American Journal of Epidemiology
© The Author 2010. Published by Oxford University Press on behalf of the Johns Hopkins Bloomberg School of Public Health. This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (http://creativecommons.org/licenses/by-nc/2.5), which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.
Vol. 171, No. 6
DOI: 10.1093/aje/kwp450
Advance Access publication:
February 23, 2010

Human Genome Epidemiology (HuGE) Review

The Association Between the Peroxisome Proliferator-Activated Receptor-γ2 (*PPARG2*) Pro12Ala Gene Variant and Type 2 Diabetes Mellitus: A HuGE Review and Meta-Analysis

Hebe N. Gouda*, Gurdeep S. Sagoo, Anne-Helen Harding, Jan Yates, Manjinder S. Sandhu, and Julian P. T. Higgins

* Correspondence to Hebe N. Gouda, Department of Public Health and Primary Care, Institute of Public Health, University of Cambridge, Forvie Site, Robinson Way, Cambridge CB2 0SR, United Kingdom (e-mail: hng20@medschl.cam.ac.uk).

Initially submitted June 15, 2009; accepted for publication December 15, 2009.

The peroxisome proliferator-activated receptor-γ gene (*PPARG*) has been implicated in the etiology of type 2 diabetes mellitus and has been investigated in numerous epidemiologic studies. In this Human Genome Epidemiology review, the authors assessed this relation in an updated meta-analysis of 60 association studies. Electronic literature searches were conducted on September 14, 2009. Population-based cohort, case-control, cross-sectional, or genome-wide association studies reporting associations between the *PPARG* Pro12Ala gene variant (rs1801282) and type 2 diabetes were included. An updated literature-based meta-analysis involving 32,849 type 2 diabetes cases and 47,456 controls in relation to the *PPARG* Pro12Ala variant was conducted. The combined overall odds ratio, calculated by per-allele genetic model random-effects meta-analysis for type 2 diabetes and the Pro12Ala polymorphism, was 0.86 (95% confidence interval: 0.81, 0.90). The analysis indicated a moderate level of heterogeneity attributable to genuine variation in gene effect size ($I^2 = 37\%$). This may reflect the variation observed between ethnic populations and/or differences in body mass index. Work on *PPARG* Pro12Ala should now focus on the observed heterogeneity in the magnitude of the association between populations. Further investigations into gene-gene and gene-environment interactions may prove enlightening.

diabetes mellitus, type 2; epidemiology; genetics; genome, human; meta-analysis; PPAR gamma; review

Abbreviations: BMI, body mass index; CI, confidence interval; EPIC, European Prospective Investigation Into Cancer and Nutrition; HuGE, Human Genome Epidemiology; PPARG, peroxisome proliferator-activated receptor-γ; SNP, single nucleotide polymorphism.

***Editor's note**: This article also appears on the Web site of the Human Genome Epidemiology Network (http://www.hugenet.org.uk/index.html).*

BACKGROUND

Gene/gene variants

The peroxisome proliferator-activated receptors (PPAR-α, PPAR-δ, and PPAR-γ) are transcription factors belonging to the nuclear hormone receptor superfamily (1). These receptors combine with the retinoid X receptors to form heterodimers that regulate various genes involved in lipid and glucose metabolism, fatty acid transport, adipocyte differentiation, carcinogenesis, and inflammation (2–4). Here we consider the relation between the peroxisome proliferator-activated receptor-γ2 (*PPARG2*) gene and type 2 diabetes.

The *PPARG* gene is located on chromosome 3p25 (OMIM number 601487) and encodes a nuclear transcription factor involved in the expression of hundreds of genes. The *PPARG* gene contains 9 exons, spans more than 100 kilobases, and, because of alternative mRNA splicing, results in the production of 2 protein isoforms, PPARG1 and PPARG2 (5). *PPARG1* is encoded by 8 exons, using

exons 1–6, A1, and A2, with *PPARG2* being encoded using exons 1–6 and B. While PPARG1 is found ubiquitously in the body, PPARG2 is largely found in adipose tissue and the large intestine (5). Several variants in the *PPARG* gene have been identified, with the Pro12Ala variant having been the most extensively examined in epidemiologic studies. The *PPARG* gene locus and gene variants have previously been addressed in a Human Genome Epidemiology (HuGE) review on the genetics of leptin and obesity (6). The association between the Pro12Ala variant (rs1801282) and type 2 diabetes has been the focus of several meta-analyses (Table 1).

Gene variant frequency

Yen et al. (7) first identified a missense mutation resulting in the alanine substitution for proline at codon 12 of the *PPARG* gene. The frequency of the 12Ala allele has been found to range from 2% to 18% in healthy people (6). We estimated Pro12Ala allele frequencies from the control groups of all studies identified for inclusion in the present review (see Web Table 1, which is posted on the *Journal*'s Web site (http://aje.oxfordjournals.org/)). Across all studies, the frequency of the 12Ala allele in control groups ranged from 1.7% to 21.6% (median, 9.5%). In the studies reporting controls as ethnically Caucasian, the 12Ala allele frequency ranged from 5.9% to 21.6% (median, 12.7%). In studies reporting controls as ethnically of East Asian descent (such as Chinese or Japanese), the 12Ala allele frequency ranged from 1.7% to 9.3% (median, 4.5%).

Disease

By the year 2030, diabetes mellitus is expected to affect almost 5% of the world's population—an estimated 366 million people (8). The vast majority of these cases will be type 2 diabetes, with current diagnoses currently accounting for approximately 90%–95% of diabetes cases. Diabetes prevalence is currently about 3% in the general population, with increased prevalence in some ethnic minority groups (such as South Asians, Africans, Afro-Caribbeans, and Chinese), as well as in less affluent populations, including those that are less physically active, have central obesity, and have a high body mass index (BMI; weight (kg)/height $(m)^2$). Some populations, such as Pima Indians, Australian Aboriginal communities, and Pacific and Indian Ocean Islanders, have a far higher prevalence, at up to 40% (9). The presence of diabetes mellitus is associated with a range of vascular complications (such as myocardial infarction, stroke, heart failure, renal failure, angina, and retinopathy) leading to a reduced life expectancy and a reduced quality of life (10). The large and increasing burden of type 2 diabetes and the potential for modifying risk through adequate treatments and lifestyle alterations make the identification of methods for the early detection of persons at greater risk an important public health challenge.

