Assessment of genetic risk for myocardial infarction

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Summary

Although lifestyle and environmental factors influence the prevalence of myocardial infarction, genetic epidemiological studies have suggested that several genetic variants increase the risk for this condition. We have performed a large-scale association study to identify gene polymorphisms for reliable assessment of the genetic risk of myocardial infarction. The study population comprised 3,483 unrelated Japanese individuals (1,913 men; 1,570 women), including 1,192 subjects with myocardial infarction and 2,291 controls. The genotypes for 164 polymorphisms of 137 candidate genes were determined with an oligonucleotide ligation assay based on analysis of fluorescent microspheres with suspension array technology. Multivariable logistic regression analysis with adjustment for age, sex, body mass index, and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia revealed that the 677C→T (Ala222Val) polymorphism of *MTHFR*, the 1595C→G (Ser447Stop) polymorphism of *LPL*, and the –108/3G→4G polymorphism of *IPF1* were significantly associated with the prevalence of myocardial infarction. A stepwise forward selection procedure demonstrated that *IPF1*, *MTHFR*, and *LPL* genotypes significantly affected the prevalence of myocardial infarction. Combined genotype analysis of these polymorphisms yielded a maximum odds ratio of 2.54 for the combined genotype of *TT* for *MTHFR*, *CC* for *LPL*, and *3G3G* for *IPF1*. The genotypes for *MTHFR*, *LPL*, and *IPF1* may prove reliable for assessment of genetic risk for myocardial infarction. Determination of the combined genotype for these genes may contribute to primary, personalized prevention of this condition.

Keywords

Genetics, polymorphism, myocardial infarction, coronary heart disease, atherosclerosis

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Introduction

Completion of the Human Genome Project has the potential to provide substantial benefits to clinical medicine, including the development of panels of genetic markers for the assessment of disease risk (1). One approach to this goal is to evaluate selected polymorphisms of genes that are possibly associated with a disease, either because they are known to encode proteins related to the disease process or because they are located within chromosomal regions identified in linkage studies.

Coronary heart disease (CHD) is the single largest killer of men and women in the United States. The total numbers of individuals affected by CHD or by myocardial infarction (MI) in 2003 were 13.2 million and 7.2 million, respectively. Despite recent advances in therapy for these conditions, nearly 480,000 and 170,000 patients die annually from CHD or MI, respectively (2). In Japan, the total number of individuals with CHD is 0.9 million and nearly 50,000 people die annually from MI (Ministry of Health, Labor, and Welfare of Japan). Disease prevention is an important strategy for reducing the overall burden of CHD and MI, and the identification of markers for disease risk is key both for risk prediction and for potential intervention to reduce the chance of future events.

Several whole-genome linkage analyses of families or sibling-pairs (3–6) and various association studies of unrelated individuals (7–15) have attempted to identify genetic variations that contribute to CHD or MI. The genetic components of these conditions have not been determined definitively, however. We have now performed a large-scale association study for 164 polymor-

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Table 1: Characteristics of the 3,483 study subjects.

Characteristic	Myocardial infarction	Controls
No. of subjects	1192	2291
Age (years)	63.7 ± 10.6*	62.5 ± 11.8
Sex (male/female, %)	77.7/22.3†	43.1/56.9
Body mass index (kg/m^2)	23.7 ± 3.2‡	23.4 ± 3.1
Current or former smoker (%)	22.6†	16.1
Hypertension (%)	73.0†	44.3
Diabetes mellitus (%)	48.7†	20.0
Hypercholesterolemia (%)	56.9†	29.6

Data for age and body mass index are means ± SD. Smoker: smoking of ≥10 cigarettes daily. Hypertension: systolic blood pressure of ≥140 mmHg or diastolic blood pressure of ≥90 mmHg (or both), or taking antihypertensive medication. Diabetes mellitus: fasting blood glucose of ≥6.93 mM (126 mg/dl) or glycosylated hemoglobin of ≥6.5% (or both), or taking antidiabetes medication. Hypercholesterolemia: serum total cholesterol of ≥5.72 mM (220 mg/dl) or taking lipid-lowering medication. *P < 0.005, †P < 0.001, ‡P < 0.01 versus controls.

phisms of 137 candidate genes and MI in 3,483 Japanese individuals. The purpose of the present study was to identify gene polymorphisms that confer susceptibility to MI and thereby to provide a basis for the primary, personalized prevention of this condition.

Materials and methods

Study population

The study population comprised 3,483 unrelated Japanese individuals (1,913 men; 1,570 women) who either visited outpatient clinics of or were admitted to one of the six participating hospitals (Gifu Prefectural Gifu, Tajimi, and Gero Hot Spring Hospitals; Hirosaki University Hospital; Reimeikyo Rehabilitation Hospital; and Yokohama General Hospital) between October 2002 and March 2005. The 1,192 subjects with a first MI (926 men; 266 women) all underwent coronary angiography and left ventriculography. The diagnosis of MI was based on typical electrocardiographic changes and increases both in the serum activities of enzymes such as creatinine kinase, aspartate aminotransferase, and lactate dehydrogenase and in the serum concentration of troponin T. The diagnosis was confirmed by the presence of a wall motion abnormality on left ventriculography and identification of the responsible stenosis in any of the major coronary arteries or in the left main trunk by coronary angiography.

The control subjects comprised 2,291 individuals (987 men; 1,304 women) who visited the outpatient clinics of participating hospitals for an annual health checkup. They had no history of CHD, peripheral arterial occlusive disease, or other atherosclerotic diseases; of ischemic or hemorrhagic stroke or other cerebral diseases; or of other thrombotic, embolic, or hemorrhagic disorders. The study protocol complied with the Declaration of Helsinki and was approved by the Committees on the Ethics of Human Research of Mie University School of Medicine, Hirosaki University School of Medicine, Gifu International Institute of Biotechnology, and participating hospitals, and written informed consent was obtained from each participant.

Selection of polymorphisms

Our aim was to identify genes associated with MI in the Japanese population in a case-control association study by examining the relation of one to three polymorphisms of each candidate gene to MI. With the use of public databases [including PubMed (NCBI), Online Mendelian Inheritance in Man (NCBI), and GeneCanvas (INSERM, Paris, France; http://ecgene.net/genecanvas/news.php)], we selected 137 candidate genes that have been characterized and were suggested to be associated with MI on the basis of a comprehensive overview of vascular biology (from the viewpoint of atherosclerosis, arterial spasm, or arterial aneurysm); platelet function; leukocyte, lymphocyte, and monocyte-macrophage biology; coagulation and fibrinolysis cascades; neurological factors (from the viewpoint of regulation of the circulation, blood pressure, or endocrine function); as well as lipid and adipose tissue metabolism, insulin and glucose metabolism, peripheral insulin sensitivity, homocysteine metabolism, and other metabolic factors. On the basis of published studies and searches of PubMed, we further selected 164 polymorphisms of these genes – most located in the promoter region, exons, or splice donor or acceptor sites of introns – that might be expected to result in changes in the function or expression of the encoded protein (see Supplementary Table 1 online at www.thrombosis-online.com). Wild-type and variant alleles of the polymorphisms were determined from the original sources.

