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Interaction between the *UCP2* –866 G > A polymorphism, diabetes, and β -blocker use among patients with acute coronary syndromes

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Abstract

Objective—*UCP2* –866G > A (rs659366) has been implicated in cardiometabolic disease and represents a novel candidate gene for β -blocker response, particularly among patients with diabetes. We assessed the function of –866G > A and its role as a modifier of β -blocker treatment outcomes by diabetes status in an acute coronary syndrome (ACS) cohort.

Methods—ACS patients with genetic samples and 12 months of follow-up for cardiac rehospitalizations or death ($n = 468$) were assessed. The influence of –866G > A on β -blocker treatment outcomes was evaluated in those with diabetes and without. To assess functional correlates of –866G > A, we compared uncoupling protein 2 (*UCP2*) expression in the skeletal muscle of obese participants by genotype and compared the activity of *UCP2* luciferase promoters with –866G and –866A alleles.

Results—An interaction between –866G > A and β -blocker treatment was found in individuals with diabetes ($P = 0.002$) but not those without ($P = 0.79$). Among G/G individuals with diabetes, discharge β -blocker use was associated with an 80% reduction in cardiac rehospitalization (adjusted hazard ratio: 0.20; 95% confidence interval: 0.04–1.02). In contrast, among A-carrier patients with

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diabetes, there was an 11-fold increase in cardiac rehospitalizations with discharge β -blocker therapy (adjusted hazard ratio: 11.75; 95% confidence interval: 1.28-108.2). Promoter activity assays showed that -866G had greater cyclic AMP response element binding protein-responsiveness compared with -866A, and compared with -866A carriers G/G individuals exhibited increased UCP2 expression in the skeletal muscle.

Conclusion—We identified a significant interaction between -866G > A and β -blocker response among ACS patients with diabetes. Furthermore, -866G conferred greater gene transcriptional activity than -866A in cell lines and in obese patients. These findings may help us gain insight into the mechanisms underlying the beneficial and detrimental effects of β -blockers in those with diabetes.

Keywords

acute coronary syndromes; gene-environment interaction; pharmacogenetics; type 2 diabetes

Introduction

Diabetes is associated with a greatly increased risk of cardiovascular disease, the leading cause of death and disability in Western societies. Macrovascular cardiovascular disease has been reported to account for 50% of all hospitalizations among individuals with diabetes [1]. Furthermore, hospital discharges with ischemic heart disease as a primary diagnosis and diabetes as a secondary diagnosis have doubled since 1980, with 545 000 discharges in 2003 [2]. Mechanisms to improve cardiovascular outcomes and reduce hospitalizations among individuals with diabetes would be of great benefit to the healthcare system and society at large.

β -blockers are now routinely used (through performance measures) in patients after myocardial infarction (MI), yet not all patients benefit. Patients with diabetes, for example, bear a great proportion of the burden of cardiovascular disease yet may be most susceptible to the adverse metabolic consequences of β -blocker therapy, thereby limiting its effectiveness. β -adrenergic receptor activation stimulates expression of a broad array of genes including uncoupling protein 2 (UCP2), which is a known target of β -adrenergic signaling cascades [3,4]. Uncoupling proteins are a family of inner mitochondrial membrane proteins that are thought to reduce the generation of reactive oxygen species, maintain fatty acid homeostasis in the mitochondria, and regulate mitochondrial biogenesis [5-7]. Alterations in these functions are likely involved in the pathogenesis of metabolic risk factors for diabetes and coronary artery disease (CAD), including increased oxidative stress, insulin resistance, and altered insulin secretion. In fact, a polymorphism in the promoter region of *UCP2*, -866 G > A (rs659366) has been associated with the risk of type 2 diabetes and CAD [8-14].