Knowledge of genetic polymorphisms that allow accurate quantification of type 2 diabetes risk will allow the development of complex models with diagnostic and prognostic potential. Several extensive reviews have summarized findings on the genetic epidemiology of type 2 diabetes (11–13). However, substantial advances have been made in the past few years, with an increase in confirmed type 2 diabetes susceptibility loci from 3 genes (*PPARG*, *KCNJ11*, and *TCF7L2*) to almost 20 (with newly identified variants in the following genes: *ADAMTS9*, *CDKAL1*, *CDKN2A/B*, *CAMK1D*, *FTO*, *HHEX/IDE*, *HNF1B*, *IGF2BP2*, *JAZF1*, *MTNR1B*, *NOTCH2*, *SLC30A8*, *THADA*, *TSPAN8*, and *WFS1*) (14–17). This major step forward has been due mainly to advances in technology, with genome-wide association scans now allowing unprecedented progress to be made in the understanding of the genetic etiology of several complex diseases, including type 2 diabetes. It is increasingly evident that the genetic basis of type 2 diabetes involves multiple genes that each have a modest effect size on diabetes susceptibility and that interaction with other susceptibility loci and/or environmental factors may result in more substantial effects.

Objectives

The association between the *PPARG2* Pro12Ala gene variant and type 2 diabetes has been investigated in numerous epidemiologic studies since it was first suggested in 1998 that carriers of the 12Ala variant showed a 75% reduction in risk of type 2 diabetes (see Web reference 1 (W1), which is listed in Web Appendix 1 on the *Journal*'s Web site (http://aje.oxfordjournals.org/)). Here we evaluate the evidence for an association between the *PPARG2* Pro12Ala (rs1801282) gene variant and type 2 diabetes using methods developed by the Human Genome Epidemiology Network and the Cochrane Collaboration (18–20). We report results from an updated meta-analysis of 60 association studies, involving a total of 32,849 type 2 diabetes cases and 47,456 controls in relation to the Pro12Ala gene variant in the *PPARG* gene. The present report primarily focuses on population association studies (including case-control, cohort, and genome-wide association studies but excluding family-based studies) and contains an investigation of potential sources of heterogeneity.

MATERIALS AND METHODS

Selection criteria and identification of studies

Eligible for inclusion were all population-based cohort, case-control, or cross-sectional studies reporting on associations between the *PPARG2* Pro12Ala variant and type 2 diabetes. We performed electronic searches, not limited to the English language, of Medline (using PubMed), EMBASE, HuGE Navigator, Web of Science, and the Science Citation Index using the search method described in Web Appendix 2 (http://aje.oxfordjournals.org/). The latest searches were undertaken on September 14, 2009. All relevant articles identified through the search were scanned on the basis of title, keywords, and abstract (where available) by one of us and were rejected in the initial screening if the article clearly did not meet the inclusion criteria. Where a title/abstract could not be rejected with certainty, we obtained the full text of the

Table 1. Findings From Previously Conducted Meta-Analyses of the Peroxisome Proliferator-Activated Receptor-γ2 (*PPARG2*) Pro12Ala Gene Variant and Type 2 Diabetes, 2000–2007

Study and Year (Ref.)	Study Details	No. of Studies	No. of Cases	No. of Controls	Main Analyses		Subanalyses		
					OR	95% CI	Subgroup	OR	95% CI
Altshuler et al., 2000 (30)	Very little detail presented.	8	NS	NS	0.79	0.70, 0.89[a]	NS		
Ek et al., 2001 (31)	Meta-analysis (using Mantel-Haenszel method) of published case-control studies.	10	3,032	3,812	0.81	0.72. 0.91	Caucasian participants	0.85	0.76, 0.96
							East Asian participants	0.42	0.26, 0.67
Lohmueller et al., 2003 (34)	Meta-analysis (using both fixed-effect and random-effects methods) of published case-control studies.	14	NS	NS	1.22[b]	1.08, 1.37	Random effects[b]	1.21	1.07, 1.37
Hara et al., 2003 (33)	Meta-analysis in a Japanese publication.	8	NS	NS	0.81[c]	0.78, 0.93[c]	NS		
Parikh and Groop, 2004 (35)	Meta-analysis (using Mantel-Haenszel method) of published studies.	23	NS	NS	0.79	NS	NS		
Vardarli, 2007 (32)	Abstract of meta-analysis (using DerSimonian and Laird (24) random-effects method) of published studies.	33	14,771	14,513	0.86	0.75, 0.97	NS		
Ludovico et al., 2007 (29)	Meta-analysis (using random-effects method and dominant genetic model) of published and unpublished studies.	43	19,250	23,660	0.81	0.75, 0.88	East Asian participants	0.65	0.54, 0.79
							North American participants	0.82	0.67, 1.01
							European participants	0.86	0.79, 0.95
							Northern Europeans	0.74	0.66, 0.83
							Central Europeans	0.90	0.82, 1.00
							Southern Europeans	1.01	0.80, 1.28

Abbreviations: CI, confidence interval; NS, not stated; OR, odds ratio.

[a] The 95% CI was estimated using the *P* value ($P = 0.0007$) from the paper by Altshuler et al. (30).

[b] The risk allele was defined as Pro12.

[c] The OR and 95% CI were estimated from Figure 9 in the paper by Hara et al. (33).

article for evaluation. We also reviewed the reference lists of retrieved articles to identify other relevant publications.

In addition, we searched specifically for genome-wide association studies of type 2 diabetes using the catalog of published genome-wide association studies (21). The latest searches were undertaken on September 14, 2009. The full text and any supplementary materials were collected for each study identified. Data for the Pro12Ala single nucleotide polymorphism (SNP) (rs1801282) were extracted from each report, and study authors were contacted if insufficient data were reported.

Data collection

The following data were extracted independently by at least 2 investigators, using a piloted data extraction form (with any discrepancies being resolved by discussion and, when necessary, adjudicated by a third reviewer): genotype frequencies by case/control status; mean ages of cases and controls; proportions of males and ethnic subgroups (defined as people of European continental ancestry, East Asian ancestry, or other); genotyping methods; and blinding of laboratory workers to participant case/control status. We calculated allele frequencies from control groups of studies that presented data on all 3 genotype groups, assuming Hardy-Weinberg equilibrium where appropriate. Where there were multiple publications from the same study group, we extracted data from each report and selected only the most complete and up-to-date data. When articles presented data for different ethnic groups or different case or control sources, results for the subgroups were considered as separate studies. Where data could not be extracted for inclusion in the meta-analysis, the investigators were contacted via letter and e-mail. Selected articles also included 4 Chinese-language publications, from which data were available from the English-language abstract (W2–W5).

Data analysis

The primary analyses for which results are presented were conducted using a per-allele inheritance model. This was the model favored by an initial analysis using the inheritance-model-free analysis of Minelli et al. (22) (data not shown). We estimated the per-allele odds ratio using logistic regression within each study. We used funnel plots and associated tests to assess assumptions involved in meta-analysis and to explore the relation between precision and magnitude of association (23). The meta-analyses used a standard approach, weighting by precision and incorporating random effects to allow for the variation in true associations across studies (24). We performed a cumulative meta-analysis to demonstrate how evidence concerning the genetic association has evolved over time.

