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Coronary Artery Disease

Noninvasive Identification of Patients With Early Coronary Atherosclerosis by Assessment of Digital Reactive Hyperemia

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OBJECTIVES We investigated the value of reactive hyperemia peripheral arterial tonometry (RH-PAT) as a noninvasive tool to identify individuals with coronary microvascular endothelial dysfunction.

BACKGROUND Coronary endothelial dysfunction, a systemic disorder, represents an early stage of atherosclerosis; RH-PAT is a technique to assess peripheral microvascular endothelial function.

METHODS Using RH-PAT, digital pulse volume changes during reactive hyperemia were assessed in 94 patients without obstructive coronary artery disease and either normal (n = 39) or abnormal (n = 55) coronary microvascular endothelial function; RH-PAT index, a measure of reactive hyperemia, was calculated as the ratio of the digital pulse volume during reactive hyperemia divided by that at baseline.

RESULTS Average RH-PAT index was lower in patients with coronary endothelial dysfunction compared with those with normal coronary endothelial function (1.27 ± 0.05 vs. 1.78 ± 0.08: $p < 0.001$). An RH-PAT index <1.35 was found to have a sensitivity of 80% and a specificity of 85% to identify patients with coronary endothelial dysfunction.

CONCLUSIONS Digital hyperemic response, as measured by RH-PAT, is attenuated in patients with coronary microvascular endothelial dysfunction, suggesting a role for RH-PAT as a noninvasive test to identify patients with this disorder. (J Am Coll Cardiol 2004;44:2137–41) © 2004 by the American College of Cardiology Foundation

Endothelial dysfunction represents an early stage of coronary artery disease (CAD) (1). The presence of endothelial dysfunction in coronary or peripheral vessels constitutes an independent predictor of cardiovascular events (2). Given that endothelial dysfunction is reversible, early detection of this disorder may have therapeutic and prognostic implications (2).

Assessment of coronary endothelial function may be considered the "gold standard" of endothelial function testing (3). However, because endothelial dysfunction is not confined to the coronary arteries, less invasive techniques for the assessment of peripheral vascular endothelial function have been developed (4,5). Although these methods are widely used research tools, their operator dependency or complexity preclude their use in clinical practice (2,5). Thus, in order to promote endothelial function testing as a screening method for individuals at increased cardiovascular risk, techniques to easily assess endothelial function are needed.

Reactive hyperemia peripheral arterial tonometry (RH-PAT) is a noninvasive technique to assess peripheral microvascular endothelial function by measuring changes in digital pulse volume during reactive hyperemia (6,7). This study was designed to investigate the relationship between coronary and peripheral microvascular endothelial function and to assess the value of RH-PAT as a tool to identify individuals with coronary endothelial dysfunction.

METHODS

Patients. This study was approved by the Mayo Clinic Institutional Review Board. Ninety-four consecutive patients, who were referred for coronary angiography to exclude CAD and were found to have no significant epicardial coronary stenoses (<30% diameter), were studied prospectively. Exclusion criteria included prior myocardial infarction; percutaneous coronary intervention; coronary artery bypass graft surgery; unstable or variant angina; an ejection fraction ≤50%; valvular heart disease; peripheral vascular disease; uncontrolled arterial hypertension; allergy to latex; and/or significant endocrine, hepatic, renal, or inflammatory disease. Cardiovascular medications were withheld for at least 48 h before cardiac catheterization. Coronary and RH-PAT studies were performed in the fasting state.

Assessment of coronary vasoreactivity. After diagnostic coronary angiography and exclusion of significant CAD, measurements of endothelium-dependent and endothelium-independent coronary flow reserve were performed as previously described (3,8,9). According to previous studies,

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Abbreviations and Acronyms

CAD	= coronary artery disease
CBF	= coronary blood flow
L-NAME	= N-nitro-L-arginine methyl ester
NO	= nitric oxide
PAT	= peripheral arterial tonometry
RH-PAT	= reactive hyperemia peripheral arterial tonometry
ROC	= receiver operating characteristic

normal coronary endothelial function was defined as an increase in coronary blood flow (CBF) of >50% in response to the maximum dose of acetylcholine (8,9).