We postulated that genetic alterations in UCP2 function could influence the clinical effect of β -blocker therapy. *UCP2* is a particularly relevant candidate gene in β -blocker response among individuals with diabetes given that free fatty acids increase UCP2 expression as a method of disposing of excess fatty acids, and β -blockers decrease UCP2 expression [15,16]. Consequently, individuals with diabetes, who are known to have elevated free fatty acid concentrations, may have variable β -blocker benefits based on their *UCP2* genotype. Accordingly, the major purpose of this study was to examine whether *UCP2* -866 G > A modulates β -blocker treatment outcomes in patients with CAD, particularly in those with type 2 diabetes because of the importance of UCP2 in regulating energy metabolism in insulin-responsive tissues. We hypothesized that *UCP2* -866 G > A is associated with decreased UCP2 expression and modifies the response of patients with acute coronary syndromes (ACS) to therapy with β -blockers. To test these hypotheses, we used a prospective cohort study of patients with ACS [INvestigation oF Outcomes from acute coronary syndRoMes study (INFORM)] from whom detailed medication histories and clinical outcomes were collected.

We also assessed UCP2 expression in skeletal muscle biopsies from obese individuals and performed in-vitro functional studies to assess the influence of *UCP2* -866 G > A on expression levels and promoter activity.

Methods

Clinical outcome study

The genetic material obtained from 468 white patients participating in INFORM [17] with up to 12-month outcomes data were evaluated. Patients admitted to one of the two Kansas City hospitals with confirmed diagnoses of acute MI or unstable angina were enrolled. MI was defined by elevated troponin and either sustained ischemic chest pain or electrocardiographic changes (ST-segment elevation or non-ST-segment elevation) consistent with MI. Unstable angina was defined by the absence of elevated troponin and any one of the following: new-onset angina (< 2 months), prolonged angina (> 20 min) at rest, recent worsening angina, or angina that occurred within 2 weeks of MI [18]. Participating patients were interviewed during their admission to obtain demographic and health status characteristics. Chart abstractions were used to obtain medical data at the time of admission and during inpatient care. Follow-up interviews were used to capture 1-year health status and hospitalization events. Mortality assessment was made by using the Social Security Administration Death Master File (<http://www.ntis.gov/products/ssa-dmf.asp>). The study was approved by the institutional review boards of both institutions and written informed consent was obtained from all participants at the time of enrollment.

Genomic DNA was isolated from whole blood samples using a commercially available kit (Gentra, Minneapolis, Minnesota, USA). Genotyping was conducted using pyrosequencing (PSQ HS 96) or TaqMan (ABI 7700). The PCR primers and conditions are published in the supplement material. The TaqMan assay, C___876035 0_10, was carried out according to manufacturer's instructions.

Transfection studies

Expression constructs driving a constitutively active form of cyclic AMP response element binding protein (CREB), Y/F-CREB, and the CREB coactivator, transducer of regulated CREB 3 (TORC3), were generously provided by Dr M. Montminy and have been described earlier [19]. SV40-driven renilla luciferase reporter DNA was originally purchased from Promega, (Madison, Wisconsin, USA). Heterologous CREB-responsive reporter constructs were generated by ligating two copies of the CREB response element in the *UCP2* gene harboring the -866 G or A allele upstream of the minimal promoter of the thymidine kinase gene and the firefly luciferase cDNA (TK-Luc). Primer sequences are published in the supplement material.

CV1 cells were used because they are known to be null for many transcription factors. Cells were maintained in sub-confluent cultures in Dulbecco's Modified Eagle Medium (DMEM) containing 10% fetal bovine serum (FBS) with antimicrobials. On reaching confluence, cells were trypsinized and then plated in 500 µl of DMEM/ 10% FBS in 24 well plates for transfection studies. Twenty-four hours later, when cells were at 85% confluency, each well of cells was cotransfected with 2 µg of luciferase reporter DNA and 500 ng of expression vector for Y/F-CREB, TORC3, or empty pcDNA 3.1 plasmid backbone (vector control). To control for transfection efficiency, 100 ng of SV40-driven renilla luciferase constructs (Promega) were transfected into all the wells. All transfections were performed in duplicate. Sixteen hours later, the transfected cells were washed twice in phosphate buffered saline and then returned to DMEM/ 10% FBS. Firefly and renilla luciferase activity was measured by using a Dual-GLO kit (Promega) and a 96 well plate luminometer according to the manufacturer's instructions.