Genotyping of polymorphisms

Venous blood (7 ml) was collected into tubes containing 50 mM ethylenediaminetetraacetic acid (disodium salt), and genomic DNA was isolated with a kit (Genomix; Talent, Trieste, Italy). Genotypes of the 164 polymorphisms were determined (G&G

Table 2: Characteristics of male and female subjects.

	Men		Women	
	Myocardial infarction	Controls	Myocardial infarction	Controls
No. of subjects	926	987	266	1304
Age (years)	62.1 ± 10.5	62.8 ± 11.7	67.0 ± 10.0*	62.9 ± 11.9
Body mass index (kg/m^2)	23.8 ± 3.1*	23.2 ± 2.8	23.3 ± 3.4	23.4 ± 3.2
Current or former smoker (%)	27.2†	32.6	6.4‡	3.6
Hypertension (%)	71.0*	50.2	80.1*	39.9
Diabetes mellitus (%)	49.0*	23.8	47.7*	17.2
Hypercholesterolemia (%)	53.5*	24.7	68.8*	33.2

Data for age and body mass index are means ± SD. *P < 0.001, †P < 0.01, ‡P < 0.05 versus corresponding controls.

Table 3: Polymorphisms related ($P < 0.05$) to myocardial infarction as revealed by the chi^2-test.

Gene symbol	Polymorphism	*P*	FDR
MTHFR	677C→T (Ala222Val)	0.0003	0.049
LPL	1595C→G (Ser447Stop)	0.0005	0.041
IPF1	–108/3G→4G	0.0007	0.038
CETP	–629C→A	0.0045	0.185
GP1BA	1018C→T (Thr145Met)	0.0052	0.171
APOE	4070C→T (Arg158Cys)	0.0062	0.170
F7	11,496G→A (Arg353Gln)	0.0074	0.173
FABP2	2445G→A (Ala54Thr)	0.0075	0.154
TNF	–863C→A	0.0084	0.153
AGER	268G→A (Gly82Ser)	0.0084	0.138
TNF	–238G→A	0.0106	0.158
AKAP10	2073A→G (Ile646Val)	0.0112	0.153
ACDC	–11,377C→G	0.0197	0.249
PAI1	A→G (Tyr243Cys)	0.0341	0.400
TNFSF4	A→G	0.0379	0.414
APOC3	–482C→T	0.0389	0.399

FDR, false discovery rate.

Science, Fukushima, Japan) by a method that combines the polymerase chain reaction and sequence-specific oligonucleotide probes with analysis by suspension array technology (Luminex 100 flow cytometer; Luminex, Austin, TX, USA). Primers, probes, and other conditions for genotyping are shown in Supplementary Table 2 (see online at www.thrombosis-online.com). Detailed methodology for genotyping was described previously (16).

Statistical analysis

Clinical data were compared between subjects with MI and controls by the unpaired Student's t-test. Qualitative data were compared by the chi^2-test. Allele frequencies were estimated by the gene counting method, and the chi^2-test was used to identify departures from Hardy-Weinberg equilibrium. In the initial screen, the genotype distribution of each autosomal polymorphism was compared between subjects with MI and controls by the chi^2-test (3×2); for polymorphisms on the X chromosome, allele frequencies were compared by the chi^2-test (2×2). The relation of polymorphisms to MI was also examined for men or women separately as well as for individuals aged ≤62 or ≥63 years separately (mean age of total population, 62.9 years). The false discovery rate (FDR) was calculated by the method of Benjamini and Hochberg (17). Calculation of the FDR is an approach to dealing with the problems associated with multiple comparisons and provides a measure of the expected proportion of false positives among data. The FDR threshold is determined from the observed *P* value distribution and is adaptive to the signal level in data. The FDR differs from a *P* value, and much higher FDRs than *P* values can be tolerated. In the present study, the chi^2-test was used as an initial screen, and multivariable logistic regression analysis and a stepwise forward selection procedure were subsequently applied in a more rigorous evaluation of association. The FDR was calculated at each step of the statistical analysis. In the initial screen (the chi^2-test), the FDR was calculated from the distribution of *P* values for the 164 polymorphisms. Polymorphisms with an FDR of <0.05 were further examined by multivariable logistic regression analysis with adjustment for covariates, with MI as a dependent variable and independent variables including age, sex (0 = woman, 1 = man), body mass index (BMI), smoking status (0 = nonsmoker, 1 = smoker), metabolic variables (0 = no history of hypertension, diabetes mellitus, or hypercholesterolemia; 1 = positive history), and genotype of each polymorphism. Metabolic variables were evaluated by measurement of parameters or on the basis of current treatment or clinical history. Each genotype was assessed according to dominant, recessive, and two additive (additive 1 and 2) genetic models, and the *P* value, odds ratio, and 95% con-

Table 4: Polymorphisms related ($P < 0.05$) to myocardial infarction in men or women as revealed by the chi^2-test.

Men				Women			
Gene	Polymorphism	*P*	FDR	Gene	Polymorphism	*P*	FDR
AGER	G→A (Gly82Ser)	0.0005	0.082	*TNF*	–863C→A	0.0023	0.377
LTA	804C→A (Thr26Asn)	0.0042	0.344	*PLAT*	–7351C→T	0.0040	0.328
GNB3	825C→T (splice variant)	0.0069	0.377	*IPF1*	–108/3G→4G	0.0082	0.448
GP1BA	1018C→T (Thr145Met)	0.0101	0.414	*UCP3*	–55C→T	0.0116	0.476
F12	46C→T	0.0111	0.364	*FABP2*	2445G→A (Ala54Thr)	0.0168	0.551
MTHFR	677C→T (Ala222Val)	0.0121	0.331	*CETP*	–629C→A	0.0190	0.519
ACDC	–11,377C→G	0.0148	0.347	*PAX4*	C→T (Arg121Trp)	0.0244	0.572
LPL	1595C→G (Ser447Stop)	0.0168	0.344	*ROS1*	G→A (Asp2213Asn)	0.0274	0.562
F3	–603A→G	0.0185	0.337	*ENG*	C→G (Asp366His)	0.0291	0.530
F7	11,496G→A (Arg353Gln)	0.0284	0.466	*MTHFR*	677C→T (Ala222Val)	0.0307	0.504
TNF	–238G→A	0.0364	0.543	*ITGA2*	1648A→G (Lys505Glu)	0.0319	0.476
TNF	–850C→T	0.0369	0.504	*ABCA1*	1051G→A (Arg219Lys)	0.0393	0.537
APOE	4070C→T (Arg158Cys)	0.0387	0.488				

FDR, false discovery rate.

Table 5: Multivariate logistic regression analysis of polymorphisms related to myocardial infarction.

