Original Article
Association of Type 2 Diabetes Mellitus related SNP genotypes with altered serum adipokine levels and metabolic syndrome phenotypes

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Abstract: The pathogenesis of T2DM involves secretion of several pro-inflammatory molecules by the dramatically increased adipocytes, both by number and size, and associated macrophages of adipose tissue. Since T2DM is usually preceded by obesity and chronic systemic inflammation, the objective of this study was to explore for any association between genetic variants of previously established 36 T2DM-associated SNPs and altered serum adipocytokine levels and metabolic syndrome phenotypes. Study consisted of 566 subjects (284 males and 282 females) of whom 147 were T2DM patients and 419 healthy controls. Study subjects were genotyped for 36 T2DM-linked single nucleotide polymorphisms (SNPs) using the KASPar SNP Genotyping System and grouped into different genotypes for each SNP. Various anthropometric and biochemical parameters were measured following standard procedures. The mean values of serum levels of individual adipocytokines and the presence/absence of metabolic syndrome phenotypes corresponding to various genotypes were compared by determining the odds ratios. Genotypic variants of five and seven of the 36 T2DM-related SNPs were significantly associated with altered serum levels of adiponectin and aPAI, respectively. Six variants of the 36 SNPs were associated with metabolic syndrome manifestations. This study identified positive associations between genotypic variants of five and seven of the 36 T2DM related SNPs and altered serum levels of adiponectin and aPAI, respectively. Six of 36 SNPs were also associated with metabolic syndrome in the studied population. The relation between specific SNPs and individual phenotypic traits may be useful in explaining the causal mechanisms of hereditary component of T2DM.

Keywords: Adiponectin, resistin, aPAI, Type 2 Diabetes Mellitus, obesity, metabolic syndrome, insulin resistance, SNP, GWAS

Introduction

Obesity is an underlying cause of chronic non-communicable diseases like Type 2 Diabetes Mellitus (T2DM). In many parts of the world, rapidly economic changes over the last few decades have drastically affected the life-style and obesity levels have now reached alarming proportions, contributing to an epidemic of related diseases, such as metabolic syndrome and T2DM [1]. Several genetic factors predispose individuals to obesity and exaggerate the effects of environmental factors such as a sedentary life style and high intake of low quality foods [2].

In addition to storing lipids and serving as an energy reservoir, adipose tissue also serves as an endocrine organ by secreting many substances, known as 'adipokines' or 'adipokines', which are involved in the regulation of several physiologic metabolic processes [3]. Increased numbers and size of adipocytes in obese individuals lead to increased synthesis and secretion of adipokines, and hence, serum levels of adipokines are influenced primarily by obesity status. Due to their influence on insulin sensitivity and glucose metabolism of various tissues and pancreatic islet beta cell function, altered adipokine levels have been suggested to be responsible for several of the manifesta-

Table 1. Clinical profile of study subjects

General Characteristics	Mean ± SD
N	566
Gender (M/F)	284/282
Age	42.4±9.0
BMI (kg/m^2)	29.6±6.3
Waist (cm)	91.6±22.1
Systolic BP (mmHg)	119.1±12.7
Diastolic BP (mmHg)	76.7±7.7
Cholesterol (mmol/l)	5.3±1.1
Glucose (mmol/l)	6.9±3.1
Triglycerides (mmol/l)	1.8±0.99
HDL (mmol/l)	0.83±0.26
LDL (mmol/l)	4.1±1.0
Adiponectin (ug/ml)	2.7 (0.02, 8.5)
Resistin (ng/ml)	17.2±4.2
aPAI (ng/ml)	11.6 (3.2, 67.5)
ANGII (ng/ml)	1.2±0.29
hsCRP (ug/ml)	3.4 (1.2, 5.8)

tions of T2DM, including systemic insulin resistance [3-7].

Decreases in adiponectin, the insulin sensitizing adipokine, have been associated with increased body fat [8], while increase in resistin, an inflammation-related adipokine has been linked to insulin resistance in humans [9]. Increased levels of activated plasminogen activator inhibitor 1 (aPAI-1) is an index of low-grade inflammatory state associated with obesity [10]. More than 75% of T2DM patients have metabolic syndrome (MetS). Combined occurrence of obesity and elevated blood pressure (BP), elevated serum triglyceride (TG) and glucose, and reduced HDL cholesterol mark MetS [25]. Also, elevated resistin and decreased adiponectin levels are risk factors for MetS [11].

Recent genome wide analysis (GWA) studies have identified reproducible associations between certain single nucleotide polymorphisms (SNPs) and incidence of T2DM [12]. Progression of T2DM parallels the increased systemic levels of several pro-inflammatory and decreased levels of anti-inflammatory cytokines and the worsening of MetS [3]. The indigenous Middle Eastern population may have an increased genetic predisposition to develop T2DM [13]. Earlier, we identified the highly heritable nature of adipokines and their pattern of co-variation with BMI from the pre-teen years in a Saudi population [14]. Following recent GWA studies in European and South Asian populations,

which have revealed 36 genetic variants related to T2DM risk [15-23], we investigated whether these SNPs were also associated with altered serum levels of adipokines and presence of MetS manifestations in a T2DM Saudi population.