UCP2 expression in obese individuals

As *UCP2* -866 G > A has been reported to affect *UCP2* expression in a tissue-specific manner, we assessed its effect on skeletal muscle *UCP2* expression in a population at high cardiometabolic risk. Forty-eight obese individuals (13 men and 35 women, mean body mass index = 36.3 ± 4.4 kg/m²), originally recruited for other studies investigating the metabolic abnormalities of obesity [20,21], participated in this study (Supplemental Table 1 <http://links.lww.com/FPC/A124>). All participants completed a medical evaluation, including a history, physical examination and routine blood tests. All participants gave written informed consent before participating in this study, which was approved by the Human Studies Committee and the General Clinical Research Center Scientific Advisory Committee of Washington University School of Medicine in St. Louis.

Participants were admitted to the General Clinical Research Center at approximately 1800 h and ate a meal containing approximately 8 kcal/kg body weight, which contained 55% of total energy as carbohydrates, 30% as fat, and 15% as protein. The next morning, after the participants had fasted overnight (12 h), blood samples were obtained for standard blood chemistry analyses (Supplemental Table 1 <http://links.lww.com/FPC/A124>). Vastus lateralis skeletal muscle biopsies were obtained by standard techniques. Muscle biopsies were immediately rinsed in sterile saline, frozen in liquid nitrogen, and stored at -80°C until RNA extraction.

Total RNA was isolated using RNeasy B (Tel-Test) as per the manufacturer's protocol. Real-time reverse transcription (RT)-PCR was performed using Applied Biosystems (Foster City, California, USA) RT kit, PowerSYBR PCR mix, and the ABI PRISM 7500 sequence detection system. Arbitrary units of target gene mRNA were corrected to 36B4 RNA content to control for loading. Primer sequences are available upon request.

Statistical analysis

Baseline characteristics were compared using student's unpaired *t*-test or χ^2 tests, as appropriate. Genotype frequency deviations from Hardy-Weinberg equilibrium were assessed using χ^2 tests. Event rates were compared using χ^2 or Fisher's exact tests, as appropriate. Kaplan-Meier curves were constructed and compared using log-rank tests. Unadjusted and adjusted Cox proportional hazards models, using the following prespecified variables as covariates, were used to evaluate the association of -866 G > A genotype with clinical endpoints: age, sex, ACS type, inpatient ACS treatment strategy (medical management, percutaneous coronary intervention, or coronary artery bypass graft), ejection fraction, history of hypertension, heart failure, COPD/asthma, diabetes (type 1 plus type 2 diabetes), discharge β -blocker treatment, *UCP2* -866 G > A genotype (according to a dominant model of inheritance), and the interaction term between genotype and discharge β -blocker treatment. The proportional hazards assumption was tested by visual inspection of the plotted residuals, and statistically using the Supremum test for proportional hazards assumption in SAS. The primary endpoint for analysis was 1-year cardiac rehospitalization. The secondary endpoint was the composite of death and cardiac rehospitalization. To minimize confounding by population stratification, all statistical analyses were limited to whites given our small number of African-Americans in the cohort. Given our hypothesis that the effects of *UCP2* might differ by diabetes status, we performed analyses stratified by the presence or absence of diabetes. SAS version 9.1 (Cary, North Carolina, USA) was used for all analyses.

