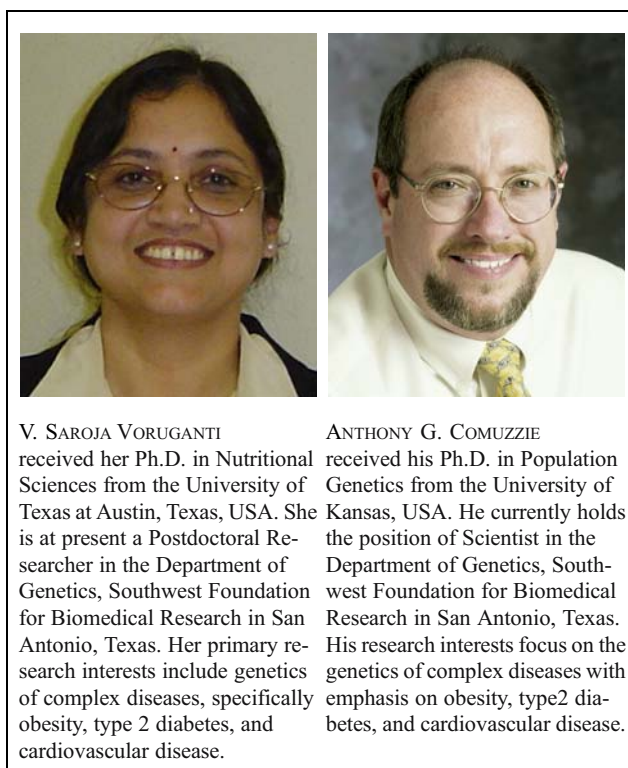


Genetics of variation in HOMA-IR and cardiovascular risk factors in Mexican-Americans

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Abstract Insulin resistance is a major biochemical defect underlying the pathogenesis of cardiovascular disease (CVD). Mexican-Americans are known to have an unfavorable cardiovascular profile. Thus, the aim of this study was to investigate the genetic effect on variation in HOMA-IR and to evaluate its genetic correlations with other phenotypes related to risk of CVD in Mexican-Americans. The homeostatic model assessment method (HOMA-IR) is one of several approaches that are used to measure insulin resistance and was used here to generate a quantitative phenotype for genetic analysis. For 644 adults who had participated in the San Antonio Family Heart Study (SAFHS), estimates of genetic contribution were computed using a variance components method implemented in SOLAR. Traits that exhibited significant heritabilities were body mass index (BMI) ($h^2 = 0.43$), waist circumference ($h^2 = 0.48$), systolic blood pressure ($h^2 = 0.30$), diastolic blood pressure ($h^2 = 0.21$), pulse pressure ($h^2 = 0.32$), triglycerides ($h^2 = 0.51$), LDL cholesterol ($h^2 = 0.31$), HDL cholesterol ($h^2 = 0.24$), C-reactive protein ($h^2 = 0.17$), and HOMA-IR ($h^2 = 0.33$). A genome-wide scan for HOMA-IR revealed significant evidence of linkage on chromosome



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12q24 (close to *PAH* (phenylalanine hydroxylase), $LOD = 3.01$, $p < 0.001$). Bivariate analyses demonstrated significant genetic correlations ($p < 0.05$) of HOMA-IR with BMI ($\rho_G = 0.36$), waist circumference ($\rho_G = 0.47$), pulse pressure ($\rho_G = 0.39$), and HDL cholesterol ($\rho_G = -0.18$). Identification of significant linkage for HOMA-IR on chromosome 12q replicates previous family-based studies reporting linkage of phenotypes associated with type 2 diabetes in the same chromosomal region. Significant genetic correlations between HOMA-IR and phenotypes

related to CVD risk factors suggest that a common set of gene(s) influence the regulation of these phenotypes.