Methods

Study subjects

Study subjects were 566 unrelated adult Saudi individuals consisting of 160 T2DM patients and 406 normal controls from the Biomarker Screening Project in Riyadh (RIYADH COHORT), a capital-wide epidemiological study involving over 17,000 consenting Saudis coming from different Primary Health Care Centres (PHCCs) of the city of Riyadh, Saudi Arabia. A generalized questionnaire aimed to seek demographic information and past medical history was given to all participating subjects. Those with morbidities that needed medical attention were excluded from the study. Written consent was obtained after orientation to the study. Ethical approval was granted by the Ethics Committee of the College of Science Research Centre, King Saud University (Riyadh, Saudi Arabia).

Participating subjects were requested to attend their assigned PHCCs after an overnight fast (> 10 hours) for anthropometry and blood withdrawal. Anthropometry included height (to the nearest 0.5 cm), weight (to the nearest 0.1 kg), body mass index (BMI) (calculated as kg/m^2), waist and hip circumference (measured utilizing a standardized measuring tape in cm), and systolic and diastolic blood pressure. Fasting serum samples were collected and stored at -20°C prior to analysis.

Genotyping

Genomic DNA was isolated from whole blood using the Blood GenomicPrep Mini Spin Kit (GE healthcare Life Sciences, Piscataway, NJ, USA). DNA concentration and purity (260/280) were checked using Nano-drop spectrophotometer. All DNA samples from T2DM patients and controls were genotyped for 36 SNPs using the KASPar SNP Genotyping System (KBioscience, Hoddesdon, UK).

Biochemical measurements

Fasting glucose and lipid profile were measured using a chemical autoanalyzer (Konelab, Vantaa, Finland) at the Biomarker Research Center (King Saud University, Riyadh, Saudi

Table 2. Relation between T2D-associated SNPs and altered serum adipocytokine levels

	Risk Allele	Model	Adiponectin Odds Ratio (95% CI)	P value	Resistin Odds Ratio (95% CI)	P value	aPAI Odds Ratio (95% CI)	P value
rs7903146	T	Additive	1.5 (0.88, 2.7)	0.11	1.1 (0.64, 1.9)	0.65	1.0 (0.53, 1.9)	0.95
		Recessive	1.6 (0.96, 2.8)	0.06	1.2 (0.70, 2.0)	0.51	1.4 (0.76, 2.5)	0.27
		Dominant	1.0 (0.71, 1.5)	0.85	0.95 (0.66, 1.3)	0.81	0.61 (0.39, 0.96)	0.03
rs13081389	A	Additive	-	-	-	-	3.2 (1.3, 7.3)	0.007
		Recessive	1.2 (0.61, 2.1)	0.64	1.6 (0.83, 3.0)	0.15	3.2 (1.3, 7.3)	0.007
		Dominant	-	-	-	-	-	-
rs1470579	C	Additive	0.71 (0.42, 1.1)	0.18	1.5 (0.95, 2.5)	0.07	2.1 (1.1, 3.8)	0.02
		Recessive	0.77 (0.48, 1.2)	0.28	1.4 (0.92, 2.3)	0.10	1.9 (1.1, 3.4)	0.02
		Dominant	0.79 (0.54, 1.1)	0.22	1.3 (0.89, 1.8)	0.18	1.3 (0.85, 2.1)	0.19
rs243021	T	Additive	1.1 (0.59, 1.8)	0.86	0.82 (0.47, 1.4)	0.49	0.48 (0.24, 0.96)	0.04
		Recessive	0.90 (0.62, 1.3)	0.59	1.0 (0.72, 1.4)	0.83	0.59 (0.38, 0.94)	0.02
		Dominant	1.1 (0.67, 1.9)	0.63	0.77 (0.46, 1.3)	0.33	0.62 (0.32, 1.2)	0.14
rs972283	G	Additive	1.6 (1.0, 2.9)	0.04	1.5 (0.87, 2.5)	0.14	1.5 (0.77, 2.9)	0.22
		Recessive	1.2 (0.81, 1.7)	0.38	1.3 (0.89, 1.8)	0.17	1.1 (0.69, 1.7)	0.71
		Dominant	1.6 (1.0, 2.7)	0.04	1.3 (0.81, 2.2)	0.25	1.5 (0.82, 2.8)	0.17
rs896854	A	Additive	2.7 (1.6, 4.6)	$1.04*10^{-4}$	0.81 (0.49, 1.3)	0.39	1.3 (0.74, 2.4)	0.32
		Recessive	1.68 (1.1, 2.5)	0.01	0.81 (0.54, 1.2)	0.31	1.1 (0.71, 1.8)	0.56
		Dominant	2.4 (1.5, 3.6)	$1.05*10^{-4}$	0.92 (0.61, 1.3)	0.67	1.3 (0.79, 2.1)	0.29
rs13292136	C	Additive	0.65 (0.18, 2.3)	0.51	0.36 (0.09, 1.4)	0.13	0.10 (0.01, 0.93)	0.03
		Recessive	0.65 (0.18, 2.3)	0.51	0.36 (0.09, 1.4)	0.13	0.11 (0.01, 0.83)	0.03
		Dominant	-	-	-	-	-	-
rs1552224	T	Additive	-	-	-	-	-	-
		Recessive	1.3 (0.59, 2.8)	0.52	0.59 (0.27, 1.2)	0.18	0.29 (0.09, 0.91)	0.03
		Dominant	-	-	-	-	-	-
rs11634397	G	Additive	1.4 (0.82, 2.3)	0.22	1.0 (0.62, 1.7)	0.87	0.76 (0.41, 1.4)	0.38
		Recessive	1.7 (1.1, 2.5)	0.01	1.2 (0.83, 1.8)	0.29	0.96 (0.59, 1.5)	0.86
		Dominant	0.94 (0.60, 1.4)	0.94	0.87 (0.56, 1.3)	0.54	0.73 (0.43, 1.2)	0.25
rs5945326	G	Additive	0.66 (0.29, 1.4)	0.32	1.5 (0.69, 3.4)	0.28	1.1 (0.48, 2.6)	0.76
		Recessive	0.72 (0.32, 1.6)	0.43	1.5 (0.67, 3.3)	0.32	1.1 (0.47, 2.5)	0.81
		Dominant	0.56 (0.35, 0.91)	0.02	1.3 (0.85, 2.1)	0.19	1.3 (0.69, 2.3)	0.43
rs163184	G	Additive	1.2 (0.71, 1.9)	0.50	0.97 (0.59, 1.6)	0.92	0.61 (0.33, 1.1)	0.11
		Recessive	1.3 (0.85, 2.0)	0.20	1.1 (0.70, 1.6)	0.74	0.85 (0.50, 1.4)	0.55
		Dominant	0.94 (0.63, 1.4)	0.79	0.90 (0.60, 1.3)	0.61	0.59 (0.36, 0.96)	0.03
rs4812829	A	Additive	0.37 (0.14, 0.98)	0.04	0.98 (0.40, 2.3)	0.96	1.2 (0.39, 3.7)	0.72
		Recessive	0.38 (0.14, 1.0)	0.05	0.90 (0.37, 2.1)	0.81	1.2 (0.38, 3.5)	0.78
		Dominant	0.81 (0.55, 1.2)	0.28	1.2 (0.85, 1.8)	0.27	1.1 (0.72, 1.7)	0.59