Consistency of the gene effect sizes across studies was assessed using a test for heterogeneity and the I^2 statistic, which describes the percentage of total variation in point estimates attributable to genuine variation rather than sampling error (25). We further explored this variation by prespecified subgrouping of studies according to sample size (<100, 100–499, or ≥ 500), ethnicity (Caucasian or East Asian), source of controls (general population or hospital), study design (retrospective or prospective), and blinding of genotyping to clinical outcome (yes, no, or unknown). We used random-effects meta-regression to explore the extent to which these subgroups could explain the between-study variance. Fixed-effect meta-analyses were conducted as sensitivity analyses. All ranges presented are 95% confidence intervals unless otherwise specified.

Only summary meta-analysis results were available from Zeggini et al. (26). Their analysis used imputed and genotyped data for the *SYN2*/*PPARG* region, including either the rs17036101 SNP or the rs1801282 SNP, as these 2 SNPs represent the same association signal. Using the rs17036101 SNP as a proxy for rs1801282, we combined a random-effects meta-analysis result from Zeggini et al. (26) with a random-effects meta-analysis of nonoverlapping studies from our own searches. We first computed the summary log odds ratio and its variance for each data source and then computed the total sum of weights and the total sum of (weight $\times$ log odds ratios) across both sources, and used these to produce an overall summary estimate and variance. This is equivalent to a standard random-effects meta-analysis, with the exception that an estimate of between-study variance among Zeggini et al.'s (26) studies is used in the weights for those studies and an estimate of between-study variance among our studies is used in the weights for our studies.

RESULTS

Characteristics of the included studies

Our literature searches yielded 1,734 reports, from which 74 eligible studies were identified, in addition to 30 genome-wide association studies (W1–W85) (Web Table 1 and Tables 2 and 3). Sixty-six studies (Web Table 1) contributed to the present review (W1–W7, W9–W14, W17, W19–W21, W23, W25, W27–W34, W36, W37, W40–W45, W47–W54, W56, W59, W61, W63, W65, W67, W68, W70, W72, W74, W79, W80, W83, W85). Nine studies from 7 papers (W35, W46, W55, W57, W77, W78, W81) (comprising approximately 7% of eligible cases and 10% of eligible controls) could not be included because of insufficient detail and lack of response from authors after our attempts at correspondence (Table 2). Researchers from the European Prospective Investigation Into Cancer and Nutrition (EPIC) provided unpublished data. Study-level characteristics for the EPIC study (27) and the EPIC nested case-control data have been previously published (supplementary materials of Zeggini et al. (26)); summary details are available in Web Table 2 (http://aje.oxfordjournals.org/) and Web Appendix 3 (http://aje.oxfordjournals.org/). We also identified 30 genome-wide association studies of type 2 diabetes in 16 papers published since 2007 (W18, W22, W24, W26, W58, W60, W62, W64, W66, W69, W71, W73, W75, W76, W82, W84) (see Table 3 and Web Table 3 (http://aje.oxfordjournals.org/)). Each of these studies included either the Pro12Ala variant (rs1801282) or a nearby tagging SNP. Zeggini et al. (26) presented results from a meta-analysis of 59,682 participants across several of these genome-wide association studies (W62, W64, W73, W84).

Table 2. Characteristics of Genetic Association Studies of the Peroxisome Proliferator-Activated Receptor-γ2 (*PPARG2*) Pro12Ala Gene Variant and Type 2 Diabetes That Were Potentially Eligible for Inclusion in the Current Analysis but Did Not Have Full Genotype Data Available, 2000–2006

Study and Year (Ref.)	No. of Cases	No. of Controls	% Male		Mean Age, years (SD)		Diagnostic Criteria	Molecular Technique	HWE Testing	Control Source	Study Design	Ethnicity/Geographic Location
			Cases	Controls	Cases	Controls						
Li, 2000 (W35)	17	138	0.0	0.0	NS	NS	NS	Probes	In HWE	Extremely obese participants	C-C	Caucasian/ United States
Mirzaei et al., 2009 (W46)	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	C-C	Caucasian/Iranian
Poulsen et al., 2003 (W55)	32	188	NS	NS	NS	NS	NS	RFLP	In HWE	Population-based twin study	Population-based twin study	Caucasian/Denmark
Radha et al. (A), 2006 (W57)	799	820	43.6	36.5	52 (11)	41 (13)	WHO	RFLP	In HWE	GP	C-C	South Asian/India
Radha et al. (B), 2006 (W57)	81	616	56.8	58.9	56 (10)	42 (13)	WHO	RFLP	In HWE	GP	C-C	South Asian/India
Radha et al. (C), 2006 (W57)	123	334	71.5	50.3	59 (8)	37 (14)	WHO	RFLP	In HWE	GP	C-C	Caucasian/United States
Vieira-Filho et al., 2004 (W77)	NS	NS	NS	NS	NS	NS	WHO	RFLP	In HWE	GP	Cohort	Parkataje Indian/Brazil
Wakil et al., 2006 (W78)	1,137	219	NS	NS	NS	NS	WHO	Probes	NS	GP—normal glucose tolerance	C-C	Saudi Arabian/ Saudi Arabia
Yanagisawa et al., 2001 (W81)	NS	NS	NS	NS	NS	NS	WHO	RFLP	NS	GP	Cohort	Palauan/Palau

Abbreviations: C-C, case-control; GP, general population; HWE, Hardy-Weinberg equilibrium; NS, not stated; RFLP, restriction fragment length polymorphism; SD, standard deviation; WHO, World Health Organization.

The identified studies were published between 1998 and 2008 and were undertaken in a wide range of geographic settings, with 66% (21,670 of 32,849) of cases having white European continental ancestry (Caucasian), 25% (8,187 of 32,849) being East Asian, and 9% (2,992 of 32,849) having other ethnic origins (including African-American, American Indian, or not stated). Nine studies were described as prospective in design, 3 were described as cross-sectional studies, 8 as genome-wide association studies, and 47 as case-control studies. Thirty-one studies involved general population-based controls, 4 involved hospital-based controls, and 1 involved health-care employees; in 13 studies, investigators did not describe the source of their controls (see Web Table 1). In 32 studies, authors reported using World Health Organization criteria for diagnosis of type 2 diabetes (28). Restriction fragment length polymorphism analysis was the most common method of genotyping used. In 23 studies, authors did not report any information on the BMI of the case group (see Web Table 1). In 11 studies, investigators reported information regarding age at disease onset. Of the 67 included studies, 60 provided data for the per-allele genetic model in our presented analyses (W1–W4, W6, W7, W9–W14, W17–W21, W25–W33, W36, W37, W40–W45, W47–W50, W52–W54, W56, W59, W61, W63–W65, W67, W71, W74, W84, W85).

Two studies were found to deviate from Hardy-Weinberg equilibrium according to the test P value ($P < 0.05$) (W2, W5). Approximately 70% of the studies had fixation coefficients with absolute values larger than 0.03 (median, −0.004; range, −0.072 to 0.185), with the studies evenly distributed around zero. Authors in 42 studies reported testing for Hardy-Weinberg equilibrium and found no deviation (see Web Table 1 and Table 2).