Keywords Insulin resistance · Variance component approach · Genetic correlations

Introduction

Insulin resistance is a condition in which glucose uptake by insulin-sensitive tissues is diminished [1]. The resultant hyperglycemia is compensated by an increased production of insulin by the pancreatic beta cells. Initially, increased secretion of insulin is effective, but gradually, beta cells are overburdened, and their function is adversely affected, leading to the development of type 2 diabetes [2]. Insulin resistance is a strong predictor of type 2 diabetes [3] and cardiovascular disease (CVD) risk [4]. Although the mechanism by which insulin resistance increases the risk of CVD is not clear, it has been hypothesized that the first step in the development of insulin resistance occurs at the level of the adipocyte. This may be the result of an interaction between a defective gene (s) and environment [5]. Once insulin resistance develops in adipocytes, uptake of free fatty acids is decreased, thereby, increasing the circulating free fatty acids [2, 5]. These free fatty acids enter the liver through the portal vein, and this results in an imbalance between high-density (HDL) and triglycerides (dyslipidemia). Hyperinsulinemia also causes an increase in very-low density lipoprotein (VLDL) secretion. Hypertriglyceridemia leads to low HDL and increased small low-density (LDL) concentrations [5]. Free fatty acids are also responsible for inducing insulin resistance in muscle, especially skeletal muscle [6]. In addition, insulin resistance is linked to hypertension through its effect on blood flow and reabsorption of water and electrolytes from kidneys [5]. Thus, insulin resistance seems to be a major biochemical defect underlying the pathogenesis of CVD [5, 7].

The risk of insulin resistance is known to increase with obesity, aging, physical inactivity, and genetic predisposition [8]. Mexican-Americans have higher rates of obesity than non-Hispanic whites [9]. Moreover, the prevalence of insulin resistance and type 2 diabetes seems to be greater in this population, and nondiabetic individuals of this ethnicity have been found to be more insulin resistant than nondiabetic non-Hispanic whites [10]. Although pattern of CVD mortality in different ethnicities is not very clear, it has been reported that Mexican-Americans have a poor cardiovascular profile compared to non-Hispanic Whites [11, 12]. In an earlier population-based study, the San Antonio Heart study (SAHS), it was found that Mexican-Americans had three times more prevalence of diabetes

than non-Hispanic Whites. In addition, they had higher levels of triglycerides and lower high-density lipoprotein (HDL) cholesterol levels [13, 14]. Surprisingly, they found that Mexican-American men had lower incidence of myocardial infarction compared to non-Hispanic Whites, although Mexican-American women were on par with their counterparts on non-Hispanic Whites. However, CVD mortality remains high in this population [14].

Evidence of a genetic component in the variation in several traits has already been demonstrated, particularly in Mexican-Americans. In this population, genetic influence on phenotypes such as body mass index (BMI), waist circumference, percent body fat, waist-hip ratio (WHR), fasting glucose, fasting insulin, blood pressure, etc. has been documented [15, 16, 17]. Continuing the same efforts, we analyzed the genetic component of insulin resistance, estimated by homeostatic model assessment method (HOMA-IR) in Mexican-Americans from the San Antonio Family Heart Study (SAFHS). In this study, our purpose was to assess the genetic influence on variation in HOMA-IR and to evaluate the genetic correlations between HOMA-IR and other CVD components. Insulin resistance, as estimated by HOMA-IR [18], correlates well with the values from insulinemic-euglycemic clamps [19] and was used here to generate a quantitative phenotype for genetic analyses.

Methodology

This study was conducted in individuals who were participating in the SAFHS, a family-based genetic study in the Hispanic community in San Antonio. Begun in 1991, it focused on the identification of genes impacting risk for the development of cardiovascular disease (CVD) in Mexican-Americans. Participants in this study were Mexican-Americans from 40 extended families. Families were recruited without any regard to disease status, with probands between 40 and 60 years of age [20]. Eligibility criteria required the proband to have at least six first-degree relatives (excluding their parents) who were 16 years or older and who resided in San Antonio, TX. Information regarding medical history, socio-demographics, and anthropometrics was obtained in the first phase of SAFHS data collection. All participants underwent physical examination, and blood was drawn after a 12-h overnight fast. Plasma was extracted and stored in aliquots at -80°C for clinical chemistries and endocrine assays. Genotyping was conducted with the DNA isolated from lymphocytes. Informed consent was obtained from all participants of this study, which was approved by the Institutional Review Board at the University of Texas Health Science Center at San Antonio.

