Endo-PAT2000
Assessing Endothelial Function: Overview

Itamar Medical
The Test

Endo-PAT tests can be carried out in both the office and hospital settings, with patients positioned either sitting or supine. Endo-PAT bio-sensors are placed on the index fingers of both arms. The test takes 15 minutes to complete, is very easy to perform, and is both operator and interpreter independent. Thermo-neutral, quiet surroundings are recommended.

Endo-PAT quantifies the endothelium-mediated changes in vascular tone, elicited by a 5-minute occlusion of the brachial artery (using a standard blood pressure cuff). When the cuff is released, the surge of blood flow causes an endothelium-dependent Flow Mediated Dilatation (FMD). The dilatation, manifested as Reactive Hyperemia, is captured by Endo-PAT as an increase in the PAT Signal amplitude. A post-occlusion to pre-occlusion ratio is calculated by the Endo-PAT software, providing the EndoScore.

Automatic Analysis

Endo-PAT software is an integral part of the Endo-PAT system. It is straight-forward and easy to use. The software is used for both on-line data acquisition as well as off-line data analysis.

The online display allows real-time viewing of events as they occur. The signals are recorded on the computer for subsequent review and automatic analysis. Since analysis is performed by the software, inter- or intra-operator interpretation variability is avoided. Analyzed test results can be exported to an Excel spreadsheet that includes multiple study parameters, calculated variables, and measures of signal quality.

PAT Technology

Peripheral Arterial Tone (PAT) signal is a proprietary technology used for non-invasively measuring arterial tone changes in peripheral arterial beds. The PAT Signal is measured from the fingertip by recording finger arterial pulsatile volume changes. Based on PAT Technology, the noninvasive Endo-PAT2000 system comprises a measurement apparatus that supports a pair of modified plethysmographic bio-sensors. The unique feature of the PAT bio-sensors is that they impart a uniform sub-diastolic pressure field to the distal two thirds of the fingers including their tips. Applying the pressure in this way is extremely important as it:

- Prevents distal venous blood pooling, that can induce a veno-arteriolar vasoconstriction reflex
- Unloads arterial wall tension, which generates a greater dynamic range of the measured PAT Signal
- Fixates the PAT bio-sensor to the finger, which reduces movement artifacts
REFERENCES

Medical Professionals with extensive experience in use and application of EndoPAT 2000:

These physicians are available for conversation regarding technology, use, application, results and treatment options. Please contact your Arterial Health representative to schedule a convenient time to speak with these individuals.

*Dr. Perry Krichmar*
Partner Elite Health in Pembroke Pines, FL
Board Certified Internal Medicine – practicing cardiologist
More than twenty-six years in practice

*Dr. Steven Lamm*
Faculty member New York University School of Medicine
Internal Medicine – Manhattan, NY
Published author, well-known speaker and media guest
Internist with tremendous experience with EndoPAT technology

Other well-known cardiologists who use the device (EndoPAT 2000) but are not generally available for reference phone calls:

*Dr. Arthur Agatston*
Board Certified – Cardiology
Southeast Preventive Cardiology, Miami Beach, FL
Baptist Health Miami, FL
Author of the Agatston Scale to gauge coronary heart disease (EBT screening)
Creator of the South Beach Diet Plan, and accompanying series of books

*Dr. Mark Houston, MS, ABAARM, FACP, FAHA, FASH*
Board Certified Internal Medicine
Saint Thomas Medical Group Nashville, TN
Vascular Institute of Saint Thomas Hospital, Nashville, TN
Vanderbilt University School of Medicine - Associate Professor
Hypertension Institute, Nashville, TN, - Director
Published author - “What Your Doctor May Not Be Telling You about Heart Disease” February 2012
Numerous lectures and published papers on CVD

*Dr. John Cooke, MD, PhD*
Stanford University - Professor of Cardiovascular Medicine
Stanford Cardiovascular Institute - Associate Director for Education and Training
Board Certified – Internal Medicine (1984)/Cardiovascular Disease (1986)
Prominent research professional targeting cardiovascular disease
Key Points on Endothelial Function and EndoPAT 2000

*What is endothelial dysfunction?*

The pathological state known as endothelial dysfunction is the earliest clinically detectable stage of cardiovascular disease (which includes heart attacks, stroke, Peripheral Arterial Disease and many other diseases). The functioning of the endothelial cells – endothelial function – is normally kept in balance. Atherosclerosis risk factors such as high cholesterol, high blood sugar, high blood pressure, smoking, aging, obesity, chronic infection, and inflammation, can all disrupt this balance and lead to endothelial dysfunction.

Endothelial dysfunction can be defined as reduced bio-availability of Nitric Oxide (NO) which plays many roles in maintaining vascular health, most importantly its role in vasomotion. Hence, endothelial dysfunction is defined as an impairment of endothelium dependent vasodilation. In their 2005 Circulation publication, Lerman et al. (1) defined endothelial dysfunction as “ultimate risk of the risk factors” a summation of the integrated affects of cardiovascular risk factor.


*What are the consequences of endothelial dysfunction?*

The main consequence of endothelial dysfunction is the initiation of an inflammatory process which leads to the formation of atherosclerosis and its late sequel, cardiovascular morbidity and mortality. Endothelial dysfunction is involved in numerous systemic disease processes such as: erectile dysfunction, metabolic syndrome, cerebrovascular diseases (stroke/TIA), pre-eclampsia toxemia, renal failure, sleep apnea, claudication, and gangrene.

*How does the EndoPAT™ measure endothelial function?*

EndoPAT™ quantifies the endothelium-mediated changes in vascular tone, elicited by a 5-minute occlusion of the brachial artery (using a standard blood pressure cuff). When the cuff is released, the surge of blood flow causes an endothelium-dependent. Flow Mediated Dilatation (FMD). The dilatation, manifested as Reactive Hyperemia, is captured by EndoPAT™ as an increase in the PAT Signal amplitude. A post-occlusion to pre-occlusion ratio is calculated by the EndoPAT™ software, providing the EndoPAT™ index.
What is the RHI and how it is calculated?

