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Relation Between Hyperinsulinemia and Nonculprit Plaque Characteristics in Nondiabetic Patients With Acute Coronary Syndromes

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OBJECTIVES We sought to assess whether hyperinsulinemia is associated with percentage lipid and coronary plaque burden in nondiabetic patients with acute coronary syndromes (ACS).

BACKGROUND Hyperinsulinemia carries an increased risk of cardiovascular disease even in pre-diabetic patients, but the precise mechanisms of its effects remain unclear.

METHODS Nonculprit coronary lesions associated with mild-to-moderate stenosis in 82 nondiabetic patients with ACS were examined by integrated backscatter intravascular ultrasound (IB-IVUS), using a 40-MHz intravascular catheter. Conventional IVUS and IB-IVUS measurements from the worst 10-mm segment (1-mm intervals) were calculated. All patients underwent a 75-g oral glucose tolerance test (OGTT) to calculate the area under the insulin concentration-time curve (AUC insulin) from 0 to 120 min.

RESULTS Patients in the high tertile of AUC insulin had a significantly greater percentage lipid area and absolute lipid volume than did patients in the intermediate and low tertiles (tertile 3 vs. tertile 2 vs. tertile 1; 37.6 ± 16.6% vs. 25.8 ± 11.9% vs. 27.5 ± 14.7%, $p < 0.01$ by analysis of variance [ANOVA], and 29.9 ± 22.6 mm^3 vs. 15.3 ± 12.6 mm^3 vs. 17.7 ± 12.7 mm^3, $p < 0.01$ by ANOVA, respectively) and a smaller percentage fibrosis area (55.0 ± 11.5% vs. 61.7 ± 9.4% vs. 60.7 ± 9.4%, $p = 0.03$ by ANOVA). Multiple regression analysis showed that the high tertile of AUC insulin was independently associated with an increased percentage lipid area ($p < 0.05$). On conventional IVUS analysis, external elastic membrane cross-sectional area was significantly increased with greater plaque volume in patients in the high tertile of AUC insulin (both $p < 0.05$ by ANOVA).

CONCLUSIONS Hyperinsulinemia is associated with an increased lipid content and a greater plaque volume of nonculprit intermediate lesions in nondiabetic patients with ACS, suggesting that plaque vulnerability is increased in this subgroup of patients. (J Am Coll Cardiol Img 2011;4:392–401)

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Diabetes mellitus (DM) is a well-established risk factor for coronary artery disease (CAD). Coronary atherosclerosis is accelerated, not only in diabetic patients, but also in pre-diabetic patients with abnormal glucose metabolism, as compared with individuals with normal glucose metabolism (1). In fact, a large cohort study has shown that abnormal glucose metabolism before overt DM is associated with a 50% to 60% higher risk of mortality (2). A better understanding of the mechanisms linking abnormal glucose metabolism to CAD is needed to develop new strategies to prevent premature mortality from cardiovascular disease in patients with milder forms of abnormal glucose metabolism.

Hyperinsulinemia, a marker of insulin resistance, is related to impaired glucose tolerance (IGT) and DM (3). Accumulating evidence suggests that insulin directly promotes atherogenesis (4). The Helsinki Policemen Study showed that hyperinsulinemia without DM predicted the risk of CAD-related events during 22 years of follow-up (5). Patients with hyperinsulinemia may thus have increased plaque vulnerability even before the onset of DM. However, few studies have directly examined the relevance of hyperinsulinemia to plaque vulnerability.

Prior studies have indicated that the degree of stenosis caused by culprit lesions in acute coronary syndromes (ACS) is not necessarily severe on coronary angiography weeks to months before the onset of symptoms (6). The disruption of vulnerable plaque associated with mild-to-moderate angiographic stenosis and subsequent thrombosis is thus thought to contribute to the onset of ACS (7). Plaque disruption and acute occlusion occur at sites of vulnerable plaque with a high lipid content covered by a thin fibrous cap (8,9).

Recently, Kawasaki et al. (10) have developed a technique for integrated backscatter intravascular ultrasound (IB-IVUS) that can identify different components of atherosclerotic plaque in human coronary arteries in vivo. This study tested the hypothesis that hyperinsulinemia is associated with an increased lipid component of atherosclerotic plaques, which may heighten plaque vulnerability. The relation between hyperinsulinemia and the tissue characteristics of nonculprit intermediate coronary plaques on IB-IVUS was examined in nondiabetic patients with ACS.

METHODS

Subjects. This study was a prospectively planned observational study of nonculprit coronary plaques in patients with ACS. Consecutive patients with ACS who underwent percutaneous coronary intervention (PCI) with IVUS guidance at the Yokohama City University Medical Center were screened for eligibility. Patients with a previous diagnosis of DM and an HbA_{1c} level of more than 6.5% were excluded because it is difficult to evaluate the insulin response to a glucose load in such patients owing to impaired insulin secretion (11). A total of 112 patients in stable condition underwent 75-g oral glucose tolerance tests (OGTT) before hospital discharge. We excluded 25 patients in whom DM was diagnosed on OGTT and 5 patients for whom IB-IVUS data were unavailable. Finally, 82 lesions with mild-to-moderate stenosis in 82 nondiabetic patients with ACS were studied. The study protocol was approved by the ethics committee of Yokohama City University Medical Center. We obtained written informed consent from all participants before initial coronary angiography.

Cardiovascular diagnosis. ACS was defined as unstable angina, ST-segment elevation myocardial infarction, or non–ST-segment elevation myocardial infarction. Unstable angina was defined as new-onset severe angina, accelerated angina, or angina at rest without a significant rise in cardiac-specific troponin T levels. New-onset angina was defined as angina in which <2 months had elapsed from the date of initial symptoms. Accelerated angina was defined as angina in which symptoms were more frequent, more severe, longer, or precipitated by distinctly less exertion than previously, while the patient was in stable condition. ST-segment elevation myocardial infarction was defined as the continuous presence of chest symptoms for more than 30 min, ST-segment elevation of >0.1 mV in 2 limb leads or >0.2 mV in 2 contiguous precordial leads, and a rise in cardiac-specific troponin T levels. Non–ST-segment elevation myocardial infarction was defined as chest pain and a rise in cardiac-specific troponin T levels without new ST-segment elevation (12).