Results

Clinical association study

The INFORM cohort was comprised of 468 white patients with 12-month follow-up data from whom genetic samples were available and genotyping was successful (98% success rate). The mean age was 62 ± 12 years, 35% were female, and 24% had diabetes (Table 1). The minor allele (A) frequency was 0.38 in the overall population, 0.39 in those with diabetes, and 0.37 in those without diabetes. Genotype frequencies did not deviate from Hardy–Weinberg equilibrium and did not differ between those with and without diabetes ($P > 0.16$). No significant differences were found among genotype groups in any of the baseline characteristics with the exception of history of MI, which was slightly more prevalent in G/G individuals than A-carrier individuals ($P = 0.048$). At study discharge, 387 (83%) patients were receiving β -blockers (Table 1). Individuals who were discharged on a β -blocker had a lower ejection fraction and tended to have an increased presentation with non-ST elevation MI or ST elevation MI over unstable angina, and increased history of prior bypass grafting compared with those who were not discharged on a β -blocker (Supplemental Table 2 <http://links.lww.com/FPC/A125>). The event rates for the overall population, those with diabetes, and those without diabetes, are shown in Table 2.

Among the 107 patients with diabetes, we identified a significant interaction between *UCP2* –866 G > A genotype and discharge β -blocker therapy on the occurrence of 1-year cardiac rehospitalization (unadjusted P for interaction = 0.005, fully adjusted P value for interaction = 0.002; Table 3 and Fig. 1). The Kaplan–Meier event plots are shown in Fig. 1a and b. Overall, there was a significant difference between G/G and A-carriers that were treated or not treated with β -blocker therapy (Fig. 1, log-rank $P = 0.004$). Among G/G individuals with diabetes, discharge β -blocker use was associated with a significant reduction in the occurrence of cardiac rehospitalizations (Fig. 1a, log-rank $P = 0.023$). Among A-carriers with diabetes, discharge β -blocker use was associated with a significant increase in cardiac rehospitalizations (Fig. 1b, log-rank $P = 0.014$). The baseline characteristics of these patients (Caucasian diabetics) are shown in Supplemental Table 3 (<http://links.lww.com/FPC/A126>).

After adjustment for all prespecified covariates (see methods section), discharge β -blocker use was associated with a nonsignificant 80% reduction in cardiac rehospitalizations among –866 G/G individuals with diabetes [adjusted hazard ratio (HR): 0.20; 95% confidence interval (CI): 0.04–1.02] compared with those not discharged on a β -blocker (Table 3). In contrast, among patients with diabetes that were –866 A-carriers, there was an 11-fold increase in cardiac rehospitalizations among those discharged on β -blocker therapy compared with those not discharged on β -blocker therapy (adjusted HR: 11.75; 95% CI: 1.28–108.2) (Table 3). The combined endpoint of death and cardiac rehospitalization also yielded similar findings wherein G/G individuals had a decrease in events with β -blocker therapy and A-carriers had an increase compared with those without discharge β -blocker therapy (Table 3). Among those without diabetes, β -blocker therapy tended to reduce the occurrence of cardiac rehospitalizations in both genotype groups, although the association was only significant in A-carrier individuals (Table 3). Importantly, we did not detect a significant interaction between genotype and β -blocker use in those without diabetes ($P = 0.47$ –0.82, Table 3).

To better disentangle the association of *UCP2* genotype and post-ACS outcomes, we examined whether or not the observed associations were because of *UCP2* genotype alone, without addressing β -blocker use. Without considering the β -blocker $\times$ genotype interaction term, *UCP2* A-variant carrier status was not associated with cardiac rehospitalization in either those with diabetes (HR: 0.79; 95% CI: 0.36–1.71) or those without (HR: 0.92; 95% CI: 0.56–1.50). However, when including the β -blocker $\times$ genotype interaction term, *UCP2* –866 A variant carrier status was associated with a reduced risk for cardiac rehospitalization (HR: 0.03; 95%

CI: 0.002–0.28) among those with diabetes. No disease-gene association was seen in those without diabetes.

