

Comparison of peripheral endothelial dysfunction and intimal media thickness in patients with suspected coronary artery disease

M-D Enderle, S Schroeder, R Ossen, C Meisner, A Baumbach, H U Haering, K R Karsch, M Pfohl

Abstract

Objective—Flow associated dilatation (FAD%) and intimal media thickness are established markers of early atherosclerosis. This study aimed to compare the ability of the non-invasive measurements FAD% and intimal media thickness to predict coronary artery disease.

Methods—FAD% and intimal media thickness were determined using high resolution ultrasound in 122 patients with clinically suspected coronary artery disease before coronary angiography. Results are given as mean (SD).

Results—Patients with coronary artery disease had reduced FAD% compared with those with angiographically normal coronary vessels (3.7 (4.1) v 7.0 (3.5)%, $p < 0.001$), whereas intimal media thickness tended to be increased in patients with coronary artery disease (0.58 (0.35) v 0.47 (0.11)mm, $p = 0.054$). There was a negative correlation between FAD% and intimal media thickness ($R = -0.317$, $p = 0.0004$). Receiver operating characteristic analysis showed that FAD% $\leq 4.5\%$ predicted coronary artery disease with a sensitivity of 0.71 (95% confidence interval 0.61 to 0.80) and a specificity of 0.81 (0.58 to 0.95). In contrast, intimal media thickness showed a positive correlation with the extent of coronary artery disease (number of vessels with a lesion $\geq 50\%$) ($R = 0.324$, $p = 0.0003$), without a clear cut off point.

Conclusions—In patients with clinically suspected coronary artery disease, FAD% discriminates between the presence or absence of coronary artery disease, whereas intimal media thickness is associated more with the extent of coronary artery disease.

(Heart 1998;80:349–354)

Keywords: coronary artery disease; endothelial dysfunction; intimal media thickness; flow associated dilatation

The most important cause of morbidity and mortality in atherosclerosis is coronary artery disease.¹ Atherosclerosis is a systemic disease and recent studies have shown a close relation between peripheral endothelial dysfunction and the presence of coronary artery disease.^{2,3} Endothelial dysfunction is regarded as an early change in the development of atherosclerosis^{4,5} and is thought to reflect a functional impair-

ment of the endothelium before morphological changes can be detected. So far, methods of detecting peripheral endothelial function have been invasive and expensive.^{6–8} Thus non-invasive techniques of determining endothelial function, such as flow associated vasodilatation (FAD%), are promising alternatives.^{9–11}

An early morphological change in atherosclerosis is the increase in intimal media thickness, which can be detected by high resolution ultrasound B mode imaging.¹² It has been shown that increased intimal media thickness in the common carotid artery is closely associated with coronary artery disease¹³ and with peripheral vascular disease.¹⁴ However, the correlation of these two features—peripheral endothelial function (that is, endothelial function of the brachial artery) and morphological changes in the common carotid artery—with the presence of coronary artery disease is not yet clearly characterised, nor has their sensitivity, specificity, and predictive value been well defined.

Our aim in this study was to analyse the diagnostic value of these non-invasive indices for the prediction of coronary artery disease. The sensitivity of FAD% and intimal media thickness was prospectively determined in patients with suspected coronary artery disease before undergoing coronary angiography.

Methods

PATIENTS

The study group consisted of 122 subjects in whom there was an indication for cardiac catheterisation for clinically suspected coronary artery disease. Thirty four were female and 88 male. Their ages ranged from 38 to 75 years (mean (SD) 60.0 (8.6) years), body mass index ranged from 20.5 to 36.7 kg/m² (27.0 (3.4) kg/m²), and lumen diameter of the brachial artery at rest was between 2.48 and 6.33 mm (4.57 (0.74) mm). The cardiovascular risk profile was as follows: *arterial hypertension* (n = 69, 57%)—systolic blood pressure 136 (19) mm Hg (92 to 190), diastolic blood pressure 86 (11) mm Hg (60 to 120); *hyperlipidaemia* (n = 91, 75%)—total plasma cholesterol 6.2 (1.1) mmol/l (3.6 to 9.7), plasma triglyceride 2.72 (2.32) mmol/l (0.62 to 17.11); *diabetes mellitus* (n = 19, 16%)—fasting blood glucose 5.7 (1.3) mmol/l (4.0 to 12.3 mmol/l); *smoking* (n = 83, 68%)—smoking duration 25.2 (11.5) years and 17.3 (16.9) pack-years; *positive family history* within first degree relatives (n = 55, 45%).