RH-PAT. The principle of peripheral arterial tonometry (PAT) has been recently described (6,7). Briefly, this system (Itamar Medical Ltd., Caesarea, Israel) comprises a finger probe to assess digital volume changes accompanying pulse waves.

The RH-PAT measurements and cardiac catheterization were performed on the same day; RH-PAT studies were carried out at least 3 h after cardiac catheterization in a thermoneutral environment. According to previous studies (6), a blood pressure cuff was placed on one upper arm (study arm), while the other arm served as a control (control arm). Peripheral arterial tonometry probes were placed on one finger of each hand for continuous recording of the PAT signal. After a 10-min equilibration period, the blood pressure cuff was inflated to suprasystolic pressures for 5 min. Then the cuff was deflated, while PAT recording continued for 10 min (Fig. 1). A total of 19 patients with normal coronary endothelial function and 17 patients with coronary endothelial dysfunction agreed to take 0.4-mg nitroglycerin sublingually to assess endothelium-independent PAT response. In these patients nitroglycerin was given 10 min after cuff deflation, and 10 min later PAT recording was stopped.

The RH-PAT data were analyzed by a computer in an operator-independent manner as previously described (6). As a measure of reactive hyperemia, RH-PAT index was calculated as the ratio of the average amplitude of the PAT signal over a 1-min time interval starting 1 min after cuff deflation divided by the average amplitude of the PAT signal of a 3.5-min time period before cuff inflation (baseline). Subsequently, RH-PAT index values from the study arm were normalized to the control arm. The choice to use the average 1-min PAT signal starting 1 min after cuff deflation to describe the magnitude of reactive hyperemia was based on the observation that this time interval provided the best information regarding detection of coronary endothelial dysfunction as determined by receiver operating characteristic (ROC) curve analysis as well as the best correlation with CBF response to acetylcholine. Hyperemic response to nitroglycerin was similarly assessed; average PAT signal amplitude of four consecutive 1-min periods starting at 5 min after administration of sublingual nitro-