RHI stands for Reactive Hyperemia Index. This is the final concluded result of the EndoPAT™ test. It is a ratio of the post-to-pre occlusion PAT amplitude of the tested arm, divided by the post-to-pre occlusion ratio of the control arm.

\[
RHI = \frac{A}{B} \times \frac{C}{D}
\]

A - Mean PAT amplitude between 90s-150s post occlusion of the occluded arm
B - Mean PAT amplitude from the baseline period of the occluded arm
C - Mean PAT amplitude between 90s-150s post occlusion of the control arm
D - Mean PAT amplitude from the baseline period of the control arm

Can the EndoPAT help predict endothelial dysfunction and cardiovascular events?

Results of a 2009 study conducted by researchers at the Mayo Clinic and Tufts reported that the EndoPAT test is “highly predictive” of a major cardiac event, such as a heart attack or stroke, for people who are considered at low or moderate risk based on their Framingham Risk Score (FRS).

The FRS is the commonly used risk predictor and was developed from the Framingham Heart Study, an ongoing longitudinal study of heart disease.

In this Mayo Clinic study, published in the Journal of the American College of Cardiology, Amir Lerman, M.D., a cardiologist at Mayo Clinic and senior author of the study, and other researchers, used an EndoPAT to measure the endothelial health of 270 patients between the ages of 42 and 66 and followed their progress between 1999 and 2007.

These patients already knew that they had low-to-medium risk of experiencing a major adverse cardiac event, or MACE, based on their FRS. Some of their risk factors included high blood pressure, high cholesterol, obesity, and a family history of heart disease.

The study results: The rate of MACE in patients who tested positive for endothelial dysfunction was 39% vs. normal endothelial function 25% (p=0.024). The study showed that patients at low FRS risk but with endothelial dysfunction were at a higher actual risk of future CV events than patients with high FRS but normal endothelial function.
Furthermore, endothelial dysfunction was found to be an independent risk factor for future MACE on multivariate analysis \((p=0.002)\).


**Is the EndoPAT test used in cardiac-related research studies?**

The EndoPAT has been used in dozens of clinical studies conducted at such renowned medical institutions as The Mayo Clinic, Johns Hopkins Medicine, Harvard Medical School, Cleveland Clinic, the Framingham Heart Study, and Mount Sinai Hospital (NY).

**How well does the EndoPAT correlate with conventional cardiovascular risk factors (Framingham Risk Score)?**

Since 2003, the Framingham Heart Study has included endothelial function measurements with EndoPAT. All three study cohorts (the original study population, the offspring, and the third generation cohort) have been tested with EndoPAT, totaling over 5,000 subjects.

Hamburg et al published a cross sectional analysis of 1,957 third generation subjects in 2008*. The study demonstrated a significant inverse relation between EndoPAT index and multiple CV risk factors, including: male sex, body mass index, total/HDL cholesterol, diabetes, smoking, and lipid-lowering treatment.


**Is there a threshold for a good EndoPAT™ result?**

Yes, the threshold for a good EndoPAT™ result is an RHI of 1.67 and above. The threshold of 1.67 was determined following the study of Bonetti et al(6) which was performed at the Mayo Clinic. In this study the EndoPAT™ was found to be correlated with the coronary endothelial function using the gold standard method of assessment which is injection of ACH during coronary catheterization.

**What are treatment options for endothelium dysfunction?**

General treatment options for endothelium dysfunction may include proper diet and exercise, and reducing or eliminating smoking and excessive alcohol consumption. Several pharmacological choices include statins, angiotensin II receptor blockers (ARB), angiotensin-converting enzymes (ACE) and β1 receptor blocker (Nebivolol) from various manufacturers. (Please refer to pharmaceutical company and FDA guidelines when choosing treatment options for your patient)

Homeopathic remedies may include L-Arginine and Niacin. Treatment strategies must take into consideration the overall medical profile of each patient, with due caution exercised as to dosage amounts and time periods for consumption.

**How can EndoPAT, which measures changes in vascular function in a fingertip, ensure that the endothelium of the entire vascular system has been checked?**

The endothelium is the same throughout the body, and when damage is noted in the fingertip, it indicates that the endothelium is damaged throughout the body—that it’s a systemic dysfunction. Endothelial dysfunction is involved in numerous systemic disease processes, including: erectile dysfunction, metabolic syndrome, renal failure, sleep apnea, and stroke.

In a study performed by Bonetti et al* at the Mayo Clinic, a group of 94 subjects underwent angiographic assessment of coronary endothelial function and subsequent EndoPAT tests. Coronary endothelial function is quantified by injection of acetylcholine during angiography.

The EndoPAT, which is a non-invasive test, was found to be highly correlative to the angiography results; it was this study that helped EndoPAT receive final FDA clearance for the detection of coronary endothelial dysfunction. An EndoScore of 1.67 provides a sensitivity of 82% and a specificity of 77% and AUC 0.82 for diagnosing coronary endothelial function.


**How reproducible are EndoPAT results?**

The EndoPAT test is both operator and interpreter independent.

Selamet et al* tested prospectively the reproducibility and feasibility of the EndoPAT. EndoPAT tests were performed on two different days separated by no more than seven days in 30 healthy fasting adolescents, ages 13 to 19 years. The authors concluded, “In healthy adolescents, Endo-PAT is feasible and has excellent reproducibility.” Moreover, the authors stated, “This technology may provide an easy and reliable means of assessing endothelial function in the pediatric population.”

What is the association between the EndoPAT™ and BAUS (Brachial Artery Ultrasound)?

BAUS is a common research method for peripheral, noninvasive assessment of endothelial function. It differs from EndoPAT™ in several ways. While the BAUS assesses a single conduit vessel, EndoPAT™ measures several vascular beds, composed of small vessels and microcirculation. Furthermore, EndoPAT™ corrects for systemic changes by a simultaneous measurement from the (un-occluded) contra-lateral arm. With minimal training necessary, EndoPAT™ is practically operator independent, while BAUS requires a trained ultrasound technician and is highly user-dependent in both data acquisition and analysis. Furthermore, the response measured with EndoPAT™ has a much larger dynamic range (hundreds of %) than the miniscule changes assessed by BAUS (around 10% for a normal response). Several studies have simultaneously measured Flow-Mediated Dilatation (FMD) with EndoPAT™ and BAUS. Kuvin et al.(8) at the Tufts Medical Center, Boston, demonstrated a significant correlation between the two methods (r=0.55, p<0.0001) in a group of 89 adult patients suffering from chest pain.


Is the EndoPAT correlative to coronary endothelial function?