Gene symbol	Polymorphism	Dominant		Recessive		Additive 1		Additive 2	
		P (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)
MTHFR	677C→T (Ala222Val)	0.0515 (0.077)		0.0006 (0.002)	1.44 (1.17–1.78)	0.3892 (0.425)		0.0006 (0.002)	1.52 (1.20–1.92)
LPL	1595C→G (Ser447Stop)	0.0132 (0.026)	0.78 (0.64–0.95)	0.1206 (0.145)		0.0289 (0.050)	0.80 (0.65–0.98)	0.0937 (0.125)	
IPF1	–108/3G→4G	0.0004 (0.005)	0.71 (0.59–0.86)	0.4941 (0.494)		0.0004 (0.002)	0.70 (0.57–0.85)	0.0079 (0.019)	0.73 (0.58–0.92)

FDR, false discovery rate; OR, odds ratio; CI, confidence interval. Multivariable logistic regression analysis was performed with adjustment for age, sex, body mass index, and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia.

fidence interval were calculated. Each genetic model comprised two groups: the combined group of variant homozygotes and heterozygotes versus wild-type homozygotes for the dominant model; variant homozygotes versus the combined group of wild-type homozygotes and heterozygotes for the recessive model; heterozygotes versus wild-type homozygotes for the additive 1 model; and variant homozygotes versus wild-type homozygotes for the additive 2 model. For combined genotype analysis, multivariable logistic regression analysis was performed with MI as a dependent variable and independent variables including age, sex, BMI, smoking status, hypertension, diabetes mellitus, hypercholesterolemia, and combined genotypes. Each genotype was assessed according to a dominant or recessive model based on statistical significance, and each combined genotype was compared with a combined genotype that confers the lowest genetic risk for MI. We also performed a stepwise forward selection procedure to examine the effects of genotypes as well as of other covariates on MI. The levels for inclusion in and exclusion from the model were 0.25 and 0.1, respectively. Given the multiple comparisons of genotypes with MI, we adopted the criterion of FDR < 0.05 for significant association at each step of the statistical analysis. For other clinical background data, we adopted the criterion of $P < 0.05$ for significance. Statistical significance was examined by two-sided tests performed with JMP version 5.1 software (SAS Institute, Cary, NC, USA).

Results

The characteristics of the 3,483 study subjects are shown in Table 1. Age, the frequency of men, BMI, and the prevalence of conventional risk factors for CHD, including smoking, hypertension, diabetes mellitus, and hypercholesterolemia, were all greater in subjects with MI than in controls. The characteristics of subjects with MI and controls separated into men or women are shown in Table 2. For men, BMI and the prevalence of hypertension, diabetes mellitus, and hypercholesterolemia were greater, whereas the prevalence of smoking was lower, in subjects with MI than in controls. For women, age and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia were greater in subjects with MI than in controls.

Evaluation of genotype distributions or allele frequencies by the chi^2-test revealed that 16 polymorphisms were related ($P < 0.05$) to the prevalence of MI (Table 3). Among these polymorphisms, the 677C→T (Ala222Val) polymorphism of the 5,10-methylenetetrahydrofolate reductase gene (*MTHFR*), the 1595C→G (Ser447Stop) polymorphism of the lipoprotein lipase gene (*LPL*), and the –108/3G→4G polymorphism of the insulin promoter factor 1 gene (*IPF1*) were significantly (FDR < 0.05) associated with MI. We also examined the relation of polymorphisms to MI for men and women separately (Table 4). Although polymorphisms related to MI appeared to differ between men and women, no polymorphism was significantly associated with MI for men or for women based on the criterion of FDR < 0.05.

Table 6: Genotype distributions of polymorphisms associated with myocardial infarction.

Gene symbol	Polymorphism	Myocardial infarction	Controls
MTHFR	677C→T (Ala222Val)		
	CC	31.5	35.1
	CT	47.8	49.5
	TT	20.7	15.4
LPL	1595C→G (Ser447Stop)		
	CC	79.6	74.2
	CG	19.4	23.9
	GG	1.0	2.0
IPF1	–108/3G→4G		
	3G3G	27.0	21.3
	3G4G	47.1	51.7
	4G4G	25.9	27.0

Table 7: Effects of genotypes and other characteristics on the prevalence of myocardial infarction as determined by a stepwise forward selection procedure.

Variable	*P*	FDR	*R2*
Sex	<0.0001	<0.001	0.0887
Hypercholesterolemia	<0.0001	<0.001	0.0631
Diabetes mellitus	<0.0001	<0.001	0.0394
Hypertension	<0.0001	<0.001	0.0194
IPF1 (*4G4G* + *3G4G* vs. *3G3G*)	0.0004	<0.001	0.0028
MTHFR (*TT* versus *CC* + *CT*)	0.0006	0.001	0.0033
Age	0.0165	0.024	0.0013
LPL (*GG* + *CG* versus *CC*)	0.0217	0.027	0.0011

FDR, false discovery rate; R^2, contribution rate.

Table 8: Assessment of genetic risk for myocardial infarction with combined genotypes for three polymorphisms.

MTHFR (0 = *CC* = *CT*, 1 = *TT*)	*LPL* (0 = CC, 1 = CG = GG)	*IPF1* (0 = *3G3G*, 1 = *3G4G* = *4G4G*)	No. of subjects with MI/controls	OR (95% CI)	*P*	FDR
1	0	0	56/59	2.54 (1.57–4.11)	0.0001	<0.001
1	1	0	13/15	2.41	0.0611	0.107
0	0	0	208/316	1.73 (1.28–2.34)	0.0004	0.001
1	0	1	145/207	1.70 (1.23–2.35)	0.0015	0.004
1	1	1	33/71	1.34	0.2834	0.331
0	0	1	540/1117	1.21	0.1335	0.187
0	1	0	45/98	1.19	0.4598	0.460
0	1	1	152/408	1.00		

OR, odds ratio; CI, confidence interval; FDR, false discovery rate. Multivariable logistic regression analysis was performed with adjustment for age, sex, body mass index, and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia.

Polymorphisms that satisfied the condition $P < 0.01$ were evaluated by multivariable logistic regression analysis with adjustment for age, BMI, and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia for men (see Supplementary Table 3 online at www.thrombosis-online.com) and for women (see Supplementary Table 4 online at www.thrombosis-online.com).

The three polymorphisms associated with MI in the entire study population were examined further by multivariable logistic regression analysis with adjustment for age, sex, BMI, and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia. The 677C→T polymorphism of *MTHFR* (recessive and additive 2 models), the 1595C→G polymorphism of *LPL* (dominant model), and the –108/3G→4G polymorphism of *IPF1* (dominant and additive 1 and 2 models) were found to be significantly (FDR < 0.05) associated with the prevalence of MI (Table 5). The 677*T* allele of *MTHFR* represented a risk factor for MI, whereas the 1595*G* allele of *LPL* and the –108/*4G* allele of *IPF1* were protective against this condition. The genotype distributions of these polymorphisms both in control subjects and in patients with MI were in Hardy-Weinberg equilibrium (Table 6).