Assessment of glucose metabolism and lipid profiles. Patients without a prior diagnosis of DM underwent a 75-g OGTT while they were in stable condition at least 4 days after admission (13). After

ABBREVIATIONS AND ACRONYMS

ACS = acute coronary syndrome(s)
ANOVA = analysis of variance
AUC = area under the curve
CAD = coronary artery disease
CSA = cross-sectional area
DM = diabetes mellitus
EEM = external elastic membrane
HOMA-IR = homeostasis model assessment of insulin resistance
IB-IVUS = integrated backscatter intravascular ultrasound
IGT = impaired glucose tolerance
IRI = immunoreactive insulin
IVUS = intravascular ultrasound
OGTT = oral glucose tolerance test
PCI = percutaneous coronary intervention
P+M = plaque plus media

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an overnight fast, venous blood samples for the measurement of plasma glucose levels and immunoreactive insulin (IRI) levels were taken at baseline, 60 min, and 120 min after the glucose load. Glucose metabolism was then classified on the basis of the results of OGTT as follows: IGT = fasting blood glucose level <126 mg/dl and 120-min post-load blood glucose level ≥140 mg/dl, but <200 mg/dl; DM = fasting blood glucose level ≥126 mg/dl or 120-min post-load blood glucose ≥200 mg/dl.

The area under the insulin concentration-time curve (AUC insulin) was calculated from the IRI levels at baseline, 60 min, and 120 min with the use of the trapezoid rule (5). The AUC insulin was used as a composite variable reflecting IRI levels. Homeostasis model assessment of insulin resistance (HOMA-IR) was calculated by the following formula: (fasting blood glucose level × fasting IRI level)/405.

IVUS and IB-IVUS examinations. Immediately after PCI, IVUS examinations were performed using a 40-MHz mechanical scanning monorail intracoronary ultrasound catheter (Boston Scientific Corporation, Natick, Massachusetts). All IVUS examinations were performed in the following manner. After the intracoronary administration of 2 to 3 mg of isosorbide dinitrate, the catheter was positioned sufficiently distal to the site of PCI. Pullback was performed automatically at 0.5 mm/s. A personal computer equipped with custom software (IB-IVUS, YD Co., Nara, Japan) was connected to the IVUS imaging system to obtain radiofrequency signal output, signal trigger output, and video image output. The IB values for each tissue component were calculated as the average power, using a fast Fourier transformation, measured in decibels, of the frequency component of backscattered signals from a small volume of tissue (10).

Analysis of conventional IVUS and IB-IVUS data. The IVUS images for the entire target vessel post-PCI were reviewed, and target plaque was determined. The target plaque had to be an intermediate lesion with no more than 50% luminal narrowing on angiography and no previous PCI. All analyzed plaques were located in the treated vessel and had to be situated at least 5 mm from the culprit lesion. Each plaque was measured by conventional IVUS and IB-IVUS at 1-mm intervals throughout the most diseased (containing the greatest plaque volume) 10-mm segment. The target plaque was selected, and the conventional grayscale IVUS and IB-IVUS data were analyzed by an investigator who was blinded to the patients' characteristics.

Conventional IVUS image analysis was conducted using commercially available software (Echo Plaque, Indec Systems, Mountain View, California). Quantitative analysis was performed according to the American College of Cardiology Clinical Expert Consensus Document on IVUS (14). The cross-sectional area (CSA) measurements included the lumen and the external elastic membrane (EEM) CSA. The luminal-intimal border was traced manually to determine the lumen CSA. EEM CSA was measured by tracing the external edge of the media-adventitia border. Plaque plus media (P+M) CSA was calculated as the EEM CSA minus the lumen CSA. Remodeling index was calculated as the minimum lumen area site EEM CSA divided by the average of the proximal and distal reference site EEM CSA, as described previously (15). The EEM, lumen, and P+M volumes were calculated by Simpson method for the integration of 1-mm-thick disks for the 10 serial CSAs (16).

Color-coded maps based on IB values were constructed for consecutive IVUS image slices of the target plaque. Because the media of the coronary arteries always presents with a low echoic band, potentially identified as "lipid" by the algorithm currently used for IB-IVUS, the luminal-intimal border and intimal-medial border were traced manually. The percentages of each plaque component (lipid, fibrosis, dense fibrosis, and calcification) were automatically computed by the IB-IVUS system after exact manual tracing. The absolute value of each plaque component was calculated (sum of each plaque component in each CSA at 1-mm intervals for the 10-mm length), and the average values of each plaque component were expressed relative to the total volume. Cross-sectional IB-IVUS analysis was also performed at the minimal lumen site.

Whole-vessel IB-IVUS analysis prior to PCI. We evaluated plaque components for all available frames (mean length of 52 mm) in the target vessel prior to PCI by IB-IVUS analysis at 1-mm intervals. We excluded 6 patients in whom pre-intervention IVUS could not be performed because the IVUS catheter could not cross the lesion. We also excluded 12 patients for whom adequate IB-IVUS data were unavailable. Therefore, IB-IVUS data prior to PCI qualified for evaluation in 64 patients.