Phenotyping

Anthropometric measurements included height, weight, waist circumference, and BMI. Weight was measured to the nearest 0.1lb and then to the nearest 0.1kg, using an ISO-9001 certified Scale-Tronix electronic scale with a capacity of 880lbs (400kg) (White Plains, NY). Standing height was measured twice, to the nearest 0.1cm, using a SECA wall-mounted stadiometer (Seca, Hanover, MD). Waist circumference was measured twice, in millimeters, at the level of the umbilicus. The hip circumference was also measured twice, in millimeters, at the widest circumference. These measurements were taken with the participant standing with feet together, arms at their side, and abdomen relaxed. The average of the two measurements for each variable was then used in analyses. Body mass index was calculated by dividing weight (kg) by height (m) squared.

Blood pressure was measured three times, with an appropriate arm cuff, using a Random-Zero sphygmomanometer, (Gelman-Hawksley, Sussex, England). The first measurement of the systolic and diastolic pressures was discarded, and the mean of the second and third readings was used in analyses. Pulse pressure was calculated as the difference between systolic and diastolic blood pressure [21].

Phenotypes that were measured using plasma included glucose, insulin, and a lipid panel. Glucose was analyzed by the glucose oxidase method with an Abbott V/P Analyzer (Abbott Laboratories, Abbott Park, IL). Plasma insulin was measured by a commercially available radioimmunoassay kit (LINCO Research, St Charles, MO). C-reactive protein was measured by enzyme-linked immunosorbent assay (ELISA) (ALPCO Diagnostics, Salem, NH). Insulin resistance was estimated by the homeostatic model assessment method (HOMA) [18], which is computed as

$$\text{HOMA-IR} = \{\text{fasting plasma insulin (mU/ml)} \times \text{fasting serum glucose (mmol/l)}\} / 22.5$$

Total cholesterol, HDL cholesterol, and triglycerides were measured with commercial reagents using Express Plus Analyzer (Bayer Diagnostics, Pittsburgh, PA). LDL cholesterol was calculated by the Friedewald equation [22].

Genotyping

Genotyping for the participants was conducted according to standard protocols. All members were genotyped for 414 short-tandem repeats (STR) markers that were spaced at an average interval of 10cM across 22 autosomes. Polymerase chain reaction (PCR) primers used to genotype each STR were from the MapPairs 6 and 8 Linkage Screening Set (Research Genetics, Huntsville, AL). Genotypes were determined using an automated DNA sequencer and

GeneScan and Genotyper software (Applied Biosystems, Foster City, CA).

Statistical analyses

A pedigree-based multipoint variance components approach was used to test for linkage between marker loci and HOMA-IR using a maximum likelihood method. This method is implemented in software program sequential oligogenic linkage analysis routines (SOLAR) [23]. According to this approach, total phenotypic variance can be partitioned into its genetic and environmental components. The fraction of total phenotypic variance (σ^2_P) due to additive genetic effects (σ^2_G) is called the heritability denoted by $h^2 = \sigma^2_G / \sigma^2_P$ [24]. To estimate the genetic component in the variation of HOMA-IR, its heritability was estimated. To find a putative quantitative trait locus (QTL) or loci (QTLs) that might be affecting HOMA-IR, a multipoint linkage analysis was conducted. This analysis is an extension of the variance component approach in which a QTL variance component is added to the basic model. The phenotypic correlations between family members can be described as the cumulative effect of a specific QTL associated with a marker and residual genetic and environmental effects. The effect of a specific QTL on the variation in phenotype can be modeled as a function of the identity by descent (IBD) relationship at the marker locus between family members [25]. A model under the null hypothesis in which the additive genetic variance for a specific QTL equals zero was tested against a model under an alternate hypothesis in which the additive variance was estimated. This is known as a likelihood ratio test, and the resultant likelihood ratio test statistic (LRT) in this particular case was distributed asymptotically as a 1/2:1/2 mixture of a χ^2 variable with 1df and a point mass at zero [26]. Traditionally, a logarithm of the odds (LOD) score, which is computed directly from the LRT, is reported in linkage analyses [23]. Generally, a LOD score of greater than three is taken as strong evidence of a putative QTL.

A variance components approach for modeling a bivariate trait was used to estimate genetic correlations between HOMA-IR and other CVD risk factors. A bivariate trait is simply one that is comprised of two constituent traits that are assumed to be jointly multivariate normal in distribution. Thus, the bivariate variance components model can be thought of as a model that incorporates what would be the regular variance components model per constituent trait of the overall bivariate trait and a component for the covariance between the constituent traits. The covariance between constituent traits can be reparameterized as a product of the constituent trait SDs, and their correlation [24, 27] was conducted.

