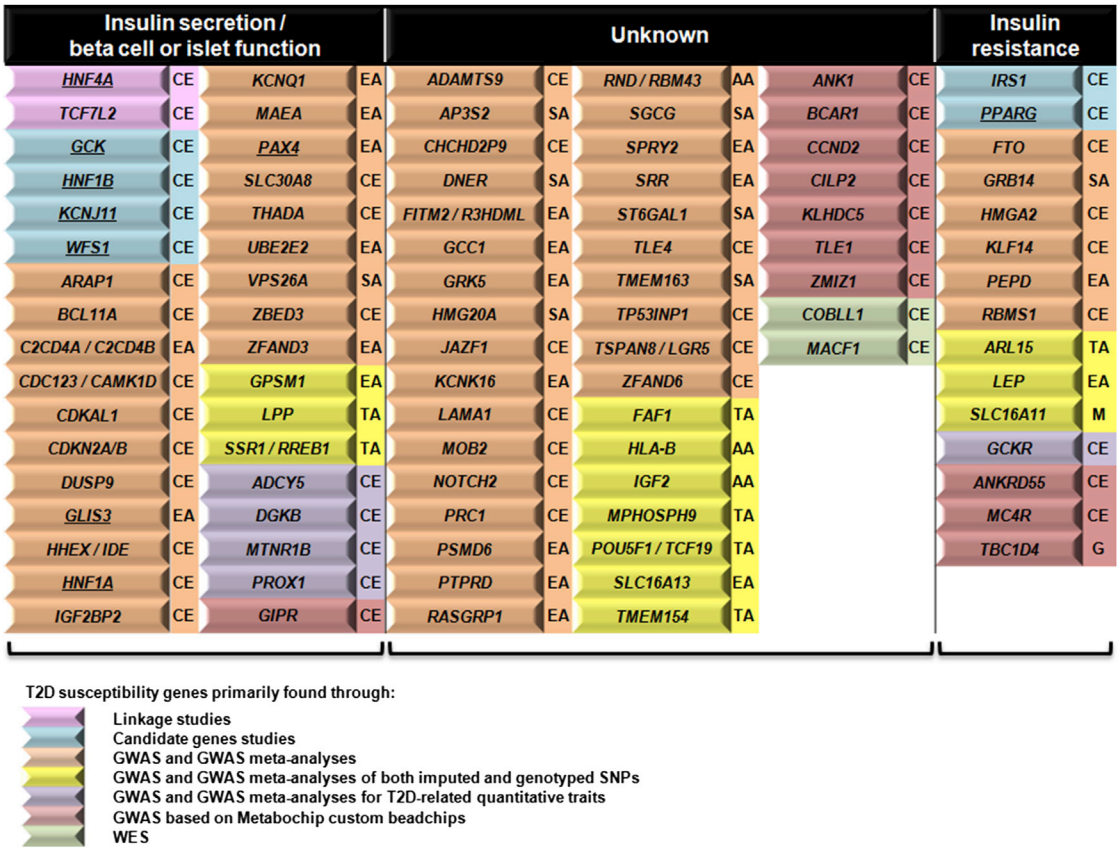

Figure 2. List of Loci Known to Harbor Common SNPs Contributing to Common T2D

Underlined loci are also known to harbor rare mutations causing monogenic forms of diabetes. Technologies used for the identification of T2D-suceptibility genes are shown below the figure. The T2D-related functions of genes have been set according to several publications (Mahajan et al., 2014; Dimas et al., 2014; Marullo et al., 2014; Moltke et al., 2014; Pasquali et al., 2014; Vassy et al., 2014). Moreover, T2D-suceptibility genes that are known to harbor rare mutations causing monogenic forms of diabetes have been classified according to their known function. The ethnicity of the population originally analyzed is also shown for each T2D-susceptibility gene. AA, African American; CE, Caucasian European; EA, East Asian; G, Greenlandic; GWAS, genome-wide association study; M, Mexican; SA, South Asian; T2D, type 2 diabetes; TA, trans-ancestry; WES, whole-exome sequencing.

success has been made possible by the Human Genome Project, subsequently exploited by HapMap for the identification and selection of tagSNPs, by a marked technological advance in the throughput for SNP genotyping through extremely reliable and inexpensive DNA microarray technology, and by robust statistical analyses enabling the careful assessment of hundreds of thousands of common SNPs in a large sample size of T2D cases and controls. Importantly, GWAS identifies disease-associated SNPs, which are conveniently named by the nearest genes; thus, it does not necessarily implicate the genes as causal.

The first T2D GWAS that analyzed normal-weight T2D individuals and normoglycemic controls of European origin demonstrated the leading contribution of *TCF7L2* to T2D and reported three new T2D-susceptibility genes: *SLC30A8* and *HHEX*, which were subsequently confirmed (Figure 2), and *EXT2-ALX4*, which has not been replicated (Sladek et al., 2007). This first study was quickly followed by several other GWAS papers that confirmed the previous data, and found new loci, most of them identified through an innovative merging strategy of data from different cohorts that was later named GWAS meta-analysis. The major goal of these meta-analyses was to optimize the identification of new signals by increasing the statistical power of the studies. This first wave of GWAS led to the identification of a dozen new T2D-associated genes in Europeans: *ADAMTS9*, *CDC123/CAMK1D*, *CDKAL1*, *CDKN2A/B*, *IGF2BP2*, *JAZF1*, *NOTCH2*, *RBMS1*, *THADA*, and *TSPAN8/LGR5* (Figure 2) (Saxena et al., 2007; Qi et al., 2010; Scott et al., 2007; Zeggini et al., 2007, 2008).

In parallel, three independent studies found an obesity-susceptibility gene that was renamed *FTO* (fat mass and obesity-associated protein) (Dina et al., 2007; Frayling et al., 2007; Scuteri et al., 2007). Frayling et al. (2007) initially discovered an association signal at *FTO* with T2D that disappeared after adjustment for BMI, suggesting an indirect effect of *FTO* on T2D risk, which was actually mediated by the excess of adiposity.

Although all these GWAS were performed in unrelated individuals, an Icelandic GWAS took into account the parental origin of transmitted alleles and identified a new T2D association signal in *MOB2* (Kong et al., 2009) (Figure 2).

A second wave of GWAS papers came from the linear analysis of T2D-related metabolic quantitative traits in non-diabetic individuals, which opposed the logistic analyses in T2D cases and controls. This strategy indirectly identified five new T2D-associated genes in Europeans: *MTNR1B*, *DGKB*, *PROX1*, and

ADCY5, which were primarily found to be associated with the variation of fasting glucose levels (Bouatia-Naji et al., 2009; Dupuis et al., 2010; Prokopenko et al., 2009), and *GCKR*, which was primarily found to be associated with the variation of fasting glucose levels, fasting insulin levels, and triglyceride levels (Saxena et al., 2007; Dupuis et al., 2010) (Figure 2). Interestingly, the overlap between loci influencing glucose- or insulin-related traits and T2D-susceptibility loci was unexpectedly limited (Dupuis et al., 2010). This result suggests that genes and related pathways that influence normal physiological levels of metabolic traits can be different from those leading to pathophysiological levels of metabolic traits that define T2D. A recent study strengthened this conclusion showing that the combination of established SNPs raising fasting glucose levels was significantly associated with the incidence of impaired fasting glucose levels over the 9-year follow-up of the study, but not with the risk of developing overt T2D (Vaxillaire et al., 2014).

In 2010, a very large meta-analysis including more than 50,000 participants found a dozen new T2D-susceptibility genes in Europeans (Figure 2) (Voight et al., 2010). Further stratification of T2D participants by BMI identified a new T2D association signal in *LAMA1* (Figure 2): a SNP at this locus associated with T2D risk in lean cases only (Perry et al., 2012).

In parallel, GWAS and meta-analyses of GWAS were performed in other ethnicities (i.e., East Asians, South Asians, African Americans) and generated multiple new T2D-associated genes (Figure 2) (Cho et al., 2012; Hanson et al., 2014; Kooner et al., 2011; Li et al., 2013; Ma et al., 2013; Palmer et al., 2012; Saxena et al., 2013; Shu et al., 2010; Tabassum et al., 2013; Tsai et al., 2010; Unoki et al., 2008; Yamauchi et al., 2010; Yasuda et al., 2008).

These meta-analyses have been limited by the fact that at best the last generation of SNP microarrays analyzes a few million common SNPs from more than 30 million SNPs discovered so far. To overcome this issue, GWAS data were imputed using whole-genome sequencing (WGS) data, mostly from the recent releases of HapMap or 1000 Genomes project (1000G). This strategy also allows the geneticists to fine-map established associations and identify the putatively causative SNPs from all SNPs that are present in a given associated DNA region to increase the power and/or to provide a uniform catalog of analyzed SNPs across studies within large meta-analyses. This strategy led to the identification of six new T2D susceptibility genes in Japanese individuals (Hara et al., 2014), in Mexicans and other Latin Americans (Williams et al., 2014), and in African Americans (Ng et al., 2014) (Figure 2). Lastly, several consortia performed a genome-wide *trans*-ancestry meta-analysis including more than 110,000 participants of various ancestries so as to boost the power and to improve the fine-mapping resolution of causal variants (Mahajan et al., 2014). Here, the imputation was mandatory to homogenize each set of SNPs between cohorts of different ancestry. They found seven new T2D susceptibility genes (Figure 2).

