

Variant Aldehyde Dehydrogenase 2 (*ALDH2*2*) Is a Risk Factor for Coronary Spasm and ST-Segment Elevation Myocardial Infarction

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Background—Mitochondrial aldehyde dehydrogenase 2 (ALDH2) plays a key role in removing toxic aldehydes. Deficient variant *ALDH2*2* genotype is prevalent in up to 40% of the East Asians and reported to be associated with acute myocardial infarction (AMI). To elucidate the mechanisms underlying the association of *ALDH2*2* with AMI, we compared the clinical features of AMI patients with *ALDH2*2* to those with wild-type *ALDH2*1/*1*.

Methods and Results—The study subjects consisted of 202 Japanese patients with acute ST-segment elevation myocardial infarction (STEMI) (156 men and 46 women; mean age, 67.3±12.0) who underwent primary percutaneous coronary intervention (PCI). In 85 patients, provocation test for coronary spasm was also done 6 month post-PCI. *ALDH2* genotyping was performed by direct application of the TaqMan polymerase chain system. Of the 202 patients, 103 (51.0%) were carriers of *ALDH2*2* and 99 (49.0%) those of *ALDH2*1/*1*. There were no differences in clinical features between *ALDH2*2* and *ALDH2*1/*1* carrier groups except higher frequencies of coronary spasm and alcohol flush syndrome (AFS) (88.6% vs 56.1%; $P=0.001$ and 94.3% vs 17.6%; $P<0.001$), less-frequent alcohol habit (14.6% vs 51.5%; $P<0.001$), and higher peak plasma creatine phosphokinase levels (2224 vs 1617 mg/dL; $P=0.002$) in the *ALDH2*2* than the *ALDH2*1/*1* carrier group.

Conclusions—*ALDH2*2* is prevalent (51.0%) among Japanese STEMI patients, and those with *ALDH2*2* had higher frequencies of coronary spasm and AFS and more-severe myocardial injury compared to those with *ALDH2*1/*1*. (*J Am Heart Assoc.* 2016;5:e003247 doi: 10.1161/JAHA.116.003247)

Key Words: acute myocardial infarction • alcohol flushing syndrome • aldehyde dehydrogenase 2 • coronary spasm • coronary spastic angina

The mitochondrial aldehyde dehydrogenase 2 (ALDH2) plays a key role in removal of not only ethanol-derived acetaldehyde, but also other toxic aldehydes such as 4-hydroxy-2-nonenal (4-HNE) derived from lipid

peroxidation.^{1,2} Up to 40% of East Asians carry a variant *ALDH2* or *ALDH2*2* (Glu504Lys) genotype with deficient activity and suffer from alcohol (ethanol) flushing syndrome (AFS) including facial flushing, headache, nausea, and palpitation, in response to a small amount of alcohol intake attributed to accumulation of acetaldehyde.^{2–5} Recent studies indicate that carriers of *ALDH2*2* genotype have a higher risk for acute myocardial infarction (AMI).^{6–11} Coronary spasm is not rarely demonstrated in AMI patients in Japan,^{12,13} and we have reported that *ALDH2*2* genotype is associated with coronary spastic angina (CSA).¹⁴ ALDH2 protects against myocardial ischemia/reperfusion (I/R) injury by removing toxic aldehydes such as 4-HNE in experimental AMI models.^{2,15–17} However, the underlying mechanisms linking ALDH2 to AMI in humans remain to be elucidated. In the present study, we examined the clinical features of AMI patients with *ALDH2*2* by comparing to those with wild-type *ALDH2*1/*1* in acute ST-segment elevation myocardial infarction (STEMI) patients who underwent reperfusion therapy with primary percutaneous coronary intervention (PCI).

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Methods

Study Subjects

Two hundred two patients (156 men and 46 women with a mean age of 67.3 ± 12.0) who underwent primary PCI with stent implantation successfully within 12 hours after symptoms and consented to be genotyped for *ALDH2* gene polymorphism were the subjects of this study. These patients were recruited from the consecutive 235 Japanese patients admitted and diagnosed as STEMI at our institutions between December 2009 and October 2015. Not included in the study were 26 patients who died during hospitalization (11 with PCI and 15 without PCI attributed to delay of admission), 10 patients with unsuccessful PCI (Thrombolysis In Myocardial Infarction [TIMI] grade <3), 2 patients with emergent coronary artery bypass graft, and 10 patients who did not give informed consent. STEMI was defined as symptoms of myocardial ischemia in association with new persistent electrocardiography (ECG) ST-segment elevation in at least 2 contiguous leads of ≥ 2 mm (0.2 mV) and a rise and/or fall of biomarkers of myocardial necrosis including cardiac troponin T (cTnT) exceeding the 99th percentile of a normal reference population.^{18,19} This study was conducted in accord with the Declaration of Helsinki and approved by the ethics committee of our institutions and written informed consent was obtained from each patient.