Arabia). Serum adiponectin, resistin and aPAI-1 were quantified using multiplex assay kits that utilize fluorescent microbead technology, allowing simultaneous quantification of several target proteins within a single serum sample of 50-100 μl. These included pre-mixed and fully customized panels that utilize the LuminexH xMAPH Technology platform (Luminex Corporation, Austin, TX, USA). Minimum detectable concentrations (MDC) were as follows: adiponectin, 145.4 pg/ml; resistin, 6.7 pg/ml and aPAI-1, 1.3 pg/ml.

Definition of MetS

The definition of MetS used was according to the National Cholesterol Education Program-Third Adult Treatment Panel (NCEP ATP III), according to which three or more of the following criteria must be fulfilled: fasting blood glucose level ≥ 5.6 mmol/l; blood pressure ≥ 130/85 mmHg; triglycerides ≥ 1.7 mmol/l; HDL-cholesterol < 1.03 mmol/l for men and < 1.29 mmol/l for women; and waist circumference > 102 cm for men and > 88 cm for women [24].

Statistical analysis

Data was analyzed using SPSS version 16.0 (Statistical package for Social Science, Inc., Chicago IL, USA). The data are presented as means ± SD. The difference between the means of 2 groups was tested using Student's

Table 3. Relation between T2D-associated SNPs and metabolic syndrome

SNPs	Risk Allele	Model	Odds Ratio (95% CI)	P
rs10440833	A	Additive	0.41 (0.20, 0.87)	0.02
		Recessive	0.43 (0.20, 0.89)	0.02
		Dominant	0.79 (0.56, 1.1)	0.20
rs11899863	G	Additive	0.20 (0.02, 1.9)	0.17
		Recessive	0.51 (0.29, 0.90)	0.02
		Dominant	0.43 (0.07, 2.6)	0.36
rs5215	T	Additive	0.83 (0.31, 2.2)	0.72
		Recessive	0.64 (0.44, 0.92)	0.01
		Dominant	0.95 (0.35, 2.5)	0.92
rs1387153	T	Additive	0.39 (0.18, 0.85)	0.01
		Recessive	0.38 (0.18, 0.82)	0.01
		Dominant	0.89 (0.63, 1.2)	0.51
rs972283	G	Additive	1.8 (1.1, 2.9)	0.02
		Recessive	1.7 (1.2, 2.5)	0.001
		Dominant	1.3 (0.83, 2.1)	0.22
rs1801214	T	Additive	1.9 (1.1, 3.0)	0.01
		Recessive	1.6 (1.1, 2.3)	0.01
		Dominant	1.5 (0.97, 2.3)	0.07

t test. Multiple group comparisons were performed using analysis of variance (ANOVA). Multinomial logistic regression was used to calculate odds ratios and 95% confidence intervals. Level of significance was given at $P \leq 0.05$.

Results

The means of anthropometric and clinical parameters of individuals enrolled in the study are given in **Table 1**. Various genetic characteristics of the 36 T2DM related SNPs are given in Supplementary Table 1. Results of analysis of associations between genotypic variants of 36 SNPs and serum adipokine levels are presented in **Table 2**. Genotypic variants of 5 and 7 of the 36 T2DM-related SNPs were significantly associated with altered blood levels of adiponectin and aPAI, respectively. Genetic variants of SNPs rs972283 [additive; OR 1.6 (1.0, 2.9)], rs896854 [additive; OR=2.7 (1.6, 4.6)], rs11634397 [recessive; OR=1.7 (1.1, 2.5)], rs5945326 [dominant; OR=0.56 (0.35,0.91)] and rs4812829 [additive; OR=0.37 (0.14, 0.98)] were associated with altered serum levels of adiponectin; rs7903146 [dominant; OR =0.61 (0.39, 0.96)], rs13081389 [dominant; OR=3.2 (1.3, 7.3)], rs1470579 [additive; OR= 2.1 (1.1, 3.8)], rs243021 [additive; OR=0.48 (0.24, 0.96)], rs13292136 [additive; OR=0.10

(0.01, 0.93)], rs1552224 [recessive; OR=0.29 (0.09, 0.91)] and rs163184 [dominant; 0.59 (0.36, 0.96)] were associated with altered serum levels of aPAI. Serum level of resistin was not related to any of the 36 SNPs.