Finally, special inexpensive DNA microarrays have also been instrumental in finding additional T2D-associated genes. The Metabochip custom genotyping arrays were designed to follow up nominal (i.e., non-genome-wide significant) signals associated with cardiometabolic traits and associated diseases or to fine-map well-established associations (Voight et al., 2012). This staged approach led to the identification of 11 new T2D-susceptibility genes in Europeans (Morris et al., 2012) and in the Greenland population (Moltke et al., 2014) (Figure 2). Paradoxically, only half of the new signals were present in the Metabochip due to their originally nominal association with T2D (*ANK1*, *BCAR1*, *CCND2*, *TLE1*, and *ZMIZ1*); the others were initially associated with postprandial glucose levels during an oral glucose tolerance test (*GIPR*), glycated hemoglobin A1c (*KLHDC5*), blood pressure (*ANKRD55*), lipids (*CILP2*), BMI (*MC4R*), and waist-to-hip ratio (*TBC1D4*). These findings confirmed the high complexity of T2D genetic architecture and the pleiotropic effect of associated genes. Illumina has recently launched an exome array that consists of ~250,000 common and much rarer SNPs located in exonic regions that were found in genetic studies on a range of diseases including cardiometabolic disorders. This new beadchip will probably soon extend the list of T2D genes.

NGS, the Next-Generation Genetic Studies in T2D

Since 2009, the development of next-generation sequencing (NGS) has revolutionized the fields of genetics and genomics in many ways. NGS studies, mainly of exomes at this time, have brought insights in several rare Mendelian disorders, including neonatal diabetes and MODY (Bamshad et al., 2011), and a substantial number of NGS studies have pointed out additional association signals. In T2D, a first multi-stage study primarily driven by low-coverage whole-exome sequencing (WES) in ~2,000 Danes highlighted two coding SNPs significantly associated with T2D located in two new genes (Figure 2) (Albrechtsen et al., 2013). In the near future, by boosting the statistical power of NGS studies through the merge of multiple WES cohort data sets, international consortia have the potential to identify new T2D susceptibility exonic mutations with a putatively marked impact on coded protein function.

Do the Genetics of Common T2D Illuminate T2D Pathophysiology?

Despite the dramatic success of GWAS and GWAS meta-analyses (Figure 2), there is a substantial gap between the discovery of many T2D-associated SNPs and the understanding of how these SNPs physiologically impact T2D pathogenesis. So far, 80% of all T2D-associated SNPs are intergenic or intronic, and the geneticists have named the closest gene on the SNP chromosome as the T2D-susceptibility gene without having strong clues about the molecular link between the gene and the variant. An associated SNP can actually interact with a distant gene located on the same chromosome, as suggested for the obesity-associated SNPs in *FTO*, which would actually impact IRX3 (Ragvin et al., 2010; Smemo et al., 2014), or even located elsewhere in the genome. Worst, it may have no functional effect, as it only tags a linkage disequilibrium (LD) block (i.e., a DNA region transmitted together from one generation to the next one). Furthermore, the individual effect of these SNPs on T2D risk is typically modest; namely, each risk allele usually increases less than 15% the risk of developing T2D (OR < 1.15). As a consequence, all common T2D-associated SNPs have captured less than 15% of T2D familial aggregation (Morris et al., 2012). Furthermore, this has important consequences for experimental studies: it is unlikely that any of the frequent DNA mutations found to associate with polygenic T2D through GWAS are sufficient to cause a major phenotype in diabetes models, as the

current functional assays lack sensitivity for subtle effects. Occasionally, the mechanisms that causally link a common T2D-associated SNP to a gene and to T2D have been elucidated. For instance, an elegant study has identified clusters of active enhancers targeted by major human islet transcription factors, using a combination of formaldehyde-assisted isolation of regulatory elements (FAIRE) technique and chromatin immunoprecipitation (ChIP), both coupled with NGS (FAIRE-seq and ChIP-seq, respectively) (Pasquali et al., 2014). The authors demonstrated that these chromosome regions serving as super enhancers of the islet genome expression carry a large load of T2D-associated SNPs, pointing out the impact of these SNPs on islet functions. In particular, SNPs in or nearby *TCF7L2*, *ZFAND3*, *CDKN2A*, *C2CD4A*, *C2CD4B*, *SLC30A8*, and *DGKB* were shown to disrupt DNA binding and islet enhancer activity (Pasquali et al., 2014).

The predominance of T2D-associated genes presumably controlling pancreatic islet and beta cell function is supported by purely genetic epidemiology studies too. Indeed, the impact of known T2D-SNPs was shown to be markedly higher in normal-weight T2D cases compared to obese T2D cases (Cauchi et al., 2008; Perry et al., 2012). These findings emphasized that many T2D-associated SNPs with strong genetic contribution primarily affect pancreatic beta cell function. Furthermore, the extensive work done by the Meta-Analyses of Glucose and Insulin-related traits Consortium (MAGIC) using biological traits (e.g., glucose, insulin, or proinsulin levels) as quantitative traits allowed the partitioning of T2D-associated SNPs according to the putative effect of the T2D risk allele on beta cell function or insulin resistance in non-diabetic individuals (Dimas et al., 2014). From these studies, more than 30% of T2D-associated SNPs are likely to affect insulin secretion or beta cell function, while 15% of these SNPs may play a role in insulin resistance (Figure 2). A recent elegant study showed that the combination of SNPs that impact beta cell function significantly predicted incident T2D, but not the combination of SNPs contributing to insulin resistance (Vassy et al., 2014). The mechanisms by which the remaining T2D-associated SNPs (i.e., more than half of the T2D-associated SNPs) affect disease pathogenesis have not been determined by genetic epidemiology studies, and further functional studies are necessary.

Taking advantage of the quick and recent progress of NGS tools, research groups have started studying the putative contribution of rare coding variants, hoping to reveal a stronger effect on the disease, with the goal to functionally investigate them and draw clear conclusions on the mutations, the genes, and the disease.

The Contribution of Rare or Low-Frequency Genetic Variants to Common T2D

Niche of T2D-Associated Rare Variants in GWAS-Identified T2D Genes

As said above, association does not mean causality. The identification of true associations between SNPs within or nearby genes and T2D does not necessarily imply that these SNPs or other variants in the same regions have a direct etiological effect on T2D. Furthermore, GWAS results can sometimes be totally misleading, as shown for the genetic link between *HK1* (encoding hexokinase 1) and glycated hemoglobin A1c: although *HK1* locus showed the strongest association with this marker of glucose control, a more careful analysis revealed that *HK1* association signal was unrelated to glucose homeostasis, but instead involved in blood red cell function (Bonnefond et al., 2009). The association with glycated hemoglobin A1c was genuine, though the initial explanation linking this enzyme with T2D was later proven wrong. A way to establish causality beyond GWAS is to identify rare coding mutations in the gene located nearby the most associated SNP, which clearly impairs protein function and modulates T2D pathophysiology. Notably, the genetic field arbitrarily defines variants with a frequency below 1% as rare, while low-frequency variants have a frequency of 1%–5%, and frequent SNPs have a minor allele frequency (MAF) above 5%.

In 2012, a causal link was demonstrated between T2D risk and the dysfunction of the melatonin receptor 1B (*MTNR1B*) due to rare loss-of-function variants (Figure 3). *MTNR1B* encodes a $G\alpha_i$-protein-coupled receptor (GPCR) and may be a promising drug target. The study combined large-scale *MTNR1B* resequencing with a systematic four-tier cell-based investigation of each identified mutation, followed by a replication step of the mutations with the most deleterious effect in additional individuals (Figure 4) (Bonnefond et al., 2012b). The two coding exons of *MTNR1B* were sequenced in 7,632 Europeans, including 2,186 T2D individuals. 40 non-synonymous mutations were identified, including 36 very rare mutations with a frequency below 0.1%, which were strongly associated with increased T2D risk (with an OR of 3.31), while the mutations with a frequency above 0.1% did not significantly contribute to T2D (Bonnefond et al., 2012b). The 40 mutations were then investigated at four levels in human cell lines: four mutants were unable to bind melatonin and to activate downstream pathways, while 10 other mutants were unable to stimulate ERK phosphorylation or activate G_i protein in the presence of melatonin (Figure 5) (Bonnefond et al., 2012b). The aggregation of these loss-of-function variants with a frequency below 0.1% yielded a T2D risk increased by greater than 5.5-fold, while the neutral very rare variants did not have any effect on T2D (Bonnefond et al., 2012b). This study is important as it established a mechanistic link between the presence of human mutations in a GWAS-identified gene and T2D risk. Since melatonin is a neurohormone that modulates circadian rhythms in the body in response to daylight, this study implies that melatonin receptor agonists, already prescribed for depression and sleep disorders, may be useful for T2D treatment (Bonnefond et al., 2012b).

More recently, and using the same kind of strategy (without a final replication step, however) (Figure 4), a study demonstrated that rare loss-of-function variants in the nuclear receptor encoded by *PPARG* increased T2D risk (Figure 3) (Majithia et al., 2014). Coding exons of *PPARG* were sequenced in ~20,000 multiethnic T2D cases/control samples as part of several candidate gene sequencing or WES studies including several consortia (Majithia et al., 2014). The authors identified a total of 52 rare non-synonymous *PPARG* variants that were not associated with T2D, even after variant stratification according to the in silico prediction of their putative functional effect (Majithia et al., 2014). Through high-throughput assays allowing the quantification of adipocyte differentiation in response to each exogenous PPAR-γ mutant, the authors found that only 12 mutations