EndoPAT provides high degrees of sensitivity and specificity when compared to the assessment of coronary artery endothelial function. Coronary endothelial function is quantified by measuring arterial diameter change and blood flow in response to graded intra-coronary infusion of acetylcholine during angiography. In a study performed by Bonetti et al* at the Mayo Clinic, a group of 94 subjects underwent angiographic assessment of coronary endothelial function and subsequent EndoPAT tests. The results of this comparative study served as the basis for the FDA clearance of the EndoPAT in the detection of coronary endothelial dysfunction. An EndoPAT index cutoff value of 1.67 provides a sensitivity of 82% and a specificity of 77% and AUC 0.82 to diagnosing coronary endothelial function.


What is the association between the EndoPAT and NO bioavailability?

Nohria and Gerhard et al* at the Brigham & Women’s Hospital, Boston, demonstrated the central role for nitric oxide (NO) in the post-occlusion vasodilatory response measured by EndoPAT. EndoPAT index (EndoScore) was measured in a group of nineteen healthy volunteers, before and after intra-arterial infusion of L-NAME (a specific inhibitor of endothelial nitric oxide synthase). Fifteen matched controls were infused with saline or phenylephrine (an endothelium independent vasoconstrictor). The study reported that L-NAME blocked 46% of the vasodilatory response (p=0.002). These results provide direct confirmation that EndoPAT indeed measures a NO-mediated endothelial response.

**Why does the EndoPAT test require using both arms?**

Think of the EndoPAT evaluation as a one-person clinical study with you comparing yourself to yourself. While endothelial function is being tested in one arm with the blood pressure cuff and finger monitor, your other arm is being used to monitor changes in blood flow that generally affects both arms. By then measuring both arms, EndoPAT automatically corrects for any systemic changes that may occur during the course of the test and calculates a final EndoScore based on information gathered from both finger monitors.

**Which arm should be used for occlusion/control?**

The non-dominant arm is recommended as the tested (occluded) arm due to a lower mass of muscle and leads to easier arterial occlusion.

**Does it matter which fingers are used for the test?**

Any finger but the thumb may be used. Placement of the sensors on the index fingers is recommended. Due to variance between fingers in blood supply, symmetrically-paired fingers on both arms should be used (i.e. either both index fingers or both middle fingers). The thumb should be avoided.

**Why does the occlusion last 5 minutes?**

Five minutes is the optimal time needed to generate the forced stimulus response of the blood rushing through the vessels in response to an occlusion. Faizi et al. (4) tested the effects of varying occlusion durations as well as the effects of occlusion location in 30 apparently healthy adult volunteers. When comparing different occlusion times (1.5, 3, 5 and 8 minutes) with the cuff placed on the forearm, they saw that the effective maximal response was reached at 5 minutes. The occlusions shorter than 5 minutes had significantly lower responses. The response to a 5 minute occlusion did not differ significantly from 8 minutes, but caused less discomfort.
Does having the blood pressure cuff inflated on the upper arm for five minutes cause any discomfort?

The five-minute blood pressure cuff inflation is an accepted standard test to cause reactive hyperemia (the increase of blood flow after a temporary restriction in blood supply) for the assessment of endothelial function. While the occlusion may cause some minor discomfort and tingling in the fingers, the test is absolutely harmless.

Where should we place the blood pressure cuff for the occlusion?

Optimal location is on the upper arm. Faizi et al. (5) tested twenty individuals with the cuff placed on their upper arm, occluding the brachial artery for 5 minutes. These results were compared to their 5 minute forearm occlusion test, showing an EndoPAT™ index of 1.88 (±0.55) for the forearm occlusion and 2.07 (±0.69) for the upper-arm occlusion (p=0.097). Forearm occlusion was reported to cause less discomfort than the upper arm.


What are the clinical setting requirements for the EndoPAT™ test?

The EndoPAT™ is compact and can be used in almost any clinical setting. Since the PAT signal reflects autonomic nervous system activity, it is recommended to create a stress free environment during the EndoPAT™ test. The room selected for the study should be in a quiet area, thermo-neutral room temperature should be maintain 21°C-24°C (70°F-75°F) and dimmed lights are preferred when performing the study. The test can be performed on a comfortable bed, exam table or armchair which will enable placing both hands at heart level in a rested position.

What is the PAT signal?

The PAT (Peripheral Arterial Tone) signal is a proprietary technology used for non-invasively measuring arterial tone changes in peripheral arterial beds. The PAT signal used in the EndoPAT is measured from the fingertip by recording finger arterial pulse volume changes. Results of the 15-minute test are automatically calculated and an EndoScore is generated, which indicates the present state of endothelial health.

EndoPAT is the only FDA-cleared device indicated for assessment of Endothelial Function. Clearance was obtained by demonstrating equivalence of PAT Technology with an invasive catheterization procedure that directly assessed endothelial dysfunction in coronary arteries. The clinical research was conducted at the Mayo Clinic.
Can the test be performed on children?

We rely on the medical judgment and expertise of the physician regarding the precise patient population. Although most of the testing on EndoPAT has been done on adults, there are many ongoing and published studies with the EndoPAT on children and adolescents. There is no special design of the biosensors for children, however. The length of the EndoPAT biosensor is 5cm, thus the length of the finger of the child must be at least as the length of the biosensor.

Selamet et al* tested prospectively the reproducibility and feasibility of the EndoPAT in adolescents. EndoPAT tests were performed on two different days separated by no more than seven days in 30 healthy fasting adolescents, ages 13 to 19 years. The authors concluded, “In healthy adolescents, Endo-PAT is feasible and has excellent reproducibility.” Moreover, the authors stated, “This technology may provide an easy and reliable means of assessing endothelial function in the pediatric population.”

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- Graduate of Vanderbilt Medical School
- Associate Clinical Professor at Vanderbilt Medical School
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- Staff physician at Saint Thomas Medical Group
- Saint Thomas Hospital, Nashville, TN
- Has presented over 10,000 lectures on hypertension
- More than 150 articles and scientific abstracts in peer-reviewed journals, textbooks, handbooks and films

Key points from a revolutionary book

P. 4

‘OLD TRUTHS’ rebutted:

1. Abnormal cholesterol levels are **not** primary causes or indicators of coronary heart disease
2. Consuming a high-cholesterol diet does **not** significantly raise blood cholesterol levels for most people
3. All LDL “bad cholesterol” is **not** harmful and does not necessarily cause coronary heart disease
4. All HDL “good” cholesterol is **not** protective - some types may promote coronary heart disease
5. Blood pressure readings taken in office may not be an accurate measurement of true blood pressure
6. A fasting morning blood sugar reading of 99 mg/dL, is **not** safe or normal
7. A normal body weight..does **not** ensure heart health as it doesn’t reflect the amount of risky visceral fat
‘NEW TRUTHS’ revealed:

P. 6

Coronary heart disease doesn’t come from cholesterol; it is the result of inflammation, oxidative stress (free radical damage) and autoimmune damage to coronary arteries and other arteries throughout the body.