In this analysis, the phenotypic correlation (ρ_P) between the phenotypes was expressed in terms of its core genetic and environmental correlations:

$$\rho_P = \rho_G(\sqrt{h_1^2}\sqrt{h_2^2}) + \rho_E(\sqrt{(1-h_1^2)}\sqrt{(1-h_2^2)})$$

h_1^2 and h_2^2 are the heritabilities of the two phenotypes being studied, and ρ_G and ρ_E are the additive genetic and environmental correlations between the traits, respectively.

A model where all the parameters are estimated was compared with a model in which the genetic correlation is constrained to zero ($\rho_G = 0$). If the result of this statistical test is significant, then we infer that the traits share the effects of a common set of genes. The genetic correlation indicates the extent of genes shared between a pair of traits (pleiotropy). The extent of the shared genetic effect is verified by a second statistical test that compares the model in which all the parameters are estimated with one in which the genetic correlation is constrained to one ($\rho_G = 1$; complete pleiotropy).

Results

Data were available for 644 participants (men = 234, women = 410). The relative pairs used for this study are listed in Table 1. Of the participants, 20% were diabetics, 28% were overweight (BMI > 25 and <30) and 30% obese (BMI > 30). One-fourth of this cohort was taking hypertension medications, and 15% were taking diabetic medications. Their phenotypic characteristics are depicted

Table 1 Relative pairs used in this study

| Relationships | Number of pairs |
|---------------------------------------|-----------------|
| Parent-offspring | 312 |
| Siblings | 340 |
| Grandparent–grandchild | 84 |
| Avuncular | 696 |
| Half-siblings | 73 |
| Grand-avuncular | 167 |
| Half-avuncular | 107 |
| First cousins | 768 |
| Half grand avuncular | 4 |
| First cousins, once removed | 802 |
| Half first cousins | 76 |
| First cousins, twice removed | 33 |
| Half first cousins, once removed | 6 |
| Second cousins | 330 |
| Double first cousins | 6 |
| Double first cousins, once removed | 25 |
| Double second cousins | 14 |
| Half first cousins and second cousins | 6 |
| Total | 3849 |

in Table 2. Sex-specific differences showed significant differences between men and women with respect to BMI, systolic and diastolic blood pressure, HDL cholesterol, triglycerides, and plasma glucose. Quantitative genetic analyses showed several CVD-related traits to be significantly heritable, as shown in Table 2. Of these, triglycerides and total cholesterol had highest heritabilities followed by waist circumference, BMI, INSULIN, HOMA-IR, pulse pressure, systolic blood pressure, HDL cholesterol, diastolic blood pressure, and C-reactive protein.

Bivariate analyses of these phenotypes demonstrated significant genetic correlations of HOMA-IR with BMI ($\rho_G = 0.36$), waist circumference ($\rho_G = 0.41$), pulse pressure ($\rho_G = 0.39$), and HDL cholesterol ($\rho_G = -0.18$) (Table 3). All the above genetic correlations were significantly different from zero ($p < 0.05$). We also calculated phenotypic correlations between these traits. Phenotypic correlations were significant and positive between HOMA-IR and BMI, waist circumference, triglycerides, C-reactive protein, and blood pressure phenotypes (systolic, diastolic, and pulse pressure). A negative correlation was obtained between HOMA-IR and HDL cholesterol. All these correlations were adjusted for age, sex, age \times sex, diabetic status, and hypertensive medication.

The best model of the genome-wide scan for HOMA-IR revealed significant evidence for linkage (empirical LOD = 3.01, $p = 0.00001$) near marker phenylalanine hydroxylase (*PAH*) at 118cM on chromosome 12, with age, sex, age \times sex, and waist circumference as covariates (Figs. 1 and 2). The 1-LOD support interval spanned a width of 16cM (112–128cM) or about 9Mb (96–105Mb). When the same scan was conducted with age, sex, age \times sex, waist circumference, diabetic status, and hypertension medication as covariates, the signal weakened to a LOD score of 2.41 ($p = 0.0003$).