Associations

In a random-effects meta-analysis with a per-allele genetic model, the combined type 2 diabetes odds ratio for the Pro12Ala polymorphism across the 66 studies was 0.86 (95% confidence interval (CI): 0.81, 0.90) (Web Figure 1 (http://aje.oxfordjournals.org/)). The fixed-effect meta-analysis was 0.85 (95% CI: 0.82, 0.88). The cumulative meta-analysis illustrates the exaggerated effect often observed in the earliest study and reveals that the accumulated evidence hovered around the conventional 5% significance level until 2004. In studies published between 2004 and 2008, the overall P value was reduced from 0.028 to 1.3×10^{-8}.

There was evidence of a moderate degree of inconsistency among these studies ($I^2 = 37\%$, 95% CI: 9, 54; $P = 0.0028$), and no evidence of funnel plot asymmetry was found either visually or using the Harbord test (23) ($P = 0.87$). Study participant ethnicity accounted for some of this heterogeneity (14% of the between-study variance). Results from subgroup analyses are presented in Figure 1. When we considered only the large (>500 cases) studies, the odds ratio was 0.84 (95% CI: 0.79, 0.90), with $I^2 = 43\%$ (95% CI: 0, 66; $P = 0.025$). When studies were subgrouped by ethnicity, the odds ratio was nearer 1 for Caucasians (odds ratio = 0.86 (95% CI: 0.81, 0.90), $I^2 = 37\%$ (95% CI: 0, 57; $P = 0.019$)) than for East Asians (odds ratio = 0.78 (95%

Table 3. Characteristics of the Genome-Wide Association Studies Conducted for Type 2 Diabetes, 2007–2009

Study and Year (Ref.)	Ethnicity/ Location	Cases						Controls				
		No. of Subjects			Age at Onset, years (SD)	Mean Age, years (SD)	BMI[a] (SD)	No. of Subjects			Mean Age, years (SD)	BMI (SD)
		Total	Males	Females				Total	Males	Females		
Takeuchi et al., 2009 (W71)	Japan	1,629	928	701	NS	Stage 1: 66.6 (9.8) Stage 2: 62.7 (11.7)	Stage 1: 24.5 (3.6) Stage 2: 23.3 (3.9)	1,517	806	711	Stage 1: 64.7 (6.8) Stage 2: 71.1 (9.6)	Stage 1: 23.3 (3.1) Stage 2: 23.0 (3.1)
Timpson et al., 2009 (W75)—see Zeggini et al., 2007 (W75)	United Kingdom											
Herder et al., 2008 (W26)	Germany	433	255	178	NS	65.2 (8.3)	30.9 (5)	1,438	693	745	61.9 (10.2)	27.7 (4.3)
Unoki et al. (A), 2008 (W76)	Japan	5,149	3,112	2,037	NS			4,176	1,988	2,188		
Unoki et al. (B), 2008 (W76)	Singapore	1,498	736	762	NS	63.9 (9.7)	25.3 (3.9)	1,881	828	1,053	35.4 (11.2)	22 (3.4)
Unoki et al. (C), 2008 (W76)	Denmark	4,085	2,423	1,662	NS	60 (9.8)	30.6 (5.6	5,302	2,456	2,846	46.9 (9.1)	25.6 (4)
Yasuda et al., 2008 (W82)	Japan											
Sladek et al., 2007 (W66)—initial sample set	France	1,380	838	542	45.0 (8.4)	60.0 (10.3)	25.8 (2.8)	1,323	532	791	53.4 (5.6)	23.2 (1.8)
Sladek et al., 2007 (W66)—replication sample set	France	2,617	1,628	989	50.4 (11.0)	62.2 (11.0)	28.9 (3.6)	2,894	1,240	1,654	56.4 (10.2)	25.3 (3.5)
Zeggini et al., 2007 (W84)—initial sample set	United Kingdom	1,924	1,118	806	50.3 (9.2)	58.6 (10.1)	NS	2,938	1,446	1,492	NS	NS
Zeggini et al., 2007 (W84)—replication sample set	United Kingdom	3,757	2,137	1,620	54.3 (9.4)	63.1 (9.7)	NS	5,346	2,719	2,627	49.9 (16.4)	NS
Steinthorsdottir et al., 2007 (W69)—initial sample set	Iceland	1,399	832	567	56.2 (12.3)	64.4 (12.8)	56.2 (12.3) [688][b]	5,275	2,743	2,532	58.3 (17.4)	27.1 (4.9) [1,751]
Steinthorsdottir et al., 2007 (W69)—replication sample set	Denmark	1,359	821	538	52.1 (10.3) [1,013]	56.8 (10.5)	29.7 (5.3) [1,343]	4,825	2,249	2,576	46.4 (8.8)	25.5 (4.1) [4,824]
Scott et al., 2007 (W64)—initial sample set	Finland	1,161	653	508	53	63.4	29.8	1,174	574	600	64	26.8
Scott et al., 2007 (W64)—replication sample set	Finland	1,215	724	491	56	60	30.1	1,258	768	490	59	26.4
Saxena et al., 2007 (W62)—initial sample set	Finland	1,007	513	494	55.5 (9.7) [946]	63.4 (9.9)	28.8 (4.6) [998]	1,038	498	540	59.0 (10.2)	26.8 (3.8) [1,032]
Saxena et al., 2007 (W62)—initial sample set	Sweden	457	228	229	59.9 (10.7)	66.0 (10.7)	28.0 (4.2)	429	209	220	58.3 (9.9)	26.4 (3.7) [420]
Saxena et al., 2007 (W62)—replication sample set	Sweden	2,830	1,667	1,163	NS	59.0 (12.0)	29.6 (5.5)	3,550	1,340	2,210	57.0 (6.0)	25.1 (3.6)
Saxena et al., 2007 (W62)—replication sample set	European descent/ United States	1,226	644	582	NS	63.0 (11.0)	32.9 (6.9)	1,226	644	582	61.0 (10.0)	27.4 (5.2)