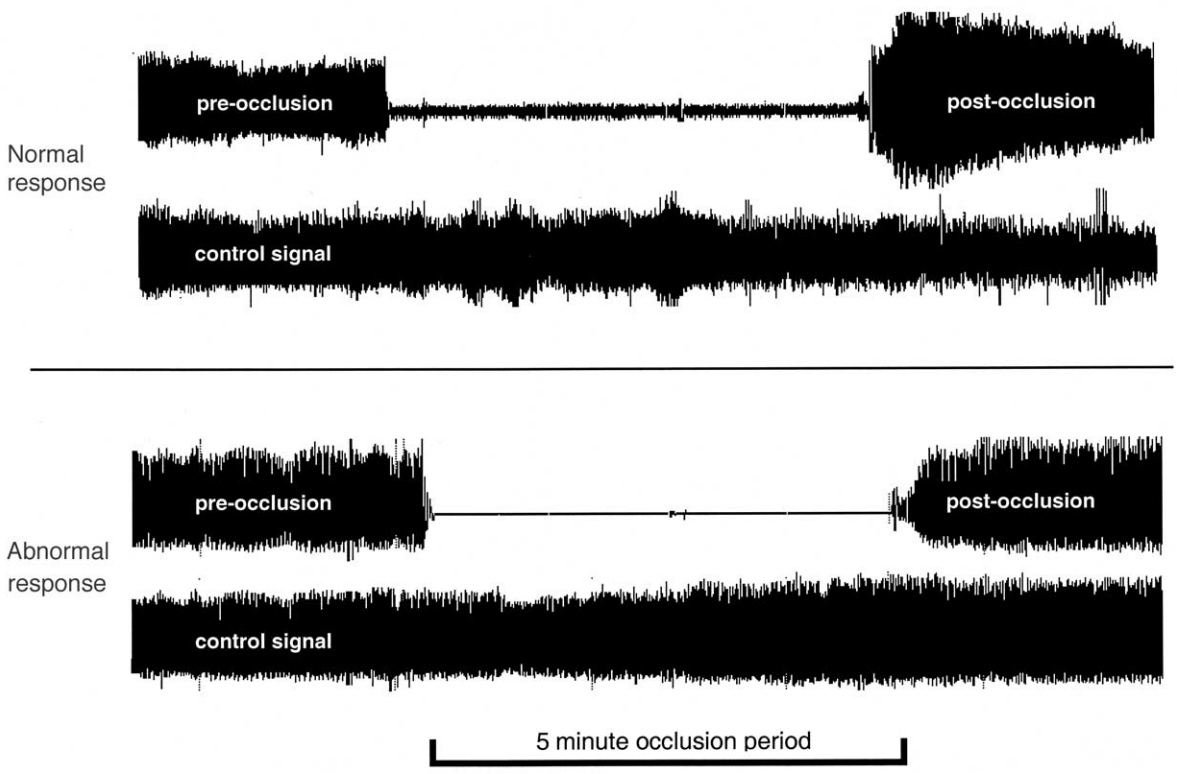

Figure 1. Representative reactive hyperemia peripheral arterial tonometry recordings of subjects with normal and abnormal reactive hyperemic response. Normal response is characterized by a distinct increase in the signal amplitude after cuff release compared with baseline.

glycerin was calculated. Peripheral arterial tonometry response to nitroglycerin was then calculated as the ratio of the PAT amplitude of the 1-min interval during which peak average PAT signal was recorded divided by the amplitude of the baseline PAT signal (nitroglycerin-PAT index).

Determination of reproducibility of RH-PAT measurements was described earlier (6).

Statistical analysis. Results are expressed as mean values ± SEM. Fisher exact test and unpaired *t* test or analysis of variance was used to compare differences between groups. The ROC curve analysis was done to identify the RH-PAT index value for optimal discrimination between presence/absence of coronary endothelial dysfunction. Simple linear regression and multivariable analysis using a backward stepwise regression model were utilized for evaluation of possible associations between RH-PAT index and various clinical variables and cardiovascular risk factors. Multivariable analysis included all variables that were tested in univariable analysis. The ROC curve analysis was done by SPSS statistical software (SPSS Inc., Chicago, Illinois). All other analyses were done by StatView statistical data analysis software (SAS Institute, Cary, North Carolina). Statistical significance was accepted for $p < 0.05$.

RESULTS

A total of 94 patients were studied; 39 had normal coronary endothelial function, and 55 had coronary endothelial dysfunction (Table 1).

Average RH-PAT index was higher in individuals with normal coronary endothelial function than in those with coronary endothelial dysfunction (1.78 ± 0.08 vs. 1.27 ± 0.05; $p < 0.001$).

Linear regression analysis identified a significant relationship between RH-PAT index and CBF response to acetylcholine ($r = 0.405$, $p < 0.001$). In addition, univariable analysis revealed significant relationships between RH-PAT index and body mass index as well as high-density lipoprotein cholesterol levels (Table 2). However, multivariable analysis identified CBF response to acetylcholine as the only independent predictor of RH-PAT index ($p = 0.006$).

By ROC curve analysis, an RH-PAT index of 1.35 was identified as the best discriminating value between individuals with normal and abnormal coronary endothelial function (Fig. 2). For an RH-PAT index value <1.35, the sensitivity and specificity for the detection of coronary endothelial dysfunction were 80% and 85%, respectively. When the patients were divided based on this cutoff value, a significant difference in the average CBF response to acetylcholine was found between patients with an RH-PAT index of ≥1.35 and those with a value of <1.35 (70.0 ± 11.9% vs. 6.5 ± 8.7%; $p < 0.001$). In contrast, there was no difference in the endothelium-independent coronary flow reserve to adenosine between these two groups (3.0 ± 0.1 vs. 3.1 ± 0.1; $p = 0.711$); PAT response to nitroglycerin was similar in patients with normal and abnormal coronary endothelial function (nitroglycerin-PAT index 1.42 ± 0.13 and 1.33 ± 0.13; $p = 0.628$).