Discussion

This study presents evidence of significant genetic influence on variation in HOMA-IR, and on its relationship with other CVD risk factors. The identification of linkage for HOMA-IR on chromosome 12q24 is significant, as this region (within one LOD score support interval) harbors several candidate genes related to obesity and diabetes. The gene for transcription factor 1 (*TCF1*), also known as the hepatocyte nuclear factor-1 alpha (*HNF-1alpha*) resides at 12q24.2 [28]. Mutations in this gene are known to cause maturity onset diabetes of the young, type 3 (MODY 3) and were also found to be related to diabetes in youth in the Japanese population [29]. In addition, insulin-like growth factor (*IGF1*) gene is located at 12q22–24.1 [30, 31]. Sun et al. [32] found significant association between *IGF1* gene (a

Table 2 Phenotypic characteristics of the participants and heritabilities of various phenotypes

| Phenotype | Men (SE) | Women (SE) | <i>p</i> value | <i>h</i> ² (SE) | <i>p</i> value |
|--------------------------------------|---------------|---------------|----------------|----------------------------|----------------|
| <i>n</i> | 234 | 410 | | | |
| Age, years | 46.7 (0.96) | 48.2 (0.72) | NS | | |
| BMI ^a , kg/m ² | 30.9 (0.42) | 32.5 (0.37) | < 0.005 | 0.43 (0.09) | <0.00001 |
| Waist circumference, cm | 104.8 (1.1) | 105.9 (0.89) | NS | 0.48 (0.09) | <0.00001 |
| HOMA ^a index | 5.84 (0.06) | 5.99 (0.05) | NS | 0.33 (0.10) | <0.00001 |
| Systolic blood pressure, mmHg | 125.8 (1.1) | 122.7 (0.99) | < 0.05 | 0.30 (0.08) | <0.0001 |
| Diastolic blood pressure, mmHg | 72.7 (0.74) | 68.7 (9.51) | < 0.001 | 0.21 (0.07) | <0.001 |
| Pulse pressure, mmHg | 53.1 (1.1) | 54.0 (1.0) | NS | 0.32 (0.08) | <0.0001 |
| Plasma glucose, mg/dl | 109.6 (2.9) | 102.1 (1.9) | < 0.05 | 0.13 (0.07) | <0.05 |
| Serum insulin, uIU/ml | 21.3 (1.3) | 22.8 (0.9) | NS | 0.35 (0.10) | <0.00001 |
| Triglycerides, mg/dl | 137.9 (7.67) | 122.8 (3.62) | <0.05 | 0.51 (0.09) | <0.00001 |
| HDL ^a cholesterol, mg/dl | 46.1 (0.91) | 50.6 (0.7) | <0.001 | 0.24 (0.09) | <0.00001 |
| LDL ^a cholesterol, mg/dl | 108.7 (0.10) | 105.74 (0.58) | NS | 0.31 (0.08) | <0.0001 |
| Total cholesterol, mg/dl | 182.4 (2.54) | 180.9 (2.0) | NS | 0.51 (0.09) | <0.0001 |
| CRP, ng/ml | 2792.3 (25.6) | 4135.4 (30.4) | <0.005 | 0.17 (0.13) | <0.05 |

^a BMI Body mass index, HOMA-IR homeostatic model assessment index, HDL high density lipoprotein, LDL low density lipoprotein, CRP C-reactive protein, *h*² heritability, SE standard error, NS not significant

growth factor primarily involved in cell growth and differentiation) marker and percent body fat, fat free mass, and changes in fat free mass. Other prominent candidate genes in this region that have been linked to adiposity are scavenger receptor class B, member 1 (*SCARB1*) [33], acetyl-coenzyme A carboxylase beta (*ACACB*) [34], and pro-melanin concentrating hormone (*PMCH*) [35].

Significant heritabilities were obtained for HOMA-IR and other CVD risk factors indicating a substantial genetic influence on their variation. These heritability estimates are in accordance with studies in other ethnicities. These include studies conducted in families from several different ethnicities (Whites, Blacks, Mexican-Americans, and Asians) [4, 36, 37]. All the above studies, including ours, confirm that all the traits that confer CVD risk are under considerable genetic influence.