Saxena et al., 2007 (W62)—replication sample set	Poland	1,009	422	587	NS	62.0 (10)	29.6 (4.8)	1,009	422	587	59.0 (7.0)	26.1 (3.6)
Wellcome Trust Case-Control Consortium, 2007 (W73)—see Zeggini et al., 2007 (W73)	United Kingdom	1,924						2,938				
Salonen et al., 2007 (W60)—initial sample set	White/ European	500	228	272	NS	NS	NS	497	NS	NS	NS	NS
Salonen et al., 2007 (W60)—replication sample set	White/ European	2,573	NS	NS	NS	NS	NS	2,776	NS	NS	NS	NS
Rampersaud et al., 2007 (W58)—initial sample set	Amish/ United States	124	41	83	NS	51.3 (10.5)	29.3 (5.8)	295	153	142	64.4 (12.9)	27.4 (4.7)
Rampersaud et al., 2007 (W58)—replication sample set	Amish/ United States							427	200	227	51.9 (11.9)	27.7 (5.0)
Hayes et al., 2007 (W24)	Mexican-American	281	108	173	45.9 (10.1)	57.9 (10.7)	31.5 (6.2)	280	69	211	NS	NS
Hanson et al., 2007 (W22)—initial sample set	American Indian	300	114	186	19.2 (4.5)	NS	38.9 (8.4)	334	160	174	55.5 (9.8)	35.4 (8.0)
Hanson et al., 2007 (W22)—replication sample set	American Indian	1,207	459	748	39.7 (10.6)	NS	38.3 (8.2)	1,627	748	879	27.7 (11.6)	35.6 (8.2)
Florez et al., 2007 (W18)—initial sample set	United States	91	NS	NS	NS	NS	NS	1,087	527	560	51.5 (9.8) [1,032]	27.5 (5.2) [1,026]
Florez et al., 2007 (W18)—replication sample set	United States	158	NS	NS	NS	NS	NS	1,465	691	774	56.1 (9.3) [1,390]	27.4 (4.8) [1,384]

Abbreviations: BMI, body mass index; NS, not stated; SD, standard deviation.
[a] Weight (kg)/height $(m)^2$.
[b] Numbers in brackets, number of subjects.

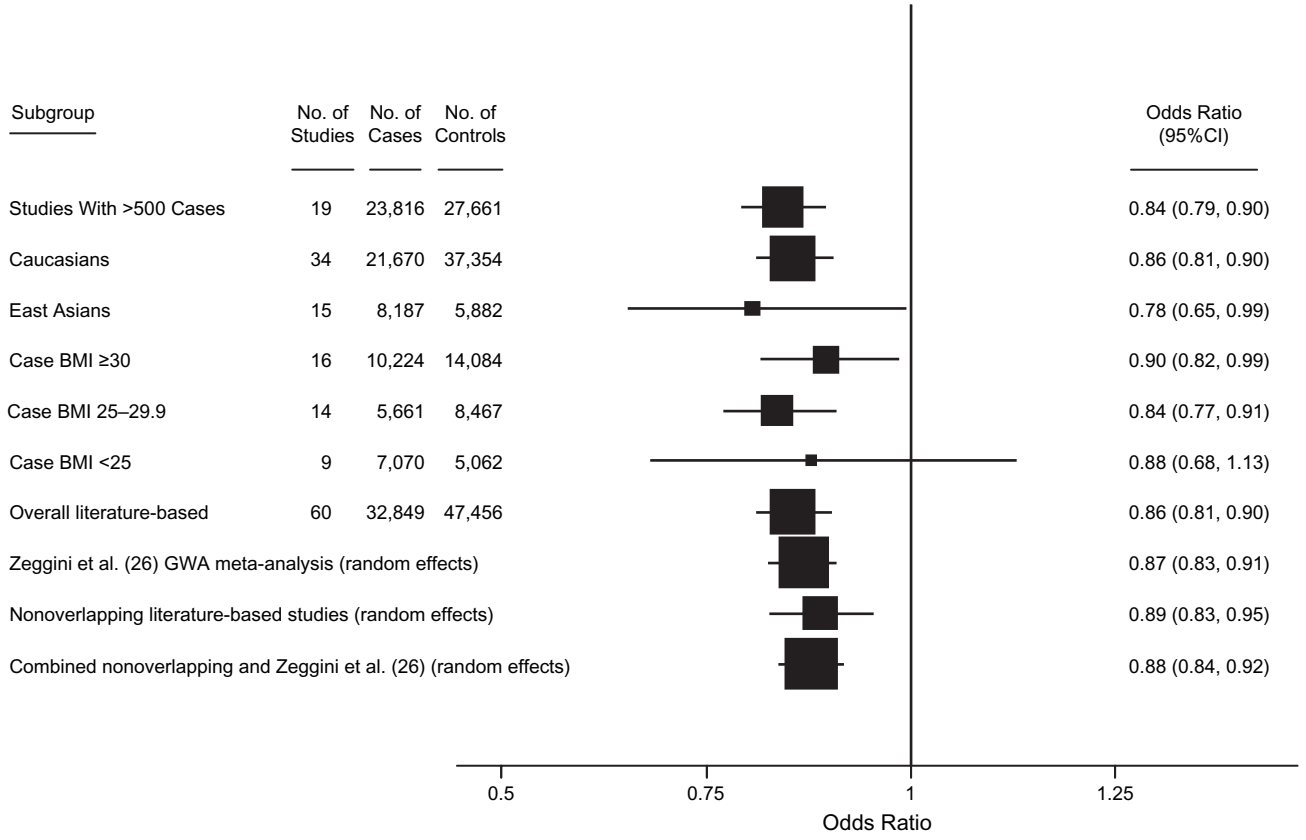

Figure 1. Results from random-effects meta-analyses of studies of the peroxisome proliferator-activated receptor-γ2 (*PPARG2*) Pro12Ala gene variant and type 2 diabetes according to various study-level characteristics. BMI, body mass index (weight (kg)/height (m)^2); CI, confidence interval; GWA, genome-wide association.

CI: 0.65, 0.99), $I^2 = 45\%$ (95% CI: 0, 69; $P = 0.031$)), although the difference was not statistically significant ($P = 0.38$). A dominant analysis of all studies composed of Asian populations produced an odds ratio of 0.80 (95% CI: 0.66, 0.97), with $I^2 = 40.5\%$ (95% CI: 0, 64).

The combined odds ratio for studies with a mean case BMI less than 25 was 0.88 (95% CI: 0.68, 1.13), with $I^2 = 57\%$ (95% CI: 0, 78; $P = 0.016$). Studies with a mean case BMI between 25 and 29.9 had an odds ratio of 0.84 (95% CI: 0.77, 0.91), with $I^2 = 0\%$ (95% CI: 0, 47; $P = 0.45$). Studies with a mean case BMI greater than or equal to 30 had an odds ratio of 0.90 (95% CI: 0.82, 0.99), with $I^2 = 46\%$ (95% CI: 0, 68; $P = 0.024$). A test for trend across these 3 groups was not statistically significant ($P = 0.49$).

No statistically significant differences were observed according to study design. Forty case-control studies had an odds ratio of 0.85 (95% CI: 0.79, 0.92), with $I^2 = 44\%$; 9 prospective studies had an odds ratio of 0.88 (95% CI: 0.74, 1.04), with $I^2 = 35\%$; and the 8 genome-wide association studies had an odds ratio of 0.84 (95% CI: 0.79, 0.89), with $I^2 = 0\%$. Only 3 cross-sectional studies were available. Only in 10 studies did authors report blinding, and among those studies that did not report blinding status, the pooled odds ratio was 0.86 (95% CI: 0.77, 0.97), with $I^2 = 63$.