Table 1. Clinical Characteristics of Patients With Normal and Abnormal Coronary Endothelial Function

	Normal Coronary Endothelial Function (n = 39)	Abnormal Coronary Endothelial Function (n = 55)
Age (yrs)	50 ± 2	49 ± 2
Male, n (%)	16 (41)	23 (42)
BMI (kg/m^2)	28.2 ± 0.8	29.5 ± 0.9
Hypercholesterolemia, n (%)	19 (49)	31 (56)
Hypertension, n (%)	21 (54)	20 (36)
Diabetes, n (%)	2 (5)	6 (11)
Smoking, n (%)	15 (38)	23 (42)
Family history of CAD, n (%)	30 (77)	41 (75)
ACE inhibitor, n (%)	7 (18)	10 (18)
Beta-blocker, n (%)	13 (33)	17 (31)
Calcium channel blocker, n (%)	19 (49)	23 (42)
Nitrate, n (%)	19 (49)	29 (53)
Lipid-lowering medication, n (%)	11 (28)	22 (40)
Total cholesterol (mmol/l)	4.8 ± 0.2	4.7 ± 0.2
LDL cholesterol (mmol/l)	2.8 ± 0.1	2.8 ± 0.1
HDL cholesterol (mmol/l)	1.4 ± 0.1	1.3 ± 0.1
Triglycerides (mmol/l)	1.4 ± 0.2	1.5 ± 0.2
Fasting blood glucose (mmol/l)	5.5 ± 0.2	5.7 ± 0.2
Heart rate (beats/min)	69 ± 2	67 ± 1
Systolic blood pressure (mm Hg)	128 ± 3	126 ± 2
Diastolic blood pressure (mm Hg)	75 ± 2	73 ± 1
Pulse pressure (mm Hg)	53 ± 3	53 ± 2
LVEF (%)	64 ± 1	63 ± 1
ΔCBF (Ach), %	107.3 ± 8.8	−12.9 ± 6.2*
ΔCAD (Ach), %	0.7 ± 1.9	−27.6 ± 4.3*
CFR	2.9 ± 0.1	3.1 ± 0.1
ΔCBF (NTG), %	22.7 ± 10.4	15.4 ± 8.9
ΔCAD (NTG), %	13.3 ± 2.7	14.8 ± 2.4

Values are mean ± SEM or n (%). *$p < 0.001$ vs. normal coronary endothelial function group.

ACE = angiotensin-converting enzyme; BMI = body mass index; CFR = endothelium-independent coronary flow reserve to adenosine; HDL = high-density lipoprotein; LDL = low-density lipoprotein; LVEF = left ventricular ejection fraction; ΔCAD (Ach) = change in coronary artery diameter in response to acetylcholine; ΔCAD (NTG) = change in coronary artery diameter in response to nitroglycerin; ΔCBF (Ach) = change in coronary blood flow in response to acetylcholine; ΔCBF (NTG) = change in coronary blood flow in response to nitroglycerin.

DISCUSSION

This study demonstrates that patients with coronary microvascular endothelial dysfunction have a lower peripheral hyperemic response, as measured by RH-PAT, than those with normal coronary endothelial function, suggesting a potential role for RH-PAT as a noninvasive test to identify individuals with coronary endothelial dysfunction.

Reactive hyperemia peripheral arterial tonometry represents a noninvasive technique for measuring digital reactive hyperemia, which is partly mediated by endothelium-derived nitric oxide (NO) (10). Thus, the magnitude of reactive hyperemia may serve as an index of peripheral microvascular endothelial function. Indeed, an excellent correlation between forearm blood flow response to reactive

Table 2. Univariate Analysis of Potential Clinical Predictors of the RH-PAT Index

Variables	r	p Value
ΔCBF (Ach)	0.405	<0.001
Age	0.129	0.215
BMI	−0.238	0.021
Height	0.091	0.379
Weight	−0.126	0.224
Heart rate	−0.128	0.217
Systolic blood pressure	0.105	0.312
Diastolic blood pressure	0.179	0.083
Pulse pressure	0.048	0.645
Total cholesterol	0.212	0.840
LDL cholesterol	−0.007	0.951
HDL cholesterol	0.255	0.014
Triglycerides	−0.055	0.600
Family history of CAD	0.040	0.703
Diabetes	0.032	0.761
Smoking	0.040	0.701

RH-PAT = reactive hyperemia peripheral arterial tonometry; other abbreviations as in Table 1.

hyperemia and that to intra-arterial infusion of acetylcholine was demonstrated (11).

Reactive hyperemia peripheral arterial tonometry measures digital pulse volume at rest and during reactive hyperemia. Although digital pulse volume is modulated by various local, systemic, and environmental factors, this parameter is also affected by the bioavailability of NO and, therefore, also depends on endothelial function (12). The role of endothelium-derived NO in the RH-PAT response was investigated in a preliminary study, in which RH-PAT testing was performed before and during brachial artery infusion of N-nitro-L-arginine methyl ester (L-NAME), an inhibitor of NO synthesis, in healthy volunteers (7). In this study, L-NAME reduced RH-PAT index significantly by 61%. Taken together, measuring reactive hyperemia by RH-PAT provides a noninvasive means for assessing peripheral microvascular endothelial function.