In addition, several studies have reported significant or suggestive linkages in the region of the one-LOD support interval on chromosome 12q and diabetes-related traits. In a study conducted in Botnia in Finland, a linkage for MODY3 was found on chromosome 12q in four families with early onset diabetes [38]. In four Finnish families that were affected by non-insulin-dependent diabetes mellitus (NIDDM), this region on chromosome 12q has been linked to NIDDM2, a type of diabetes that is similar to MODY3 in all respects except that it is less severe in its insulin secretion defect [39]. In another study in a larger Finnish population with about 58 families, the same region on 12q was linked to type 2 diabetes (LOD = 2.1) [40]. Similar results were presented in Caucasian sib pairs [41] with diabetic nephropathy. Rich et al. [42] reported linkage of acute insulin response to this region in African-American

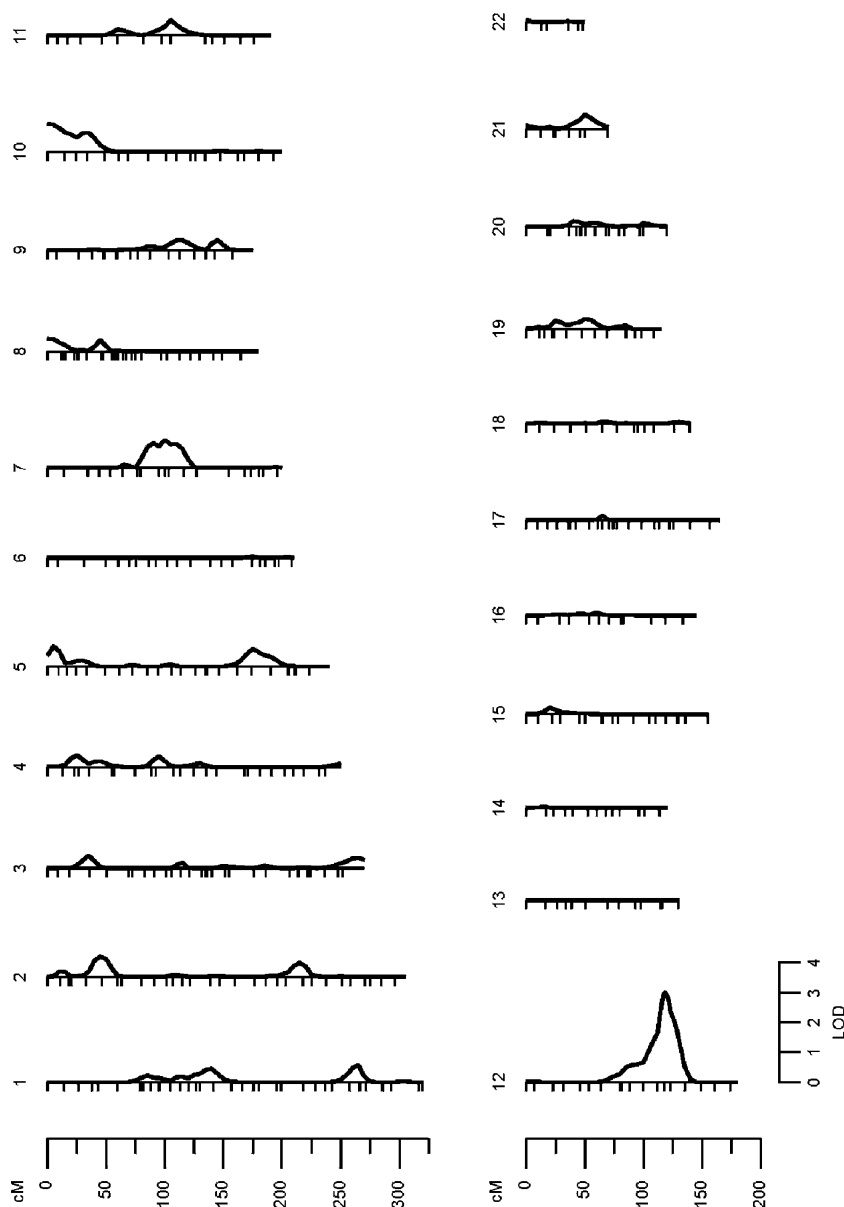
Table 3 Genetic and phenotypic correlations of HOMA-IR with cardiovascular risk factors