These are the elements you should be attempting to control.

P. 18

For years we believed that heart disease began with atherosclerosis...That idea is now obsolete. *Today we understand that heart disease begins with an injury to the endothelium.* (Italics added)

Tests to detect endothelial dysfunction:

- **Carotid Duplex Scans**
- **Carotid Intima-Media Thickness**
- **Ankle-Brachial Index**
- **EndoPAT** (measuring blood flow via Peripheral Arterial Tonometry)
Noninvasive Identification of Patients With Early Coronary Atherosclerosis by Assessment of Digital Reactive Hyperemia

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Rochester, Minnesota; and Boston, Massachusetts

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,
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).

**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

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

<table>
<thead>
<tr>
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<th>Normal Coronary Endothelial Function (n = 39)</th>
<th>Abnormal Coronary Endothelial Function (n = 55)</th>
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<tbody>
<tr>
<td>Age (yrs)</td>
<td>50 ± 2</td>
<td>49 ± 2</td>
</tr>
<tr>
<td>Male, n (%)</td>
<td>16 (41)</td>
<td>23 (42)</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>28.2 ± 0.8</td>
<td>29.5 ± 0.9</td>
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<tr>
<td>Hypercholesterolemia, n (%)</td>
<td>19 (49)</td>
<td>31 (56)</td>
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<td>Hypertension, n (%)</td>
<td>21 (54)</td>
<td>20 (36)</td>
</tr>
<tr>
<td>Diabetes, n (%)</td>
<td>2 (5)</td>
<td>6 (11)</td>
</tr>
<tr>
<td>Smoking, n (%)</td>
<td>15 (38)</td>
<td>23 (42)</td>
</tr>
<tr>
<td>Family history of CAD, n (%)</td>
<td>30 (77)</td>
<td>41 (75)</td>
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<tr>
<td>ACE inhibitor, n (%)</td>
<td>7 (18)</td>
<td>10 (18)</td>
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<tr>
<td>Beta-blocker, n (%)</td>
<td>13 (33)</td>
<td>17 (31)</td>
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<tr>
<td>Calcium channel blocker, n (%)</td>
<td>19 (49)</td>
<td>23 (42)</td>
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<td>Nitrate, n (%)</td>
<td>19 (49)</td>
<td>29 (53)</td>
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<tr>
<td>Lipid-lowering medication, n (%)</td>
<td>11 (28)</td>
<td>22 (40)</td>
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<tr>
<td>Total cholesterol (mmol/l)</td>
<td>4.8 ± 0.2</td>
<td>4.7 ± 0.2</td>
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<tr>
<td>LDL cholesterol (mmol/l)</td>
<td>2.8 ± 0.1</td>
<td>2.8 ± 0.1</td>
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<tr>
<td>HDL cholesterol (mmol/l)</td>
<td>1.4 ± 0.1</td>
<td>1.3 ± 0.1</td>
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<td>Triglycerides (mmol/l)</td>
<td>1.4 ± 0.2</td>
<td>1.5 ± 0.2</td>
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<td>Fasting blood glucose (mmol/l)</td>
<td>5.5 ± 0.2</td>
<td>5.7 ± 0.2</td>
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<tr>
<td>Heart rate (beats/min)</td>
<td>69 ± 2</td>
<td>67 ± 1</td>
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<td>Systolic blood pressure (mm Hg)</td>
<td>128 ± 3</td>
<td>126 ± 2</td>
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<td>Diastolic blood pressure (mm Hg)</td>
<td>75 ± 2</td>
<td>73 ± 1</td>
</tr>
<tr>
<td>Pulse pressure (mm Hg)</td>
<td>53 ± 3</td>
<td>53 ± 2</td>
</tr>
<tr>
<td>LVEF (%)</td>
<td>64 ± 1</td>
<td>63 ± 1</td>
</tr>
<tr>
<td>ΔCBF (Ach), %</td>
<td>107.3 ± 8.8</td>
<td>-12.9 ± 6.2*</td>
</tr>
<tr>
<td>ΔCAD (Ach), %</td>
<td>0.7 ± 1.9</td>
<td>-27.6 ± 4.3*</td>
</tr>
<tr>
<td>CFR</td>
<td>2.9 ± 0.1</td>
<td>3.1 ± 0.1</td>
</tr>
<tr>
<td>ΔCBF (NTG), %</td>
<td>22.7 ± 10.4</td>
<td>15.4 ± 8.9</td>
</tr>
<tr>
<td>ΔCAD (NTG), %</td>
<td>13.3 ± 2.7</td>
<td>14.8 ± 2.4</td>
</tr>
</tbody>
</table>

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

ΔACE = 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.
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.

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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REFERENCES

11. Higashi Y, Sasaki S, Nakagawa K, Matsuura H, Kajiyama G, Oshima T. A noninvasive measurement of reactive hyperemia that can be used to assess resistance artery endothelial function in humans. Am J Cardiol 2001;87:121–5.
Assessment of endothelial function by non-invasive peripheral arterial tonometry predicts late cardiovascular adverse events

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Aims
There is growing need for the identification of novel non-invasive methodologies for the identification of individuals at risk for adverse cardiovascular (CV) events. We examined whether endothelial dysfunction, as detected by non-invasive peripheral arterial tonometry (EndoPAT), can predict late CV events.

Methods
Reactive hyperaemia (RH) was induced following upper arm occlusion of systolic blood pressure in 270 outpatients (54±12 years, 48% female). The natural logarithmic scaled RH index (L_RHI) was calculated from the ratio between the digital pulse volume during RH and at baseline. The patients were followed for CV adverse events (AE: cardiac death, myocardial infarction, revascularization or cardiac hospitalization) during a 7-year follow-up (inter-quartile range = 4.4–8). Cox models were used to estimate the association of EndoPAT results with AE adjusted for age.