Genotyping

The details of the method were reported.²⁰ Briefly, single-nucleotide polymorphism genotyping of *ALDH2* (Glu504Lys; rs671) was performed using the TaqMan assay on an ABI 7300 Real Time polymerase chain reaction (PCR) System (Applied Biosystems, Foster City, CA) without DNA extraction on whole blood. The mixture was 20 μ L and consisted of 10 μ L of a Thunderbird Probe qPCR Mix (QPS-101; Toyobo, Osaka, Japan), 0.4 μ L of a 50 \times ROX reference dye (Toyobo), 1 μ L of a 20 \times *ALDH2* TaqMan Probe & *ALDH2* Primer Mix (C_11703892_10, Applied Biosystems), 2 μ L of PCR product, and 6.6 μ L of distilled water. Thermal cycling process was performed according to the Applied Biosystems PCR conditions: 2 minutes at 50°C; 10 minutes at 95°C; 40 cycles of denaturation at 95°C for 15 seconds; and annealing and extension at 60°C for 1 minute. Results were analyzed by Applied Biosystems Prism 7300 SDS software. Genotyping was performed with the identification of the study subjects blinded.

Angiographic Documentation of Coronary Spasm

Provocation test for coronary spasm was also performed in 85 of the study patients after obtaining informed consent around 6 months after the attack. Patients with organic stenosis of $\geq 75\%$ (n=19), 3-vessel organic disease (n=8), restenosis

(n=8), left main trunk lesion (n=1), uncontrolled arrhythmias (n=2), heart failure (n=10), resting hypertension $>180/110$ mm Hg (n=6) acute systemic illness, and hepatic or renal insufficiency or other severe conditions (n=12) were excluded from the study. All vasoactive medications, including calcium-channel blockers, beta-receptor blockers, angiotensin-converting enzyme inhibitors, angiotensin II receptors blockers, and statins, were withdrawn for at least 3 days before angiography except for nitroglycerin used for attacks. Coronary spasm was defined as a transient total or subtotal occlusion or severe diffuse vasoconstriction of an epicardial coronary artery associated with ischemic changes on ECG with or without chest discomfort. The final diagnosis was determined with the consensus of 3 investigators blinded to genotype/flushing phenotype. Coronary spasm was induced by the intracoronary injection of acetylcholine (ACh; Daiichi-Sankyo Co., Tokyo, Japan) after diagnostic catheterization in the morning.¹⁴ Details of the provocation test were previously reported.^{13,14} Briefly, ACh dissolved in 5 mL of warmed 0.9% saline was infused manually in 20 seconds into the left coronary artery (LCA) in incremental doses of 10, 20, 50, and 100 μ g and then 10, 20, and 50 μ g into the right coronary artery (RCA) depending on the vascular reactivity under continuous monitoring of 12-lead ECG and blood pressure with a temporary pacemaker in place in the morning. Coronary angiography was performed 1 minute after each ACh injection or when ischemic ECG changes appeared. Figure 1 shows a typical case of coronary spasm with stent implantation in the provocation test by ACh 6 months post-AMI.

Coronary spasm induced by this method usually disappeared spontaneously within 1 to 2 minutes and both the LCA and RCA could be examined separately, unless severe spasm occurred in the LCA and necessitated the prompt injection of nitroglycerin (GTN) or isosorbide dinitrate (ISDN) into the artery. Finally, GTN or ISDN was infused to relieve spasm and examine organic lesions. Significant organic coronary stenosis was defined as $>50\%$ luminal diameter. The number of study patients needed was calculated based on an uncorrected chi-squared statistic at the 2-tailed test 5% significance level and 80% power, assuming the prevalence of coronary spasm to be 80% in the variant *ALDH2*2* MI patient group and 50% in the wild *ALDH2*1/*1* MI patient group based on our previous work and others.^{12–14}

Questionnaire Survey

Subjects were asked to fill out a simple questionnaire concerning alcohol flushing on alcohol intake, alcohol drinking habit, and smoking. Habitual drinker was defined as an alcohol drinker more than 5 days a week. Alcohol flushing was defined as a current or a history of facial flushing