We also investigated potential variation arising from the use of different sources of controls. In 42 studies, investigators reported the use of a general population cohort as their control group; the combined odds ratio was 0.86 (95% CI: 0.81, 0.91), with $I^2 = 38\%$. In only 4 studies did authors report the use of hospital-based controls; in those studies, the odds ratio was 1.09 (95% CI: 0.86, 1.37), with $I^2 = 0\%$ ($P = 0.06$ in comparison with general population controls), while in those studies for which authors did not state the source of their controls ($n = 11$), the pooled odds ratio was 0.77 (95% CI: 0.64, 0.93), with $I^2 = 21.1\%$ ($P = 0.26$ in comparison with general population controls).

We combined the random-effects meta-analysis of Zeggini et al. (26) with our own random-effects meta-analysis of nonoverlapping published literature-based studies to produce an overall pooled odds ratio of 0.88 (95% CI: 0.84, 0.92) involving a total of 116,040 participants (see Figure 1).

DISCUSSION

Main findings

This HuGE association review involved an updated meta-analysis of the relation between the *PPARG* Pro12Ala

polymorphism and type 2 diabetes among 32,849 cases and 47,456 controls in 60 studies (counting every study's cases and controls only once). The *PPARG* 12Ala polymorphism was associated with a reduction in type 2 diabetes risk (odds ratio = 0.86, 95% CI: 0.81, 0.90), and this confirms findings from previous meta-analyses (29–35). When only the largest studies (>500 cases) were considered, the association remained stable (odds ratio = 0.84, 95% CI: 0.79, 0.90). This work further confirms the association between the *PPARG* Pro12Ala polymorphism and type 2 diabetes.

The results of the Zeggini et al. (26) meta-analysis of genome-wide association studies were compatible with our own literature-based meta-analysis. The Zeggini et al. analysis provided an estimate that minimized publication bias because of the availability of genotype data from a consortium-based approach. We pooled this estimate with our own estimate from nonoverlapping published literature association studies to produce an estimate for 116,040 participants. Proposed guidelines for assessing the strength of evidence from gene-disease association studies (36) would designate the findings for Pro12Ala and type 2 diabetes as "strong" evidence (see Web Table 4 (http://aje.oxfordjournals.org/)).

Limitations

When interpreting these results, the potential limitations of such a report should be considered. First, the key threat to any literature-based review and meta-analysis is that of reporting bias (where only the most exciting findings are available in the published literature). Although our own assessments did not generally suggest the presence of material publication bias, it is not possible to rule it out entirely (37, 38). Given the consistency of findings between our main analysis and our analysis of only the large (>500 cases) studies, which should have been less prone to selective reporting, we may place reasonable confidence in our observations. Our findings were also consistent with the results from Zeggini et al.'s meta-analysis of genome-wide association studies (26).

Consistency across studies

Overall, I^2 was estimated to be 37%, with statistically significant evidence of heterogeneity. This represents a moderate level of inconsistency attributable to genuine variation in gene effect size. It has been hypothesized in the past that this may reflect variation between ethnic populations. We observed a slightly higher magnitude of association in East Asians compared with Caucasians, but the difference was not statistically significant. Polymorphism frequencies are known to vary by ethnicity, but the effect of this on risk remains insufficiently studied. In the studies we identified, the frequency of the 12Ala polymorphism in controls was observed to be greater in Caucasian populations than in East Asian populations.

The observed difference in the magnitude of association between these populations may also be explained partly by differences in BMI. In a study of French Caucasians conducted by Ghoussaini et al. (W20), the presence of the Pro12 variant was observed to double the risk of type 2 diabetes in the obese subpopulation. However, the Ala12 variant has also been shown to be weakly associated with higher BMI, as confirmed in a meta-analysis conducted in 2003 by Masud et al. (39).

A meta-analysis conducted by Ludovico et al. (W37) found that the alanine polymorphism conferred significantly greater protection against type 2 diabetes among Asians than among Caucasians. Using 10 studies of Asian populations, their results revealed an odds ratio of 0.65 (95% CI: 0.54, 0.79). When the analysis was adjusted for BMI in the controls, this statistical significance was lost. Furthermore, taking into account body fat content and distribution, Radha et al. (W57) found no protective effect of the Pro12Ala polymorphism in South Asians.

We conducted a similar analysis of results from 20 relevant studies (using a dominant model) which showed an odds ratio of 0.80 (95% CI: 0.66, 0.97) and suggested no difference between Asians and Caucasians ($P = 0.59$). Studies in which cases had a lower mean BMI observed a slightly greater protective effect of the 12Ala polymorphism on type 2 diabetes risk, but this was not statistically significant ($P = 0.57$; see also Figure 1).

In 2001, Luan et al. (40) hypothesized a gene-nutrient interaction based on the ratio of polyunsaturated fats to saturated fats which determined the association between the Pro12Ala polymorphism and BMI. This study may contribute towards an explanation of the role played by ethnicity and differences in dietary habits. However, more recently, Robitaille et al. (41) suggested an interaction in the opposite direction, highlighting the need to carefully assess possible interactions.

Another potential source of variation has been identified by Hegele et al. (W25) and Radha et al. (W57): These authors noted differences in the association between Pro12Ala and type 2 diabetes between men and women. More recently, Ali et al. (W1), Morini et al. (42), and Mattevi et al. (43) observed a relation between the polymorphism and higher BMI in men which was absent in women. Since few investigators publish their data stratified by sex, we were unable to investigate these issues in detail. Although these investigations have been inconsistent in the past, differences in the phenotypic consequences of the Pro12Ala polymorphism among males and females warrant further investigation. Further studies of gene-gene and gene-environment interaction are necessary to better understand these relations.

Biology

The substitution from proline to alanine at codon 12 has been found to modulate transcriptional activity (44, 45). This substitution is close to the NH_2-terminus of the protein in the ligand-independent activation domain, the activity of which is potentiated through phosphorylation by insulin. The structure and consequently the function of the protein may be affected by this amino acid change, since alanine favors the formation of α-helices while proline prevents it (46). The alanine isoform leads to the less efficient stimulation of PPARG target genes and predisposes people to lower levels of adipose tissue mass accumulation, which

in turn may improve insulin sensitivity—thus supporting the protective effect observed for 12Ala carriers, since decreased insulin sensitivity plays a central role in the pathogenesis of type 2 diabetes.

Potential public health impact and other implications of results

There is evidence that *PPARG* supports the ''thrifty gene hypothesis'' in that the wild-type genotype optimizes the building of fat deposits as energy reserves and thus favored human survival in times when food was either limited, sporadically available, or poor in quality (47–50). Because today's lifestyle is much more relaxed and sedentary and is characterized by a diet that is rich in carbohydrates and fats and poor in fiber, these once favorable genetic factors have now become detrimental, leading to an increase in the risk of developing chronic diseases such as type 2 diabetes. PPARG is a known target for thiazolidinediones, antidiabetic drugs which have been shown to improve insulin sensitivity and to reduce plasma glucose and blood pressure in persons with type 2 diabetes (51). Thiazolidinediones are highly specific ligands for PPARG and lead to the activation of the nuclear receptor on binding (52).