During the follow-up, AE occurred in 86 patients (31%). Seven-year AE rate was 48% in patients with L_RHI < 0.4 vs. 28% in those with L_RHI ≥ 0.4 (P = 0.03). Additional univariate predictors of AE were advancing age (P = 0.02) and prior coronary bypass surgery (P = 0.01). The traditional Framingham risk score was not higher in patients with AE. Multivariate analysis identified L_RHI < 0.4 as an independent predictor of AE (P = 0.03).

Conclusion
A low RH signal detected by EndoPAT, consistent with endothelial dysfunction, was associated with higher AE rate during follow-up. L_RHI was an independent predictor of AE. Non-invasive assessment of peripheral vascular function may be useful for the identification of patients at risk for cardiac AEs.

Keywords
Endothelial function • Outcome • Peripheral arterial tonometry • Reactive hyperaemia

Background
Coronary heart disease is the leading cause of morbidity and mortality in most industrialized societies.1 In spite of comprehensive treatment and modification of conventional risk factors, there is still high incidence of cardiovascular (CV) events rate.2 Thus, there is a need to identify a more individualized functional risk profile in order to personalize treatment.3

It has been suggested that cardiac risk factors can cause impairment of coronary vasomotor function of both the epicardial arteries and the microcirculation,4–8 which is considered an important phase in atherogenesis.9–11 It has been demonstrated that coronary endothelial dysfunction in humans may be associated with myocardial ischaemia.12,13

Coronary endothelial dysfunction is considered an early stage of atherosclerosis14 and has been shown to be associated with an increased risk of ischaemic CV outcome events and stroke.15–18 Assessment of coronary microcirculatory vasomotor function (especially in patients without obstructive coronary artery disease) may therefore allow the identification of patients in the early stages of coronary atherosclerosis and at risk for CV events.

However, one of the main obstacles in using peripheral endothelial function for individualized assessment of CV risk is the lack of standardization of these tests.3,19

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Measurement of peripheral vasodilator response as a measure for endothelial dysfunction is correlated with adverse outcome events and measurements of this response using fingertip pulse amplitude tonometry (peripheral arterial tonometry-PAT) may emerge as a useful method for non-invasive assessment of vascular health. Impairment of pulse amplitude hyperemic response is associated with the presence of coronary artery endothelial dysfunction, and we have previously reported the correlation between abnormal PAT results and coronary microvascular endothelial dysfunction as detected by invasive evaluation of the coronary endothelial function. Reactive hyperaemia (RH) response (with PAT) as detected by the RH index (RHI) has recently been shown to be related to multiple traditional and metabolic risk factors. However, it is unknown whether this non-invasive approach can predict adverse CV outcome events beyond the traditional risk assessment.

The purpose of this study was to examine whether endothelial dysfunction, as detected by non-invasive PAT, may have a role in predicting late adverse CV events.

Methods

Patient selection

Between August 1999 and August 2007, 329 symptomatic outpatients (with unexplained chest pain (low-risk findings during stress testing) and/or the absence of new obstructive lesions by an invasive coronary angiogram) underwent evaluation of endothelial function using RH with non-invasive PAT at the centres for coronary physiology at Mayo Clinic in Rochester, MN, and in Tufts Medical Center in Boston, MA. Five patients were excluded from the current investigation on the basis of poor quality signal (n = 3) or lack of research authorization as required by Minnesota law (n = 2). In 54 patients (17%), the follow-up and phone interview could not be completed. The final study group consisted of 270 patients.

Reactive hyperaemia by peripheral arterial tonometry

All vasoactive medications were discontinued at least 24 h prior to testing. Peripheral arterial tonometry signals were obtained using the EndoPAT 2000 device (Itamar Medical Inc., Caesarea, Israel), which has been validated and used previously to assess peripheral arterial tone in other populations. Specially designed finger probes were placed on the middle finger of each subject's hand. These probes comprised a system of inflatable latex air cuffs connected by pneumatic tubes to an inflating device controlled through a computer algorithm. A constant counter pressure (pre-determined by baseline signal and indexed to the contra lateral arm) was applied through the air cushions. This algorithm automatically normalizing for baseline signal and indexed to the contra lateral arm. The calculated ratio reflects the RHI.

The natural logarithmic scaled RHI (L_RHI) was calculated from the ratio of the PAT signal after cuff release compared with baseline.

Assessment of clinical status and outcome events

Vital status was determined by review of medical records, social security death index, and reviews of death certificates supplemented by a phone interview to determine patients’ clinical status and occurrence of CV outcome events since PAT examination date. Clinical data were determined by an investigator blinded to PAT data. The following clinical parameters were assessed.

Baseline characteristics

Baseline characteristics including risk factors status during the week in which PAT was performed: (i) Hypertension (blood pressure of >140/90 or treatment with anti-hypertensive medications), (ii) diabetes mellitus (patient history and/or treatment with insulin or oral hypoglycaemic agents), (iii) family history of coronary artery disease in first-degree relatives <55 (male) or <65 (female) years of age, (iv) hyperlipidaemia (total serum cholesterol level >240 mg/dL or treatment with lipid-lowering drugs), (v) smoking history (previous or current cigarette smoking), (vi) coronary artery disease, which was defined as any coronary stenosis >50% diagnosed by coronary angiography or documented prior myocardial infarction (MI) (defined according to standard definition (serum cardiac biomarker elevation with symptoms of ischaemia and/or ECG changes indicative of new ischaemia/infarction)), (vii) angiographically diagnosed peripheral vascular disease, and (viii) patient medications (at baseline).

We also collected the arterial blood pressure measurements and lipid profile (total, HDL and LDL cholesterol and level of triglycerides, all expressed in mg/dL) at the time of PAT study. The data from the risk factors in conjunction with the level of total and HDL cholesterol and the systolic blood pressure were then used to calculate the Framingham risk score (FRS) which presented as 10 years risk (in per cent).