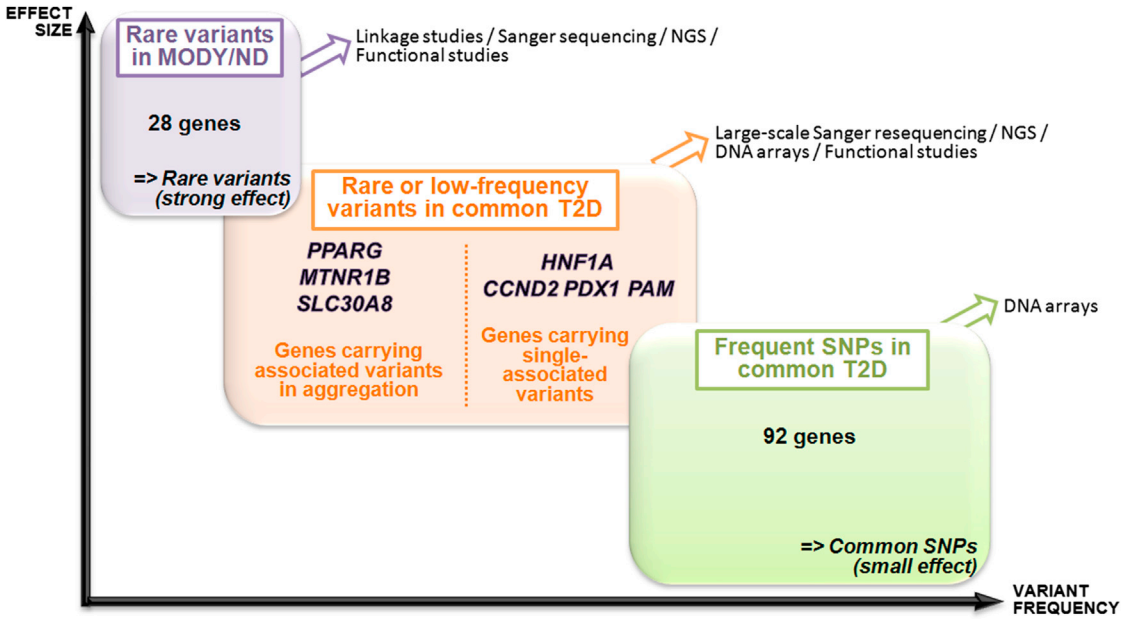

Figure 3. Distribution of Both Effect Size and Frequency of Variants/Mutations Contributing to Monogenic Forms of Diabetes or to Common T2D

The effect size of genetic variants in diabetes-associated genes is reported on the x axis, while the frequency of these variants is reported on the y axis. The arrows show the technologies/experiments used for the identification of causal or associated variants in each group of variants (purple, rare variants involved in monogenic diabetes; orange, rare or low-frequency variants involved in common T2D; green, frequent SNPs involved in common T2D). The rectangles show the number or the name of genes that carry rare variants involved in monogenic diabetes (purple), rare or low-frequency variants involved in common T2D (orange), or frequent SNPs involved in common T2D (green). MODY, maturity-onset diabetes of the young; ND, neonatal diabetes; NGS, next-generation sequencing; SNP, single nucleotide polymorphism; T2D, type 2 diabetes.

significantly reduced the stimulation of adipocyte differentiation, which significantly yielded in aggregation an increased T2D risk of greater than 7-fold, while the combined neutral variants did not have any effect on T2D (Majithia et al., 2014). The carriers of loss-of-function variants in *PPARG* did not present with extreme insulin resistance phenotypes (Majithia et al., 2014), as originally observed in families with severe lipodystrophy (Barroso et al., 1999), although three of these deleterious variants had already been reported in such families. Therefore, the penetrance and the phenotypic expressivity of loss-of-function *PPARG* mutations are variable.

Another GWAS-identified T2D gene was found to harbor rare mutations associated with T2D: *SLC30A8* (Figure 3) (Flannick et al., 2014). *SLC30A8* encodes a zinc transporter that is located in the insulin granules in pancreatic beta cells. The study was primarily driven by NGS targeting specifically 115 genes that lay either within GWAS-identified T2D-associated loci or genes containing rare mutations known to cause monogenic diabetes, in 758 Europeans. The authors pointed out a nonsense mutation in *SLC30A8*, which was more present in controls than in individuals with T2D (Flannick et al., 2014). As the p value of the protection of this mutation against T2D was only nominal, they sought additional evidence through (1) the genotyping of this mutation in ~50,000 Europeans, which replicated the protection of the nonsense variant against T2D; (2) WGS and imputation in 71,000 Icelanders, which identified a frameshift mutation that was enriched in controls compared to T2D cases; (3) the genotyping of this frameshift variant in 5,000 Norwegians, which was only found in controls; (4) the functional assays, which showed that the overexpression of both nonsense and frameshift SLC30A8 mutants markedly decreased SLC30A8 expression; and (5) WES in 12,000 individuals from various ethnicities, which identified 10 additional rare *SLC30A8* variants that protect against T2D when analyzed in aggregation. The meta-analysis of all these studies showed a significant protection of rare *SLC30A8* variants against T2D with an OR of 0.34 (Flannick et al., 2014). Unfortunately, the functional studies were only performed for 2 out of 12 identified mutations. Additional functional investigations of human variants in different models would be useful to understand the precise molecular mechanisms of SLC30A8 dysfunction underlying the unexpected protective effects against T2D. Indeed, a recent comprehensive review pointed out conflicting data from animal and cellular studies on the zinc transporter SLC30A8 role in insulin secretion, and more genetic studies would help to bring definite responses (Davidson et al., 2014).

A study has recently identified a low-frequency loss-of-function variant in *HNF1A* encoding the homeodomain-containing transcription factor HNF-1α, which strongly increased T2D risk in Mexicans and Latin Americans (Figure 3) (Estrada et al., 2014). Through WES in 3,756 individuals of Mexican origin, the study found that the *HNF1A* missense variant (p.E508K, with a frequency of ~0.2% in controls) yielded an increased T2D risk of about 5-fold. This result was replicated in more than 3,000 Mexicans. Rare mutations in *HNF1A* had already been known to cause MODY-3 (Figure 1) (Vaxillaire et al., 1995). Here, the authors demonstrated that p.E508K mutation impacted the HNF-1α transactivation activity, although to a

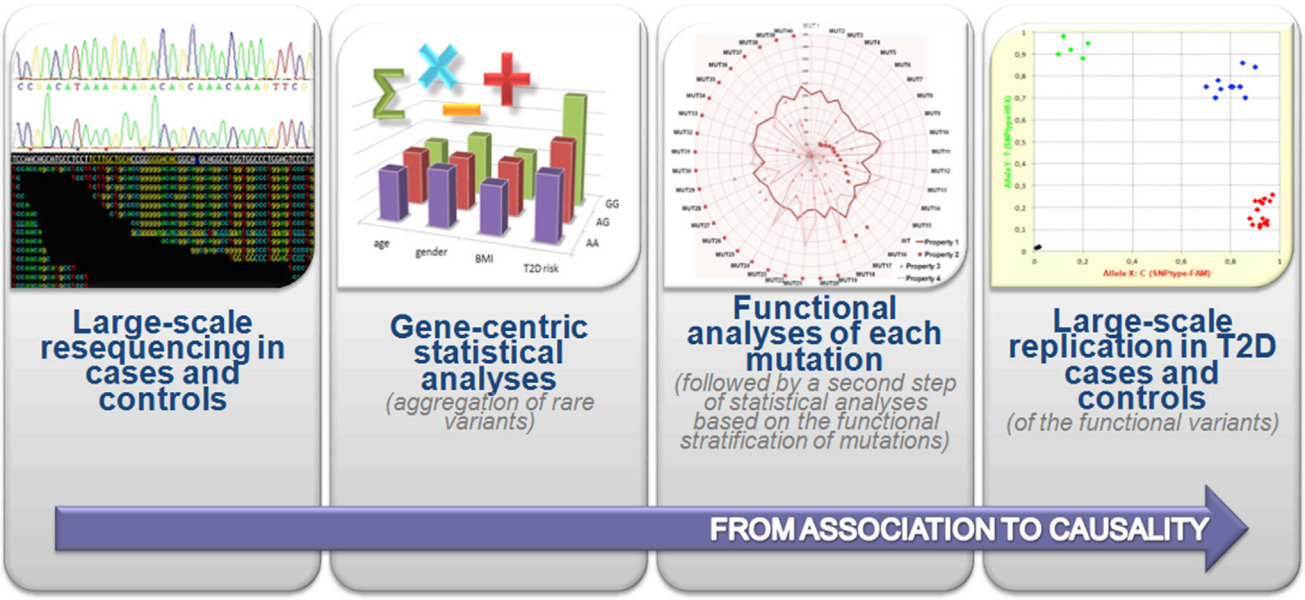

Figure 4. Typical Experimental Design to Draw a Causal Link between Rare Variants and T2D
This figure shows how a causal link can be established between rare variants and T2D. The first step is a large-scale resequencing of genes (or even WES or WGS) in cases and controls. The second step is to look for a significant enrichment in cases of rare variants in aggregation (in each gene). When a suggestive enrichment of rare variants within a gene is identified in cases, the third step is the analysis of functional consequences of each variant using various models, so as to know whether the functional variants (and not the neutral ones) are significantly enriched in cases. A final step of replication can be performed to confirm the association of functional variants with disease risk in additional individuals. T2D, type 2 diabetes.

lesser extent than three mutants known to cause MODY-3. Furthermore, compared with wild-type HNF-1α, the p.E508K mutant equally bound to DNA, but its localization was not bordered to the nucleus. The age at onset of T2D in the carriers of p.E508K was similar in other T2D Mexican individuals. Importantly, this mutation had previously been reported in only two MODY-3 European families (Bellanné-Chantelot et al., 2008; Forlani et al., 2010) and in one isolated T2D European patient (Flannick et al., 2013). Therefore, this mutation seems very rare but more penetrant in the European ethnicity. Similar striking ethnic contrast in mutation penetrance and expressivity was previously observed for heterozygous *MC4R* mutations that are less penetrant for obesity in Pakistan than in Europe (Saeed et al., 2012). Importantly, the identification of *HNF1A* p.E508K in T2D patients may have clinical implications toward personalized diabetic medicine, as it has been evidenced that MODY-3 patients treated with oral sulfonylureas show a better glycemic control when compared with those treated with insulin in the long term (Shepherd et al., 2003), and they may be good responders to GLP-1 analogs (Østoft et al., 2014).