Statistical analysis. Statistical analysis was performed with StatView 5.0 software (SAS Institute,

Cary, North Carolina). Qualitative data are presented as numbers (%). Normally distributed, continuous variables are expressed as mean ± SD, and continuous variables with skewed distributions (triglycerides and C-reactive protein) are expressed as median values (interquartile range). Categorical variables were compared by the chi-square test. Continuous clinical and IVUS variables among the 3 groups were compared by analysis of variance (ANOVA), with pairwise post hoc comparisons adjusted by the Bonferroni method. For continuous variables with skewed distributions, the Kruskal-Wallis test was used to examine differences in median values, with pairwise post hoc comparisons by the Mann-Whitney *U* test with the Bonferroni adjustment. Correlations of plaque contents, as assessed by IB-IVUS, with the high tertile of AUC insulin and clinical and laboratory variables were evaluated by simple regression analysis. Multiple linear regression analysis was used to determine the best predictors of the percentage lipid area. Logarithms (base 2) of triglycerides and C-reactive protein were used in all the regression analyses to account for the skewed distributions of these variables. Univariate predictors of percentage lipid area with p values of <0.2 were entered into the multivariate model. Independent predictors and their regression coefficients were calculated. Differences with p values of <0.05 were considered statistically significant, except when employing the Bonferroni adjustment. In the analysis with Bonferroni adjustment, p values of <0.0167 were considered significant.

RESULTS

Subjects and baseline characteristics. The mean age of the patients was 64 ± 12 years. Seventy-nine percent of the study group were men, 45% had a history of hypertension, and 55% were current smokers. At admission, statins were being used by 18% of the patients. The criteria for IGT was met in 65%. The AUC insulin, calculated from the results of a 75-g OGTT, ranged from 1,998 to 28,080 IU/l × min in this patient cohort. Unstable angina was diagnosed in 15% of the patients, non–ST-segment elevation myocardial infarction in 24%, and ST-segment elevation myocardial infarction in 61%. The prevalence of IGT was similar among the 3 groups (64% vs. 65% vs. 67%, p = 0.98). The AUC insulin tended to be higher in patients with unstable angina than in those with non–ST-segment elevation myocardial infarction and ST-segment elevation myocardial infarction (10,087 ± 6,958 IU/l × min vs. 8,071 ± 3,568 IU/l × min vs. 7,182 ± 3,496 IU/l × min, p = 0.097 by ANOVA).

The patients were divided into 3 groups according to the tertile of AUC insulin at baseline: tertile 1 (low tertile), 27 patients with AUC insulin of $<$5,300 IU/l × min; tertile 2 (intermediate tertile), 27 patients with AUC insulin of 5,300 to 9,000 IU/l × min; and tertile 3 (high tertile), 28 patients with AUC insulin of $>$ 9,000 IU/l × min. The baseline characteristics of the subjects are shown in Table 1. Body mass index and waist circumference were significantly greater in patients in the high tertile of AUC insulin than those in the intermediate or low tertiles. Lipid profiles and the prevalence of hypertension, current smokers, and statin use at admission did not differ among the tertiles. Clinical presentation, the proportions of patients with multivessel disease and stent implantation, and target plaque locations were also similar. As expected, fasting IRI levels were significantly higher in the high tertile of AUC insulin than in the other tertiles. Serum triglycerides and the prevalence of IGT were significantly higher in the high tertile than in the low tertile.

Quantitative measures of conventional and IB-IVUS. Table 2 shows the results of conventional grayscale IVUS analyses. The EEM, lumen, and P+M volumes were greater in the high tertile of AUC insulin than in the intermediate and low tertiles ($p < 0.05$ by ANOVA). The remodeling index and the proportion of target plaques proximal to the PCI site were similar in the 3 groups.

On IB-IVUS analysis, patients in the high tertile of AUC insulin had a significantly greater percentage lipid area than those in the intermediate and low tertiles (tertile 3 vs. tertile 2 vs. tertile 1; 37.6 ± 16.6% vs. 25.8 ± 11.9% vs. 27.5 ± 14.7%, $p < 0.01$ by ANOVA) (Fig. 1A), as well as a smaller percentage fibrosis area (55.0 ± 11.5% vs. 61.7 ± 9.4% vs. 60.7 ± 8.9%, p = 0.03 by ANOVA) (Fig. 1B) and percentage dense fibrosis area (5.5 ± 5.0% vs. 9.4 ± 5.1% vs. 8.7 ± 6.6%, p = 0.02 by ANOVA) (Fig. 1C). The percentage calcification area was similar in the 3 groups (p = 0.14 by ANOVA) (Fig. 1D). The absolute lipid volume was significantly greater in the high tertile of AUC insulin than in the intermediate and low tertiles (tertile 3 vs. tertile 2 vs. tertile 1: 29.9 ± 22.5 mm^3 vs. 15.3 ± 12.6 mm^3 vs. 17.7 ± 12.7 mm^3, $p < 0.01$ by ANOVA) (Fig. 2A). The absolute fibrosis volume, dense fibrosis volume,

Table 1. Baseline Clinical Characteristics of Patients by Tertiles of AUC Insulin

	Tertile 1 (n = 27)	Tertile 2 (n = 27)	Tertile 3 (n = 28)	p Value
Age, yrs	67 ± 11	63 ± 12	60 ± 12	0.10
Male sex	21 (78)	19 (70)	25 (89)	0.22
Target plaque location				0.15
Left anterior descending	11 (41)	15 (56)	14 (50)	
Left circumflex	0 (0)	3 (11)	4 (14)	
Right	16 (59)	9 (33)	10 (36)	
Clinical presentation				0.41
Unstable angina	3 (11)	3 (11)	6 (21)	
Non–ST-segment elevation MI	6 (22)	5 (19)	9 (33)	
ST-segment elevation MI	18 (67)	19 (70)	13 (46)	
Stent implantation	26 (96)	25 (93)	28 (100)	0.34
BMS	19 (70)	19 (70)	21 (75)	0.91
DES	7 (26)	6 (23)	7 (25)	0.95
Previous PCI	1 (4)	0 (0)	2 (7)	0.38
Multivessel disease	12 (44)	10 (37)	13 (46)	0.77
Body mass index, kg/m^2	22.4 ± 2.9	23.6 ± 3.6	26.4 ± 3.9†	<0.01
Waist circumference, cm	84 ± 7	85 ± 9	94 ± 11†	<0.01
Hypertension	9 (33)	13 (48)	15 (54)	0.30
Current smoker	15 (56)	17 (63)	13 (46)	0.47
Blood lipid levels, mg/dl				
LDL cholesterol	134 ± 30	149 ± 39	140 ± 26	0.20
HDL cholesterol	47 ± 9	52 ± 15	48 ± 14	0.26
Triglycerides	92 (55–126)	101 (57–199)	174 (133–234)*	<0.01
Statin use at admission	5 (19)	4 (15)	6 (21)	0.82
hs-CRP, mg/dl	0.163 (0.062–0.996)	0.187 (0.093–0.355)	0.130 (0.091–0.585)	0.95
IGT	13 (48)	17 (63)	23 (82)	0.03
HemoglobinA_{1c}, %	5.2 ± 0.4	5.4 ± 0.4	5.3 ± 0.3	0.35
Fasting blood glucose, mg/dl	98 ± 7	101 ± 8	103 ± 7	0.07
Fasting IRI, IU/l	4.9 ± 2.0	6.3 ± 1.7	10.8 ± 4.7†	<0.01
AUC insulin, IU/l × min	3,876 ± 889	7,068 ± 1,244*	12,360 ± 3,769†	<0.01