We next performed a stepwise forward selection procedure to examine the effects of genotypes for the 677C→T polymorphism of *MTHFR*, the 1595C→G polymorphism of *LPL*, and the –108/3G→4G polymorphism of *IPF1* as well as of age, sex, BMI, and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia on MI (Table 7). Sex, hypercholesterolemia, diabetes mellitus, hypertension, *IPF1* genotype (dominant model), *MTHFR* genotype (recessive model), age, and *LPL* genotype (dominant model), in descending order of statistical significance, were significant and independent (FDR < 0.05) determinants of the prevalence of MI.

We performed multivariable logistic regression analysis of combined genotypes for assessment of genetic risk for MI. Combined genotype analysis of three polymorphisms (677C→T of *MTHFR*, 1595C→G of *LPL*, and –108/3G→4G of *IPF1*) revealed that the maximal odds ratio of 2.54 was obtained for the combined genotype of *TT* for *MTHFR*, *CC* for *LPL*, and *3G3G* for *IPF1*, whereas the lowest genetic risk was apparent with the combined genotype of *CC* or *CT* for *MTHFR*, *CG* or *GG* for *LPL*, and *3G4G* or *4G4G* for *IPF1* (Table 8).

Finally, we examined the effect of age on the association of the three polymorphisms with MI. Given that the mean age of the total population was 62.9 years, we divided subjects into those aged ≤62 years and those aged ≥63 years. The chi^2-test revealed that the 1595C→G polymorphism of *LPL* and the –108/3G→4G polymorphism of *IPF1*, but not the 677C→T polymorphism of *MTHFR*, were associated (FDR < 0.05) with MI in individuals aged ≤62 years, whereas the 677C→T polymorphism of *MTHFR* and the 1595C→G polymorphism of *LPL*, but not the –108/3G→4G polymorphism of *IPF1*, were associated with MI in individuals aged ≥63 years (Table 9).

Polymorphisms that satisfied FDR < 0.05 in the chi^2-test were further examined by multivariable logistic regression analysis with adjustment for age, sex, BMI, and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia. Among individuals aged ≤62 years, the –108/3G→4G polymorphism of *IPF1* (dominant and additive 1 and 2 models) was significantly (FDR < 0.05) associated with MI, whereas the 1595C→G polymorphism of *LPL* was not (Table 10). Among individuals aged ≥63 years, the 677C→T polymorphism of *MTHFR* (recessive and additive 2 models) was significantly associated with MI, whereas the 1595C→G polymorphism of *LPL* was not.

Table 9: Relation of polymorphisms of *MTHFR*, *LPL*, and *IPF1* to myocardial infarction in individuals aged ≤62 or ≥63 years as revealed by the chi^2-test.

		≤62 years (*n* = 530/1106)*		≥63 years (*n* = 662/1185)*	
Gene	Polymorphism	*P*	FDR	*P*	FDR
MTHFR	677C→T (Ala222Val)	0.1418	0.170	0.0017	0.005
LPL	1595C→G (Ser447Stop)	0.0148	0.030	0.0245	0.037
IPF1	–108/3G→4G	0.0004	0.002	0.1775	0.178

FDR, false discovery rate. *Number of subjects with MI/controls.

Discussion

We have examined the relation of 164 polymorphisms in 137 candidate genes to MI. Our large-scale association study with 3,483 subjects revealed that the 677C→T polymorphism of *MTHFR*, the 1595C→G polymorphism of *LPL*, and the –108/3G→4G polymorphism of *IPF1* were significantly associated with the prevalence of MI in a Japanese population. Com-

Table 10: Multivariate logistic regression analysis of polymorphisms related to myocardial infarction in individuals aged ≤62 or ≥63 years.

Gene symbol	Polymorphism	Dominant		Recessive		Additive 1		Additive 2	
		P (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)
≤62 years									
LPL	1595C→G (Ser447Stop)	0.0855 (0.195)		0.1125 (0.164)		0.1498 (0.200)		0.0991 (0.198)	
IPF1	−108/3G→4G	0.0002 (0.003)	0.58 (0.43–0.77)	0.2511 (0.309)		0.0005 (0.004)	0.57 (0.42–0.78)	0.0030 (0.010)	0.58 (0.41–0.83)
≥63 years									
MTHFR	677C→T (Ala222Val)	0.0995 (0.177)		0.0009 (0.005)	1.57 (1.20–2.06)	0.5795 (0.580)		0.0015 (0.006)	1.64 (1.21–2.22)
LPL	1595C→G (Ser447Stop)	0.0695 (0.185)		0.3349 (0.357)		0.1040 (0.166)		0.2838 (0.324)	

FDR, false discovery rate; OR, odds ratio; CI, confidence interval. Multivariable logistic regression analysis was performed with adjustment for age, sex, body mass index, and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia.

bined genotype analysis of these three polymorphisms yielded a maximal odds ratio of 2.54 for predisposition to MI. The association of these three polymorphisms with MI was affected by age: Among individuals aged ≤62 years or ≥63 years, the –108/3G→4G polymorphism of *IPF1* and the 677C→T polymorphism of *MTHFR*, respectively, were associated with MI. Although polymorphisms related to MI appeared to differ between men and women, no polymorphism was significantly associated with this condition in men or in women separately.

Homocysteine is a sulfur-containing amino acid that plays a pivotal role in methionine metabolism. 5,10-Methylenetetrahydrofolate reductase (MTHFR) catalyzes the reduction of 5,10-methylenetetrahydrofolate to 5-methylenetetrahydrofolate, a reaction that provides a substrate for the methylation of homocysteine to methionine catalyzed by methionine synthase. Individuals with the 677C→T (Ala222Val) substitution of *MTHFR* manifest reduced MTHFR activity and higher homocysteine levels compared with those without it (18–20). Association of the 677C→T polymorphism of *MTHFR* with CHD or MI has been described (21–24). Other studies, however, did not support such an association (20, 25, 26). These apparently contradictory results are attributable, at least in part, to differences in intake of folate and other B vitamins (27). A meta-analysis of the association of the 677C→T polymorphism of *MTHFR* with the risk of CHD in 11,162 cases and 12,758 controls from 40 studies revealed that individuals with the *TT* genotype had an odds ratio of 1.16 for CHD compared with those with the *CC* genotype (28). These observations suggest that impaired folate metabolism, resulting in high homocysteine concentrations, is an important determinant of CHD. Another meta-analysis of the association of the 677C→T polymorphism of *MTHFR* with CHD in 26,000 cases and 31,183 controls from 80 studies yielded an overall odds ratio of 1.14 for the *TT* genotype versus the *CC* genotype; odds ratios for Europe, Australia, and North America were about 1.0, whereas those for the Middle East and Asia were 2.61 and 1.23, respectively (29). These results indicate that the 677C→T polymorphism of *MTHFR* is associated with CHD in the Middle East and Asia, but not in Europe, North America, or Australia, with this geographic variability possibly reflecting higher folate intake in the latter regions (29). These previous observations support our present results showing that the 677C→T polymorphism of *MTHFR* was significantly associated with the prevalence of MI in Japanese, with the *TT* genotype being a risk factor for this condition.