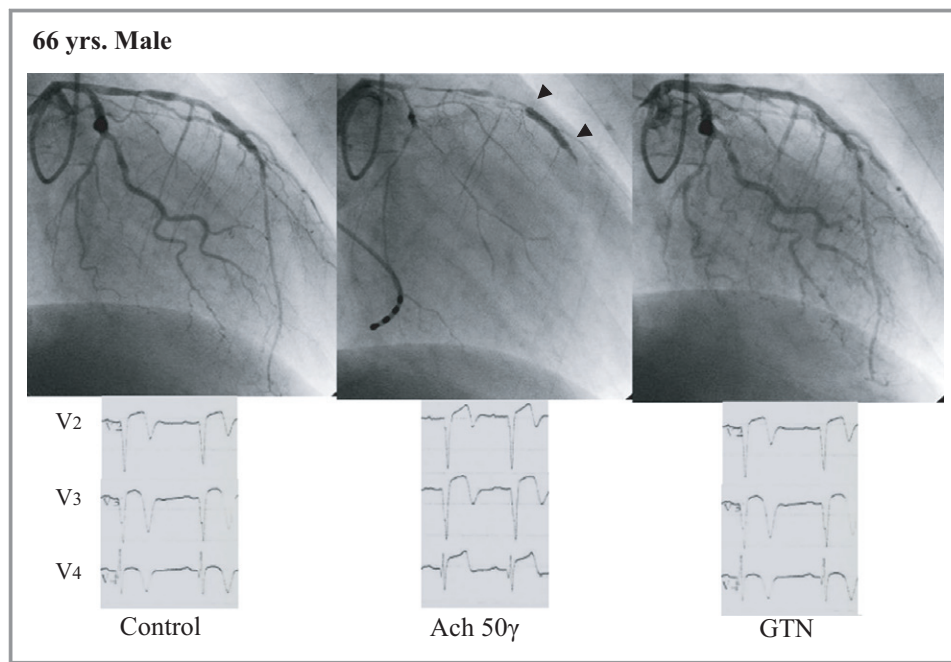


Figure 1. A representative case of coronary spasm induced by intracoronary ACh infusion. Severe diffuse spasm appeared associated with ST-segment elevation on ECG post-ACh infusion at the distal segments of stent implanted in the left anterior descending artery and disappeared after nitroglycerin. Arrow heads indicate stent. Ach indicates acetylcholine; ECG, electrocardiogram; GTN, nitroglycerin.

immediately after drinking a glass of beer. Smokers were defined as current and past smokers.

Blood Chemistry Measurements

Blood samples for measurement of clinical chemistry and other data were collected on admission and repeatedly at least 3 to 4 times later with patients in the supine position. Biochemical and other analyses were done using standard laboratory procedures. Total creatine phosphokinase (CPK) was measured with a standard enzymatic method on a UniCel DxC800 Multi analyzer 2700 (Beckman Coulter Clinical Diagnostics, Brea, CA). cTnT was assessed by an electrochemoluminescence immunoassay using the Cobas system troponin T kit provided by the manufacturer on a Cobas e 411 disk system analyzer (Roche Diagnostics, Indianapolis, IN). Blood samples for total CPK and lactate dehydrogenase (LDH) were measured on admission and repeated every 4 hours during the first 12 hours and every 12 hours up to 72 hours. cTnT was measured on admission and repeated 3 to 6 hours later and further several times for 1 to 5 days depending on patient conditions.

Statistical Analysis

Allele frequencies were determined by direct gene counting, and genotype distributions were checked for departure from

Hardy-Weinberg equilibrium using the Pearson chi-square test. Baseline clinical data were expressed as mean±SD or median (25th or 75th percentile) for continuous variables, and within-group differences were evaluated with an unpaired *t* test or the Mann-Whitney rank-sum test. For discrete variables, data were expressed as counts and percentages and analyzed with the chi-square or Fisher's exact test. A 2-tailed value of $P<0.05$ was considered to be statistically significant. Analyses were done using the STATA software program (STATA 11.0; StataCorp, College Station, TX).