Assessment of the effect of *UCP2* –866 G > A on transcriptional response to CREB

In-silico assessment of *UCP2* –866 G > A showed that the G allele created a better match to an ideal CREB response element as compared with the A allele (<http://jaspar.genereg.net/>). To test this hypothesis, heterologous reporter constructs containing two copies of the CREB response element in the *UCP2* gene promoter containing either the G or A allele were generated (see Supplemental Methods for sequence information <http://links.lww.com/FPC/A127>) and tested in cotransfection assays in CV1 cells. The G allele exhibited enhanced CREB responsiveness upon cotransfection with either constitutively active CREB (Y/F CREB) or the CREB coactivator TORC3 compared with that of the constructs containing the A allele (Fig. 2; $P < 0.05$ for allele-specific differences in CREB responsiveness). Next, the expression of *UCP2* mRNA was evaluated in the skeletal muscle of obese, insulin-resistant volunteers using quantitative RT-PCR. Consistent with the promoter-reporter studies, *UCP2* –866 A-carrier status was associated with decreased *UCP2* mRNA expression compared with –866 G/G genotype in the skeletal muscle (Fig. 3). Specifically, –866 A-carriers exhibited 30% lower skeletal *UCP2* expression than –866 G/G ($P < 0.05$).

Discussion

This study is the first pharmacogenetic evaluation of the effect of a polymorphism in *UCP2* on the clinical response to β -blocker therapy in patients with ACS. Our data show that patients hospitalized with ACS who have diabetes and the *UCP2* –866 G/G genotype benefited from β -blocker therapy, whereas A-carriers did not. In contrast, this effect was absent in the nondiabetic cohort. In addition, we showed the functional importance of the *UCP2* gene promoter SNP, –866 G > A, in studies conducted in cell lines and in obese subjects. Briefly, constructs containing the –866 A allele had reduced CREB-responsiveness and patients carrying the –866 A allele had reduced skeletal muscle *UCP2* expression compared with individuals with the –866 G/G genotype. These results suggest a complex interaction between *UCP2* function, response to β -blocker therapy, and the presence or absence of diabetes in patients with ACS, and underscore the importance of genetic variability in determining the clinical response to medical therapy.

One interpretation of our data is that risk is already low in patients with diabetes who are –866 A-carriers, which prevents these patients from achieving a full therapeutic effect of β -blocker therapy after an ACS. In fact, A-carriers with diabetes who were not discharged on a β -blocker had the lowest rehospitalization rates among all groups studied (Fig. 2). Moreover, our data suggest that β -blocker therapy was deleterious in diabetic A-carriers and resulted in a higher cardiac rehospitalization rate than in those who were not treated with β -blockers after an ACS. We speculate that the benefit-to-risk ratio of β -blockers in the presence of diabetes may be altered by *UCP2* –866 G > A genotype with A-carrier individuals having more risk, perhaps because of increased susceptibility to the adverse metabolic consequences of β -blockers, and G/G individuals having more benefit. This hypothesis is supported by recent studies highlighting the adverse metabolic consequences of β -blockers in some, but not all, at-risk patients [22,23]. Alternatively, however, it is possible that β -blocker therapy masked an increased risk for adverse outcomes experienced by –866 G/G individuals with diabetes and that *UCP2* –866 G > A alters the disease process itself, and not β -blocker response specifically. Whether –866 A-carriers represent a subgroup of patients in whom β -blockers should be avoided because of increased metabolic side effects needs to be further explored.

The interaction we identified between *UCP2* genotype and β -blocker response was not evident in individuals without diabetes, suggesting that the metabolic derangements of diabetes create