Lipoprotein lipase (LPL) is the rate-limiting enzyme in lipolysis of triglyceride-rich lipoproteins in the circulation. It is synthesized in parenchymal cells of adipose tissue as well as in skeletal and cardiac muscle, and it is then transferred to heparan sulfate binding sites of the vascular endothelium (30). The hydrolytic function of LPL is important for the processing of triglyceride-rich chylomicrons and very low density lipoproteins to remnant particles as well as for the transfer of phospholipids and apolipoproteins to high density lipoproteins. LPL also plays an important role in the receptor-mediated removal of lipoproteins from the circulation (31). *LPL* is polymorphic, with amino acid substitutions affecting triglyceride and HDL-cholesterol levels, which are implicated in atherosclerosis risk (32). The 1595C→G (Ser447Stop) polymorphism of *LPL* results in carboxyl-terminal truncation of LPL by two amino acids. This change is thought to increase the binding affinity of the protein for receptors or to facilitate or otherwise affect its formation of dimers (32). The *G* allele of the 1595C→G (Ser447Stop) polymorphism has also been shown to be related to decreased plasma triglyceride or increased HDL-cholesterol levels, or both (31–37). In addition, the *G* (Stop) allele of this polymorphism was found to be associated with a reduced risk of CHD or MI (32, 38). The previous observations suggest that the catalytic activity and stability of the truncated variant of LPL may be largely normal, but that it may be present at higher concentrations in the circulation, resulting in a higher level of LPL activity (31, 39–41). Our present results indicate that the 1595C→G (Ser447Stop) polymorphism of *LPL* is associated with the prevalence of MI, with the *G* (Stop) allele protecting against this condition, consistent with the previous observations (32, 38).

Insulin promoter factor 1 (IPF1) is a homeodomain-containing protein that is a key regulator of the insulin gene in pancreatic β cells (42, 43) and plays an important role in development of the pancreas (44, 45). IPF1-deficient mice thus selectively lack the pancreas at birth (44), and a patient with pancreatic agenesis and insulin-deficient diabetes was found to have a single nucleotide deletion in codon 63 of *IPF1* that caused a frameshift in the transactivation domain (45). A 3G→4G polymorphism of *IPF1* was identified 108 bp upstream of the translation start site in the

Japanese population but was found not to be related to the prevalence of type 2 diabetes mellitus (46, 47). Our results indicate that the 3G→4G polymorphism of *IPF1* was significantly associated with MI, with the *4G* allele protecting against this condition. This is the first demonstration of an association of this polymorphism in *IPF1* with MI, although the underlying molecular mechanism remains to be elucidated.

Given the multiple comparisons of genotypes with MI in the present study, we adopted the criterion of FDR < 0.05 for significant association in each step of the statistical analysis. It is not possible, however, to exclude completely potential statistical errors such as false positives. Validation of our findings will require their replication with independent subject panels. It is also possible that one or more of the polymorphisms associated with MI in our study are in linkage disequilibrium with polymorphisms of other nearby genes that are actually responsible for the development of this condition. Furthermore, the relevance of the identified polymorphisms to gene transcription or to protein structure or function was not determined in the present study.

In conclusion, our present results suggest that *MTHFR*, *LPL*, and *IPF1* are susceptibility loci for MI in the Japanese population. Determination of combined genotypes for these polymorphisms may prove informative for assessment of the genetic risk for MI and may contribute to the primary, personalized prevention of this condition.

Appendix

In addition to the authors, the following investigators participated in the study: Y. Matsuno and M. Tomita (Gifu Prefectural Gifu Hospital, Gifu); T. Kameyama and M. Oguri (Gifu Prefectural Tajimi Hospital, Tajimi); S. Tanihata (Gifu Prefectural Gero Hot Spring Hospital, Gero); M. Hiramoto (Yokohama General Hospital, Yokohama); and nursing and laboratory staff at the participating hospitals.

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Supplementary Tables to Yamada et al. "Genetic risk for myocardial infarction" TH06-02-0117

Supplementary Table 1. The 164 polymorphisms examined in the study.

Locus	Gene	Symbol	Polymorphism	dbSNP*
1p36.3	5,10-Methylenetetrahydrofolate reductase	*MTHFR*	677C→T (Ala222Val)	rs1801133
1p36.2	Natriuretic peptide precursor A	*NPPA*	664G→A (Val7Met)	rs5063
1p36	Urotensin II	*UTS2*	347G→A (Ser89Asn)	rs2890565
1p35.1	Gap junction protein, alpha-4	*GJA4*	1019C→T (Pro319Ser)	rs1764391
1p35.1	Gap junction protein, beta-4	*GJB4*	C→T (Arg103Cys)	rs9426009
1p34.3	Ischemia/reperfusion inducible protein	*FLJ23476*	C→A (Pro55Gln)	rs16824518
1p34.3	Ischemia/reperfusion inducible protein	*FLJ23476*	T→C (Met769Thr)	rs11488569
1p34.2	Polycystic kidney disease 1-like	*PKD1-like*	G→A (Gly243Asp)	rs1635712
1p34.1-p32	Proprotein convertase, subtilisin/kexin-type, 9	*PCSK9*	23968A→G (Glu670Gly)	rs505151
1p22-p21	Coagulation factor III	*F3*	-603A→G	rs1361600
1p22.1	Glutamate-cysteine ligase, modifier subunit	*GCLM*	-588C→T	(U72210)
1q21-q23	C-reactive protein, pentraxin-related	*CRP*	1444C→T	rs1130864
1q23-q25	Selectin E	*SELE*	561A→C (Ser128Arg)	rs5361
1q23-q25	Selectin P	*SELP*	G→T (Val640Leu)	rs6133
1q25	Tumor necrosis factor ligand superfamily, member 4	*TNFSF4*	A→G	rs3850641
1q31-q32	Interleukin 10	*IL10*	-819T→C	rs1800871
1q31-q32	Interleukin 10	*IL10*	-592A→C	rs1800872
1q42-q43	Angiotensinogen	*AGT*	-6G→A	rs5051
2q14	Interleukin 1-beta	*IL1B*	-511C→T	rs16944
2q31	Collagen, type III, alpha-1	*COL3A1*	2209G→A (Ala698Thr)	rs1800255
2q31	Collagen, type III, alpha-1	*COL3A1*	3730A→G (Ile1205Val)	rs2271683
2q36	Insulin receptor substrate 1	*IRS1*	3931G→A (Gly972Arg)	rs1801278
2q37.3	Calpain 10	*CAPN10*	4852G→A	rs3792267
3pter-p21	Chemokine, CX3C motif, receptor 1	*CX3CR1*	926C→T (Thr280Met)	rs3732378
3p25	Peroxisome proliferator-activated receptor-gamma	*PPARG*	-681C→G	rs10865710
3p25	Peroxisome proliferator-activated receptor-gamma	*PPARG*	34C→G (Pro12Ala)	rs1801282
3p22	Transforming growth factor-beta receptor, type II	*TGFBR2*	1167C→T (Asn389Asn)	rs2228048
3p22-p21.3	Phospholipase C, delta-1	*PLCD1*	864G→A (Arg257His)	rs933135
3p21.3	Glutathione peroxidase	*GPX1*	C→T (Pro198Leu)	rs1050450