HNF1A is not the only gene harboring rare loss-of-function mutations causing a spectrum of diabetes-related phenotypes. Several mutations in *INS*, *KCNJ11*, or *ABCC8* can cause neonatal diabetes, MODY, common T2D, mild hyperglycemia, or even associate with apparent normal glucose tolerance in individuals sharing the same allele (Bonnefond et al., 2012a; Meur et al., 2010; Riveline et al., 2012). Most of these specific mutations were proven to be deleterious. Indeed, diabetic carriers of a mutation in *KCNJ11* or *ABCC8* (which encode subunits of ATP-dependent potassium channel in pancreatic beta cells) can optimally be treated with oral sulfonylureas (Pearson et al., 2006), and the switch from insulin to oral sulfonylureas in diabetic carriers of these specific mutations able to cause a spectrum of diabetes-related phenotypes was actually very successful (Bonnefond et al., 2012a; Riveline et al., 2012).

A lesson from genetic studies is that mutations that have been found to cause monogenic diabetes are not totally penetrant. In this respect, a study has reported that nearly 1% of the general population carried rare nonsynonymous variants in a MODY gene, which had been previously identified in MODY probands or were likely to be deleterious according to in silico functional prediction (Flannick et al., 2013). Actually, prediction algorithms should always be considered with healthy skepticism, as they often bring erroneous conclusions (Bonnefond et al., 2012b, 2013; Majithia et al., 2014). The identification of a rare MODY mutation in a single patient with a putative MODY phenotype is not sufficient to establish causality. Ideally the causal link should be reached through the study of the whole MODY family, cell-based or animal-based functional studies, or a successful switch from insulin therapy to oral sulfonylureas in MODY patients with a mutation in *HNF1A*, *KCNJ11*, or *ABCC8*. It would be erroneous to underestimate the deleterious impact of true monogenic diabetes mutations, as they have been rarely detected in general population studies that are usually poorly phenotyped. The main conclusion is the lack of true clinical interest for sequencing the genome of healthy individuals, apart from having catalogs of human genetic diversity. Even if putative loss-of-function mutations are found through NGS, there is no way to assess their physiological relevance for the future of the healthy individual. In contrast, generalizing NGS in patients with serious disorders, including diabetes, is probably relevant for patients' stratification and for precision medicine.

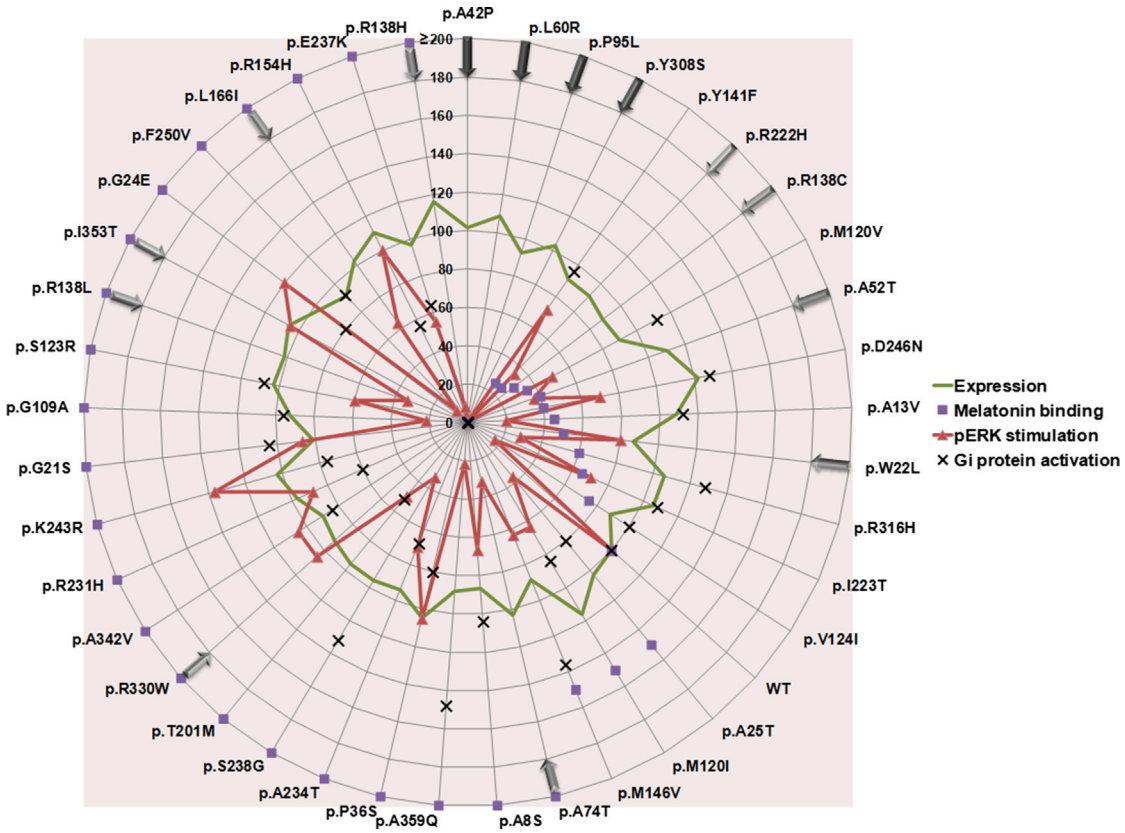

Figure 5. Four-Tier Functional Investigation of Each Mutation Identified in *MTNR1B*

40 mutations were investigated at four levels in human cell lines: expression of each mutant at the cell membrane (green line), binding of the melatonin in each mutant (purple squares), and melatonin-dependent activation of G_i protein (black crosses) or ERK (red triangles) pathway by each mutant. Black arrows indicate total loss-of-function mutants (i.e., unable to bind the melatonin), while gray arrows indicate partial loss-of-function mutants (i.e., unable to activate downstream pathways). The unit is arbitrary.

Rare or Low-Frequency Variants Associated with T2D Risk in Novel T2D Genes

Using WGS of 2,630 Icelanders and imputation into 11,000 cases and 267,000 controls of Icelandic origin, followed by a replication step in Danish and Iranian T2D case-control samples, a study has identified low-frequency or rare variants independently contributing to T2D risk in two novel common T2D-susceptibility genes (*CCND2*, *PAM*) and in one MODY gene (*PDX1*) (Figure 3) (Steinthorsdottir et al., 2014). In *CCND2*, a low-frequency intronic variant significantly protected against T2D (OR ~0.5), increased insulin release from pancreatic beta cells in nondiabetic participants, and was associated with increased *CCND2* expression in white blood cells and adipocytes (Steinthorsdottir et al., 2014). *CCND2* encodes cyclin D2 that regulates G1/S cell-cycle transition in multiple cell types including pancreatic beta cells (Steinthorsdottir et al., 2014). In *PAM*, one low-frequency and one rare missense variant were shown to moderately increase T2D risk (OR < 1.5) (Steinthorsdottir et al., 2014). The rare variant was also associated with reduced insulin release from pancreatic beta cells (Steinthorsdottir et al., 2014). *PAM* encodes peptidylglycine α-amidating monooxygenase, which plays a crucial role in post-translational modifications of multiple peptides. In *PDX1*, a rare frameshift variant strongly increased risk of T2D (OR ~2.3) (Steinthorsdottir et al., 2014). *PDX1* encodes a transcription factor with a key role in pancreatic beta cell development, and mutations in this gene cause MODY-4 or neonatal diabetes associated with pancreatic agenesis when mutations are homozygous (Figure 1) (Vaxillaire et al., 2012). *PDX1* enriches the list of genes that can harbor rare mutations causing a spectrum of diabetes subtypes like *HNF1A*, *ABCC8*, *KCNJ11*, *PPARG*, *INS*, and *WFS1* (see above). A limitation of this study, however, remains the lack of functional experiments demonstrating the causality of these rare variants (Steinthorsdottir et al., 2014).

Conclusions and Future Directions

GWAS and GWAS meta-analyses have by far been the most efficient way to identify new T2D genes (Figure 2), but their predictive value for future occurrence of T2D has been very limited compared to classic risk factors such as obesity and fasting glucose levels (Walford et al., 2014). Although it might be good news that our genome does not fully dictate our future, the knowledge of its specificities may help us to improve our health. Early genetic studies showed that the higher risk for T2D conferred by *TCF7L2* variant can be reversed by lifestyle intervention (Florez et al., 2006), opening avenues for strategies targeted on genetically selected individuals with pre-diabetes. *TCF7L2* has also been shown to be associated with a lower

efficiency of oral sulfonylureas in newly diagnosed T2D patients (Pearson et al., 2007), but a more recent Danish study suggested that in contrast to clinical markers, all known T2D-associated variants do not significantly affect the time to prescription of the first drug after disease onset (Hornbak et al., 2014). In other words, frequent SNPs are not helpful to predict patients' futures, though the good use of genetic data may contribute to provide better care to newly diagnosed T2D patients who are currently all treated the same (with metformin).

To be fair, the T2D missing heritability may explain part of the weak predictive power of current T2D SNPs. The ''black matter'' issue has raised much interest within geneticists. Many explanations have been proposed, including the putative overestimation of T2D heritability (Groop and Pociot, 2014). Genetic or epigenetic mechanisms other than SNP or mutation may also contribute to T2D genetic risk. For instance, constitutional chromosome variations, called gene copy-number variants (CNVs) have been found to significantly contribute to obesity risk (Falchi et al., 2014; Walters et al., 2010), but not to T2D risk so far.

The biggest opportunity for T2D genetics may be in assisting experimental biologists and physiologists to establish the most important pathways that are causatively associated with T2D in humans, so as to design efficient drug therapies. Functional genomics tools should be systematically used to bring clues about the biological role of newly discovered genes (Bonnefond et al., 2012b; Majithia et al., 2014) and also to validate the impact of variants that are not all deleterious, as shown above in contrast to generous algorithm predictions. The rapidly emerging induced pluripotent stem cell (iPSC) biology opens unexplored avenues in T2D modeling in the own patients' cells (or cells from controls with causal mutations introduced by genome editing), which could be derived into pancreatic beta cells, hepatocytes, adipocytes, or muscle cells (Inoue et al., 2014). Notably, the use of iPSCs is expected to be very useful in drug discovery and in clinical trials, enabling the assessment of drug responsiveness in each patient (drug responders versus non-responders) through the analysis of the drug efficiency on iPSC-derived cells (Inoue et al., 2014).