Assessment of subsequent interventions and outcome events during follow-up

Analysis of the patient medical records which were supplemented by phone interview (as mentioned above) was performed to detect the occurrence of any of the following outcome events: (i) all-cause death, (ii) CV death (all death not known to be definitely non-CV), (iii) MI, (iv) percutaneous coronary intervention, (v) coronary artery bypass grafting, (vi) diagnosed ischaemic or haemorrhagic stroke or transient ischaemic attack (TIA). All cerebrovascular events were confirmed by neurologists to meet the criteria for stroke or TIA through appropriate combination of medical history, examination, and/or neuroimaging, and (vii) hospitalization for any cardiac cause, defined as hospitalization for any of the following symptoms: chest pain, dyspnoea, palpitations, or syncope. The number of cardiac hospitalizations
during follow-up period was also recorded. The combined endpoint of cardiac adverse event (AE) was defined as the occurrence of CV death, MI, revascularization, or cardiac hospitalizations.

Death certificates were reviewed to verify the date and cause of all death events occurred during the follow-up period.

Statistical analysis
The statistical analysis was performed by an independent observer (R.J.L.). Continuous variables are summarized as mean ± SD, unless otherwise specified. Discrete variables are summarized as frequency (percentage). Kaplan–Meier methods were used to estimate survival and event-free survival rates. Cox proportional hazards models were used to estimate unadjusted and adjusted hazard ratios with corresponding 95% confidence intervals and P-values. Multiple Cox regression models were computed to estimate the partial effect of L_RHI. Three covariate sets were investigated: (1) age, (2) age and prior CABG, (3) the FRS (modelled as a three-degree-of-freedom spline). Sets 1 and 2 were chosen because they were significantly associated with AE. However, both gave similar results. The FRS was chosen for clinical relevance (and incorporates age into its calculation).

The best cut-off points for the PAT results for predicting future outcomes was identified and P-values were adjusted for the multiple tests done to identify the cut-off point. The ability of the cut-point to discriminate between high- and low-risk patients was estimated by a modified c-index statistic, similar to the traditional area under the ROC curve, but specifically for time-to-event endpoints which preclude ROC calculations.

One thousand bootstrap samples were created to estimate the optimism in the estimated association between L_RHI and follow-up AE. The optimism measure indicates how much better the model (or cut-point) works in the data set on which it was derived vs. other similar data sets. In each bootstrap sample, the best cut-off for L_RHI was determined as in the observed sample. The optimism measure was subtracted from the observed estimate to get a corrected measure of the effect. The same standard error was used to generate confidence intervals about the corrected point estimate, and a Wald test was used to calculate a corrected P-value.

Results
As mentioned above, a detailed follow-up including a telephone interview was completed in 270 patients (83%). Baseline characteristics are presented in Table 1. There were missing laboratory data on the following parameters: creatinine (n = 4), total and HDL cholesterol (n = 39), triglycerides (n = 38), LDL cholesterol (n = 31), systolic BP (n = 12), diastolic BP (n = 14).

Analysis of reactive hyperaemia–peripheral arterial tonometry
Reactive hyperaemia index was calculated from the ratio of the digital pulse volume during RH and baseline in 270 patients, and the mean (natural logarithmic) L_RHI was 0.5 ± 0.4 (range 0.7 to 1.8).

Clinical outcome
Patients were followed for a mean follow-up of 5.8 years (median = 5.8 years, IQR 4.4, 8.0) during which 98 patients had an AE. Adverse event as defined (CV death/MI/revascularization or CV hospitalization) occurred in 86 patients.

Nine patients died during follow-up: three died from a definite CV cause (two patients who died of MI and one patient due to heart failure). Eight patients experienced MI during the follow-up period (and two of them died later as mentioned above).

Twenty-eight patients underwent a coronary revascularization procedure (18 percutaneous intervention and 10 coronary artery bypass surgery). Definite stroke was diagnosed in 10 patients. Sixty-nine patients overall were hospitalized for a cardiac cause during the follow-up period. Careful assessment of all cardiac hospitalizations identified exacerbation of chest pain (angina) and suspected acute coronary syndrome as the most frequent admission diagnosis (in >90% of cases).

<table>
<thead>
<tr>
<th>Table 1 Baseline characteristics of 270 patients undergoing reactive hyperaemia–peripheral arterial tonometry</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (mean ± SD)</td>
</tr>
<tr>
<td>Females</td>
</tr>
<tr>
<td>Hypertension</td>
</tr>
<tr>
<td>Diabetes mellitus</td>
</tr>
<tr>
<td>Smoking history</td>
</tr>
<tr>
<td>Hyperlipidaemia</td>
</tr>
<tr>
<td>Family history of coronary artery disease</td>
</tr>
<tr>
<td>Proven peripheral vascular disease</td>
</tr>
<tr>
<td>Body mass index (kg/m²) (mean ± SD)</td>
</tr>
<tr>
<td>10 years Framingham risk in % (range)</td>
</tr>
<tr>
<td>History of prior myocardial infarction</td>
</tr>
<tr>
<td>Known coronary artery disease (previous MI or revascularization)</td>
</tr>
</tbody>
</table>

Discharge medications
Beta-blockers                                                  | 117 (44%)         |
Calcium channel blockers                                        | 107 (40%)         |
Anti-platelet medications                                       | 126 (47%)         |
ACE-inhibitors or ARBs                                         | 123 (46%)         |
Lipid-lowering drugs                                            | 169 (63%)         |
Nitrates                                                       | 119 (44%)         |
Total cholesterol (mg/dL) (mean ± SD)                          | 182 ± 41          |
HDL cholesterol (mg/dL) (mean ± SD)                            | 51 ± 18           |
LDL cholesterol (mg/dL) (mean ± SD)                            | 103 ± 34          |
Triglycerides (mg/dL) (mean ± SD)                              | 147 ± 109         |
Systolic BP in mmHg (mean ± SD) (range)                        | 126 ± 18 (90–190) |
Diastolic BP in mmHg (mean ± SD) (range)                       | 76 ± 11 (50–109)  |
Mean L_RHI                                                     | 0.5 ± 0.4         |
due to a significantly higher rate (≥50% increased rate) of CV hospitalizations (Table 2). Patients with L_RHI < 0.4 were not only more likely to be hospitalized but were more likely to experience repeated hospitalizations. Thus, the total number of hospitalizations during the follow-up period was significantly higher in patients with L_RHI < 0.4, with an incidence estimate of 0.19/ year (135 hospitalizations during a total of 718.8 follow-up years in this group), when compared with 0.05/year (overall 44 hospitalizations during a total of 826.4 follow-up years) in the group with L_RHI ≥ 0.4.