3p21	Chemokine, CC motif, receptor 2	*CCR2*	190G→A (Val64Ile)	rs1799864
3p21	Chemokine, CC motif, receptor 5	*CCR5*	59029G→A	rs1799987
3q21-q25	Angiotensin receptor 1	*AGTR1*	1166A→C	rs5186
3q21-q25	Angiotensin receptor 1	*AGTR1*	G→A (Ala163Thr)	rs12721226
3q24-q25	Purinergic receptor P2Y, G protein-coupled, 12	*P2RY12*	744T→C	(NC_000003)
3q26.1-q26.2	Butyrylcholinesterase	*BCHE*	1615G→A (Ala539Thr)	rs1803274
3q26.3-q27	Thrombopoietin	*THPO*	5713A→G	rs6141
3q27	Adipocyte, C1Q, and collagen domain containing	*ACDC*	-11377C→G	rs266729
3q28	Adaptor-related protein complex 2, MU-1 subunit	*AP2M1*	62G→T	rs1501299
4p15.1	Peroxisome proliferator-activated receptor-gamma, coactivator 1	*PPARGC1*	1564G→A (Gly482Ser)	rs8192678
4q22-q24	Microsomal triglyceride transfer protein, 88-kD	*MTP*	-493G→T	rs1800591
4q26-q28	Annexin A5	*ANXA5*	-1C→T	rs11575945
4q28	Fibrinogen, B beta polypeptide	*FGB*	-455G→A	rs1800790
4q28	Fibrinogen, B beta polypeptide	*FGB*	8059G→A (Arg448Lys)	rs4220
4q28-q31	Fatty acid-binding protein 2	*FABP2*	2445G→A (Ala54Thr)	rs1799883
4q31	Uncoupling protein 1	*UCP1*	-112A→C	rs10011540
4q31.22	Endothelin receptor, type A	*EDNRA*	-231A→G	rs1801708
5q12	Phosphodiesterase 4D, cAMP-specific	*PDE4D*	TAAA→- (3'-UTR)	rs3839219
5q13	Thrombospondin IV	*THBS4*	1186G→C (Ala387Pro)	rs1866389
5q13	Phosphatidylinositol 3-kinase, regulatory, 1	*PIK3R1*	1020G→A (Met326Ile)	rs3730089
5q23-q31	Integrin, alpha-2	*ITGA2*	1648A→G (Lys505Glu)	rs10471371
5q32-q34	Beta-2-adrenergic receptor	*ADRB2*	46A→G (Arg16Gly)	rs1042713
5q32-q34	Beta-2-adrenergic receptor	*ADRB2*	79C→G (Gln27Glu)	rs1042714
5q33-qter	Factor XII	*F12*	46C→T	rs17876008
6p24-p23	Endothelin 1	*EDN1*	5665G→T (Lys198Asn)	rs5370
6p21.3	Lymphotoxin-alpha	*LTA*	804C→A (Thr26Asn)	rs2229093
6p21.3	Tumor necrosis factor	*TNF*	-863C→A	rs1800630
6p21.3	Tumor necrosis factor	*TNF*	-850C→T	rs1799724
6p21.3	Tumor necrosis factor	*TNF*	-238G→A	rs361525
6p21.3	Advanced glycosylation end product-specific receptor	*AGER*	268G→A (Gly82Ser)	rs2070600
6p21.2-p21.1	Peroxisome proliferator-activated receptor-delta	*PPARD*	294T→C	rs2016520
6p21.2-p12	Phospholipase A2, group VII	*PLA2G7*	994G→T (Val279Phe)	rs16874954
6p21	Solute carrier family 26 (sulfate transporter), member 8	*SLC26A8*	A→G (Ile639Val)	rs2295852
6p12	Glutamate-cysteine ligase, catalytic subunit	*GCLC*	-129C→T	rs17883901
6p12	Vascular endothelial growth factor	*VEGF*	936C→T	rs3025039

6q22	v-Ros avian UR2 sarcoma virus oncogene homolog 1	*ROS1*	G→A (Asp2213Asn)	rs529038
6q22-q23	Ectonucleotide pyrophosphatase/phosphodiesterase 1	*ENPP1*	97A→C (Lys121Gln)	rs1044498
6q25.1	Estrogen receptor 1	*ESR1*	-1989T→G	rs2071454
6q27	Thrombospondin II	*THBS2*	3949T→G	rs8089
7p21	Interleukin 6	*IL6*	-572G→C	rs1800796
7p15-p13	Glucokinase	*GCK*	-30G→A	rs1799884
7q11.2	Syntaxin 1A	*STX1A*	205T→C (Asp68Asp)	rs2293485
7q11.2	Elastin	*ELN*	1264G→A (Gly422Ser)	rs2071307
7q11.2	CD36 antigen	*CD36*	30294G→C	rs1049673
7q11.23-q21.11	Protein phosphatase 1, regulatory subunit 3A	*PPP1R3A*	2647G→T (Ser883Arg)	(X78578)
7q11.23-q21.11	Protein phosphatase 1, regulatory subunit 3A	*PPP1R3A*	2711G→T (Tyr905Asp)	rs1799999
7q21.3	Paraoxonase 1	*PON1*	-162G→A	rs705381
7q21.3	Paraoxonase 1	*PON1*	532A→G (Arg160Gly)	rs13306698
7q21.3	Paraoxonase 1	*PON1*	584G→A (Gln192Arg)	rs662
7q21.3	Paraoxonase 2	*PON2*	475C→G (Ala148Gly)	rs11545941
7q21.3-q22	Plasminogen activator inhibitor 1	*PAI1*	-668/4G→5G	rs1799768
7q21.3-q22	Plasminogen activator inhibitor 1	*PAI1*	A→G (Tyr243Cys)	rs13306846
7q22.1	Collagen, type I, alpha-2	*COL1A2*	G→C (Ala459Pro)	rs42524
7q32	Paired box gene 4	*PAX4*	567C→T (Arg121Trp)	(AF043978)
7q36	Nitric oxide synthase 3	*NOS3*	-786T→C	rs2070744
8p22	Lipoprotein lipase	*LPL*	1595C→G (Ser447Stop)	rs328
8p21-p12	Epoxide hydrolase 2, cytosolic	*EPHX2*	G→A (Arg287Gln)	rs751141
8p12	Plasminogen activator, tissue	*PLAT*	-7351C→T	rs2020918
8p12-p11.2	Beta-3-adrenergic receptor	*ADRB3*	190T→C (Trp64Arg)	rs4994
8p12-p11.2	RecQ protein-like 2	*RECQL2*	47765T→C (Cys1367Arg)	rs1346044
9q22-q31	ATP-binding cassette, subfamily A, member 1	*ABCA1*	1051G→A (Arg219Lys)	rs2230806
9q22-q31	ATP-binding cassette, subfamily A, member 1	*ABCA1*	2583A→G (Ile823Met)	rs4149313
9q34.1	Endoglin	*ENG*	1691C→G (Asp366His)	rs1800956
9q34.2-q34.3	Prostaglandin D2 synthase, brain	*PTGDS*	4111A→C	rs6926
10q11.2	Arachidonate 5-lipoxygenase	*ALOX5*	G→A (Glu254Lys)	rs2228065
10q24-q26	Beta-1-adrenergic receptor	*ADRB1*	1165G→C (Gly389Arg)	rs1801253
11p15.5	Insulin	*INS*	-23T→A	rs689
11p15.1	Potassium chnnel, inwardly rectifying, subfamily J, member 11	*KCNJ11*	276A→G (Glu23Lys)	rs5219
11p15.1	ATP-binding cassette, subfamily C, member 8	*ABCC8*	3857G→A (Arg1273Arg)	rs4148643
11q13	Uncoupling protein 2	*UCP2*	-866G→A	rs659366