T2D human genetics has other potentials; for instance, NGS large screen for mutations in new putative drug targets can be very helpful in validating them (for example, if causing insulin secretion defects in beta cells) and in determining drugs side effect in humans carrying loss- or gain-of-function mutations. T2D genetics has shown biologists the results of the experiments of nature that have mutated genes and non-coding regions, with direct effects on T2D risk. It revealed in human the role of key pathways involved in glucose homeostasis, opening novel therapeutic avenues. As modern genomics is not biased by prior researchers' hypotheses, but aims to assess all genome influences comprehensively, it brings unique integrated information that deserves to be tested by experimental biologists in animal models and cell lines. Biologists ought not to fear the plethora of T2D genes. On the contrary, together with geneticists, they should formulate innovative strategies for exploiting genetic discoveries in order to answer the following question in humans: among the numerous described metabolic pathways, what is essential to glucose control? What is causing early diabetic abnormalities? And what should be targeted to prevent or even cure T2D?

REFERENCES

Albrechtsen, A., Grarup, N., Li, Y., Sparsø, T., Tian, G., Cao, H., Jiang, T., Kim, S.Y., Korneliussen, T., Li, Q., et al.; D.E.S.I.R. Study Group; DIAGRAM Consortium (2013). Exome sequencing-driven discovery of coding polymorphisms associated with common metabolic phenotypes. Diabetologia *56*, 298–310.

Almgren, P., Lehtovirta, M., Isomaa, B., Sarelin, L., Taskinen, M.R., Lyssenko, V., Tuomi, T., and Groop, L.; Botnia Study Group (2011). Heritability and familiality of type 2 diabetes and related quantitative traits in the Botnia Study. Diabetologia *54*, 2811–2819.

Almind, K., Bjørbaek, C., Vestergaard, H., Hansen, T., Echwald, S., and Pedersen, O. (1993). Aminoacid polymorphisms of insulin receptor substrate-1 in non-insulin-dependent diabetes mellitus. Lancet *342*, 828–832.

American Diabetes Association (2013). Diagnosis and classification of diabetes mellitus. Diabetes Care *36* (1), S67–S74.

Bamshad, M.J., Ng, S.B., Bigham, A.W., Tabor, H.K., Emond, M.J., Nickerson, D.A., and Shendure, J. (2011). Exome sequencing as a tool for Mendelian disease gene discovery. Nat. Rev. Genet. *12*, 745–755.

Barroso, I., Gurnell, M., Crowley, V.E., Agostini, M., Schwabe, J.W., Soos, M.A., Maslen, G.L., Williams, T.D., Lewis, H., Schafer, A.J., et al. (1999). Dominant negative mutations in human PPARgamma associated with severe insulin resistance, diabetes mellitus and hypertension. Nature *402*, 880–883.

Bellanné-Chantelot, C., Carette, C., Riveline, J.-P., Valéro, R., Gautier, J.-F., Larger, E., Reznik, Y., Ducluzeau, P.-H., Sola, A., Hartemann-Heurtier, A., et al. (2008). The type and the position of HNF1A mutation modulate age at diagnosis of diabetes in patients with maturity-onset diabetes of the young (MODY)-3. Diabetes *57*, 503–508.

Boj, S.F., van Es, J.H., Huch, M., Li, V.S.W., José, A., Hatzis, P., Mokry, M., Haegebarth, A., van den Born, M., Chambon, P., et al. (2012). Diabetes risk gene and Wnt effector Tcf7l2/TCF4 controls hepatic response to perinatal and adult metabolic demand. Cell *151*, 1595–1607.

Bonnefond, A., Vaxillaire, M., Labrune, Y., Lecoeur, C., Chèvre, J.-C., Bouatia-Naji, N., Cauchi, S., Balkau, B., Marre, M., Tichet, J., et al. (2009). Genetic variant in HK1 is associated with a proanemic state and A1C but not other glycemic control-related traits. Diabetes *58*, 2687–2697.

Bonnefond, A., Philippe, J., Durand, E., Dechaume, A., Huyvaert, M., Montagne, L., Marre, M., Balkau, B., Fajardy, I., Vambergue, A., et al. (2012a). Whole-exome sequencing and high throughput genotyping identified KCNJ11 as the thirteenth MODY gene. PLoS ONE *7*, e37423.

Bonnefond, A., Clément, N., Fawcett, K., Yengo, L., Vaillant, E., Guillaume, J.-L., Dechaume, A., Payne, F., Roussel, R., Czernichow, S., et al.; Meta-Analysis of Glucose and Insulin-Related Traits Consortium (MAGIC) (2012b). Rare MTNR1B variants impairing melatonin receptor 1B function contribute to type 2 diabetes. Nat. Genet. *44*, 297–301.

Bonnefond, A., Raimondo, A., Stutzmann, F., Ghoussaini, M., Ramachandrappa, S., Bersten, D.C., Durand, E., Vatin, V., Balkau, B., Lantieri, O., et al. (2013). Loss-of-function mutations in SIM1 contribute to obesity and Prader-Willi-like features. J. Clin. Invest. *123*, 3037–3041.

Bonnycastle, L.L., Chines, P.S., Hara, T., Huyghe, J.R., Swift, A.J., Heikinheimo, P., Mahadevan, J., Peltonen, S., Huopio, H., Nuutila, P., et al. (2013). Autosomal dominant diabetes arising from a Wolfram syndrome 1 mutation. Diabetes *62*, 3943–3950.

Bottazzo, G.F., Florin-Christensen, A., and Doniach, D. (1974). Islet-cell antibodies in diabetes mellitus with autoimmune polyendocrine deficiencies. Lancet *2*, 1279–1283.

Bouatia-Naji, N., Bonnefond, A., Cavalcanti-Proença, C., Sparsø, T., Holmkvist, J., Marchand, M., Delplanque, J., Lobbens, S., Rocheleau, G., Durand, E., et al. (2009). A variant near MTNR1B is associated with increased fasting plasma glucose levels and type 2 diabetes risk. Nat. Genet. *41*, 89–94.

Cauchi, S., Nead, K.T., Choquet, H., Horber, F., Potoczna, N., Balkau, B., Marre, M., Charpentier, G., Froguel, P., and Meyre, D. (2008). The genetic susceptibility to type 2 diabetes may be modulated by obesity status: implications for association studies. BMC Med. Genet. *9*, 45.

Cho, Y.S., Chen, C.-H., Hu, C., Long, J., Ong, R.T.H., Sim, X., Takeuchi, F., Wu, Y., Go, M.J., Yamauchi, T., et al.; DIAGRAM Consortium; MuTHER Consortium

(2012). Meta-analysis of genome-wide association studies identifies eight new loci for type 2 diabetes in east Asians. Nat. Genet. *44*, 67–72.

Davidson, H.W., Wenzlau, J.M., and O'Brien, R.M. (2014). Zinc transporter 8 (ZnT8) and β cell function. Trends Endocrinol. Metab. *25*, 415–424.

Deeb, S.S., Fajas, L., Nemoto, M., Pihlajamäki, J., Mykkänen, L., Kuusisto, J., Laakso, M., Fujimoto, W., and Auwerx, J. (1998). A Pro12Ala substitution in PPARgamma2 associated with decreased receptor activity, lower body mass index and improved insulin sensitivity. Nat. Genet. *20*, 284–287.

Dimas, A.S., Lagou, V., Barker, A., Knowles, J.W., Mägi, R., Hivert, M.-F., Benazzo, A., Rybin, D., Jackson, A.U., Stringham, H.M., et al.; MAGIC Investigators (2014). Impact of type 2 diabetes susceptibility variants on quantitative glycemic traits reveals mechanistic heterogeneity. Diabetes *63*, 2158–2171.

Dina, C., Meyre, D., Gallina, S., Durand, E., Körner, A., Jacobson, P., Carlsson, L.M.S., Kiess, W., Vatin, V., Lecoeur, C., et al. (2007). Variation in FTO contributes to childhood obesity and severe adult obesity. Nat. Genet. *39*, 724–726.

Dupuis, J., Langenberg, C., Prokopenko, I., Saxena, R., Soranzo, N., Jackson, A.U., Wheeler, E., Glazer, N.L., Bouatia-Naji, N., Gloyn, A.L., et al.; DIAGRAM Consortium; GIANT Consortium; Global BPgen Consortium; Anders Hamsten on behalf of Procardis Consortium; MAGIC investigators (2010). New genetic loci implicated in fasting glucose homeostasis and their impact on type 2 diabetes risk. Nat. Genet. *42*, 105–116.

Estrada, K., Aukrust, I., Bjørkhaug, L., Burtt, N.P., Mercader, J.M., García-Ortiz, H., Huerta-Chagoya, A., Moreno-Macías, H., Walford, G., Flannick, J., et al.; SIGMA Type 2 Diabetes Consortium (2014). Association of a low-frequency variant in HNF1A with type 2 diabetes in a Latino population. JAMA *311*, 2305–2314.

Falchi, M., El-Sayed Moustafa, J.S., Takousis, P., Pesce, F., Bonnefond, A., Andersson-Assarsson, J.C., Sudmant, P.H., Dorajoo, R., Al-Shafai, M.N., Bottolo, L., et al. (2014). Low copy number of the salivary amylase gene predisposes to obesity. Nat. Genet. *46*, 492–497.

Flannick, J., Beer, N.L., Bick, A.G., Agarwala, V., Molnes, J., Gupta, N., Burtt, N.P., Florez, J.C., Meigs, J.B., Taylor, H., et al. (2013). Assessing the phenotypic effects in the general population of rare variants in genes for a dominant Mendelian form of diabetes. Nat. Genet. *45*, 1380–1385.