Six variants of the 36 SNPs were associated with metabolic syndrome manifestations (**Table 3**). These were: rs10440833 [additive; OR=0.41 (0.20, 0.87)], rs11899863 [recessive; OR=0.51 (0.29, 0.90)]; rs5215 [recessive; OR=0.64 (0.44, 0.92)], rs1387153 [additive; OR=0.39 (0.18, 0.85)]; rs972283 [additive; OR=1.8 (1.1, 2.9)] and rs1801214 [additive; OR=1.9 (1.1, 3.0)].

Discussion

Previous GWA studies have identified 36 SNPs strongly associated with T2DM in European and South Asian populations. Since T2DM develops and progresses readily, and is difficult to control in obese people adipokines secreted by adipocytes may be involved in the pathology of T2DM. Therefore, this study explored for any associations between genotypic variants of T2DM-related SNPs and serum adipokine levels and presence of metabolic syndrome phenotypes. Allelic variants of seven and five of the 36 SNPs were significantly associated with altered levels of adiponectin and aPAI, respectively. Allelic variants of six of the 36 SNPs showed significant association with metabolic syndrome.

Several traits like obesity, insulin resistance and increased levels of proinflammatory cytokines precede and accompany T2DM [25]. On the other hand, it is widely accepted that T2DM has a hereditary component. Hence, it is natural to expect patients suffering from hereditarily transferred predisposition to T2DM to have one or more SNPs responsible for accelerated progression of underlying traits. The indigenous Saudi population, having an increased genetic predisposition to develop T2DM [13], may be considered appropriate for this kind of study.

In a large-scale GWA study involving 34,840 individuals with T2DM and 114,981 controls, McCarthy and colleagues identified pathways associated with cell cycle regulation, adipokine

protein signaling, and CREB-BP-related transcription involved in diabetes pathogenesis [26]. More studies are expected to detect new variants that will explain a larger proportion of the heritability of obesity, and hence, T2DM. Region/population-specific genetic studies are expected to yield useful insights, since the distribution of SNPs is a race- and region-dependent genetic variation. In this respect, risk allele frequencies of TCF7L2 SNPs showing the strongest effect on T2DM in European populations are very few in the Japanese (~5%) compared to populations of European descent (~40%) [12]. Resistin is a polypeptide hormone that has been associated with insulin resistance, inflammation and risk of T2DM [27]. The variant rs12779790, mapped to intergenic region between CDC123 and/CAMK1D, was associated with T2DM vulnerability, a finding that has been replicated [22]. However, in a German population, the rs12779790 mutation was not associated with T2DM or prediabetic phenotypes related to insulin secretion or sensitivity [28, 29]. However, the rs11899863 variant, located in the intron region of THADA gene, was associated significantly with aPAI in our study. Moreover, the rs5945326 SNP, located near DUSP9 on the X-chromosome, was associated with increased resistin levels in our study. The latter SNP was only modestly related to T2DM in a Chinese population [30]. Another resistin-associated SNP, rs1801214, mapped to WFS1 gene coding region, was related to T2DM in an African American population [31]. The rs243021 mutation, an intergenic SNP located between EIF3FP3-BCL11A and reproducibly associated with T2DM [21] was also related to serum aPAI of T2DM patients in our study.

Adiponectin is negatively correlated with body mass index [32], and is decreased in the presence of insulin resistance and T2DM [33]. Meta-analyses have shown that hypoadiponectinemia and hyperresistinemia are strongly associated with increased risk of insulin resistance in T2DM [34]. One of the variants of the 36 SNPs examined in the current study was associated with decreased adiponectin levels. Previously we showed population-based differences in the association of adiponectin gene variants with metabolic phenotypes in a study involving T2DM Saudi patients [35].

The modest effect sizes of individual common susceptibility variants seen in our study resem-

ble several GWA studies. For example, the largest allelic odds ratio of any established common variant for T2DM was ~1.35 (TCF7L2), with the nine other validated associations to common variants having allelic odds ratios between 1.1 and 1.2 [19]. The heterogeneous nature of pathogenesis of T2DM and the range of underlying traits may explain the modest contribution of individual genetic factors. These include environmental factors, dietary habits and levels of physical activity, which have a significant effect on T2DM and on the clinical, metabolic and inflammatory traits preceding T2DM, and hence, may be expected to affect adipokine levels.

The authors acknowledge certain limitations of this study. Confirmation of common variants in the human genome with modest effects on common disease risk, even if real, need large sample sizes to overcome the influence of many genetic and environmental modifiers. Also, our study subjects consisted of Saudi ethnicity, and thus, the generalizability to other ethnicities is unknown.

In conclusion, we investigated 36 previously confirmed T2DM-associated loci in a Saudi population and identified nine of them to be also significantly associated with alterations in the levels of serum adipokines and another five related to the presence of metabolic syndrome. Risk allele scores defined by the five loci were also associated with T2DM-related phenotypes, including systolic and diastolic blood pressure, cholesterol, triglycerides and fasting glucose levels. Replication of these results in additional studies/other populations may reveal the molecular mechanisms underlying specific T2DM precursor traits and may allow early detection and preventive measures.