Values are mean ± SD or n (%). p = chi-square test, analysis of variance, or Kruskal-Wallis test among the 3 tertiles; values are mean ± SD or median values (25th to 75th percentiles) or numbers of patients (percentages). *p < 0.0167 by Bonferroni adjustment (vs. tertile 1); †p < 0.0167 by Bonferroni adjustment (vs. tertile 1 and tertile 2).
ACS = acute coronary syndrome; AUC = area under curve; BMS = bare-metal stent; DES = drug-eluting stent; HDL = high-density lipoprotein; hs-CRP = high-sensitivity C-reactive protein; IGT = impaired glucose tolerance; IRI = immunoreactive insulin; LDL = low-density lipoprotein; PCI = percutaneous coronary intervention; MI = myocardial infarction.

and calcification volume were similar among the 3 groups. At the minimal lumen site, similar results were obtained (tertile 3 vs. tertile 2 vs. tertile 1: 3.4 ± 2.9 mm^2 vs. 1.7 ± 1.6 mm^2 vs. 2.1 ± 1.5 mm^2, p < 0.01 by ANOVA) (Fig. 2B). Figure 3 shows representative conventional IVUS images and IB-IVUS color-coded maps of a nonculprit coronary plaque in a patient belonging to the high tertile of AUC insulin and those in a patient belonging to the low tertile of AUC insulin.

Table 2. Data for Conventional Intravascular Ultrasound

	Tertile 1 (n = 27)	Tertile 2 (n = 27)	Tertile 3 (n = 28)	p Value
EEM volume, mm^3	134 ± 42	129 ± 42	166 ± 50*	<0.05
Lumen volume, mm^3	74 ± 30	76 ± 24	93 ± 34	<0.05
P+M volume, mm^3	59 ± 27	56 ± 25	73 ± 28	<0.05
Remodeling index	0.94 ± 0.12	0.99 ± 0.17	0.96 ± 0.12	0.45
Proximal to the PCI site, %	78	59	75	0.28

p = analysis of variance, values are means ± standard deviations. *p < 0.0167 by Bonferroni adjustment (vs. tertile 1 and tertile 2).
EEM = external elastic membrane; P+M = plaque plus media; PCI = percutaneous coronary intervention.

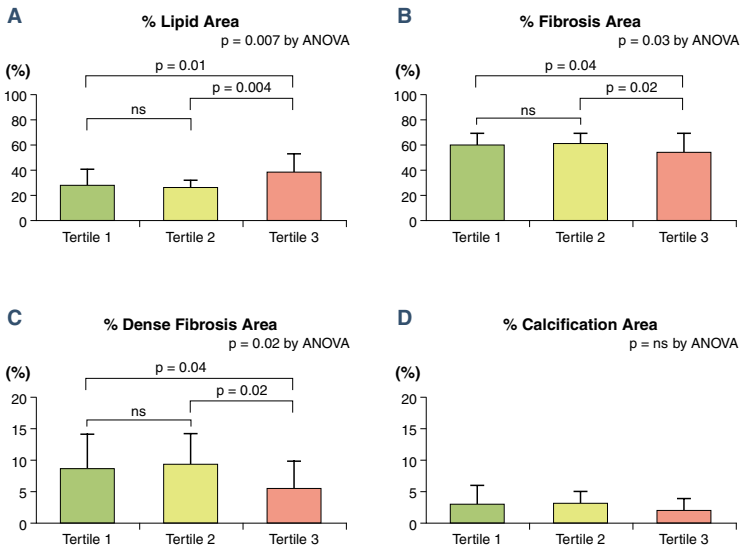

Figure 1. Relations Between AUC Insulin and Each Relative Plaque Component

Nonculprit plaques in patients belonging to the high tertile of area under the curve (AUC) insulin had a significantly increased percentage lipid area **(A)** as compared with those in patients belonging to the intermediate and low tertiles (tertile 3 vs. tertile 2 vs. tertile 1: 37.6 ± 16.6% vs. 25.8 ± 11.9% vs. 27.5 ± 14.7%, p = 0.007 by analysis of variance [ANOVA]) and a decreased percentage fibrosis area **(B)** (55.0 ± 11.5% vs. 61.7 ± 9.4% vs. 60.7 ± 8.9%, p = 0.03 by ANOVA) and percentage dense fibrosis area **(C)** (5.5 ± 5.0% vs. 9.4 ± 5.1% vs. 8.7 ± 6.6%, p = 0.02 by ANOVA). The percentage calcification area **(D)** did not differ significantly among the 3 groups.