Flannick, J., Thorleifsson, G., Beer, N.L., Jacobs, S.B.R., Grarup, N., Burtt, N.P., Mahajan, A., Fuchsberger, C., Atzmon, G., Benediktsson, R., et al.; Go-T2D Consortium; T2D-GENES Consortium (2014). Loss-of-function mutations in SLC30A8 protect against type 2 diabetes. Nat. Genet. *46*, 357–363.

Florez, J.C., Jablonski, K.A., Bayley, N., Pollin, T.I., de Bakker, P.I.W., Shuldiner, A.R., Knowler, W.C., Nathan, D.M., and Altshuler, D.; Diabetes Prevention Program Research Group (2006). TCF7L2 polymorphisms and progression to diabetes in the Diabetes Prevention Program. N. Engl. J. Med. *355*, 241–250.

Forlani, G., Zucchini, S., Di Rocco, A., Di Luzio, R., Scipione, M., Marasco, E., Romeo, G., Marchesini, G., and Mantovani, V. (2010). Double heterozygous mutations involving both HNF1A/MODY3 and HNF4A/MODY1 genes: a case report. Diabetes Care *33*, 2336–2338.

Frayling, T.M., Timpson, N.J., Weedon, M.N., Zeggini, E., Freathy, R.M., Lindgren, C.M., Perry, J.R.B., Elliott, K.S., Lango, H., Rayner, N.W., et al. (2007). A common variant in the FTO gene is associated with body mass index and predisposes to childhood and adult obesity. Science *316*, 889–894.

Froguel, P., Vaxillaire, M., Sun, F., Velho, G., Zouali, H., Butel, M.O., Lesage, S., Vionnet, N., Clément, K., Fougerousse, F., et al. (1992). Close linkage of glucokinase locus on chromosome 7p to early-onset non-insulin-dependent diabetes mellitus. Nature *356*, 162–164.

Gloyn, A.L., Pearson, E.R., Antcliff, J.F., Proks, P., Bruining, G.J., Slingerland, A.S., Howard, N., Srinivasan, S., Silva, J.M.C.L., Molnes, J., et al. (2004). Activating mutations in the gene encoding the ATP-sensitive potassium-channel subunit Kir6.2 and permanent neonatal diabetes. N. Engl. J. Med. *350*, 1838–1849.

Grant, S.F.A., Thorleifsson, G., Reynisdottir, I., Benediktsson, R., Manolescu, A., Sainz, J., Helgason, A., Stefansson, H., Emilsson, V., Helgadottir, A., et al. (2006). Variant of transcription factor 7-like 2 (TCF7L2) gene confers risk of type 2 diabetes. Nat. Genet. *38*, 320–323.

Groop, L., and Pociot, F. (2014). Genetics of diabetes—are we missing the genes or the disease? Mol. Cell. Endocrinol. *382*, 726–739.

Hani, E.H., Boutin, P., Durand, E., Inoue, H., Permutt, M.A., Velho, G., and Froguel, P. (1998). 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 *41*, 1511–1515.

Hanis, C.L., Boerwinkle, E., Chakraborty, R., Ellsworth, D.L., Concannon, P., Stirling, B., Morrison, V.A., Wapelhorst, B., Spielman, R.S., Gogolin-Ewens, K.J., et al. (1996). A genome-wide search for human non-insulin-dependent (type 2) diabetes genes reveals a major susceptibility locus on chromosome 2. Nat. Genet. *13*, 161–166.

Hanson, R.L., Muller, Y.L., Kobes, S., Guo, T., Bian, L., Ossowski, V., Wiedrich, K., Sutherland, J., Wiedrich, C., Mahkee, D., et al. (2014). A genome-wide association study in American Indians implicates DNER as a susceptibility locus for type 2 diabetes. Diabetes *63*, 369–376.

Hara, K., Fujita, H., Johnson, T.A., Yamauchi, T., Yasuda, K., Horikoshi, M., Peng, C., Hu, C., Ma, R.C.W., Imamura, M., et al.; DIAGRAM consortium (2014). Genome-wide association study identifies three novel loci for type 2 diabetes. Hum. Mol. Genet. *23*, 239–246.

Hornbak, M., Allin, K.H., Jensen, M.L., Lau, C.J., Witte, D., Jørgensen, M.E., Sandbæk, A., Lauritzen, T., Andersson, Å., Pedersen, O., and Hansen, T. (2014). A combined analysis of 48 type 2 diabetes genetic risk variants shows no discriminative value to predict time to first prescription of a glucose lowering drug in Danish patients with screen detected type 2 diabetes. PLoS ONE *9*, e104837.

Inoue, H., Tanizawa, Y., Wasson, J., Behn, P., Kalidas, K., Bernal-Mizrachi, E., Mueckler, M., Marshall, H., Donis-Keller, H., Crock, P., et al. (1998). A gene encoding a transmembrane protein is mutated in patients with diabetes mellitus and optic atrophy (Wolfram syndrome). Nat. Genet. *20*, 143–148.

Inoue, H., Nagata, N., Kurokawa, H., and Yamanaka, S. (2014). iPS cells: a game changer for future medicine. EMBO J. *33*, 409–417.

Kong, A., Steinthorsdottir, V., Masson, G., Thorleifsson, G., Sulem, P., Besenbacher, S., Jonasdottir, A., Sigurdsson, A., Kristinsson, K.T., Jonasdottir, A., et al.; DIAGRAM Consortium (2009). Parental origin of sequence variants associated with complex diseases. Nature *462*, 868–874.

Kooner, J.S., Saleheen, D., Sim, X., Sehmi, J., Zhang, W., Frossard, P., Been, L.F., Chia, K.-S., Dimas, A.S., Hassanali, N., et al.; DIAGRAM; MuTHER (2011). Genome-wide association study in individuals of South Asian ancestry identifies six new type 2 diabetes susceptibility loci. Nat. Genet. *43*, 984–989.

Li, H., Gan, W., Lu, L., Dong, X., Han, X., Hu, C., Yang, Z., Sun, L., Bao, W., Li, P., et al.; DIAGRAM Consortium; AGEN-T2D Consortium (2013). A genome-wide association study identifies GRK5 and RASGRP1 as type 2 diabetes loci in Chinese Hans. Diabetes *62*, 291–298.

Ma, R.C.W., Hu, C., Tam, C.H., Zhang, R., Kwan, P., Leung, T.F., Thomas, G.N., Go, M.J., Hara, K., Sim, X., et al.; DIAGRAM Consortium; MuTHER Consortium (2013). Genome-wide association study in a Chinese population identifies a susceptibility locus for type 2 diabetes at 7q32 near PAX4. Diabetologia *56*, 1291–1305.

Mahajan, A., Go, M.J., Zhang, W., Below, J.E., Gaulton, K.J., Ferreira, T., Horikoshi, M., Johnson, A.D., Ng, M.C., Prokopenko, I., et al.; DIAbetes Genetics Replication And Meta-analysis (DIAGRAM) Consortium; Asian Genetic Epidemiology Network Type 2 Diabetes (AGEN-T2D) Consortium; South Asian Type 2 Diabetes (SAT2D) Consortium; Mexican American Type 2 Diabetes (MAT2D) Consortium; Type 2 Diabetes Genetic Exploration by Nex-generation sequencing in muylti-Ethnic Samples (T2D-GENES) Consortium (2014). Genome-wide trans-ancestry meta-analysis provides insight into the genetic architecture of type 2 diabetes susceptibility. Nat. Genet. *46*, 234–244.

Majithia, A.R., Flannick, J., Shahinian, P., Guo, M., Bray, M.-A., Fontanillas, P., Gabriel, S.B., Rosen, E.D., and Altshuler, D.GoT2D Consortium; NHGRI JHS/FHS Allelic Spectrum Project; SIGMA T2D Consortium; T2D-GENES Consortium (2014). Rare variants in PPARG with decreased activity in adipocyte differentiation are associated with increased risk of type 2 diabetes. Proc. Natl. Acad. Sci. USA *111*, 13127–13132.

Marullo, L., El-Sayed Moustafa, J.S., and Prokopenko, I. (2014). Insights into the genetic susceptibility to type 2 diabetes from genome-wide association studies of glycaemic traits. Curr. Diab. Rep. *14*, 551.

März, W., Nauck, M., Hoffmann, M.M., Nagel, D., Boehm, B.O., Koenig, W., Rothenbacher, D., and Winkelmann, B.R. (2004). G(-30)A polymorphism in the pancreatic promoter of the glucokinase gene associated with angiographic

coronary artery disease and type 2 diabetes mellitus. Circulation *109*, 2844–2849.

Meur, G., Simon, A., Harun, N., Virally, M., Dechaume, A., Bonnefond, A., Fetita, S., Tarasov, A.I., Guillausseau, P.-J., Boesgaard, T.W., et al. (2010). Insulin gene mutations resulting in early-onset diabetes: marked differences in clinical presentation, metabolic status, and pathogenic effect through endoplasmic reticulum retention. Diabetes *59*, 653–661.

Meyre, D., Bouatia-Naji, N., Tounian, A., Samson, C., Lecoeur, C., Vatin, V., Ghoussaini, M., Wachter, C., Hercberg, S., Charpentier, G., et al. (2005). Variants of ENPP1 are associated with childhood and adult obesity and increase the risk of glucose intolerance and type 2 diabetes. Nat. Genet. *37*, 863–867.

Moltke, I., Grarup, N., Jørgensen, M.E., Bjerregaard, P., Treebak, J.T., Fumagalli, M., Korneliussen, T.S., Andersen, M.A., Nielsen, T.S., Krarup, N.T., et al. (2014). A common Greenlandic TBC1D4 variant confers muscle insulin resistance and type 2 diabetes. Nature *512*, 190–193.