Results

Genotype Distribution of *ALDH2*

Genotype distributions did not depart from the Hardy-Weinberg equilibrium for *ALDH2* genes in the whole group ($\chi^2=0.057$; $P=0.972$). Frequencies of *ALDH2**1/*1, *ALDH2**1/*2, and *ALDH2**2/*2 genotype were 49.0% (99 of 202), 42.3% (86 of 202), and 8.4% (17 of 202), respectively. Frequency of the *ALDH2**2 allele was thus 29.7% (120 of 404) in the STEMI patients in this study. This is high, as compared with that of the general population in Japan (23.5% [247 of 1050] in Kumamoto and 24.3% [183 of 752] in Tokyo, Japan).³ We have recently reported that frequencies of *ALDH2**1/*1, *ALDH2**1/*2, and *ALDH2**2/*2 genotype were 57.5% (272

of 473), 36.6% (173 of 473), and 5.9% (28 of 473), respectively, and thus frequency of the *ALDH2*2* allele was 24.2% (229 of 946) in subjects who had no histories or ECG signs of ischemic heart disease recruited from a health screening program, with age and sex comparable to those of STEMI patients in the present study (age: 68.8 ± 7.6 vs 67.3 ± 12.0 ; $P=0.054$ and male ratio: 72.7% [344 of 473] vs 77.2% [156 of 202]; $P=0.222$).²¹ Frequency of the *ALDH2*2* allele was therefore significantly higher in STEMI patients than in those without apparent ischemic heart disease (29.7% [120 of 404] vs 24.2% [229 of 946]; odds ratio [OR]=1.32 [95% CI, 1.01–1.73]; $P=0.035$). The findings are consistent with those of previous studies^{6–11} and indicate that *ALDH2*2* is associated with AMI, being randomly assigned at conception independently of the possible confounding factors (Mendelian randomization).²² The study also indicates that the variant *ALDH2*2* genotype exists mainly as the heterozygote (*ALDH2*1/*2*), and we combined heterozygote (*ALDH2*1/*2*) and homozygote (*ALDH2*2/*2*) as a single category of variant *ALDH2*2* and compared them with the wild homozygote *ALDH2*1/*1* in the following analysis. Accordingly, 51.0% (103 of 202) of STEMI patients (82 men and 21 women; mean age, 67.1 ± 12.2) had variant *ALDH2*2* and the remaining 99 (74 men and 25 women; mean age, 67.5 ± 11.8) or 49.0% wild-type *ALDH2*1/*1* as shown in Table.

Comparison of Clinical Characteristics Between the *ALDH2*2* and *ALDH2*1/*1* Groups

Table compares the clinical characteristics of patients between the variant *ALDH2*2* group and wild *ALDH2*1/*1* group. There were no differences in age, sex, body mass index (BMI), blood pressure, heart rate, cardiac index, pulmonary capillary wedge pressure (PCWP), smoking habit, and plasma levels of low-density lipoprotein (LDL) cholesterol, triglycerides, glucose, C-reactive protein, infarct-related artery, percentage of multivessel disease, and other variables between the two groups. However, the frequencies of AFS and coronary spasm were higher (94.3% vs 17.6%; $P<0.001$ and 88.6% vs 56.1%; $P=0.001$, respectively) and alcohol habit was lower (14.6% vs 51.5%; $P<0.001$) in the *ALDH2*2* group than in the *ALDH2*1/*1* group (Table and Figure 2A). Thus, *ALDH2*2* is associated with coronary spasm as well as AFS (OR=6.1; 95% CI [1.8–23.4] and OR=77.8; 95% CI [17.9–441.8], respectively), being randomly assigned at conception independently of the possible confounding factors (Mendelian randomization).²² Accordingly, AFS had 94.3% (50 of 53) sensitivity and 82.4% (42 of 51) specificity for *ALDH2**. Conversely, *ALDH2*2* had an 84.7% (50 of 59) sensitivity and 93.3% (42 of 45) specificity for AFS and 62.9% (39 of 62) sensitivity and 78.2% (18 of 23) specificity for CSA,

respectively. AFS may therefore be a useful clinical marker for *ALDH2*2* in the absence of genotyping.

We performed sex-specific analyses to make sure that we did not miss a sex-specific effect. In men, coronary spasm were higher (91.9% vs 59.4%; OR=7.8; 95% CI [1.8–46.3]; $P=0.001$) in the *ALDH2*2* group than in the *ALDH2*1/*1* group. However, the number of women was small ($n=46$) for statistical significance (71.4% vs 44.4%; $P=0.28$).

Peak plasma levels of total CPK were significantly higher ($P=0.002$) (Table and Figure 2B) and those of LDH, cTnT, and B-type natriuretic peptide (BNP) tended to be higher and left ventricular ejection fraction (LVEF) tended to lower in the *ALDH2*2* than in the *ALDH2*1/*1* carrier group (Table). This suggested that myocardial injury may be more severe in AMI patients with *ALDH2*2* as compared to those with *ALDH2*1*1*.