The estimated 7-year AE rate was 48% in patients with L_RHI < 0.4 vs. 28% of patients with L_RHI ≥ 0.4 (P = 0.030; c-index = 0.57; corrected c-index = 0.55) and the Kaplan–Meier curves continued to separate during the follow-up period (Figure 1).

MI, revascularization, and stroke rates were not significantly higher in the patient group with L_RHI < 0.4. The comparison of the significance of this cut-point vs. others to differentiate patients by risk of AE is shown in Figure 2 as the relation between the (inverted) P-values and the L_RHI.

Most baseline characteristics were not different between the two groups. The mean FRS was also similar in both groups (Table 3).

Multivariate analysis showed that, for the observed data set, L_RHI < 0.4 was independently associated with increased AE rate (HR = 1.79, 95% CI 1.16–2.76, P = 0.008) during follow-up and identified advancing age as a predictor of AE (HR = 1.2, 95% CI 1.013–1.45, P = 0.035). L_RHI remained an independent predictor of AE even when included in a model with the FRS (HR = 1.68, 95% CI 1.02–2.78, P = 0.043) (Table 4).

Discussion

The main finding of our study was that low RH value derived from the EndoPAT signal was associated with a higher incidence of AEs during the follow-up period and especially was predictive of significant symptoms (mostly chest pain) during follow-up, requiring repeated hospitalizations for suspected acute coronary syndrome. The combined endpoint of CV death, MI, revascularization, and CV hospitalizations was not only more frequent during follow-up in

![Figure 1](https://example.com/figure1.png)

Figure 1 Cardiovascular adverse events: a combination of cardiac death/myocardial infarction/coronary revascularization and cardiac hospitalizations in patients with and without low L_RHI (<0.4).

![Figure 2](https://example.com/figure2.png)

Figure 2 Plot of the (inverted) P-values vs. the L_RHI cut-points comparing the usefulness of the cut-point of L_RHI 0.4 vs. other cut-points to risk stratify patients for adverse events.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Patients with L_RHI &lt; 0.4 (n = 130)</th>
<th>Patients with L_RHI ≥ 0.4 (n = 140)</th>
<th>HR (95% CI)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>CV death</td>
<td>3.9%</td>
<td>0.0%</td>
<td>∞ (1.32, ∞)</td>
<td>0.032</td>
</tr>
<tr>
<td>Myocardial infarction</td>
<td>3.4%</td>
<td>3.7%</td>
<td>1.06 (0.27, 4.27)</td>
<td>0.93</td>
</tr>
<tr>
<td>Revascularization</td>
<td>12.7%</td>
<td>11.4%</td>
<td>1.21 (0.55, 2.65)</td>
<td>0.64</td>
</tr>
<tr>
<td>Stroke</td>
<td>5.3%</td>
<td>3.1%</td>
<td>1.6 (0.45, 5.68)</td>
<td>0.46</td>
</tr>
<tr>
<td>CV hospitalizations</td>
<td>30.5%</td>
<td>18.7%</td>
<td>2.06 (1.26, 3.38)</td>
<td>0.018*</td>
</tr>
<tr>
<td>AE</td>
<td>48%</td>
<td>28%</td>
<td>1.83 (1.18, 2.81)</td>
<td>0.030*</td>
</tr>
</tbody>
</table>

*P-values adjusted for the multiple tests done to identify the optimal cut-point.
patients with a low L_RHI than in patients with higher L_RHI, but also was able to predict future AE (as defined) beyond the traditional FRS. This may suggest a role for individualized risk assessment using non-invasive evaluation of the endothelial function by PAT.

Endothelial cells dysfunction is a key component of atherogenesis and contributes to the development of clinical CV diseases. In the presence of known vascular risk factors, endothelial cells undergo phenotypic changes resulting in decreased nitric oxide bioactivity, thereby promoting vasoconstriction, inflammation, and thrombosis. In human studies, CV risk factors have been associated with impaired vasomotor function, and individuals with abnormal vasodilator function were shown to have higher rate of CV events. Furthermore, modifications of CV risk factors that contribute to endothelial dysfunction improve patient clinical outcomes disproportionately to the improvement in coronary atherosclerosis, thus implying that these beneficial effects may be mediated in part through improvement in endothelial function.

A key player in the endothelial cell response to various stimuli is nitric oxide.

Nitric oxide has been shown to be an important factor contributing to the augmentation of the PAT pulse amplitude after ischaemia, and administration of an endothelial nitric oxide inhibitor blunted the hyperaemic response as detected by PAT. Hence, the hyperaemic response detected by PAT reflects endothelial function.

Coronary endothelial dysfunction as evaluated by invasive methods predict CV events and stroke and correlate with abnormal PAT results and coronary endothelial dysfunction as detected by the invasive evaluation of coronary endothelial function. Brachial artery flow-mediated dilation was also shown recently to predict future adverse outcome events in patients with diagnosed obstructive coronary artery disease. The current study shows that PAT has a similar predictive power.

Overall, PAT is becoming a useful method to evaluate vascular health in various disease states. For example, PAT detected improved endothelial function following treatment with enhanced external counter pulsation in patients with refractory angina pectoris. Abnormal endothelial function by PAT was also shown to be prevalent in adolescents with type 1 diabetes mellitus. Thus, the concept of using a non-invasive method to evaluate endothelial function in an office based manner is an appealing concept in primary (or secondary) prevention. The potential significance of the assessment of endothelial function is underscored by previous studies that demonstrated that lack of improvement of endothelial function by conventional therapy is associated with CV events. Therefore, the non-invasive evaluation of endothelial function may serve as a clinical tool, not only for prediction of events but also for the assessment of therapy. While the main limitation of peripheral endothelial testing was reproducibility of the results, PAT may have improved reproducibility, especially because of ongoing monitoring allowing for complete arterial occlusion and indexing the RH score to the contralateral arm.

Our finding that PAT results were predictive of outcome events beyond the traditional FRS is encouraging given the established predictive value of the FRS. However, in selected groups such as young adults and females, the FRS may have a lower predictive value, and its usefulness for risk stratification in the individual patient may still be limited. Thus, there is a dire need for better risk prediction tools, and individualized evaluation of endothelial dysfunction may allow a more personalized risk assessment. PAT may therefore be an important risk stratification tool in addition to the traditional FRS.