Morris, A.P., Voight, B.F., Teslovich, T.M., Ferreira, T., Segrè, A.V., Steinthorsdottir, V., Strawbridge, R.J., Khan, H., Grallert, H., Mahajan, A., et al.; Wellcome Trust Case Control Consortium; Meta-Analyses of Glucose and Insulin-related traits Consortium (MAGIC) Investigators; Genetic Investigation of ANthropometric Traits (GIANT) Consortium; Asian Genetic Epidemiology Network–Type 2 Diabetes (AGEN-T2D) Consortium; South Asian Type 2 Diabetes (SAT2D) Consortium; DIAbetes Genetics Replication And Meta-analysis (DIAGRAM) Consortium (2012). Large-scale association analysis provides insights into the genetic architecture and pathophysiology of type 2 diabetes. Nat. Genet. *44*, 981–990.

Neel, J.V. (1976). Diabetes Mellitus — A Geneticist's Nightmare. In The Genetics of Diabetes Mellitus, W. Creutzfeldt, J. Köbberling, and J.V. Neel, eds. (Berlin, Heidelberg: Springer Berlin Heidelberg), pp. 1–11.

Ng, M.C.Y., Shriner, D., Chen, B.H., Li, J., Chen, W.-M., Guo, X., Liu, J., Bielinski, S.J., Yanek, L.R., Nalls, M.A., et al.; FIND Consortium; eMERGE Consortium; DIAGRAM Consortium; MuTHER Consortium; MEta-analysis of type 2 DIabetes in African Americans Consortium (2014). Meta-analysis of genome-wide association studies in African Americans provides insights into the genetic architecture of type 2 diabetes. PLoS Genet. *10*, e1004517.

Nobrega, M.A. (2013). TCF7L2 and glucose metabolism: time to look beyond the pancreas. Diabetes *62*, 706–708.

Østoft, S.H., Bagger, J.I., Hansen, T., Pedersen, O., Holst, J.J., Knop, F.K., and Vilsbøll, T. (2014). Incretin effect and glucagon responses to oral and intravenous glucose in patients with maturity-onset diabetes of the young—type 2 and type 3. Diabetes *63*, 2838–2844.

Palmer, N.D., McDonough, C.W., Hicks, P.J., Roh, B.H., Wing, M.R., An, S.S., Hester, J.M., Cooke, J.N., Bostrom, M.A., Rudock, M.E., et al.; DIAGRAM Consortium; MAGIC Investigators (2012). A genome-wide association search for type 2 diabetes genes in African Americans. PLoS ONE *7*, e29202.

Pasquali, L., Gaulton, K.J., Rodríguez-Seguí, S.A., Mularoni, L., Miguel-Escalada, I., Akerman, I., Tena, J.J., Morán, I., Gómez-Marín, C., van de Bunt, M., et al. (2014). Pancreatic islet enhancer clusters enriched in type 2 diabetes risk-associated variants. Nat. Genet. *46*, 136–143.

Pearson, E.R., Flechtner, I., Njølstad, P.R., Malecki, M.T., Flanagan, S.E., Larkin, B., Ashcroft, F.M., Klimes, I., Codner, E., Iotova, V., et al.; Neonatal Diabetes International Collaborative Group (2006). Switching from insulin to oral sulfonylureas in patients with diabetes due to Kir6.2 mutations. N. Engl. J. Med. *355*, 467–477.

Pearson, E.R., Donnelly, L.A., Kimber, C., Whitley, A., Doney, A.S.F., McCarthy, M.I., Hattersley, A.T., Morris, A.D., and Palmer, C.N.A. (2007). Variation in TCF7L2 influences therapeutic response to sulfonylureas: a GoDARTs study. Diabetes *56*, 2178–2182.

Perry, J.R.B., Voight, B.F., Yengo, L., Amin, N., Dupuis, J., Ganser, M., Grallert, H., Navarro, P., Li, M., Qi, L., et al.; MAGIC; DIAGRAM Consortium; GIANT Consortium (2012). Stratifying type 2 diabetes cases by BMI identifies genetic risk variants in LAMA1 and enrichment for risk variants in lean compared to obese cases. PLoS Genet. *8*, e1002741.

Pirart, J. (1959). [Treatment of fat diabetes; importance of strict diet & slimming treatment]. Brux. Méd. *39*, 751–765.

Poulsen, P., Kyvik, K.O., Vaag, A., and Beck-Nielsen, H. (1999). Heritability of type II (non-insulin-dependent) diabetes mellitus and abnormal glucose tolerance—a population-based twin study. Diabetologia *42*, 139–145.

Prokopenko, I., Langenberg, C., Florez, J.C., Saxena, R., Soranzo, N., Thorleifsson, G., Loos, R.J.F., Manning, A.K., Jackson, A.U., Aulchenko, Y., et al. (2009). Variants in MTNR1B influence fasting glucose levels. Nat. Genet. *41*, 77–81.

Qi, L., Cornelis, M.C., Kraft, P., Stanya, K.J., Linda Kao, W.H., Pankow, J.S., Dupuis, J., Florez, J.C., Fox, C.S., Paré, G., et al.; Meta-Analysis of Glucose and Insulin-related traits Consortium (MAGIC); Diabetes Genetics Replication and Meta-analysis (DIAGRAM) Consortium (2010). Genetic variants at 2q24 are associated with susceptibility to type 2 diabetes. Hum. Mol. Genet. *19*, 2706–2715.

Ragvin, A., Moro, E., Fredman, D., Navratilova, P., Drivenes, Ø., Engström, P.G., Alonso, M.E., de la Calle Mustienes, E., Gómez Skarmeta, J.L., Tavares, M.J., et al. (2010). Long-range gene regulation links genomic type 2 diabetes and obesity risk regions to HHEX, SOX4, and IRX3. Proc. Natl. Acad. Sci. USA *107*, 775–780.

Riveline, J.-P., Rousseau, E., Reznik, Y., Fetita, S., Philippe, J., Dechaume, A., Hartemann, A., Polak, M., Petit, C., Charpentier, G., et al. (2012). Clinical and metabolic features of adult-onset diabetes caused by ABCC8 mutations. Diabetes Care *35*, 248–251.

Rosamond, W., Flegal, K., Furie, K., Go, A., Greenlund, K., Haase, N., Hailpern, S.M., Ho, M., Howard, V., Kissela, B., et al.; American Heart Association Statistics Committee and Stroke Statistics Subcommittee (2008). Heart disease and stroke statistics—2008 update: a report from the American Heart Association Statistics Committee and Stroke Statistics Subcommittee. Circulation *117*, e25–e146.

Rung, J., Cauchi, S., Albrechtsen, A., Shen, L., Rocheleau, G., Cavalcanti-Proença, C., Bacot, F., Balkau, B., Belisle, A., Borch-Johnsen, K., et al. (2009). Genetic variant near IRS1 is associated with type 2 diabetes, insulin resistance and hyperinsulinemia. Nat. Genet. *41*, 1110–1115.

Saeed, S., Butt, T.A., Anwer, M., Arslan, M., and Froguel, P. (2012). High prevalence of leptin and melanocortin-4 receptor gene mutations in children with severe obesity from Pakistani consanguineous families. Mol. Genet. Metab. *106*, 121–126.

Sandhu, M.S., Weedon, M.N., Fawcett, K.A., Wasson, J., Debenham, S.L., Daly, A., Lango, H., Frayling, T.M., Neumann, R.J., Sherva, R., et al. (2007). Common variants in WFS1 confer risk of type 2 diabetes. Nat. Genet. *39*, 951–953.

Saxena, R., Voight, B.F., Lyssenko, V., Burtt, N.P., de Bakker, P.I., Chen, H., Roix, J.J., Kathiresan, S., Hirschhorn, J.N., Daly, M.J., et al.; Diabetes Genetics Initiative of Broad Institute of Harvard and MIT, Lund University, and Novartis Institutes of BioMedical Research (2007). Genome-wide association analysis identifies loci for type 2 diabetes and triglyceride levels. Science *316*, 1331–1336.

Saxena, R., Hivert, M.-F., Langenberg, C., Tanaka, T., Pankow, J.S., Vollenweider, P., Lyssenko, V., Bouatia-Naji, N., Dupuis, J., Jackson, A.U., et al.; GIANT consortium; MAGIC investigators (2010). Genetic variation in GIPR influences the glucose and insulin responses to an oral glucose challenge. Nat. Genet. *42*, 142–148.

Saxena, R., Saleheen, D., Been, L.F., Garavito, M.L., Braun, T., Bjonnes, A., Young, R., Ho, W.K., Rasheed, A., Frossard, P., et al.; DIAGRAM; MuTHER; AGEN (2013). Genome-wide association study identifies a novel locus contributing to type 2 diabetes susceptibility in Sikhs of Punjabi origin from India. Diabetes *62*, 1746–1755.

Scott, L.J., Mohlke, K.L., Bonnycastle, L.L., Willer, C.J., Li, Y., Duren, W.L., Erdos, M.R., Stringham, H.M., Chines, P.S., Jackson, A.U., et al. (2007). A genome-wide association study of type 2 diabetes in Finns detects multiple susceptibility variants. Science *316*, 1341–1345.

Scuteri, A., Sanna, S., Chen, W.-M., Uda, M., Albai, G., Strait, J., Najjar, S., Nagaraja, R., Orrú, M., Usala, G., et al. (2007). Genome-wide association scan shows genetic variants in the FTO gene are associated with obesity-related traits. PLoS Genet. *3*, e115.

Shao, W., Wang, D., Chiang, Y.-T., Ip, W., Zhu, L., Xu, F., Columbus, J., Belsham, D.D., Irwin, D.M., Zhang, H., et al. (2013). The Wnt signaling pathway effector TCF7L2 controls gut and brain proglucagon gene expression and glucose homeostasis. Diabetes *62*, 789–800.