Simple and multiple regression analysis for percentage lipid area as the dependent variable. The high tertile of AUC insulin was used as a variable for simple and multiple regression analyses for percentage lipid area because the high tertile of AUC insulin had significantly increased percentage lipid area compared with intermediate and low tertiles (p = 0.004 and p = 0.01, respectively, by the Bonferroni adjustment). On simple regression analysis, the high tertile of AUC insulin (r = 0.34, p < 0.01), younger age (r = −0.25, p = 0.03), male sex (r = 0.33, p < 0.01), and the presence of IGT (r = 0.27, p = 0.01) correlated with a greater percentage lipid area (Table 3). Multiple regression analysis showed that the high tertile of AUC insulin and the prevalence of IGT were independently associated with an increased percentage lipid area (Table 3).

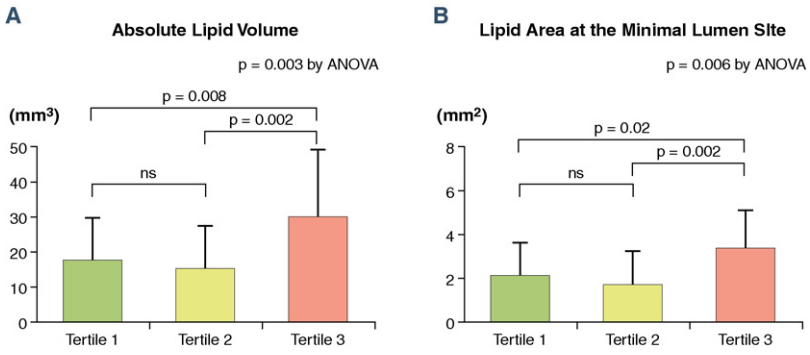

Figure 2. Relations Between AUC Insulin and Absolute Lipid Volume and Lipid Area at Minimal Lumen Site

Nonculprit plaques in patients belonging to the high tertile of area under the curve (AUC) insulin had an increased absolute lipid volume and lipid area at minimal lumen site as compared with those in patients belonging to the intermediate and low tertiles (tertile 3 vs. tertile 2 vs. tertile 1: 29.9 ± 22.5 mm^3 vs. 15.3 ± 12.6 mm^3 vs. 17.7 ± 12.7 mm^3, p = 0.003 by analysis of variance [ANOVA] **[A]** and 3.4 ± 1.5 mm^2 vs. 1.7 ± 1.6 mm^2 vs. 2.1 ± 1.5 mm^2, p = 0.006 by ANOVA **[B]**, respectively).

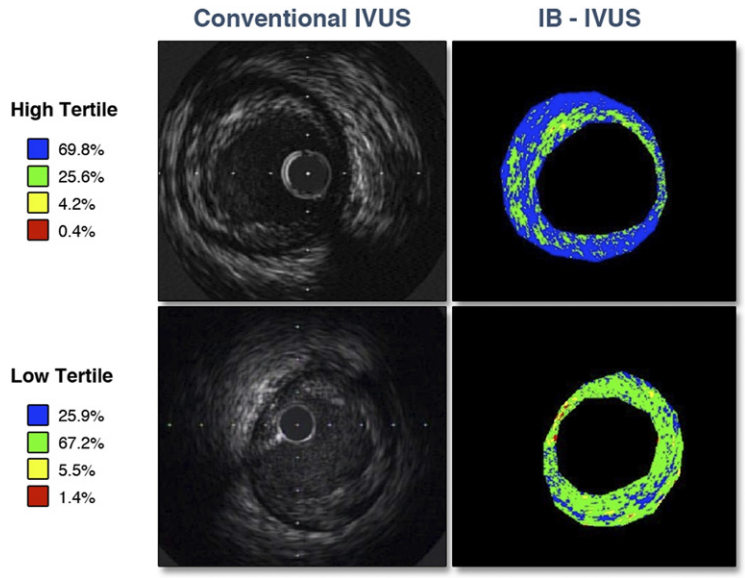

Figure 3. Representative Images of Conventional IVUS and IB-IVUS Color-Coded Maps of Coronary Artery Plaques

The percentage lipid area, percentage fibrosis area, and percentage dense fibrosis area of coronary artery plaque were 69.8%, 27.0%, and 1.8%, respectively, in a patient belonging to the high tertile of area under the curve (AUC) insulin (AUC insulin, 13,290; **top panels**). The percentage lipid area, percentage fibrosis area, and percentage dense fibrosis area were 25.9%, 67.0%, and 5.7%, respectively, in a patient belonging to the low tertile of AUC insulin (AUC insulin, 4,746; **bottom panels**). **Blue** = lipid area, **green** = fibrosis area, **yellow** = dense fibrosis area, **red** = calcification area. IB-IVUS = integrated backscatter intravascular ultrasound.

Relations between IGT and HOMA-IR and plaque components. IGT is reported to be associated with hyperinsulinemia (3). Therefore, we evaluated the relation between IGT and percentage lipid area. Patients with IGT had a significantly greater percentage lipid area as compared with those with normal glucose tolerance (IGT vs. normal glucose tolerance: 33.4 ± 15.6% vs. 24.7 ± 13.3%, p = 0.01) (Fig. 4).

In addition, we evaluated the association between HOMA-IR and plaque components, since HOMA-IR is a simple method for assessing insulin resistance (17). We divided the patients into the 3 tertiles according to the HOMA-IR levels: tertile 1, 27 patients with HOMA-IR of <1.23; tertile 2, 27 patients with HOMA-IR of 1.23 to 1.93; and tertile 3, 28 patients with HOMA-IR of >1.93. There were no significant differences in percentage lipid area among the 3 tertiles (p = 0.20 by ANOVA). On simple regression analysis, the high tertile of HOMA-IR showed a trend toward an increased percentage lipid area (r = 0.19, p = 0.09).

Subgroup analysis of the whole target vessel prior to PCI. In 64 patients in whom whole target vessels prior to PCI were analyzed, patients with the high tertile of AUC insulin had the highest percentage lipid area (tertile 3 vs. tertile 2 vs. tertile 1: 38.7 ± 13.5% vs. 30.7 ± 14.2% vs. 26.1 ± 9.3%, p < 0.01 by ANOVA) (Fig. 5).