11q13	Uncoupling protein 3	*UCP3*	-55C→T	rs1800849
11q22.2-q22.3	Matrix metalloproteinase 12	*MMP12*	-82A→G	rs2276109
11q22-q23	Matrix metalloproteinase 1	*MMP1*	-1607/1G→2G	rs1799750
11q23	Apolipoprotein A-I	*APOA1*	-75G→A	rs670
11q23	Apolipoprotein A-I	*APOA1*	84T→C	rs5070
11q23	Apolipoprotein A-V	*APOA5*	-1131T→C	rs662799
11q23	Apolipoprotein C-III	*APOC3*	-482C→T	rs2854117
11q23	Apolipoprotein C-III	*APOC3*	1100C→T	rs4520
11q23	Matrix metalloproteinase 3	*MMP3*	-1171/5A→6A	rs3025058
11q23	Matrix metalloproteinase 3	*MMP3*	A→G (Lys45Glu)	rs679620
11q23.3-q25	Heat-shock 70-kD protein 8	*HSPA8*	-110A→C	rs1008438
12p13	Guanine nucleotide-binding protein, beta-3	*GNB3*	825C→T (splice variant)	rs5443
12p13-p12	Low density lipoprotein, oxidized, receptor 1	*OLR1*	501G→C (Lys167Asn)	rs11053646
13q12.1	Insulin promoter factor 1	*IPF1*	-108/3G→4G	(S82168)
13q14.11	Carboxypeptidase B2, plasma	*CPB2*	529G→A (Ala147Thr)	rs3742264
13q14.11	Carboxypeptidase B2, plasma	*CPB2*	T→C (Ile347Thr)	rs1926447
13q34	Factor VII	*F7*	11496G→A (Arg353Gln)	rs6046
13q34	Protein Z	*PROZ*	79G→A	rs3024735
14q11.2	Cathepsin G	*CTSG*	2108A→G (Asn125Ser)	(J04990)
14q32.1	Alpha-1-antichymotrypsin	*AACT*	50G→A (Ala15Thr)	rs4934
14q32.1-q32.2	Bradykinin receptor B2	*BDKRB2*	C→T (Arg14Cys)	rs1046248
15q21.1	Fibrillin 1	*FBN1*	1875T→C	rs25458
15q21-q23	Lipase, hepatic	*LIPC*	-250G→A	rs2070895
16p13.11	Hypertension-associated SA, rat, homolog of	*SAH*	A→G (-7 from exon 13)	rs13306607
16p13	Major histocompatibility complex, class II, transactivator	*MHC2TA*	-168A→G	rs3087456
16q13	Matrix metalloproteinase 2	*MMP2*	-1306C→T	rs243865
16q21	Cholesteryl ester transfer protein, plasma	*CETP*	-629C→A	rs1800775
16q21	Cholesteryl ester transfer protein, plasma	*CETP*	1061A→G (Ile405Val)	rs5882
16q24	Cytochrome b(-245), alpha subunit	*CYBA*	242C→T (His72Tyr)	rs4673
17pter-p12	Glycoprotein Ib, platelet, alpha polypeptide	*GP1BA*	-5T→C	rs2243093
17pter-p12	Glycoprotein Ib, platelet, alpha polypeptide	*GP1BA*	1018C→T (Thr145Met)	rs6065
17p13	Chemokine, CXC motif, ligand 16	*CXCL16*	C→T (Ala181Val)	rs2277680
17p11.2	Sterol regulatory element-binding transcription factor 1	*SREBF1*	-36G→-	(AX977070)
17p11.1	A-kinase anchoring protein 10	*AKAP10*	2073A→G (Ile646Val)	rs203462
17q11.2-q12	Chemokine, CC motif, ligand 5	*CCL5*	-28C→G	rs2280788

17q11.2-q12	Chemokine, CC motif, ligand 5	*CCL5*	-403G→A	rs2107538
17q21.1-q21.2	Chemokine, CC motif, ligand 11	*CCL11*	G→A (Ala23Thr)	rs3744508
17q23	Angiotensin I- converting enzyme	*ACE*	-240A→T	rs4291
17q23	Platelet-endothelial cell adhesion molecule 1	*PECAM1*	1454C→G (Leu125Val)	rs668
17q23-qter	Apolipoprotein H	*APOH*	341G→A (Ser88Asn)	rs1801692
18q21.1	Lipase, endothelial	*LIPG*	584C→T (Thr111Ile)	rs2000813
19p13.3	Resistin	*RETN*	-420C→G (C-180G)	rs1862513
19p13.3	Resistin	*RETN*	-180C→G	rs1862513
19p13.3	Resistin	*RETN*	+62G→A	rs3745368
19p13.3-p13.2	Intercellular adhesion molecule 1	*ICAM1*	1462G→A (Glu469Lys)	rs5498
19p13.2	Insulin receptor	*INSR*	7067365C→A	rs2860172
19p13.2	Low density lipoprotein receptor	*LDLR*	1184G→A (Ala370Thr)	rs11669576
19q13.1	Transforming growth factor, beta-1	*TGFB1*	-509C→T	rs1800469
19q13.2	Apolipoprotein E	*APOE*	-219G→T	rs405509
19q13.2	Apolipoprotein E	*APOE*	3932T→C (Cys112Arg)	rs429358
19q13.2	Apolipoprotein E	*APOE*	4070C→T (Arg158Cys)	rs7412
19q13.3	Glycogen synthase 1	*GYS1*	260A→G (Met416Val)	rs5447
19q13.4	Glycoprotein VI, platelet	*GP6*	13254T→C (Ser219Pro)	rs1613662
20p11.2	Thrombomodulin	*THBD*	2136C→T (Ala455Val)	rs1042579
20q11.2-q13.1	Matrix metalloproteinase 9	*MMP9*	855G→A (Arg279Gln)	rs2664538
20q12-q13.1	Hepatocyte nuclear factor 4-alpha	*HNF4A*	A→G	rs2425640
20q13.11-q13.13	Prostaglandin I2 synthase	*PTGIS*	1117C→A	rs6095558
20q13.31	Phosphoenolpyruvate carboxykinase 1, soluble	*PCK1*	-232C→G	rs2071023
21q22.3	Integrin, beta-2	*ITGB2*	1323C→T	rs235326
22q11.2	Catechol-O-methyltransferase	*COMT*	G→A (Val158Met)	rs4680
22q12	Heme oxygenase 1	*HMOX1*	-413T→A	rs2071746
22q12	Heme oxygenase 1	*HMOX1*	99G→C (Asp7His)	rs2071747
22q12-q13	Lectin, garactoside-binding, soluble, 2	*LGALS2*	3279C→T (intron 1)	rs7291467
Xq22-q23	Angiotensin II receptor, type 2	*AGTR2*	1675G→A	rs1403543
Xq22-q23	Angiotensin II receptor, type 2	*AGTR2*	3123C→A	rs11091046

*In instances in which rs numbers in dbSNP were not detected, NCBI GenBank accession numbers are shown in parentheses.