Discussion

East Asians show rapid, intense AFS after drinking a small amount of alcohol in up to 40% of the population.^{2–5} This is caused by the presence of the variant of mitochondrial *ALDH2* genotype with a substitution of glutamate to lysine at position 504 (Glu504Lys) or *ALDH2*2*, which is present only in East Asians but is virtually absent in other populations of the world.^{2–5} *ALDH2*2* exerts a dominant negative effect over wild-type homozygote *ALDH2*1/*1*, and heterozygote *ALDH2*1/*2* shows a severely reduced and homozygote *ALDH2*2/*2* negligible ALDH2 activity.⁴ Carriers of *ALDH2*2* thus manifest the characteristic AFS caused by accumulation of ethanol-derived acetaldehyde. ALDH2 is a key enzyme that removes toxic aldehydes such as 4-HNE derived from lipid peroxidation as well as acetaldehyde.^{1,2}

Previous studies reported that *ALDH2*2* genotype was associated with AMI in East Asians including Japanese, Koreans, and Chinese.^{6–10} Takeuchi et al. identified the genetic locus of *ALDH2*2* (*rs671*) as the strongest predictor for AMI on a genome-wide association study in Japanese,¹⁰ and their findings were confirmed recently by the study of Hirokawa et al.¹¹

The present study shows that *ALDH2*2* genotype is significantly associated with STEMI, and the results are consistent with those of previous studies.^{6–11} Because the genotype is assigned randomly at conception independently of the possible confounding factors according to Mendel's law (Mendelian randomization),²² it is reasonable to think that *ALDH2*2* genotype is causally associated with STEMI. The study therefore identifies deficient ALDH2 activity and hence increased reactive aldehydes as a causative risk factor for STEMI.

However, the underlying mechanisms linking *ALDH2*2* to STEMI remain to be elucidated. STEMI is assumed to be

Table. Comparison of Clinical Characteristics Between Variant and Wild Type of *ALDH2*

Variables	Total Subjects (n=202)	Variant <i>ALDH2*2</i> (n=103)	Wild <i>ALDH2*1/*1</i> (n=99)	P Value
Age, y	67.3±12.0	67.1±12.2	67.5±11.8	0.787
Sex (male), n (%)	156 (77.2)	82 (79.6)	74 (74.7)	0.410
BMI, kg/m ²	24.0±3.3	23.8±2.9	24.1±3.6	0.564
Systolic BP, mm Hg	127.1±24.1	126.1±22.9	128.2±25.4	0.529
Diastolic BP, mm Hg	73.5±15.3	72.9±14.8	74.1±15.9	0.588
Heart rate, beat/min	75 (66, 82)	74 (69, 81)	76 (65, 82)	0.844
Cardiac index, L/min per m ²	2.89 (2.26, 2.95)	2.60 (2.16, 2.79)	3.17 (2.26, 3.07)	0.819
PCWP, mm Hg	13.3 (9.1, 15.5)	13.0 (9.1, 15.5)	13.5 (9.0, 17.0)	0.957
hs-CRP, mg/L	0.63 (0.30, 1.9)	0.68 (0.30, 2.20)	0.60 (0.30, 1.70)	0.752
Glucose, mmol/L	5.92 (5.05, 6.38)	5.74 (5.05, 6.16)	6.09 (5.05, 6.55)	0.130
Total-C, mmol/L	4.73 (3.98, 5.38)	4.69 (3.98, 5.30)	4.78 (3.93, 5.53)	0.673
Triglycerides, mmol/L	1.52 (0.91, 1.85)	1.46 (0.93, 1.68)	1.58 (0.89, 1.93)	0.491
HDL-C, mmol/L	1.23 (0.98, 1.42)	1.21 (0.96, 1.45)	1.24 (1.06, 1.42)	0.106
LDL-C, mmol/L	2.75 (2.12, 3.34)	2.84 (2.15, 3.34)	2.70 (2.07, 3.34)	0.196
Uric acid, μmol/L	338 (291, 393)	339 (291, 393)	338 (286, 387)	0.761
eGFR, mL/min per 1.73 m ²	67.8±18.2	66.1±16.4	69.5±19.9	0.234
Leukocyte, /μL	6466 (5500, 7300)	6500 (5600, 7400)	6300 (5200, 7300)	0.425
Hemoglobin, g/dL	13.2±1.8	13.2±1.8	13.2±1.8	0.997
Platelets, ×10 ⁴ /μL	22.6±7.5	23.2±7.3	22.0±7.8	0.250
Smokers, n (%)	129 (63.9)	67 (65.0)	62 (62.6)	0.720
Current smokers, n (%)	58 (28.7)	29 (28.2)	29 (29.3)	0.858
Alcohol habit, n (%)	66 (32.7)	15 (14.6)	51 (51.5)	<0.001
Alcohol flushing, n (%)	59/104 (56.7)	50/53 (94.3)	9/51 (17.6)	<0.001
0 Vessel disease, n (%)	3 (1.5)	1 (1.0)	2 (2.0)	0.972
Multivessels, n (%)	51 (25.2)	25 (24.3)	26 (26.3)	0.745
Infarct-related artery				
LAD, n (%)	93 (46.0)	49 (47.6)	44 (44.4)	0.915
RCA, n (%)	79 (39.1)	40 (38.8)	39 (39.4)	0.935
LCX, n (%)	29 (14.4)	14 (13.6)	15 (15.2)	0.752
LMT, n (%)	1 (0.0)	0 (0.0)	1 (1.0)	0.984
Coronary spasm, n (%)	62/85 (72.9)	39/44 (88.6)	23/41 (56.1)	0.001
Peak CPK, IU/L	1995 (1066, 2942)	2224 (1431, 3478)	1617 (850, 2538)	0.002
Peak LDH, IU/L	581 (426, 779)	606 (447, 1091)	570 (381, 699)	0.079
cTnT (admission), ng/mL	3.09 (0.93, 7.22)	3.99 (0.86, 7.68)	2.96 (0.93, 5.63)	0.254
BNP (discharge), pg/mL	162 (33, 170)	181 (42, 200)	141 (29, 156)	0.127
Echo EF (discharge), %	57.4 (52.6, 64.0)	56.6 (52.0, 62.3)	58.2 (53.6, 65.0)	0.112