Since the Pro12 allele is present in at least 80% of humans, the population attributable risk of type 2 diabetes associated with this polymorphism is as high as 25% (30). This gene is a confirmed type 2 diabetes susceptibility locus and is now one of almost 20 type 2 diabetes susceptibility loci identified over the last few years. With other type 2 diabetes susceptibility loci, research on how best to use this information in a translational context is needed. Given the possible heterogeneity in the magnitude of the association among populations and with environmental factors, a more systematic assessment locus should also be used as a genetic factor with which to investigate both gene-gene and gene-environment interactions with type 2 diabetes.

ACKNOWLEDGMENTS

Author affiliations: Department of Public Health and Primary Care, Institute of Public Health, University of Cambridge, Cambridge, United Kingdom (Hebe N. Gouda, Jan Yates); Strangeways Research Laboratory, Institute of Public Health, University of Cambridge, Cambridge, United Kingdom (Gurdeep S. Sagoo, Manjinder S. Sandhu, Julian P. T. Higgins); MRC Biostatistics Unit, Institute of Public Health, University of Cambridge, Cambridge, United Kingdom (Gurdeep S. Sagoo, Julian P. T. Higgins); and MRC Epidemiology Unit, Institute of Public Health, University of Cambridge, Cambridge, United Kingdom (Anne-Helen Harding).

The work of Drs. Gurdeep S. Sagoo and Julian P. T. Higgins was funded by a grant from the PHG Foundation. Dr. Julian P. T. Higgins was supported by Medical Research Council grant U.1052.00.011.

The authors thank Drs. Phillippe Amouyel, Leslie J. Baier, Jens P. Berg, Michael Boehnke, Raffaella Buzetti, Young Min Cho, Coleen Damcott, Ian Deary, Paul Franks, Philippe Froguel, David Flavell, Jose Florez, Maya Ghoussaini, Leif Groop, Robert Hegele, Christian Herder (and all collaborators in the KORA genome-wide association study), Joel Hirschorn, Wendy Johnson, Takashi Kadowaki, Tuomas Kilpelainen, Ornella Ludovico, Valeriya Lyssenko, Maciek Malecki, James Meigs, Aline Meirhaeghe-Hurez, Reza Meshkani, Luis Gallert Millan, Yunhua Muller, Jill Norris, Kyong Soo Park, Oluf Borbye Pederson, Fabio Pellegrini, Raúl Pisabarro, Alan Shuldiner, Inmaculada Simon, Soren Snitker, Daniela Sramkova, Heather Stringham, Vlad Tavares, Andrew Thomas, Per Thorsby, Vincenzo Trischitta, Olga Vaccaro, and Eleftheria Zeggini for correspondence and for contributing data.

Conflict of interest: none declared.

REFERENCES

1. Michalik L, Auwerx J, Berger JP, et al. International Union of Pharmacology. LXI. Peroxisome proliferator-activated receptors. *Pharmacol Rev.* 2006;58(4):726–741.
2. Spiegelman BM. PPAR-γ: adipogenic regulator and thiazolidinedione receptor. *Diabetes*. 1998;47(4):507–514.
3. Dubuquoy L, Dharancy S, Nutten S, et al. Role of peroxisome proliferator-activated receptor γ and retinoid X receptor heterodimer in hepatogastroenterological diseases. *Lancet*. 2002; 360(9343):1410–1418.
4. Mukherjee R, Jow L, Croston GE, et al. Identification, characterization, and tissue distribution of human peroxisome proliferator-activated receptor (PPAR) isoforms PPARγ2 versus PPARγ1 and activation with retinoid X receptor agonists and antagonists. *J Biol Chem.* 1997;272(12):8071–8076.
5. Fajas L, Auboeuf D, Raspé E, et al. The organization, promoter analysis, and expression of the human PPARγ gene. *J Biol Chem.* 1997;272(30):18779–18789.
6. Paracchini V, Pedotti P, Taioli E. Genetics of leptin and obesity: a HuGE review. *Am J Epidemiol*. 2005;162(2): 101–114.
7. Yen CJ, Beamer BA, Negri C, et al. Molecular scanning of the human peroxisome proliferator activated receptor γ (hPPARγ) gene in diabetic Caucasians: identification of a Pro12Ala PPARγ 2 missense mutation. *Biochem Biophys Res Commun*. 1997;241(2):270–274.
8. Wild S, Roglic G, Green A, et al. Global prevalence of diabetes: estimates for the year 2000 and projections for 2030. *Diabetes Care*. 2004;27(5):1047–1053.
9. Zimmet P, Alberti KG, Shaw J. Global and societal implications of the diabetes epidemic. *Nature*. 2001;414(6865): 782–787.
10. UK Prospective Diabetes Study (UKPDS) Group. Intensive blood-glucose control with sulphonylureas or insulin compared with conventional treatment and risk of complications in patients with type 2 diabetes (UKPDS 33). *Lancet*. 1998; 352(9131):837–853.
11. Freeman H, Cox RD. Type-2 diabetes: a cocktail of genetic discovery. *Hum Mol Genet.* 2006;15(spec no. 2):R202–R209.
12. Gloyn AL. The search for type 2 diabetes genes. *Ageing Res Rev.* 2003;2(2):111–127.
13. McCarthy MI. Progress in defining the molecular basis of type 2 diabetes mellitus through susceptibility-gene identification. *Hum Mol Genet.* 2004;13(spec no. 1):R33–R41.