Shepherd, M., Pearson, E.R., Houghton, J., Salt, G., Ellard, S., and Hattersley, A.T. (2003). No deterioration in glycemic control in HNF-1alpha maturity-onset

diabetes of the young following transfer from long-term insulin to sulphonylureas. Diabetes Care *26*, 3191–3192.

Shields, B.M., Hicks, S., Shepherd, M.H., Colclough, K., Hattersley, A.T., and Ellard, S. (2010). Maturity-onset diabetes of the young (MODY): how many cases are we missing? Diabetologia *53*, 2504–2508.

Shu, X.O., Long, J., Cai, Q., Qi, L., Xiang, Y.-B., Cho, Y.S., Tai, E.S., Li, X., Lin, X., Chow, W.-H., et al. (2010). Identification of new genetic risk variants for type 2 diabetes. PLoS Genet. *6*, e1001127.

Silander, K., Mohlke, K.L., Scott, L.J., Peck, E.C., Hollstein, P., Skol, A.D., Jackson, A.U., Deloukas, P., Hunt, S., Stavrides, G., et al. (2004). Genetic variation near the hepatocyte nuclear factor-4 alpha gene predicts susceptibility to type 2 diabetes. Diabetes *53*, 1141–1149.

Sladek, R., Rocheleau, G., Rung, J., Dina, C., Shen, L., Serre, D., Boutin, P., Vincent, D., Belisle, A., Hadjadj, S., et al. (2007). A genome-wide association study identifies novel risk loci for type 2 diabetes. Nature *445*, 881–885.

Smemo, S., Tena, J.J., Kim, K.-H., Gamazon, E.R., Sakabe, N.J., Gómez-Marín, C., Aneas, I., Credidio, F.L., Sobreira, D.R., Wasserman, N.F., et al. (2014). Obesity-associated variants within FTO form long-range functional connections with IRX3. Nature *507*, 371–375.

Steinthorsdottir, V., Thorleifsson, G., Sulem, P., Helgason, H., Grarup, N., Sigurdsson, A., Helgadottir, H.T., Johannsdottir, H., Magnusson, O.T., Gudjonsson, S.A., et al. (2014). Identification of low-frequency and rare sequence variants associated with elevated or reduced risk of type 2 diabetes. Nat. Genet. *46*, 294–298.

Stone, L.M., Kahn, S.E., Fujimoto, W.Y., Deeb, S.S., and Porte, D., Jr. (1996). A variation at position -30 of the beta-cell glucokinase gene promoter is associated with reduced beta-cell function in middle-aged Japanese-American men. Diabetes *45*, 422–428.

Tabassum, R., Chauhan, G., Dwivedi, O.P., Mahajan, A., Jaiswal, A., Kaur, I., Bandesh, K., Singh, T., Mathai, B.J., Pandey, Y., et al.; DIAGRAM; INDICO (2013). Genome-wide association study for type 2 diabetes in Indians identifies a new susceptibility locus at 2q21. Diabetes *62*, 977–986.

Tsai, F.-J., Yang, C.-F., Chen, C.-C., Chuang, L.-M., Lu, C.-H., Chang, C.-T., Wang, T.-Y., Chen, R.-H., Shiu, C.-F., Liu, Y.-M., et al. (2010). A genome-wide association study identifies susceptibility variants for type 2 diabetes in Han Chinese. PLoS Genet. *6*, e1000847.

Unoki, H., Takahashi, A., Kawaguchi, T., Hara, K., Horikoshi, M., Andersen, G., Ng, D.P.K., Holmkvist, J., Borch-Johnsen, K., Jørgensen, T., et al. (2008). SNPs in KCNQ1 are associated with susceptibility to type 2 diabetes in East Asian and European populations. Nat. Genet. *40*, 1098–1102.

Vassy, J.L., Hivert, M.-F., Porneala, B., Dauriz, M., Florez, J.C., Dupuis, J., Siscovick, D.S., Fornage, M., Rasmussen-Torvik, L.J., Bouchard, C., and Meigs, J.B. (2014). Polygenic type 2 diabetes prediction at the limit of common variant detection. Diabetes *63*, 2172–2182.

Vaxillaire, M., Boccio, V., Philippi, A., Vigouroux, C., Terwilliger, J., Passa, P., Beckmann, J.S., Velho, G., Lathrop, G.M., and Froguel, P. (1995). A gene for maturity onset diabetes of the young (MODY) maps to chromosome 12q. Nat. Genet. *9*, 418–423.

Vaxillaire, M., Bonnefond, A., and Froguel, P. (2012). The lessons of early-onset monogenic diabetes for the understanding of diabetes pathogenesis. Best Pract. Res. Clin. Endocrinol. Metab. *26*, 171–187.

Vaxillaire, M., Yengo, L., Lobbens, S., Rocheleau, G., Eury, E., Lantieri, O., Marre, M., Balkau, B., Bonnefond, A., and Froguel, P. (2014). Type 2 diabetes-related genetic risk scores associated with variations in fasting plasma glucose and development of impaired glucose homeostasis in the prospective DESIR study. Diabetologia *57*, 1601–1610.

Vftter, K. (1986). Diabetes mellitus. WHO Technical Report Series 727. 113 Seiten. World Health Organization, Geneva 1985. Preis: 9.00 Sw. fr. Nahrung, 30, 700. http://dx.doi.org/10.1002/food.19860300713.

Vionnet, N., Hani, E.H., Dupont, S., Gallina, S., Francke, S., Dotte, S., De Matos, F., Durand, E., Leprêtre, F., Lecoeur, C., et al. (2000). 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. *67*, 1470–1480.

Voight, B.F., Scott, L.J., Steinthorsdottir, V., Morris, A.P., Dina, C., Welch, R.P., Zeggini, E., Huth, C., Aulchenko, Y.S., Thorleifsson, G., et al.; MAGIC investigators; GIANT Consortium (2010). Twelve type 2 diabetes susceptibility loci identified through large-scale association analysis. Nat. Genet. *42*, 579–589.

Voight, B.F., Kang, H.M., Ding, J., Palmer, C.D., Sidore, C., Chines, P.S., Burtt, N.P., Fuchsberger, C., Li, Y., Erdmann, J., et al. (2012). The metabochip, a custom genotyping array for genetic studies of metabolic, cardiovascular, and anthropometric traits. PLoS Genet. *8*, e1002793.

Walford, G.A., Porneala, B.C., Dauriz, M., Vassy, J.L., Cheng, S., Rhee, E.P., Wang, T.J., Meigs, J.B., Gerszten, R.E., and Florez, J.C. (2014). Metabolite traits and genetic risk provide complementary information for the prediction of future type 2 diabetes. Diabetes Care *37*, 2508–2514.

Walters, R.G., Jacquemont, S., Valsesia, A., de Smith, A.J., Martinet, D., Andersson, J., Falchi, M., Chen, F., Andrieux, J., Lobbens, S., et al. (2010). A new highly penetrant form of obesity due to deletions on chromosome 16p11.2. Nature *463*, 671–675.

Williams, A.L., Jacobs, S.B., Moreno-Macías, H., Huerta-Chagoya, A., Churchhouse, C., Márquez-Luna, C., García-Ortíz, H., Gómez-Vázquez, M.J., Burtt, N.P., Aguilar-Salinas, C.A., et al.; SIGMA Type 2 Diabetes Consortium (2014). Sequence variants in SLC16A11 are a common risk factor for type 2 diabetes in Mexico. Nature *506*, 97–101.

Winckler, W., Weedon, M.N., Graham, R.R., McCarroll, S.A., Purcell, S., Almgren, P., Tuomi, T., Gaudet, D., Boström, K.B., Walker, M., et al. (2007). Evaluation of common variants in the six known maturity-onset diabetes of the young (MODY) genes for association with type 2 diabetes. Diabetes *56*, 685–693.

World Health Organization (2013). http://www.who.int/mediacentre/factsheets/fs312/. Fact sheet N°312.

Yamauchi, T., Hara, K., Maeda, S., Yasuda, K., Takahashi, A., Horikoshi, M., Nakamura, M., Fujita, H., Grarup, N., Cauchi, S., et al. (2010). A genome-wide association study in the Japanese population identifies susceptibility loci for type 2 diabetes at UBE2E2 and C2CD4A-C2CD4B. Nat. Genet. *42*, 864–868.

Yasuda, K., Miyake, K., Horikawa, Y., Hara, K., Osawa, H., Furuta, H., Hirota, Y., Mori, H., Jonsson, A., Sato, Y., et al. (2008). Variants in KCNQ1 are associated with susceptibility to type 2 diabetes mellitus. Nat. Genet. *40*, 1092–1097.

Zeggini, E., Weedon, M.N., Lindgren, C.M., Frayling, T.M., Elliott, K.S., Lango, H., Timpson, N.J., Perry, J.R.B., Rayner, N.W., Freathy, R.M., et al.; Wellcome Trust Case Control Consortium (WTCCC) (2007). Replication of genome-wide association signals in UK samples reveals risk loci for type 2 diabetes. Science *316*, 1336–1341.

Zeggini, E., Scott, L.J., Saxena, R., Voight, B.F., Marchini, J.L., Hu, T., de Bakker, P.I.W., Abecasis, G.R., Almgren, P., Andersen, G., et al.; Wellcome Trust Case Control Consortium (2008). Meta-analysis of genome-wide association data and large-scale replication identifies additional susceptibility loci for type 2 diabetes. Nat. Genet. *40*, 638–645.

Zhou, Y., Park, S.-Y., Su, J., Bailey, K., Ottosson-Laakso, E., Shcherbina, L., Oskolkov, N., Zhang, E., Thevenin, T., Fadista, J., et al. (2014). TCF7L2 is a master regulator of insulin production and processing. Hum. Mol. Genet. *23*, 6419–6431.

Zimmet, P. (1982). Type 2 (non-insulin-dependent) diabetes—an epidemiological overview. Diabetologia *22*, 399–411.