AFS indicates alcohol flushing syndrome; ALDH2, aldehyde dehydrogenase type 2; BMI, body mass index; BNP, B-type natriuretic peptide; CPK, creatine phosphokinase; hs-CRP, high sensitivity C-reactive protein; cTnT, cardiac troponin T; DBP, diastolic blood pressure; EF, ejection fraction; eGFR, estimated glomerular filtration rate; HDL-C, high-density lipoprotein-cholesterol; LAD, left anterior descending artery; LCX, left circumflex artery; LDH, lactate dehydrogenase; LDL-C, low-density lipoprotein-cholesterol; LMT, left main trunk; RCA, right coronary artery; SBP, systolic blood pressure; Total-C, total-cholesterol.

triggered by plaque rupture or erosion resulting in coronary thrombotic occlusion.^{23–25} The present study also shows that *ALDH2*2* is causally associated with coronary spasm as well

as AFS in patients with STEMI, which is in agreement with our previous study in patients with CSA.¹⁴ Coronary spasm is associated with increased reactive oxygen species (ROS),

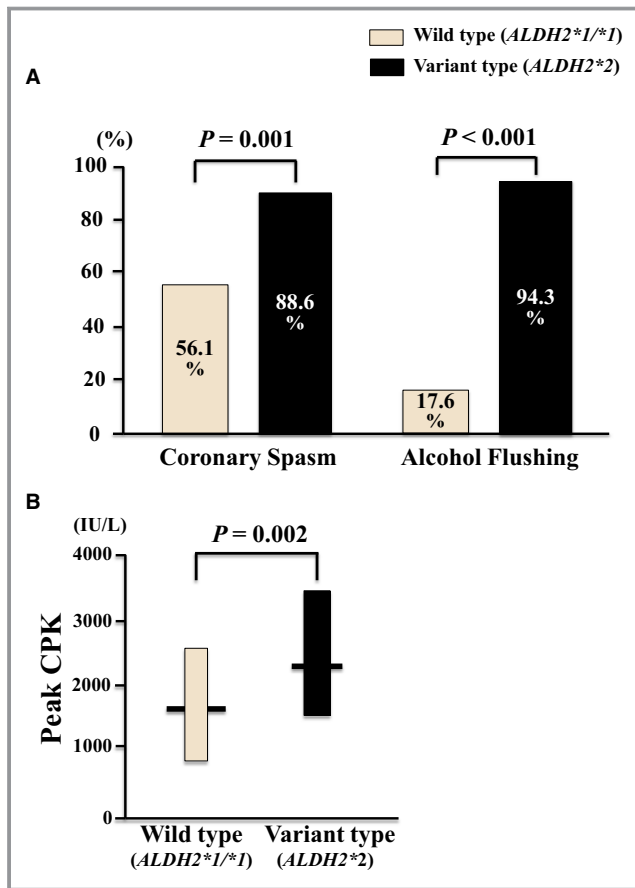


Figure 2. Comparison of (A) frequency of CS and AFS by ALDH2 genotype and (B) levels of peak CPK. Frequencies of CS and AFS (A) and peak CPK levels (B) were higher in *ALDH2*2* compared with *ALDH2*1/*1* carriers. Horizontal bar indicates a median and the vertical bar 25th and 75th percentile, respectively (B). AFS indicates alcohol flushing syndrome; ALDH2, aldehyde dehydrogenase 2; CPK, creatine phosphokinase.