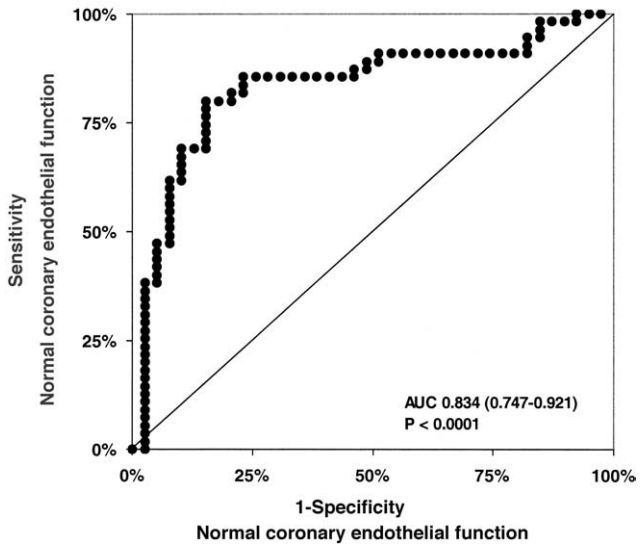

Figure 2. Receiver operating characteristic curve for the reactive hyperemia peripheral arterial tonometry (RH-PAT) index to identify patients with normal coronary endothelial function. An RH-PAT index of 1.35 discriminates best between presence/absence of coronary endothelial dysfunction. AUC = area under the curve.

Average RH-PAT index was significantly lower in individuals with coronary endothelial dysfunction. Moreover, and similar to a study by Anderson et al. (13), we found a significant correlation between RH-PAT index and the CBF response to acetylcholine. This moderate correlation may be secondary to the differential response of vascular beds to different stimuli. Therefore, defining a cutoff value may represent a more accurate method to compare endothelial function between peripheral and coronary vessels. Indeed, using ROC curve analysis, we found a sensitivity of 80% and a specificity of 85% for an RH-PAT index <1.35 to identify patients with coronary endothelial dysfunction.

The similar PAT response to nitroglycerin in individuals with normal and abnormal coronary endothelial function supports the concept that the RH-PAT index represents a measure of endothelial function. This is underscored by the similar endothelium-independent coronary flow reserve in both groups when patients were divided based on an RH-PAT index of 1.35.

To minimize the impact of confounding factors on the RH-PAT results, a two-pronged approach was used. First, the reactive hyperemic response was referenced to a baseline derived from the same finger in order to eliminate local finger-related effects. Second, the effect of systemic factors was minimized by normalizing the RH-PAT value of the study arm to the corresponding PAT signal of the control arm. Other factors affecting peripheral vascular tone, like temperature, were less of a concern in our study because environmental conditions during testing were kept equal for all patients.

The present study has several limitations. Only patients with chest pain undergoing cardiac catheterization were included. Similar to a previous study (8), distribution of traditional risk factors was similar among patients with normal and abnormal coronary endothelial function. This may explain the absence of a significant relationship between traditional risk factors and RH-PAT index and may also suggests the possibility of a selection bias that may limit translation of the results to a general population. Another potential limitation pertains to the definition of coronary endothelial dysfunction used. The role of coronary endothelial dysfunction as an independent risk factor for cardiovascular events is well established (2). Thus, our definition of coronary endothelial dysfunction was based on previous studies demonstrating the adverse prognostic impact of an increase in CBF to acetylcholine of <50% (8,9). Finally, our results cannot be transferred to patients with heart failure or autonomous nervous system dysfunction who may show alterations of the peripheral circulation. Given these limitations, our results require independent confirmation and further validation in different populations.

In summary, our study demonstrates that digital reactive hyperemia, as measured by RH-PAT, is attenuated in

patients with coronary endothelial dysfunction compared with individuals with normal coronary endothelial function. This suggests a role for RH-PAT as a noninvasive tool to identify patients during the early stage of CAD.

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