14. Zeggini E. A new era for type 2 diabetes genetics. *Diabet Med*. 2007;24(11):1181–1186.
15. Frazer KA, Murray SS, Schork NJ, et al. Human genetic variation and its contribution to complex traits. *Nat Rev Genet.* 2009;10(4):241–251.
16. Frayling TM. Genome-wide association studies provide new insights into type 2 diabetes aetiology. *Nat Rev Genet.* 2007; 8(9):657–662.
17. McCarthy MI. Casting a wider net for diabetes susceptibility genes. *Nat Genet.* 2008;40(9):1039–1040.
18. Higgins JP, Green S, eds. *Cochrane Handbook for Systematic Reviews of Interventions 5.0.1 (Updated September 2008)*. London, United Kingdom: John Wiley & Sons Ltd; 2008.
19. Sagoo GS, Little J, Higgins JP. Systematic reviews of genetic association studies [electronic article]. *PLoS Med*. 2009; 6(3):e28.
20. Little J, Higgins JP, eds. *The HuGENet™ HuGE Review Handbook, Version 1.0*. Ottawa, Ontario, Canada: HuGENet Canada Coordinating Centre; 2008. (http://hugenet.ca/). (Accessed November 29, 2008).
21. Hindorff LA, Junkins HA, Mehta JP, et al. *A Catalog of Published Genome-Wide Association Studies*. Bethesda, MD: Office of Population Genomics, National Human Genome Research Institute; 2008. (www.genome.gov/26525384). (Accessed November 11, 2008).
22. Minelli C, Thompson JR, Tobin MD, et al. An integrated approach to the meta-analysis of genetic association studies using Mendelian randomization. *Am J Epidemiol*. 2004;160(5): 445–452.
23. Harbord RM, Egger M, Sterne JA. A modified test for small-study effects in meta-analyses of controlled trials with binary endpoints. *Stat Med*. 2006;25(20):3443–3457.
24. DerSimonian R, Laird N. Meta-analysis in clinical trials. *Control Clin Trials*. 1986;7(3):177–188.
25. Higgins JP, Thompson SG, Deeks JJ, et al. Measuring inconsistency in meta-analyses. *BMJ*. 2003;327(7414):557–560.
26. Zeggini E, Scott LJ, Saxena R, et al. Meta-analysis of genome-wide association data and large-scale replication identifies additional susceptibility loci for type 2 diabetes. *Nat Genet.* 2008;40(5):638–645.
27. Day N, Oakes S, Luben R, et al. EPIC-Norfolk: study design and characteristics of the cohort. European Prospective Investigation of Cancer. *Br J Cancer.* 1999;80(suppl 1):95–103.
28. Department of Noncommunicable Disease Surveillance, World Health Organization. *Diagnosis and Classification of Diabetes Mellitus: Definition, Diagnosis and Classification of Diabetes Mellitus and Its Complications. Report of a WHO Consultation*. Geneva, Switzerland: World Health Organization; 1999.
29. Ludovico O, Pellegrini F, Di Paola R, et al. Heterogeneous effect of peroxisome proliferator-activated receptor γ2 Ala12 variant on type 2 diabetes risk. *Obesity (Silver Spring)*. 2007; 15(5):1076–1081.
30. Altshuler D, Hirschhorn JN, Klannemark M, et al. The common PPARγ Pro12Ala polymorphism is associated with decreased risk of type 2 diabetes. *Nat Genet.* 2000;26(1):76–80.
31. Ek J, Andersen G, Urhammer SA, et al. Studies of the Pro12Ala polymorphism of the peroxisome proliferator-activated receptor-γ2 (PPAR-γ2) gene in relation to insulin sensitivity among glucose tolerant Caucasians. *Diabetologia*. 2001;44(9):1170–1176.
32. Vardarli I. Is the Pro12Ala polymorphism of the peroxisome proliferator-activated receptor gamma gene associated with type 2 diabetes? A meta-analysis [abstract]. *Diabetologia*. 2007;50(suppl):S538.
33. Hara K, Yamauchi T, Kubota N, et al. The role of PPARγ in the onset of type 2 diabetes [in Japanese]. *Nippon Yakurigaku Zasshi*. 2003;122(4):317–324.
34. Lohmueller KE, Pearce CL, Pike M, et al. Meta-analysis of genetic association studies supports a contribution of common variants to susceptibility to common disease. *Nat Genet.* 2003;33(2):177–182.
35. Parikh H, Groop L. Candidate genes for type 2 diabetes. *Rev Endocr Metab Disord*. 2004;5(2):151–176.
36. Ioannidis JP, Boffetta P, Little J, et al. Assessment of cumulative evidence on genetic associations: interim guidelines. *Int J Epidemiol*. 2008;37(1):120–132.
37. Colhoun HM, McKeigue PM, Davey Smith G. Problems of reporting genetic associations with complex outcomes. *Lancet*. 2003;361(9360):865–872.
38. Ioannidis JP, Trikalinos TA, Ntzani EE, et al. Genetic associations in large versus small studies: an empirical assessment. *Lancet*. 2003;361(9357):567–571.
39. Masud S, Ye S. Effect of the peroxisome proliferator activated receptor-γ gene Pro12Ala variant on body mass index: a meta-analysis. *J Med Genet.* 2003;40(10):773–780.
40. Luan J, Browne PO, Harding AH. Evidence for gene-nutrient interaction at the PPARγ locus. *Diabetes*. 2001;50(3): 686–689.
41. Robitaille J, Després JP, Pérusse L, et al. The PPAR-gamma P12A polymorphism modulates the relationship between dietary fat intake and components of the metabolic syndrome: results from the Québec Family Study. *Clin Genet.* 2003; 63(2):109–116.
42. Morini E, Tassi V, Capponi D, et al. Interaction between PPARγ2 variants and gender on the modulation of body weight. *Obesity (Silver Spring)*. 2008;16(6):1467–1470.
43. Mattevi VS, Zembrzuski VM, Hutz MH, et al. Effects of a PPARG gene variant on obesity characteristics in Brazil. *Braz J Med Biol Res*. 2007;40(7):927–932.
44. Deeb SS, Fajas L, Nemoto M, et al. A Pro12Ala substitution in PPARγ2 associated with decreased receptor activity, lower body mass index and improved insulin sensitivity. *Nat Genet.* 1998;20(3):284–287.
45. Masugi J, Tamori Y, Mori H, et al. Inhibitory effect of a proline-to-alanine substitution at codon 12 of peroxisome proliferator-activated receptor-γ 2 on thiazolidinedione-induced adipogenesis. *Biochem Biophys Res Commun*. 2000;268(1):178–182.
46. Werman A, Hollenberg A, Solanes G, et al. Ligand-independent activation domain in the N terminus of peroxisome proliferator-activated receptor γ (PPARγ). Differential activity of PPARγ1 and -2 isoforms and influence of insulin. *J Biol Chem.* 1997;272(32):20230–20235.
47. Argmann CA, Cock TA, Auwerx J. Peroxisome proliferator-activated receptor γ: the more the merrier? *Eur J Clin Invest*. 2005;35(2):82–92.
48. Joffe B, Zimmet P. The thrifty genotype in type 2 diabetes: an unfinished symphony moving to its finale? *Endocrine*. 1998; 9(2):139–141.
49. Neel JV. Diabetes mellitus: a "thrifty" genotype rendered detrimental by "progress"? *Am J Hum Genet.* 1962;14(4):353–362.
50. Ruiz-Narváez E. Is the Ala12 variant of the PPARG gene an "unthrifty allele"? *J Med Genet.* 2005;42(7):547–550.
51. Day C. Thiazolidinediones: a new class of antidiabetic drugs. *Diabet Med*. 1999;16(3):179–192.
52. Lehmann JM, Moore LB, Smith-Oliver TA, et al. An antidiabetic thiazolidinedione is a high affinity ligand for peroxisome proliferator-activated receptor γ (PPARγ). *J Biol Chem.* 1995;270(22):12953–12956.