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Disclosure of conflict of interest

None.

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References

[1]   Al-Daghri NM, Al-Attas OS, Alokail MS, Alkharfy KM, Yousef M, Sabico SL and Chrousos GP. Diabetes mellitus type 2 and other chronic non-communicable diseases in the central region, Saudi Arabia (Riyadh cohort 2): a decade of an epidemic. BMC Med 2011; 9: 76.

[2]   Basic M, Butorac A, Landeka Jurcevic I and Bacun-Druzina V. Obesity: genome and environment interactions. Arh Hig Rada Toksikol 2012; 63: 395-405.

[3]   Bluher M. Clinical relevance of adipokines. Diabetes Metab J 2012; 36: 317-327.

[4]   Rabe K, Lehrke M, Parhofer KG and Broedl UC. Adipokines and insulin resistance. Mol Med 2008; 14: 741-751.

[5]   Kadowaki T, Yamauchi T, Kubota N, Hara K, Ueki K and Tobe K. Adiponectin and adiponectin receptors in insulin resistance, diabetes, and the metabolic syndrome. J Clin Invest 2006; 116: 1784-1792.

[6]   Gulcelik NE, Usman A and Gurlek A. Role of adipocytokines in predicting the development of diabetes and its late complications. Endocrine 2009; 36: 397-403.

[7]   Dunmore SJ and Brown JE. The role of adipokines in beta-cell failure of type 2 diabetes. J Endocrinol 2013; 216: T37-45.

[8]   Gilardini L, McTernan PG, Girola A, da Silva NF, Alberti L, Kumar S and Invitti C. Adiponectin is a candidate marker of metabolic syndrome in obese children and adolescents. Atherosclerosis 2006; 189: 401-407.

[9]   McTernan CL, McTernan PG, Harte AL, Levick PL, Barnett AH and Kumar S. Resistin, central obesity, and type 2 diabetes. Lancet 2002; 359: 46-47.

[10]   Dandona P, Aljada A, Chaudhuri A, Mohanty P and Garg R. Metabolic syndrome: a comprehensive perspective based on interactions between obesity, diabetes, and inflammation. Circulation 2005; 111: 1448-1454.

[11]   Lau CH and Muniandy S. Novel adiponectin-resistin (AR) and insulin resistance (IRAR) indexes are useful integrated diagnostic biomarkers for insulin resistance, type 2 diabetes and metabolic syndrome: a case control study. Cardiovasc Diabetol 2011; 10: 8.

[12]   Imamura M and Maeda S. Genetics of type 2 diabetes: the GWAS era and future perspectives [Review]. Endocr J 2011; 58: 723-739.

[13]   Elhadd TA, Al-Amoudi AA and Alzahrani AS. Epidemiology, clinical and complications profile of diabetes in Saudi Arabia: a review. Ann Saudi Med 2007; 27: 241-250.

[14]   Al-Daghri NM, Al-Attas OS, Alokail MS, Alkharfy KM, Yakout SM, Sabico SB, Gibson GC, Chrousos GP and Kumar S. Parent-offspring transmission of adipocytokine levels and their associations with metabolic traits. PLoS One 2011; 6: e18182.

[15]   Kooner JS, Saleheen D, Sim X, Sehmi J, Zhang W, Frossard P, Been LF, Chia KS, Dimas AS, Hassanali N, Jafar T, Jowett JB, Li X, Radha V, Rees SD, Takeuchi F, Young R, Aung T, Basit A, Chidambaram M, Das D, Grundberg E, Hedman AK, Hydrie ZI, Islam M, Khor CC, Kowlessur S, Kristensen MM, Liju S, Lim WY, Matthews DR, Liu J, Morris AP, Nica AC, Pinidiyapathirage JM, Prokopenko I, Rasheed A, Samuel M, Shah N, Shera AS, Small KS, Suo C, Wickremasinghe AR, Wong TY, Yang M, Zhang F, Abecasis GR, Barnett AH, Caulfield M, Deloukas P, Frayling TM, Froguel P, Kato N, Katulanda P, Kelly MA, Liang J, Mohan V, Sanghera DK, Scott J, Seielstad M, Zimmet PZ, Elliott P, Teo YY, McCarthy MI, Danesh J, Tai ES and Chambers JC. Genome-wide association study in individuals of South Asian ancestry identifies six new type 2 diabetes susceptibility loci. Nat Genet 2011; 43: 984-989.

[16]   Palmer ND, Hester JM, An SS, Adeyemo A, Rotimi C, Langefeld CD, Freedman BI, Ng MC and Bowden DW. Resequencing and analysis of variation in the TCF7L2 gene in African Americans suggests that SNP rs7903146 is the causal diabetes susceptibility variant. Diabetes 2011; 60: 662-668.

[17]   Rung J, Cauchi S, Albrechtsen A, Shen L, Rocheleau G, Cavalcanti-Proenca C, Bacot F, Balkau B, Belisle A, Borch-Johnsen K, Charpentier G, Dina C, Durand E, Elliott P, Hadjadj S, Jarvelin MR, Laitinen J, Lauritzen T, Marre M, Mazur A, Meyre D, Montpetit A, Pisinger C, Posner B, Poulsen P, Pouta A, Prentki M, Ribel-Madsen R, Ruokonen A, Sandbaek A, Serre D, Tichet J, Vaxillaire M, Wojtaszewski JF, Vaag A, Hansen T, Polychronakos C, Pedersen O, Froguel P and Sladek R. Genetic variant near IRS1 is associated with type 2 diabetes, insulin resistance and hyperinsulinemia. Nat Genet 2009; 41: 1110-1115.