a unique milieu for the observed pharmacogenetic association. This finding is not surprising given the experimental data suggesting that the metabolic consequences of UCP2 function are likely to be context-specific and highly dependent on disease and environmental factors [5,6]. It is possible that the known activating effects of fatty acids on UCP activity lead to heightened respiratory uncoupling activity in diabetes. Given increased lipid delivery to the diabetic muscle and heart, β -blocker therapy may reverse this dysregulatory effect and confer differential benefit depending on $-866\text{ G} > \text{A}$ genotype. The results of earlier studies evaluating the functional impact of $-866\text{ G} > \text{A}$ have been discordant, possibly related to the cell type assessed [12-14,24]. The results of our assessment of UCP2 expression as a function of $-866\text{ G} > \text{A}$ genotype in obese patients are consistent with studies indicating that individuals with the variant -866A allele have decreased *UCP2* gene expression compared with the -866G allele, as would be predicted by the integrity of the consensus sequence for CREB binding and supported by our transcriptional studies. These data add to the current body of knowledge regarding the transcriptional regulation of UCP2 and the functional significance of the $-866\text{ G} > \text{A}$ SNP by identifying a functional CREB site containing the $-866\text{ G} > \text{A}$ SNP and showing allele-specific CREB responsiveness.

The differences in UCP2 expression that we saw by $-866\text{ G} > \text{A}$ genotype in obese individuals (who were similar to diabetic patients in many ways) suggest the intriguing possibility that diabetic patients in our ACS cohort with the -866A allele may not have benefited from β -blocker therapy because of lower UCP2 expression. Indeed, β -blockers have been shown experimentally to attenuate adrenergically-mediated upregulation of UCP. As an extension, lower skeletal UCP2 expression in obese -866 A -carriers may result in an attenuated responsiveness to adrenergic stimulation (i.e. a pseudo- β -blocked state). Thus, it is conceivable that patients in whom adrenergic responsiveness is diminished would be less likely to experience benefit from the addition of β -blockers, as was seen in the ACS patients with diabetes and the -866A allele that were discharged on a β -blocker.

UCP2 $-866\text{ G} > \text{A}$ has primarily been studied with regard to the development and progression of diabetes and obesity phenotypes. We are not aware of any studies that have evaluated the impact of *UCP2* variation in response to medical therapy. Although earlier genetic association studies with $-866\text{ G} > \text{A}$ have not looked at interactions with β -blocker therapy, some have found associations congruent with our findings. Recently, in a clinical trial of patients with type 2 diabetes, -866 A -carrier individuals were found to have a decreased risk of developing CAD compared with G/G individuals, HR: 0.88, 95% CI: 0.88–0.96 [8]. In contrast, two other studies have found an increased CAD or asymptomatic atherosclerosis risk for A/A individuals compared with other genotypes [9,13]. The basis for these discrepant results is unclear, but the emerging picture suggests that UCP2's function is affected by environmental factors, such as diabetes. Furthermore, we show that failure to consider the interaction between *UCP2* genotype and β -blocker use biased disease-gene associations towards the null (i.e. no association between *UCP2* genotype and outcomes was seen without incorporation of a genotype $\times$ β -blocker interaction term).

Limitations

Our study has several limitations which merit discussion. First, participants with diabetes in our study only made up about a quarter of the overall population and stratification by β -blocker use and genotype makes these numbers even smaller. Therefore, it will be important to replicate these findings in larger populations. That notwithstanding, we were still able to detect a statistically significant association and provide molecular evidence for the validation of these potentially very clinically important findings. The possibility of a type I error, given that we did not adjust for multiple comparisons, does exist. Our analysis was conducted to test a specific hypothesis regarding *UCP2* $-866\text{ G} > \text{A}$ and β -blocker response in diabetics versus

nondiabetics. Therefore, we did not adjust for multiple comparisons. If we had, it is likely the gene-environment interaction we found in diabetic individuals would have remained significant, but the *P* values for β -blocker use in G/G and A-carriers would not have remained significant. This limitation emphasizes the provocative, yet preliminary nature of our findings until they can be replicated in other cohorts. A second limitation of our study is that we were only able to assess the association between *UCP2* genotype and β -blocker treatment outcomes in white individuals as our other racial/ethnic groups were too small. Therefore, whether these findings hold true in other populations is unclear. Last, although we have identified some initial molecular mechanisms for the observed associations, further experiments will be required using additional tissue types and additional transcriptional regulatory studies to better elucidate all of the mechanisms underlying these findings.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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Fig. 1.