### Study limitations

The study may be influenced by selection bias as the patients referred for tertiary centres may be a selected group, perhaps with more significant symptoms but with less overt obstructive coronary disease. The inclusion of patients with known coronary

### Table 3  Selected baseline characteristics and laboratory values in relation to the natural logarithmic scaled reactive hyperaemia index

<table>
<thead>
<tr>
<th>Parameter</th>
<th>L_RHI &gt; 0.4 (n = 140)</th>
<th>L_RHI &lt; 0.4 (n = 130)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>53 ± 12</td>
<td>54 ± 13</td>
<td>0.28</td>
</tr>
<tr>
<td>Total cholesterol</td>
<td>185 ± 36</td>
<td>178 ± 45</td>
<td>0.14</td>
</tr>
<tr>
<td>HDL cholesterol</td>
<td>55 ± 18</td>
<td>46 ± 17</td>
<td>0.0001</td>
</tr>
<tr>
<td>LDL cholesterol</td>
<td>104 ± 31</td>
<td>103 ± 36</td>
<td>0.57</td>
</tr>
<tr>
<td>Systolic blood pressure (mmHg)</td>
<td>127 ± 17</td>
<td>125 ± 18</td>
<td>0.57</td>
</tr>
<tr>
<td>Diastolic blood pressure (mmHg)</td>
<td>76 ± 10</td>
<td>75 ± 12</td>
<td>0.4</td>
</tr>
<tr>
<td>Framingham risk (10 years, %)</td>
<td>6.3 ± 4.7</td>
<td>8.3 ± 8.3</td>
<td>0.28</td>
</tr>
<tr>
<td>L_RHI</td>
<td>0.8 ± 0.3</td>
<td>0.1 ± 0.2</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

### Table 4  Hazard ratios for adverse event from multiple Cox regression models

<table>
<thead>
<tr>
<th>Variable</th>
<th>Estimate</th>
<th>Hazard ratio</th>
<th>95% CI</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>L_RHI &lt; 0.4</td>
<td>0.6004</td>
<td>1.82</td>
<td>(1.18, 2.81)</td>
<td>0.007</td>
</tr>
<tr>
<td>Corrected</td>
<td>0.4472</td>
<td>1.56</td>
<td>(1.01, 2.41)</td>
<td>0.030</td>
</tr>
<tr>
<td>Model 2: adjusted for age (corrected-statistic 0.57)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>L_RHI &lt; 0.4</td>
<td>0.5831</td>
<td>1.79</td>
<td>(1.16, 2.76)</td>
<td>0.008</td>
</tr>
<tr>
<td>Corrected</td>
<td>0.4301</td>
<td>1.54</td>
<td>(1.00, 2.37)</td>
<td>0.052</td>
</tr>
<tr>
<td>Age, per decade</td>
<td>0.1918</td>
<td>1.21</td>
<td>(1.01, 1.45)</td>
<td>0.036</td>
</tr>
<tr>
<td>Model 3: adjusted for Framingham risk (corrected-statistic 0.59)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>L_RHI &lt; 0.4</td>
<td>0.5197</td>
<td>1.68</td>
<td>(1.02, 2.78)</td>
<td>0.043</td>
</tr>
<tr>
<td>Corrected</td>
<td>0.3420</td>
<td>1.41</td>
<td>(0.85, 2.33)</td>
<td>0.18</td>
</tr>
</tbody>
</table>

Corrected for bias in choosing a cut-point that fits the observed data best via a bootstrap algorithm. Corrected P-value according to Contal and O’Quigley.
artery disease may have increased the likelihood for future events but the L_RHI remained an independent predictor of events even when adjusted to the presence of known coronary artery disease. Additionally, we have not been able to demonstrate an independent role of PAT in the prediction of MI alone, and subsequent CV hospitalizations were the most common AEs. Nonetheless, in symptomatic patients with non-obstructive coronary artery disease, repeated cardiac hospitalizations may be a marker of other CV AEs, especially in women, and the events may be a result of coronary microvascular dysfunction or endothelial dysfunction. Moreover, CV hospitalizations for acute coronary syndromes are also a significant endpoint because they are associated with significant costs (average cost of $16,842 per hospitalization in the USA during 2006). This is especially important given the significant higher incidence of repeated hospitalizations observed among patients with a low L_RHI.

Conclusions

In conclusion, a low RH signal as detected by the EndoPAT, and consistent with endothelial dysfunction, was associated with higher AE rate during follow-up. L_RHI was an independent predictor of AE beyond the traditional FRS. Non-invasive assessment of peripheral vascular function in addition to the FRS may be useful for individualized identification of patients at risk for cardiac AEs.

Supplementary material

Supplementary material is available at European Heart Journal online.

Conflict of interest: A.L. is a member of the advisory board of Itamar Medical. All other authors have no conflicts of interest to disclose.

References

Contrast echocardiography guidance for alcohol septal ablation of hypertrophic obstructive cardiomyopathy

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A 63-year-old man was admitted due to worsening dyspnoea and faintness related to hypertrophic obstructive cardiomyopathy. Resting left ventricular outflow tract obstruction on echocardiography was 100 mmHg. Angiography showed normal coronary arteries, with one main septal perforator artery suitable for alcohol ablation (Panel A, black arrow). After installation of a 0.014 in. wire, a 1.5 mm over-the-wire angioplasty balloon was placed and inflated (Panel B, black arrow). Injection of the echographic contrast agent (Sonovue®, Bracco Imaging) through the catheter showed that the septal artery supplied a myocardial area distal to the subaortic bulge, extending to the tricuspid subvalvular apparatus (Panel C, white arrow). Thus, ablation of this septal artery was given up. Careful review of the baseline angiogram showed the presence of another small, hardly visible, septal perforator artery, 1 cm proximal to the previous one (Panel A, white arrow). It was catheterized by a wire and a balloon was installed (Panel D, white arrow). Contrast injection opacified the septal bulge (Panel E), confirming that it was the target vessel for ablation. Two millilitres of pure ethanol were infused over a period of 15 min. Immediate haemodynamic result was good, with disappearance of either resting or provoked left ventricular outflow tract gradient. Clinical outcome was uneventful.

Alcohol septal ablation has emerged as an effective method to treat symptomatic hypertrophic obstructive cardiomyopathy refractory to medical therapy. However, questions about its acute and long-term safety are still pending. Guidance of the procedure by contrast echocardiography can avoid acute complications which cannot be anticipated by angiography.

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