[18]   Scott LJ, Mohlke KL, Bonnycastle LL, Willer CJ, Li Y, Duren WL, Erdos MR, Stringham HM, Chines PS, Jackson AU, Prokunina-Olsson L, Ding CJ, Swift AJ, Narisu N, Hu T, Pruim R, Xiao R, Li XY, Conneely KN, Riebow NL, Sprau AG, Tong M, White PP, Hetrick KN, Barnhart MW, Bark CW, Goldstein JL, Watkins L, Xiang F, Saramies J, Buchanan TA, Watanabe RM, Valle TT, Kinnunen L, Abecasis GR, Pugh EW, Doheny KF, Bergman RN, Tuomilehto J, Collins FS and Boehnke M. A genome-wide association study of type 2 diabetes in Finns detects multiple susceptibility variants. Science 2007; 316: 1341-1345.

[19]   Sladek R, Rocheleau G, Rung J, Dina C, Shen L, Serre D, Boutin P, Vincent D, Belisle A, Hadjadj

S, Balkau B, Heude B, Charpentier G, Hudson TJ, Montpetit A, Pshezhetsky AV, Prentki M, Posner BI, Balding DJ, Meyre D, Polychronakos C and Froguel P. A genome-wide association study identifies novel risk loci for type 2 diabetes. Nature 2007; 445: 881-885.

[20] Unoki H, Takahashi A, Kawaguchi T, Hara K, Horikoshi M, Andersen G, Ng DP, Holmkvist J, Borch-Johnsen K, Jorgensen T, Sandbaek A, Lauritzen T, Hansen T, Nurbaya S, Tsunoda T, Kubo M, Babazono T, Hirose H, Hayashi M, Iwamoto Y, Kashiwagi A, Kaku K, Kawamori R, Tai ES, Pedersen O, Kamatani N, Kadowaki T, Kikkawa R, Nakamura Y and Maeda S. SNPs in KCNQ1 are associated with susceptibility to type 2 diabetes in East Asian and European populations. Nat Genet 2008; 40: 1098-1102.

[21] Voight BF, Scott LJ, Steinthorsdottir V, Morris AP, Dina C, Welch RP, Zeggini E, Huth C, Aulchenko YS, Thorleifsson G, McCulloch LJ, Ferreira T, Grallert H, Amin N, Wu G, Willer CJ, Raychaudhuri S, McCarroll SA, Langenberg C, Hofmann OM, Dupuis J, Qi L, Segre AV, van Hoek M, Navarro P, Ardlie K, Balkau B, Benediktsson R, Bennett AJ, Blagieva R, Boerwinkle E, Bonnycastle LL, Bengtsson Bostrom K, Bravenboer B, Bumpstead S, Burtt NP, Charpentier G, Chines PS, Cornelis M, Couper DJ, Crawford G, Doney AS, Elliott KS, Elliott AL, Erdos MR, Fox CS, Franklin CS, Ganser M, Gieger C, Grarup N, Green T, Griffin S, Groves CJ, Guiducci C, Hadjadj S, Hassanali N, Herder C, Isomaa B, Jackson AU, Johnson PR, Jorgensen T, Kao WH, Klopp N, Kong A, Kraft P, Kuusisto J, Lauritzen T, Li M, Lieverse A, Lindgren CM, Lyssenko V, Marre M, Meitinger T, Midthjell K, Morken MA, Narisu N, Nilsson P, Owen KR, Payne F, Perry JR, Petersen AK, Platou C, Proenca C, Prokopenko I, Rathmann W, Rayner NW, Robertson NR, Rocheleau G, Roden M, Sampson MJ, Saxena R, Shields BM, Shrader P, Sigurdsson G, Sparso T, Strassburger K, Stringham HM, Sun Q, Swift AJ, Thorand B, Tichet J, Tuomi T, van Dam RM, van Haeften TW, van Herpt T, van Vliet-Ostaptchouk JV, Walters GB, Weedon MN, Wijmenga C, Witteman J, Bergman RN, Cauchi S, Collins FS, Gloyn AL, Gyllensten U, Hansen T, Hide WA, Hitman GA, Hofman A, Hunter DJ, Hveem K, Laakso M, Mohlke KL, Morris AD, Palmer CN, Pramstaller PP, Rudan I, Sijbrands E, Stein LD, Tuomilehto J, Uitterlinden A, Walker M, Wareham NJ, Watanabe RM, Abecasis GR, Boehm BO, Campbell H, Daly MJ, Hattersley AT, Hu FB, Meigs JB, Pankow JS, Pedersen O, Wichmann HE, Barroso I, Florez JC, Frayling TM, Groop L, Sladek R, Thorsteinsdottir U, Wilson JF, Illig T, Froguel P, van Duijn CM, Stefansson K, Altshuler D, Boehnke M and McCarthy MI. Twelve type 2 diabetes susceptibility loci identified through large-scale association analysis. Nat Genet 2010; 42: 579-589.