Table 3. Simple and Multiple Regression Analysis With % Lipid Area as the Dependent Variable

Variable	Simple Regression		Multiple Regression	
	Regression Coefficient	p Value	Regression Coefficient	p Value
Age	−0.33	0.03	−0.18	0.27
Male	12.43	<0.01	6.72	0.13
Body mass index	0.53	0.23		
Unstable angina	3.53	0.46		
Non–ST-segment elevation MI	−3.53	0.37		
ST-segment elevation MI	0.88	0.80		
Hypertension	−0.12	0.97		
Current smoker	4.89	0.15	3.19	0.38
LDL cholesterol	−0.04	0.42		
HDL cholesterol	−0.23	0.07	−0.13	0.34
Triglycerides	2.33	0.19	−2.13	0.24
Statin use at admission	2.52	0.57		
hs-CRP	0.87	0.24		
IGT	8.69	0.01	6.60	0.04
Fasting blood glucose	0.43	0.06	0.40	0.06
The high tertile of AUC insulin	10.95	<0.01	7.91	0.03

Abbreviations as in Table 1.

DISCUSSION

Our IB-IVUS study assessed the impact of hyperinsulinemia, expressed as the AUC insulin during a 75-g OGTT, on the tissue characteristics of coronary plaques associated with angiographically mild-to-moderate coronary lesions. We found that hyperinsulinemia was related to a significantly increased lipid content and a greater plaque volume of nonculprit intermediate lesions in nondiabetic patients with ACS. These results provide important insight into the relation between hyperinsulinemia and plaque stability.

It has been reported that disruption of vulnerable plaques and subsequent thrombus formation are the most frequent causes of ACS (7). Lipid incorporation into the arterial wall is the key event in the initiation of atherosclerosis, and the formation of a soft lipid core is an important determinant of spontaneous plaque rupture (18). Necropsy studies have suggested that the presence of a large lipid-rich core is associated with a high risk of ACS after spontaneous or mechanically induced rupture because of the thrombogenicity of the lipid content (19). Previous conventional IVUS studies have shown that large eccentric plaques containing an echolucent zone can have an increased risk of instability even if the lumen area is preserved at the time of initial study, indicating that tissue characteristics have a great impact on coronary plaque vulnerability (8). Recent preliminary in vitro studies have shown that IB values reflect the structural and biochemical compositions of atherosclerotic lesions and can differentiate fibrofatty, fatty, and calcified lesions of arterial walls (20). Okubo et al. (21) used IB values to analyze lipid-rich, fibrotic, and fibrocalcific coronary plaque components and obtained high predictive accuracies of 90%, 93%, and 96%, respectively, as compared with the corresponding histological images. Therefore, IB-IVUS is a well-validated and established method that provides accurate information on plaque components.

DM is well recognized to be a major risk factor for CAD, and hyperinsulinemia is an indicator of insulin resistance that precedes the development of DM (3). Several studies have demonstrated that hyperinsulinemia is associated with an increased risk of CAD events in pre-diabetic patients (5,22,23). These findings suggest that macrovascular disease starts before the development of DM in patients with hyperinsulinemia. In the present study, hyperinsulinemia was significantly related to an increased plaque lipid content in nondiabetic patients with ACS. Our results are supported by the findings of previous IVUS tissue characterization studies showing that plaque vulnerability is increased in patients with DM as well as those with insulin resistance. Recently published virtual histology IVUS studies have reported that the prevalence of a necrotic core is significantly higher in patients with DM, especially in those with a longer duration of DM (24), than in patients without DM (25,26). Marso et al. (27) have shown that low adiponectin levels, which correlate with high insulin resistance, are associated with increased plaque volume and lipid-rich plaque in nondiabetic coronary arteries.

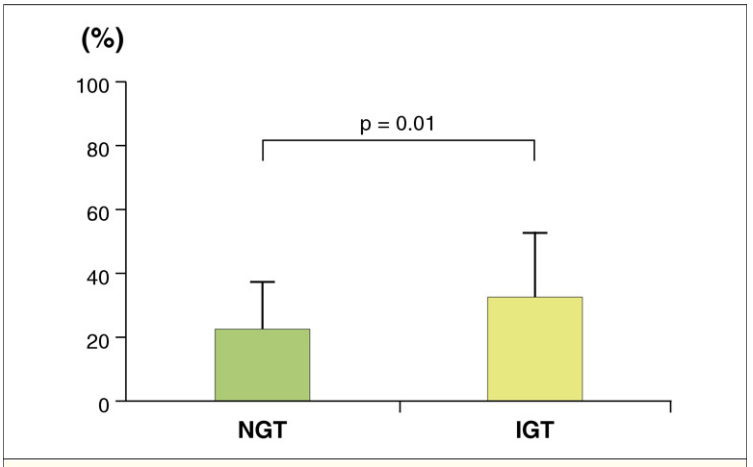

Figure 4. Relation Between IGT and Percentage Lipid Area

Patients with impaired glucose tolerance (IGT) showed a significant increase in percentage lipid area as compared with those with normal glucose tolerance (NGT).

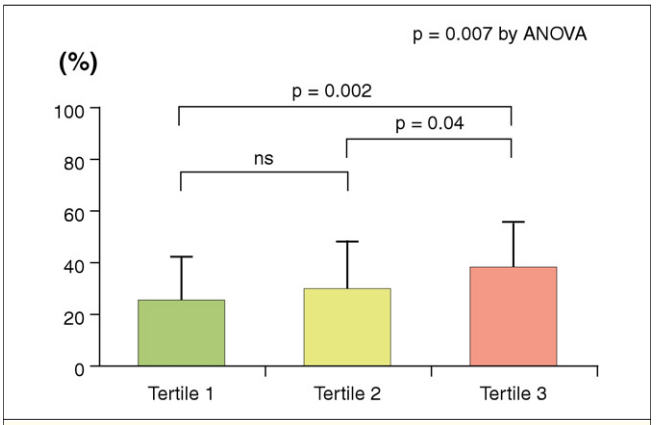

Figure 5. Relation Between AUC Insulin and Percentage Lipid Area in the Whole Target Vessel Prior to PCI

In the whole target vessel prior to percutaneous coronary intervention (PCI), percentage lipid area was highest in the high tertile of AUC insulin as compared with the intermediate and low tertiles (tertile 3 vs. tertile 2 vs. tertile 1: 38.7 ± 13.5% vs. 30.7 ± 14.2% vs. 26.1 ± 9.3%, $p < 0.01$ by ANOVA). Abbreviations as in Figures 1 and 3.