Supplementary Table 2. Primers, probes, and other PCR conditions for genotyping

Gene Symbol	Polymorphism	Sense primer	Antisense primer	Probe 1	Probe 2	Annealing (ºC)	Cycles (times)
MTHFR	677C→T (Ala222Val)	gAggCTgACCTgAAgCACTTg	CAAAgCggAAgAATgTgTCAgC	CTgCgggAgCCgATTTCATCA	gATgATgAAATCgACTCCCg	60	50
LPL	1595C→G (Ser447Stop)	gCAgAAAggAAAggCACCTgC	TAgggTgCAAgCTCAggATgC	TgCTCACCAgCCTgACTTCTTA	CTCTgAATAAgAAgTgAggCT	60	50
IPF1	-108/3G→4G	TggCTgTgggTTCCCTCTgAg	gATTTggCACTgTgTggCgTTC	CgAgCAggggTggCgCC	ggCgCCACCCTgCTCgCT	60	50
CETP	-629C→A	TCCCggAggCAgCCAATgATC	TATgTAgACTTTCCTTgATATgCATAA	AggCTgTATACCCCCCCAgAgT	AggCTgTATACCCACCCAgA	60	50
GP1BA	1018C→T (Thr145Met)	ggCgAACTCCAAgAgCTCTAC	AgCggggAgCTCAgTCAAgT	AgggCTCCTgACgCCCACA	TTgggTgTgggCATCAggAg	60	50
APOE	4070C→T (Arg158Cys)	CCACCTgCgCAAgCTgCgTA	gCTCgCggATggCgCTgAg	TgACCTgCAgAAgCgCCTgg	gTACACTgCCAggCACTTCTg	60	50
F7	11496G→A (Arg353Gln)	CggCTACTCggATggCAgCA	CCAAAgTggCCCACggTTgC	TACCACgTgCCCCggTAgTg	gCCACCCACTACCAgggCA	60	50
FABP2	2445G→A (Ala54Thr)	AgCTgACAATTACACAAgAAggAA	gTTgTAATTAAAggTgACACCAAg	AATgTTTCgAAAAgCgCTTgATT	TCAAAgAATCAAgCACTTTTCgA	60	50
TNF	-863C→A	ggAgATgTgACCACAgCAATgg	ggTCCTggAggCTCTTTCACT	ggACCCCCACTTAACgAAg ggACCCCCCCTTAACgAAg	ggACCCCCACTTAATgAAgAC gACCCCCCCTTAATgAAgAC	60	50
AGER	268G→A (Gly82Ser)	ACAggCCggACAgAAgCTTgg	CATTCCTgTTCATTgCCTggCAC	gAAgAgggAgCCgTTgggAA	gTCCTTCCCAACAgCTCCCT	60	50
TNF	-283G→A	gTCCTACACACAAATCAgTCAgT	gACACACAAgCATCAAggATACC	CTCCCTgCTCCgATTCCgT	CCCTCggAATCAgAgCAgg	60	50
AKAP10	2073A→G (Ile646Val)	ggCCCAggAAgAgCTAgCTTg	gTAgATTTCTCTAACggTTgATCAT	gATAgTCAgTgACATTATgCAg	CCTgCTgCATAACgTCACTg	60	50
ACDC	-11377C→G	TAATTCATCAgAATgTgTggCTTg	TTAggCTTgAAgTggCAACATTC	gCTCAgATCCTgCCCTTCAAA	gTTTTgTTTTTgAAgCgCAggAT	60	50
PAI1	A→G (Tyr243Cys)	TCTTgTCgTCTTCACAgCTgAgT	AggggCAgCAATgAACATgCTg	CAggATgTCgTAgTAATggC	CAggATgTCgTAgCAATggC	60	50
TNFSF4	A→G	TAATTgCCTgATCAAACACATTAC	ACTTTgAAgCTTTgAgTCACTgAT	CTggTCTACCCATTgTgATAg	CTggTCTACCCACTgTgATAg	60	
APOC3	-482C→T	AggggCTgTgAgAgCTCAgC	AggggCTTCTTCAgACTTgAgA	ggCACAgAAgACCAggCATCA	gCCACTgATgCCCggTCTTC	60	50

Supplementary Table 3. Multivariable logistic regression analysis of polymorphisms related to myocardial infarction in men.

Gene	Polymorphism	Dominant model		Recessive model		Additive 1 model		Additive 2 model	
		P (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)
AGER	G→A (Gly82Ser)	0.4326 (0.519)		0.0043 (0.017)	0.28 (0.11–0.64)	0.1316 (0.197)		0.0061 (0.018)	0.30 (0.12–0.67)
LTA	804C→A (Thr26Asn)	0.0030 (0.018)	1.36 (1.11–1.67)	0.6354 (0.693)		0.0028 (0.034)	1.39 (1.12–1.73)	0.0936 (0.160)	
GNB3	825C→T (splice variant)	0.3828 (0.510)		0.0072 (0.017)	1.38 (1.09–1.73)	0.9894 (0.989)		0.0261 (0.052)	1.38 (1.04–1.82)

FDR, false discovery rate; OR, odds ratio; CI, confidence interval. Multivariable logistic regression analysis was performed with adjustment for age, BMI, and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia.

Supplementary Table 4. Multivariate logistic regression analysis of polymorphisms related to myocardial infarction in women.

Gene	Polymorphism	Dominant model		Recessive model		Additive 1 model		Additive 2 model	
		P (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)	*P* (FDR)	OR (95% CI)
TNF	–863C→A	0.0072 (0.029)	0.61 (0.43–0.87)	0.3526 (0.385)		0.0114 (0.034)	0.62 (0.43–0.89)	0.2677 (0.321)	
PLAT	–7351C→T	0.0153 (0.037)	0.69 (0.51–0.93)	0.0771 (0.103)		0.0463 (0.069)	0.73 (0.54–0.99)	0.0343 (0.059)	0.47 (0.22–0.91)
IPF1	–108/3G→4G	0.0013 (0.008)	0.58 (0.42–0.81)	0.6481 (0.648)		0.0012 (0.014)	0.55 (0.39–0.79)	0.0223 (0.045)	0.63 (0.42–0.93)

FDR, false discovery rate; OR, odds ratio; CI, confidence interval. Multivariable logistic regression analysis was performed with adjustment for age, BMI, and the prevalence of smoking, hypertension, diabetes mellitus, and hypercholesterolemia.