endothelial impairment, and thrombogenicity.^{26–31} It is therefore likely that coronary spasm may trigger plaque rupture or erosion and eventually result in coronary occlusion and STEMI,^{18,19,23–25} particularly in those with *ALDH2*2*. It is to be noted in this connection that coronary spasm has a circadian variation with a peak incidence occurring in the early morning, which corresponds to that of STEMI.^{31,32} It is also possible that STEMI with stent implantation may conversely predispose to coronary spasm.^{12,13} Interestingly, there are currently almost no genetic data pertaining directly to STEMI except for ABO blood groups in the populations other than East Asians.³³ Previous studies treated the phenotypes of myocardial infarction (MI) and coronary artery disease (CAD) as synonymous on the basis that MI is precipitated by rupture of an atheromatous plaque followed by thrombotic occlusion superimposed on significant coronary atherosclerosis.³⁴ The association of genes with both CAD and MI are mainly

accounted for by significant atherosclerosis rather than MI.³³ However, the genetic variants contributing to plaque rupture or thrombosis may be distinct from those contributing to coronary atherosclerosis.³⁴

Timely reperfusion by direct PCI is mandatory to salvage ischemic myocardium from infarction, but reperfusion per se paradoxically contributes to myocardial injury.^{19,35,36} Toxic aldehydes such as 4-HNE are abundantly produced as a result of myocardial I/R injury, and ALDH2 activity protects against I/R injury by eliminating toxic aldehydes in experimental models.^{2,15,16} In the present study, peak plasma total CPK levels were significantly higher, those of LDH, cTnT, and BNP tended to be higher, and LVEF lower in patients with *ALDH2*2* than those with *ALDH2*1/*1*. These findings suggest that the carriers of *ALDH2*2* suffer more-severe myocardial injury post-AMI as compared to those of *ALDH2*1/*1*. It is intriguing in this connection to note that the in-hospital mortality rate for AMI is the highest in Japan among the developed countries of the Organization for Economic Cooperation and Development (OECD) member nations³⁷ and that the adjusted in-hospital mortality rates for AMI are higher in Asian-Americans than white Americans.³⁸ However, recent studies indicate that 4-HNE may paradoxically evoke hormetic protective effects against oxidative injury in the relatively low concentrations through activating antioxidative systems such as the Nrf2 pathway in experimental models.^{16,39–42} Thus, further studies are required to determine whether carriers of *ALDH2*2* suffer more-severe myocardial injury post-AMI.

Clinical Implications

Oxidative degradation of lipid membrane (lipid peroxidation) generates numerous reactive aldehydes and causes oxidative damage in various organs and tissues including the cardiovascular system.^{1,2,15,16,39–42} ALDH2 removes not only acetaldehyde but also other toxic aldehydes, including 4-HNE and malondialdehyde from lipid peroxidation or acrolein in tobacco smoke, and thereby protects tissues and cells from oxidative damage.^{1,2} Conversely, ALDH2 activity is suppressed by ROS and/or aldehydes.^{2,41,42} It is thus likely that carriers of *ALDH2*2* have increased reactive aldehydes as risk factors for coronary spasm and AMI. The present study therefore identifies reactive aldehydes as the target to be intervened for the treatment and prevention of coronary spasm and AMI. Using genetically modified ALDH2 models, Ren et al. have shown that ALDH2 also has a cardioprotective role through counteracting cardiac remodeling and myocardial dysfunction after alcohol intake.⁴³ ALDH2 may thus possess important therapeutic potential against alcoholic and other forms of myocardial damage as well.⁴³

The study also indicates that AFS is a sensitive clinical marker for *ALDH2*2*. These concepts are illustrated in