UCP2 -866 G > A variant carrier status confers differential cardiac rehospitalization rates among patients with diabetes discharged with and without β -blocker therapy. Panel 'a' is G/G individuals and panel 'b' is A-carrier individuals. Solid line is β -blocker treated and hatched line is no discharge β -blocker treatment. Overall P value comparing all four groups = 0.0040. GG no BB versus A-carrier no BB $P = 0.0003$, GG no BB versus GG BB $P = 0.0233$, A-carrier BB versus A-carrier no BB $P = 0.0142$. BB, β -blocker treated; non-BB, non- β -blocker treated.

Fig. 2.

UCP2 -866 G > A is associated with differential cyclic AMP response element binding protein (CREB)-responsiveness in CV1 cells. Graph depicts mean relative luciferase units of cells transfected with heterologous reporter constructs driven by the CREB response element harboring the -866 G or A allele upstream of the minimal promoter of the thymidine kinase gene and the firefly luciferase cDNA (TK-Luc) (-866A 2X TKLuc or -866G 2X TKLuc). Cells were cotransfected with expression constructs for constitutive active CREB (Y/F CREB), the CREB coactivator TORC3, or empty vector control (pcDNA) as indicated at the bottom. Values were normalized to the activity of cells transfected with the -866A 2X TKLuc reporter and empty expression vector (= 1.0). * $P < 0.05$ versus activity -866A 2X TKLuc with same expression vector.

Fig. 3. UCP2 mRNA expression in skeletal muscle of obese subjects by $-866\text{ G} > \text{A}$ genotype. Graph depicts mean levels of UCP2 mRNA normalized to 36B4 in skeletal muscle of participants after an overnight fast. * $P < 0.05$ for G/G versus A-carrier. Overall P across three genotype groups = 0.09.

Table 1

Baseline characteristics in Caucasian cohort with complete *UCP2* –866 G>A genotype data and compared by genotype

	INFORM (n =468)	G/G (n= 189)	A carrier (n=279)	P value *
Age, mean (SD) (years)	62.0 (12.5)	62.9 (12.2)	62.2 (11.8)	0.55
Women	165 (35%)	62 (33%)	103 (37%)	0.36
BP, mean (SD) (mmHg)				
Systolic	135 (25)	134 (24)	137 (25)	0.11
Diastolic	72 (15)	73 (15)	73 (14)	0.86
Heart rate, mean (SD) (beats per min)	76.8 (18.1)	76.2 (21.2)	77.2 (15.8)	0.57
Triglycerides (SD) (mg/dl) ^a	188.5 (144.8)	176.5 (146.3)	197.1 (143.6)	0.25
HDL cholesterol (SD) (mg/dl) ^a	40.3 (12.1)	40.2 (12.3)	40.3 (12.0)	0.92
LDL cholesterol (SD) (mg/dl) ^a	103.9 (40.0)	101.5 (40.9)	105.6 (39.5)	0.50
BMI, mean (SD) (kg/m ²)	29.4 (5.9)	28.9 (5.3)	29.6 (5.9)	0.19
EF, mean (SD) (%) ^b	47.5 (12.3)	47.7 (11.6)	48.0 (12.6)	0.80
ACS type				
ST elevation MI	147 (31%)	70 (37%)	77 (28%)	0.08
Non-ST elevation MI	149 (32%)	58 (31%)	91 (33%)	
Unstable angina	171 (37%)	60 (32%)	111 (40%)	
Old LBBB/unknown	1 (0.2%)	1 (0.5%)	0 (0%)	
Past medical history				
Myocardial infarction	142 (30%)	67 (35%)	75 (27%)	0.048
Prior PCI	163 (35%)	63 (33%)	100 (36%)	0.58
Prior CABG	96 (21%)	36 (19%)	60 (22%)	0.52
Hypertension	299 (64%)	127 (67%)	172 (62%)	0.22
Heart failure	21 (4%)	10 (5%)	11 (4%)	0.49
Smoking				
Past	186 (40%)	76 (40%)	110 (40%)	0.98
Within 30 days	143 (31%)	57 (30%)	86 (31%)	
Never	138 (30%)	56 (30%)	82 (30%)	
Diabetes	107 (23%)	37 (20%)	70 (25%)	0.16
Hypercholesterolemia	296 (63%)	125 (66%)	171 (61%)	0.29
Renal failure	5 (1%)	0 (0%)	5 (2%)	0.06
Discharge β-blocker	387 (83%)	154 (81%)	233 (84%)	0.57