[22] Zeggini E, Scott LJ, Saxena R, Voight BF, Marchini JL, Hu T, de Bakker PI, Abecasis GR, Almgren P, Andersen G, Ardlie K, Bostrom KB, Bergman RN, Bonnycastle LL, Borch-Johnsen K, Burtt NP, Chen H, Chines PS, Daly MJ, Deodhar P, Ding CJ, Doney AS, Duren WL, Elliott KS, Erdos MR, Frayling TM, Freathy RM, Gianniny L, Grallert H, Grarup N, Groves CJ, Guiducci C, Hansen T, Herder C, Hitman GA, Hughes TE, Isomaa B, Jackson AU, Jorgensen T, Kong A, Kubalanza K, Kuruvilla FG, Kuusisto J, Langenberg C, Lango H, Lauritzen T, Li Y, Lindgren CM, Lyssenko V, Marvelle AF, Meisinger C, Midthjell K, Mohlke KL, Morken MA, Morris AD, Narisu N, Nilsson P, Owen KR, Palmer CN, Payne F, Perry JR, Pettersen E, Platou C, Prokopenko I, Qi L, Qin L, Rayner NW, Rees M, Roix JJ, Sandbaek A, Shields B, Sjogren M, Steinthorsdottir V, Stringham HM, Swift AJ, Thorleifsson G, Thorsteinsdottir U, Timpson NJ, Tuomi T, Tuomilehto J, Walker M, Watanabe RM, Weedon MN, Willer CJ, Illig T, Hveem K, Hu FB, Laakso M, Stefansson K, Pedersen O, Wareham NJ, Barroso I, Hattersley AT, Collins FS, Groop L, McCarthy MI, Boehnke M and Altshuler D. 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.

[23] Zeggini E, Weedon MN, Lindgren CM, Frayling TM, Elliott KS, Lango H, Timpson NJ, Perry JR, Rayner NW, Freathy RM, Barrett JC, Shields B, Morris AP, Ellard S, Groves CJ, Harries LW, Marchini JL, Owen KR, Knight B, Cardon LR, Walker M, Hitman GA, Morris AD, Doney AS, McCarthy MI and Hattersley AT. Replication of genome-wide association signals in UK samples reveals risk loci for type 2 diabetes. Science 2007; 316: 1336-1341.

[24] Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults. Executive Summary of The Third Report of The National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, And Treatment of High Blood Cholesterol In Adults (Adult Treatment Panel III). JAMA 2001; 285: 2486-2497.

[25] Cefalu WT. Inflammation, insulin resistance, and type 2 diabetes: back to the future? Diabetes 2009; 58: 307-308.

[26] Morris AP, Voight BF, Teslovich TM, Ferreira T, Segre AV, Steinthorsdottir V, Strawbridge RJ, Khan H, Grallert H, Mahajan A, Prokopenko I, Kang HM, Dina C, Esko T, Fraser RM, Kanoni S, Kumar A, Lagou V, Langenberg C, Luan J, Lindgren CM, Muller-Nurasyid M, Pechlivanis S,

Rayner NW, Scott LJ, Wiltshire S, Yengo L, Kinnunen L, Rossin EJ, Raychaudhuri S, Johnson AD, Dimas AS, Loos RJ, Vedantam S, Chen H, Florez JC, Fox C, Liu CT, Rybin D, Couper DJ, Kao WH, Li M, Cornelis MC, Kraft P, Sun Q, van Dam RM, Stringham HM, Chines PS, Fischer K, Fontanillas P, Holmen OL, Hunt SE, Jackson AU, Kong A, Lawrence R, Meyer J, Perry JR, Platou CG, Potter S, Rehnberg E, Robertson N, Sivapalaratnam S, Stancakova A, Stirrups K, Thorleifsson G, Tikkanen E, Wood AR, Almgren P, Atalay M, Benediktsson R, Bonnycastle LL, Burtt N, Carey J, Charpentier G, Crenshaw AT, Doney AS, Dorkhan M, Edkins S, Emilsson V, Eury E, Forsen T, Gertow K, Gigante B, Grant GB, Groves CJ, Guiducci C, Herder C, Hreidarsson AB, Hui J, James A, Jonsson A, Rathmann W, Klopp N, Kravic J, Krjutskov K, Langford C, Leander K, Lindholm E, Lobbens S, Mannisto S, Mirza G, Muhleisen TW, Musk B, Parkin M, Rallidis L, Saramies J, Sennblad B, Shah S, Sigurethsson G, Silveira A, Steinbach G, Thorand B, Trakalo J, Veglia F, Wennauer R, Winckler W, Zabaneh D, Campbell H, van Duijn C, Uitterlinden AG, Hofman A, Sijbrands E, Abecasis GR, Owen KR, Zeggini E, Trip MD, Forouhi NG, Syvanen AC, Eriksson JG, Peltonen L, Nothen MM, Balkau B, Palmer CN, Lyssenko V, Tuomi T, Isomaa B, Hunter DJ, Qi L, Shuldiner AR, Roden M, Barroso I, Wilsgaard T, Beilby J, Hovingh K, Price JF, Wilson JF, Rauramaa R, Lakka TA, Lind L, Dedoussis G, Njolstad I, Pedersen NL, Khaw KT, Wareham NJ, Keinanen-Kiukaanniemi SM, Saaristo TE, Korpi-Hyovalti E, Saltevo J, Laakso M, Kuusisto J, Metspalu A, Collins FS, Mohlke KL, Bergman RN, Tuomilehto J, Boehm BO, Gieger C, Hveem K, Cauchi S, Froguel P, Baldassarre D, Tremoli E, Humphries SE, Saleheen D, Danesh J, Ingelsson E, Ripatti S, Salomaa V, Erbel R, Jockel KH, Moebus S, Peters A, Illig T, de Faire U, Hamsten A, Morris AD, Donnelly PJ, Frayling TM, Hattersley AT, Boerwinkle E, Melander O, Kathiresan S, Nilsson PM, Deloukas P, Thorsteinsdottir U, Groop LC, Stefansson K, Hu F, Pankow JS, Dupuis J, Meigs JB, Altshuler D, Boehnke M and McCarthy MI. Large-scale association analysis provides insights into the genetic architecture and pathophysiology of type 2 diabetes. Nat Genet 2012; 44: 981-990.