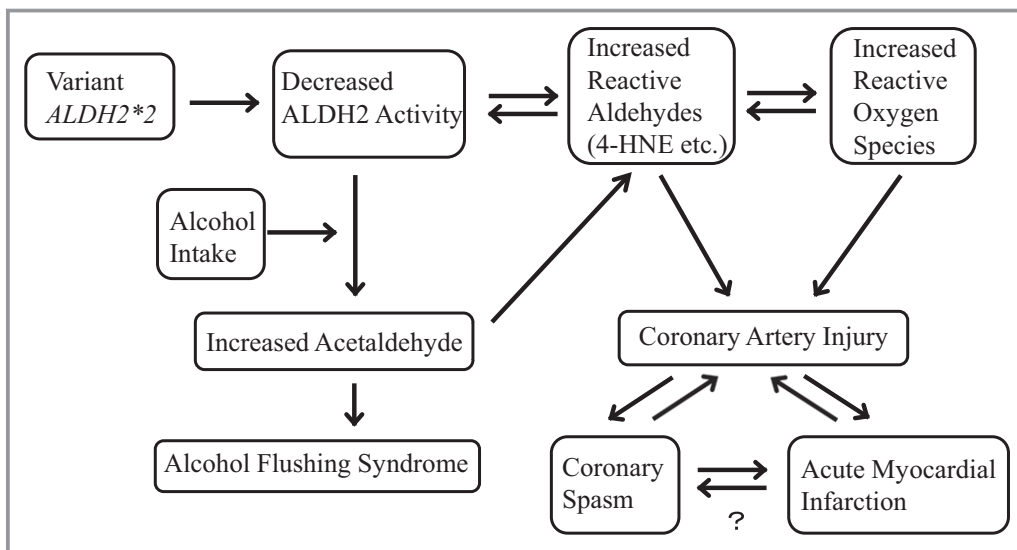


Figure 3. Association of variant *ALDH2*2* with coronary spasm and acute myocardial infarction. Variant *ALDH2*2* with deficient ALDH2 activity leads to increased reactive aldehydes, such as 4-HNE, associated with increased reactive oxygen species. Increased reactive aldehydes and reactive oxygen species cause coronary artery injury, which may lead to coronary spasm and acute myocardial infarction. Carriers of variant *ALDH2*2* also have increased acetaldehyde caused by deficient ALDH2 activity on alcohol intake and may thereby suffer from alcohol flushing syndrome. 4-HNE indicates 4-hydroxynonenal; ALDH2, aldehyde dehydrogenase 2.

Figure 3. We thus propose that all patients should be screened for AFS or at least have it documented in the patient's health records.

ALDH2 also plays an essential role in bioactivation of GTN widely used for treatment of ischemic heart disease.^{2,44,45} However, continued administration of GTN leads to tolerance or even cardiac events through inactivation of ALDH2 enzyme and an increased ROS.^{2,44–46} Accordingly, carriers of *ALDH2*2* genotypes are less responsive to GTN and are also susceptible to GTN tolerance and oxidative damage.^{2,44–46}

Calcium-channel blockers are the mainstay in treatment of coronary spasm at present.^{31,47} The results of the present study thus imply that calcium-channel blockers also should be considered for treatment of post-AMI patients, particularly those with *ALDH2*2*.^{12,13,47} Recently, Chen et al. showed that a novel small molecule activator of ALDH2, Alda-1, enhanced activity of ALDH2 and effectively restored activity of variant *ALDH2*2* to wild-type levels in animal models.^{2,15} Accordingly, it is expected that this class of drug may serve as new therapeutics for AMI and coronary spasm, particularly in those with *ALDH2*2* in the future.

Limitations

The number of study patients was small because the study included also the invasive intervention. The study subjects were a select population of STEMI who underwent PCI with stent implantation, and this may have altered the relationship

with the ALDH2 variant. Thus, the association found in this study needs confirmation in other patient groups. The study subjects were limited to Japanese AMI patients because of the genetic association study,²² and thus the results of this study may not be necessarily applicable to other populations. Frequency of AFS was assessed on questionnaire survey, and recall subjective biases may have influenced the results. Aldehydes such as 4-HNE were not measured because aldehydes are highly reactive and large parts of them exist as adducts with proteins, DNA, or lipids in tissues,^{1,41,42} and clinically applicable methods are not yet available. In the present study, the patients with severe coronary atherosclerosis and those with severe hypertension were excluded, although the rs671 variant has been reported to associate with both CAD and essential hypertension in the previous studies.^{9–11,48} However, the risk factors for, and the predilection sites of, coronary spasm are distinct from those of coronary atherosclerosis and hypertension is not a risk factor for coronary spasm.^{49,50} We have shown that stable angina, a prototype of advanced coronary atherosclerosis, is not associated with *ALDH2*2* (rs671), but MI is.²¹

Conclusions

The variant *ALDH2*2* genotype was causally associated with coronary spasm and AMI. The study identified deficient ALDH2 activity and hence reactive aldehydes as the risk factors to be targeted and intervened for treatment and

prevention of coronary spasm and AMI. AFS is a sensitive clinical marker for *ALDH2*2* genotype.

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Disclosures

None.

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