ACS, acute coronary syndrome; BMI, body mass index; BP, blood pressure; CABG, coronary artery bypass graft; INFORM, Investigation of Outcomes from ACSs study; LBBB, left bundle branch block; MI, myocardial infarction; PCI, percutaneous coronary intervention.

^a n= 271 for triglycerides, n= 195 for HDL, n =182 for LDL.

^b n=449 (n= 182 G/G and n= 267 A carriers).

* P value comparing –866 G/G and A carriers.

Table 2
Cardiac rehospitalization event rates in the overall population and by diabetes status

	All whites		Diabetic whites		Nondiabetic whites	
	No BB	BB	No BB	BB	No BB	BB
A/A	12/72 (17%)		3/14 (21%)		9/58 (16%)	
G/A	50/207 (24%)		19/56 (34%)		31/151 (21%)	
G/G	45/189 (24%)		15/37 (41%)		30/132 (20%)	
<i>P</i> *	0.39		0.44		0.71	
A/A	3/15 (20%)	9/57 (16%)	0/4 (0%)	3/10 (30%)	3/11 (27%)	6/47 (13%)
G/A	8/31 (26%)	42/176 (24%)	1/12 (8%)	18/44 (41%)	7/19 (37%)	24/132 (18%)
G/G	13/35 (37%)	32/154 (21%)	6/8 (75%)	9/29 (31%)	7/27 (26%)	23/125 (18%)
<i>P</i> *	0.40		0.005		0.71	
					0.66	

BB, β -blocker treated; non-BB, non- β -blocker treated.

* *P* value comparing event rates across genotype groups.

Table 3HR 95% confidence intervals for β -blocker therapy versus no β -blocker therapy by *UCP2* -866 G>A genotype

Outcome	-866 G/G (<i>n</i> =37)	-866 A-carrier (<i>n</i> = 70)	Genotype $\times$ β -blocker interaction <i>P</i> value
Patients with diabetes, unadjusted model			
Cardiac rehospitalization	0.32 (0.11–0.90)	8.22 (1.10–61.26)	0.004
Death or cardiac rehospitalization	0.45 (0.17–1.20)	5.32 (1.27–22.36)	0.005
Patients with diabetes, fully adjusted model ^a			
Cardiac rehospitalization	0.20 (0.04–1.02)	11.75 (1.28–108.2)	0.002
Death or cardiac rehospitalization	0.42 (0.12–1.52)	5.75 (1.31–25.31)	0.005
Patients without diabetes, unadjusted model			
Cardiac rehospitalization	0.67 (0.29–1.56)	0.45 (0.22–0.92)	0.47
Death or cardiac rehospitalization	0.72 (0.33–1.57)	0.54 (0.27–1.08)	0.58
Patients without diabetes, fully adjusted model ^a			
Cardiac rehospitalization	0.67 (0.27–1.68)	0.58 (0.26–1.26)	0.79
Death or cardiac rehospitalization	0.85 (0.37–1.97)	0.65 (0.30–1.38)	0.82

ACS, acute coronary syndrome.

^a Adjusted for: age, sex, ACS type, inpatient ACS treatment strategy (medical management, percutaneous coronary intervention, or coronary artery bypass graft), ejection fraction, history of hypertension, heart failure, COPD/asthma, diabetes (type 1 plus type 2 diabetes), discharge β -blocker treatment, *UCP2* -866 G> A genotype (according to a dominant model of inheritance).