| Phenotype ^a | ρ_G (SE) | <i>p</i> value ($\rho_G=0$) | <i>p</i> value ($\rho_G=1$) | ρ_P (calculated from ρ_G and ρ_E) | <i>p</i> value |
|------------------------------|---------------|-------------------------------|-------------------------------|---|-----------------------|
| BMI ^b | 0.36 (0.16) | <0.05 | <0.001 | 0.50 | 1.7×10^{-36} |
| Waist circumference | 0.41 (0.16) | <0.05 | <0.001 | 0.47 | 1.3×10^{-32} |
| Triglycerides | 0.04 (0.19) | NS | <0.001 | 0.25 | 3.2×10^{-10} |
| HDL ^b cholesterol | -0.18 (0.08) | <0.05 | <0.05 | - 0.40 | 1.3×10^{-68} |
| LDL ^b cholesterol | -0.32 (0.22) | NS | <0.001 | 0.05 | NS |
| Total cholesterol | -0.27 (0.20) | NS | <0.005 | 0.04 | NS |
| Systolic blood pressure | 0.32 (0.20) | NS | <0.001 | 0.15 | 4.2×10^{-4} |
| Diastolic blood pressure | 0.09 (0.28) | NS | <0.001 | 0.11 | 5.8×10^{-3} |
| Pulse pressure | 0.61 (0.20) | <0.05 | <0.05 | 0.11 | 1.7×10^{-2} |
| CRP ^b | -0.68 (0.51) | NS | <0.05 | 0.12 | 3.7×10^{-2} |

^a All phenotypes have been log transformed and adjusted for age, sex, age × sex, diabetic status, and hypertension medication; SE Standard error, ρ_G genetic correlation, ρ_P phenotypic correlation

^b BMI Body mass index, HOMA-IR homeostatic model assessment index, HDL high density lipoprotein, LDL low density lipoprotein, CRP C-reactive protein

Fig. 1 Map showing string plot of HOMA-IR (map distance in cM is depicted on X-axis and LOD scores on Y-axis)



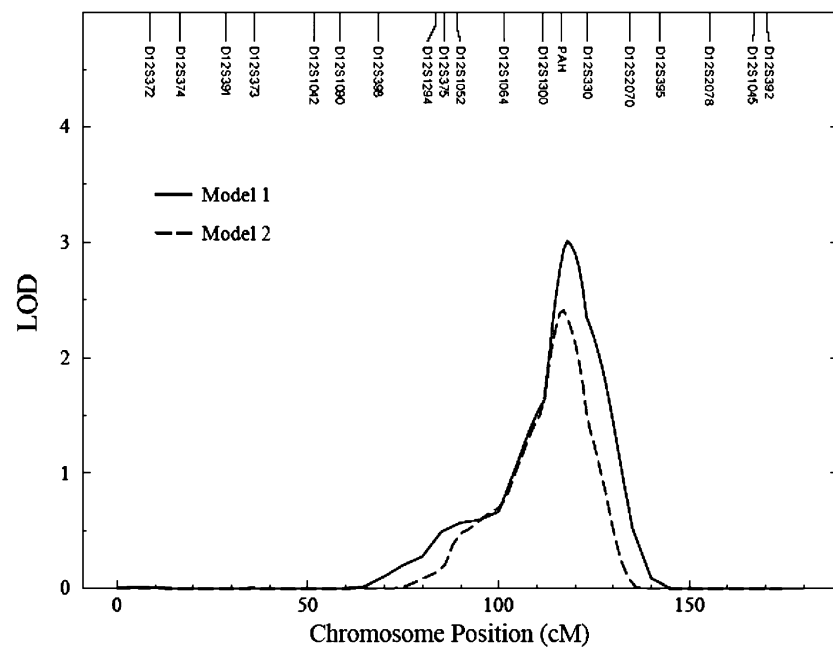
families from the Insulin Resistance Atherosclerosis Study (IRAS) Family study. Ehm et al. [43] found evidence of suggestive linkage of impaired glucose homeostasis to this region on 12q, in a population of Mexican-Americans and non-Hispanic Whites. The study by van Tilburg et al. [44] found a suggestive linkage for type 2 diabetes in obese, diabetic individuals in a region that was proximal to 12q24. In addition, in a study in African-American and non-Hispanic White women (the HyperGEN study), 12q24 was linked to BMI and percent body fat [45]. These studies emphasize the importance of this region in diabetes-related phenotypes.