Amano et al. (28) have reported that IGT and DM are independent predictors of lipid-rich coronary plaque and that increased HOMA-IR, a simple index of insulin resistance, is associated with an elevated lipid component. Thus, diabetic status, hyperinsulinemia, and insulin resistance may be related to the increased plaque vulnerability underlying the poor long-term outcomes in diabetic and pre-diabetic patients.

On conventional IVUS analysis, EEM was significantly increased with greater P+M volume in patients in the high tertile of hyperinsulinemia. These findings suggested that coronary vessels in patients with hyperinsulinemia might be compensatorily enlarged to prevent atheroma from encroaching on the lumen. On IB-IVUS analysis, plaques in patients with hyperinsulinemia had a significantly increased proportion of lipid area. Previous studies have reported that positively remodeled vessels with greater plaque volume (29) and increased lipid-rich components (8,9) are relevant to plaque vulnerability, resulting in increased coronary events. Taken together, these findings might explain how the development of atherosclerosis contributes to increased plaque vulnerability in patients with hyperinsulinemia.

The development of hyperinsulinemia, a measure of insulin resistance, is associated with the progression of metabolic syndrome and DM (30), both of which are associated with coronary atherosclerosis. Moreover, hyperinsulinemia and insulin resistance probably have several atherogenic effects, including the promotion of inflammation and endothelial dysfunction (31). In addition, accumulating evidence from experimental studies has indicated that insulin in physiological concentrations stimulates cholesterol synthesis and low-density lipoprotein binding in arterial smooth muscle cells in animal models (32,33). Krone et al. (34) have demonstrated that insulin stimulates the specific binding, accumulation, and degradation of low-density lipoprotein in human mononuclear leukocytes. These findings provide evidence supporting a direct role of insulin in the development of lipid-rich atherosclerosis. Nevertheless, whether or not insulin has direct atherogenic effects remains controversial. Shamir et al. (35) demonstrated that insulin administration in apolipoprotein E-null mice suppresses cholesterol synthesis, reduces oxidative damage due to lipid and superoxide production by macrophages, and decreases the severity and extent of atherosclerotic plaques. Dandona et al. (36) have shown in obese subjects with IGT that vascular endothelial growth factor, which accelerates the progression of atherosclerosis, is suppressed by insulin. Our results suggest that hyperinsulinemia is associated with lipid-rich plaque and greater plaque burden, which may lead to increased plaque vulnerability. However, whether insulin increases lipid component and plaque burden remains to be established.

Study limitations. First, lipid-rich plaque as defined in the present study might not be a major marker of plaque vulnerability. Although previous studies have reported that vulnerable plaques in patients with ACS are related to an increased percentage lipid area (6–9), large, long-term studies are needed to precisely define the value of lipid-rich plaque for predicting future coronary events. Second, we excluded patients with DM. Insulin response to a glucose load might differ in patients with DM because of their impaired insulin secretion. Further studies in patients with DM are required to assess the relation between hyperinsulinemia and plaque components. Third, AUC insulin in this study was provided by only 3 measurements. It may be preferable to calculate AUC insulin from several measurements to evaluate it more accurately. Finally, we did not perform 3-vessel IVUS in this study because it is generally considered unacceptable to perform IVUS interrogations of nonculprit vessels in the setting of ACS. However, similar findings were obtained when we evaluated whole coronary vessels prior to PCI by IB-IVUS analysis.

CONCLUSIONS

Hyperinsulinemia was associated with an increased lipid content and a greater plaque volume of nonculprit intermediate lesions in nondiabetic patients with ACS. Our findings suggest that the evaluation of hyperinsulinemia may help identify patients at higher risk and may assist us in making decisions for prevention and treatment in this population.