[27] Osawa H, Ochi M, Tabara Y, Kato K, Yamauchi J, Takata Y, Nishida W, Onuma H, Shimizu I, Fujii Y, Miki T, Ohashi J and Makino H. Serum resistin is positively correlated with the accumulation of metabolic syndrome factors in type 2 diabetes. Clin Endocrinol (Oxf) 2008; 69: 74-80.

[28] Staiger H, Machicao F, Kantartzis K, Schafer SA, Kirchhoff K, Guthoff M, Silbernagel G, Stefan N, Fritsche A and Haring HU. Novel meta-analysis-derived type 2 diabetes risk loci do not determine prediabetic phenotypes. PLoS One 2008; 3: e3019.

[29] Schleinitz D, Tonjes A, Bottcher Y, Dietrich K, Enigk B, Koriath M, Scholz GH, Bluher M, Zeggini E, McCarthy MI, Kovacs P and Stumvoll M. Lack of significant effects of the type 2 diabetes susceptibility loci JAZF1, CDC123/CAMK1D, NOTCH2, ADAMTS9, THADA, and TSPAN8/LGR5 on diabetes and quantitative metabolic traits. Horm Metab Res 2010; 42: 14-22.

[30] Bao XY, Peng B and Yang MS. Replication study of novel risk variants in six genes with type 2 diabetes and related quantitative traits in the Han Chinese lean individuals. Mol Biol Rep 2012; 39: 2447-2454.

[31] Long J, Edwards T, Signorello LB, Cai Q, Zheng W, Shu XO and Blot WJ. Evaluation of genome-wide association study-identified type 2 diabetes loci in African Americans. Am J Epidemiol 2012; 176: 995-1001.

[32] Arita Y, Kihara S, Ouchi N, Takahashi M, Maeda K, Miyagawa J, Hotta K, Shimomura I, Nakamura T, Miyaoka K, Kuriyama H, Nishida M, Yamashita S, Okubo K, Matsubara K, Muraguchi M, Ohmoto Y, Funahashi T and Matsuzawa Y. Paradoxical decrease of an adipose-specific protein, adiponectin, in obesity. 1999. Biochem Biophys Res Commun 2012; 425: 560-564.

[33] Hotta K, Funahashi T, Arita Y, Takahashi M, Matsuda M, Okamoto Y, Iwahashi H, Kuriyama H, Ouchi N, Maeda K, Nishida M, Kihara S, Sakai N, Nakajima T, Hasegawa K, Muraguchi M, Ohmoto Y, Nakamura T, Yamashita S, Hanafusa T and Matsuzawa Y. Plasma concentrations of a novel, adipose-specific protein, adiponectin, in type 2 diabetic patients. Arterioscler Thromb Vasc Biol 2000; 20: 1595-1599.

[34] Li S, Shin HJ, Ding EL and van Dam RM. Adiponectin levels and risk of type 2 diabetes: a systematic review and meta-analysis. JAMA 2009; 302: 179-188.

[35] Al-Daghri NM, Al-Attas OS, Alokail MS, Alkharfy KM, Hussain T, Yakout S, Vinodson B and Sabico S. Adiponectin gene polymorphisms (T45G and G276T), adiponectin levels and risk for metabolic diseases in an Arab population. Gene 2012; 493: 142-147.

Supplementary Table 1. General Characteristics of T2DM Associated SNPs

SNP	Chromosome	Nearest Gene	Risk Allele
rs10923931	1	NOTCH2	T
rs11899863	2	THADA	C
rs243021	2	BCL11A	A
rs3923113	2	GRB14	A
rs7578326	2	IRS1	A
rs13081389	3	PPARG	A
rs6795735	3	ADAMTS9	C
rs16861329	3	ST6GAL1	G
rs1801214	4	WFS1	T
rs4457053	5	ZBED3	G
rs10440833	6	CDKAL1	A
rs849134	7	JAZF1	A
rs972283	7	KLF14	G
rs896854	8	TP53INP1	T
rs3802177	8	SLC30A8	G
rs10965250	9	CDKN2A/B	G
rs13292136	9	CHCD9	C
rs12779790	10	CDC123	G
rs1802295	10	VPS26A	A
rs5015480	10	HHEX	C
rs7903146	10	TCF7L2	T
rs231362	11	KCNQ1	G
rs163184	11	KCNQ1	G
rs5215	11	KCNJ11	C
rs1552224	11	CENTD2	A
rs1387153	11	MTNR1B	T
rs1531343	12	HMGA2	C
rs4760790	12	TSPAN8	A
rs7957197	12	HNF1A	T
rs7178572	15	HMG20A	G
rs11634397	15	ZFAND6	G
rs2028299	15	AP3S2	C
rs8042680	15	PRC1	A
rs11642841	16	FTO	A
rs4430796	17	HNF1B	G
rs4812829	20	HNF4A	A
rs5945326	23	DUSP9	A