As HOMA-IR is calculated incorporating glucose and insulin values, we conducted linkage analysis with glucose, insulin, and glucose/insulin as separate phenotypes. How-

ever, none of them had linkage in the same region, which implies that this signal seems to be more specific for HOMA-IR than glucose or insulin alone. In a previous study that analyzed data from an earlier phase of SAFHS, glucose and insulin were localized to chromosomes 8 and 13 [46]. In this study, Cai et al. [46] found a suggestive linkage for corrected insulin response (CIR) in 12q24.2, but it is about 20cM distant to our signal.

After establishing that variation in HOMA-IR and other CVD risk factors is regulated by genetic factors, we conducted bivariate analysis to check for pleiotropy between these traits. In a study conducted in Mexican-Americans (The San Antonio Heart Study), HOMA-IR was found to increase the risk of CVD [47], and as it has already been shown in earlier studies in Mexican-Americans that

Fig. 2 Map showing linkage of HOMA-IR on chromosome 12 (map distance in cM is depicted on X-axis and LOD scores on Y-axis)



Model 1: adjusted by age, sex, age*sex, waist circumference

Model 2: adjusted by age, sex, age*sex, waist circumference, diabetic status and hypertension medication

CVD risk factors have higher prevalence among this population [16], particularly obesity and type 2 diabetes [16, 48], we investigated the genetic influence on the relationship between HOMA-IR and other CVD risk factors. Strong genetic correlations between HOMA-IR and BMI, waist circumference, HDL cholesterol, and pulse pressure confirmed the relationship between HOMA-IR and CVD risk. As it is known that adiposity plays a major role in the association of insulin resistance with blood pressure, we tested the genetic correlations between HOMA-IR and pulse pressure after adjusting for either BMI or waist circumference. The correlation remained significant, however, indicating that this correlation is independent of adiposity. In this study, we tested two hypotheses; one which stated that there were no shared genetic effects between the two given traits ($\rho_G = 0$) and that there was a complete overlap of the genes that control the expression of the two given traits ($\rho_G = 1$). The hypothesis that there is no sharing of genetic effects ($\rho_G = 0$) was rejected only for BMI, waist circumference, HDL cholesterol, and pulse pressure. However, the hypothesis that there is complete overlap of genes between traits was rejected for all traits. This means that common set of genes (shared genetic effect) might be regulating HOMA-IR and BMI, waist circumference, HDL cholesterol, and pulse pressure, but these traits are not regulated by one gene or the same set of genes.

A study conducted in Mexican-Americans, participating in the first phase of SAFHS, found similar relationships between fasting insulin and 2-h insulin and BMI. In this study, fasting insulin was used as a surrogate measure for insulin resistance and had weak genetic correlations with

systolic blood pressure [15]. In our study, strong genetic correlations between HOMA-IR and pulse pressure may have been due to the difference in the parameter that was used to estimate insulin resistance. The positive correlations, found in this study, between HOMA-IR and CVD risk factors such as BMI, waist circumference, and pulse pressure indicate that the genes associated with increased HOMA-IR are also associated with increase in these phenotypes. Similarly, the negative relationship between HOMA-IR and HDL cholesterol indicates that genes associated with elevated HOMA-IR are associated with lower HDL cholesterol.

As very few studies report genetic correlations, we also computed phenotypic correlations. Phenotypic correlations are calculated using genetic and environmental correlations and indicate overall correlations between two traits. All the CVD phenotypes had significant correlation with HOMA-IR, except LDL and total cholesterol. Bonora et al. [49] found HOMA-IR to be significantly correlated with most of the CVD risk factors and also found that insulin-resistant individuals had an increased risk for CVD than others. Similar reports have been shown by other studies [50, 51]. A study by Bertoni et al. [52], however, did not find any significant association between HOMA-IR and risk of CVD. The link between insulin resistance and CVD risk factors, as evidenced in this study, suggests that insulin resistance not only reflects defect in glucose metabolism but also increases the risk for CVD.

In summary, (1) the genetic analysis of HOMA-IR revealed a region on 12q that has tremendous importance in relation to diabetes and related phenotypes in our

Mexican-American study population. Thus, the next logical step would be to investigate whether variants in the two positional candidate genes (*TCF1* and *IGF1*) in this region account for the variation in HOMA-IR and other CVD risk factors. (2) Common set of genes regulate variation in both insulin resistance (as measured by HOMA) and BMI, waist circumference, HDL cholesterol, and pulse pressure, all of them important components of the CVD.

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