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REFERENCES

1. Coutinho M, Gerstein HC, Wang Y, Yusuf S. The relationship between glucose and incident cardiovascular events. A metaregression analysis of published data from 20 studies of 95,783 individuals followed for 12.4 years. Diabetes Care 1999;22:233–40.
2. Barr EL, Zimmet PZ, Welborn TA, et al. Risk of cardiovascular and all-cause mortality in individuals with diabetes mellitus, impaired fasting glucose, and impaired glucose tolerance: the Australian Diabetes, Obesity, and Lifestyle Study (AusDiab). Circulation 2007;116:151–7.
3. Reusch JE, Draznin BB. Atherosclerosis in diabetes and insulin resistance. Diabetes Obes Metab 2007;9:455–63.
4. Stout RW. Insulin and atheroma. 20-yr perspective. Diabetes Care 1990;13:631–54.
5. Pyorala M, Miettinen H, Laakso M, Pyorala K. Hyperinsulinemia predicts coronary heart disease risk in healthy middle-aged men: the 22-year follow-up results of the Helsinki Policemen Study. Circulation 1998;98:398–404.
6. Little WC, Constantinescu M, Applegate RJ, et al. Can coronary angiography predict the site of a subsequent myocardial infarction in patients with mild-to-moderate coronary artery disease? Circulation 1988;78:1157–66.
7. Fuster V, Badimon L, Badimon JJ, Chesebro JH. The pathogenesis of coronary artery disease and the acute coronary syndromes (1). N Engl J Med 1992;326:242–50.
8. Varnave AM, Mills PG, Davies MJ. Relationship between coronary artery remodeling and plaque vulnerability. Circulation 2002;105:939–43.
9. Yamagishi M, Terashima M, Awano K, et al. Morphology of vulnerable coronary plaque: insights from follow-up of patients examined by intravascular ultrasound before an acute coronary syndrome. J Am Coll Cardiol 2000;35:106–11.
10. Kawasaki M, Takatsu H, Noda T, et al. In vivo quantitative tissue characterization of human coronary arterial plaques by use of integrated backscatter intravascular ultrasound and comparison with angioscopic findings. Circulation 2002;105:2487–92.
11. Ferrannini E, Mari A. How to measure insulin sensitivity. J Hypertens 1998;16:895–906.
12. Alpert JS, Thygesen K, Antman E, Bassand JP. Myocardial infarction redefined—a consensus document of The Joint European Society of Cardiology/American College of Cardiology Committee for the redefinition of myocardial infarction. J Am Coll Cardiol 2000;36:959–69.
13. Norhammar A, Tenerz A, Nilsson G, et al. Glucose metabolism in patients with acute myocardial infarction and no previous diagnosis of diabetes mellitus: a prospective study. Lancet 2002;359:2140–4.
14. Mintz GS, Nissen SE, Anderson WD, et al. American College of Cardiology clinical expert consensus document on standards for acquisition, measurement and reporting of intravascular ultrasound studies (IVUS). A report of the American College of Cardiology Task Force on Clinical Expert Consensus Documents. J Am Coll Cardiol 2001;37:1478–92.
15. Hibi K, Ward MR, Honda Y, et al. Impact of different definitions on the interpretation of coronary remodeling determined by intravascular ultrasound. Catheter Cardiovasc Interv 2005;65:233–9.
16. Matar FA, Mintz GS, Farb A, et al. The contribution of tissue removal to lumen improvement after directional coronary atherectomy. Am J Cardiol 1994;74:647–50.
17. Ascaso JF, Pardo S, Real JT, et al. Diagnosing insulin resistance by simple quantitative methods in subjects with normal glucose metabolism. Diabetes Care 2003;26:3320–5.
18. Davies MJ, Richardson PD, Woolf N, Katz DR, Mann J. Risk of thrombosis in human atherosclerotic plaques: role of extracellular lipid, macrophage, and smooth muscle cell content. Br Heart J 1993;69:377–81.
19. Fernández-Ortiz A, Badimon JJ, Falk E, et al. Characterization of the relative thrombogenicity of atherosclerotic plaque components: implications for consequences of plaque rupture. J Am Coll Cardiol 1994;23:1562–9.
20. Barzilai B, Saffitz JE, Miller JG, Sobel BE. Quantitative ultrasonic characterization of the nature of atherosclerotic plaques in human aorta. Circ Res 1987;60:459–63.
21. Okubo M, Kawasaki M, Ishihara Y, et al. Development of integrated backscatter intravascular ultrasound for tissue characterization of coronary plaques. Ultrasound Med Biol 2008;34:655–63.
22. Fontbonne A, Charles MA, Thibult N, et al. Hyperinsulinemia as a predictor of coronary heart disease mortality in a healthy population: the Paris Prospective Study, 15-year follow-up. Diabetologia 1991;34:356–61.
23. Yanase M, Takatsu F, Tagawa T, et al. Insulin resistance and fasting hyperinsulinemia are risk factors for new cardiovascular events in patients with prior coronary artery disease and normal glucose tolerance. Circ J 2004;68:47–52.
24. Lindsey JB, House JA, Kennedy KF, Marso SP. Diabetes duration is associated with increased thin-cap fibroatheroma detected by intravascular ultrasound with virtual histology. Circ Cardiovasc Interv 2009:2:543–8.
25. Nasu K, Tsuchikane E, Katoh O, et al. Plaque characterisation by virtual histology intravascular ultrasound analysis in patients with type 2 diabetes. Heart 2008;94:429–33.
26. Hong YJ, Jeong MH, Choi YH, et al. Plaque characteristics in culprit lesions and inflammatory status in diabetic acute coronary syndrome patients. J Am Coll Cardiol Img 2009;2:339–49.
27. Marso SP, Mehta SK, Frutkin A, et al. Low adiponectin levels are associated with atherogenic dyslipidemia and lipid-rich plaque in nondiabetic coronary arteries. Diabetes Care 2008;31:989–94.
28. Amano T, Matsubara T, Uetani T, et al. Abnormal glucose regulation is associated with lipid-rich coronary plaque: relationship to insulin resistance. J Am Coll Cardiol Img 2008;1:39–45.
29. von Birgelen C, Klinkhart W, Mintz GS, et al. Plaque distribution and vascular remodeling of ruptured and nonruptured coronary plaques in the same vessel: an intravascular ultrasound study in vivo. J Am Coll Cardiol 2001;37:1864–70.
30. Reaven GM. Banting lecture 1988. Role of insulin resistance in human disease. Diabetes 1988;37:1595–607.
31. Bansilal S, Farkouh ME, Fuster V. Role of insulin resistance and hyperglycemia in the development of atherosclerosis. Am J Cardiol 2007;99:6B–14B.
32. Stout RW. The effect of insulin and glucose on sterol synthesis in cultured rat arterial smooth muscle cells. Atherosclerosis 1977;27:271–8.
33. Young IR, Stout RW. Effects of insulin and glucose on the cells of the arterial wall: interaction of insulin with dibutyryl cyclic AMP and low density lipoprotein in arterial cells. Diabete Metab 1987;13:301–6.
34. Krone W, Naegele H, Behnke B, Greten H. Opposite effects of insulin and catecholamines on LDL-receptor activity in human mononuclear leukocytes. Diabetes 1988;37:1386–91.
35. Shamir R, Shehadeh N, Rosenblat M, et al. Oral insulin supplementation attenuates atherosclerosis progression in apolipoprotein E-deficient mice. Arterioscler Thromb Vasc Biol 2003;23:104–10.
36. Dandona P, Aljada A, Mohanty P, et al. Insulin suppresses plasma concentration of vascular endothelial growth factor and matrix metalloproteinase-9. Diabetes Care 2003;26:3310–4.

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