Department of
Endocrinology,
University of
Tübingen, Tübingen,
Germany
M-D Enderle
R Ossen
H U Haering
M Pfohl

Department of
Cardiology, University
of Tübingen
S Schroeder
A Baumbach
K R Karsch

Institute for Medical
Information
Processing, University
of Tübingen
C Meisner

Correspondence to:
Dr Markus-D Enderle,
Eberhard-Karls-Universität
Tübingen, Medizinische
Klinik und Poliklinik, Abt
Innere Medizin IV,
Otfried-Müller-Str 10,
D-72076 Tübingen,
Germany.

Accepted for publication
26 June 1998

Table 1 Baseline characteristics of 122 patients subdivided into five groups according to coronary angiography results

	No CAD	CAD-0 Stenosis < 50%	CAD-1 Stenosis > 50%	CAD-2 Stenosis > 50%	CAD-3 Stenosis > 50%	p value
Number	21	30	26	21	24	
Age (years)	57.3 (8.2)	61.4 (7.7)	59.6 (9.7)	59.9 (8.2)	62.8 (8.8)	0.263
Sex (M/F)	13/8	23/7	17/9	19/2	16/8	0.210
BMI (kg/m ²)	27.3 (3.3)	26.7 (3.1)	26.5 (3.6)	27.9 (3.9)	26.9 (3.3)	0.678
Lumen diameter (mm)	4.4 (0.8)	4.7 (0.6)	4.6 (0.7)	4.8 (0.7)	4.4 (0.9)	0.209
SBP (mm Hg)	129.9 (13.1)	138.1 (21.3)	134.6 (20.4)	139.7 (19.0)	136.5 (17.9)	0.457
DBP (mm Hg)	85.6 (11.5)	87.7 (10.1)	85.6 (12.5)	86.5 (10.3)	83.4 (10.1)	0.633
Smoking (% (n))	57 (12/21)	70 (21/30)	62 (16/26)	76 (16/21)	75 (18/24)	0.578
Diabetes (% (n))	5 (1/21)	10 (3/30)	15 (4/26)	29 (6/21)	25 (6/24)	0.092
Hypertension (% (n))	43 (9/21)	47 (14/30)	58 (15/26)	62 (13/21)	75 (18/24)	0.173
Hyperlipidaemia (% (n))	52 (11/21)	75 (22/30)	85 (22/26)	81 (17/21)	79 (19/24)	0.108
Total cholesterol (mmol/l)	5.7 (0.9)	6.2 (1.1)	6.3 (1.0)	6.3 (0.5)	6.3 (1.4)	0.597
Triglycerides (mmol/l)	1.59 (0.66)	3.65 (3.78)	2.67 (1.54)	2.56 (1.47)	2.68 (1.75)	0.042
History of CHD (% (n))	43 (9/21)	50 (14/29)	38 (10/26)	29 (6/21)	63 (15/24)	0.181

Values are mean (SD) unless otherwise specified.

Coronary artery disease classification: CAD-0, vessel alterations but lesion < 50%; CAD-1, lesion > 50% in one vessel; CAD-2, lesion > 50% in two vessels; CAD-3, lesion > 50% in three vessels.

BMI, body mass index; SBP/DBP, systolic/diastolic blood pressure; CHD, coronary heart disease.

The diagnosis of coronary artery disease was based on either typical clinical symptoms (n = 112, 92%), or atypical chest pain with pathological resting or stress electrocardiography, or both (n = 84, 69%). Arterial hypertension was defined as systolic blood pressure > 160 mm Hg, diastolic blood pressure > 95 mm Hg (documented at least four times), a history of arterial hypertension treated with antihypertensive agents, or combinations of these. Diabetes mellitus was defined as the use of oral hypoglycaemic agents or insulin, or as raised blood glucose concentrations according to the WHO criteria.¹⁵ Hyperlipidaemia was defined as either being on lipid lowering drugs, or having a raised plasma total cholesterol of > 5.2 mmol/l, raised plasma triglycerides of > 2.26 mmol/l, or both (on at least two measurements). Smoking habits were documented by a standardised interview; current smoking of more than five cigarettes a day for more than three months was defined as a positive smoking history.

Patients with chronic congestive heart failure, renal insufficiency, or valvar heart disease were excluded, as were those on chronic haemodialysis or intravenous anticoagulants. All subjects were studied on their standard treatment regimens which included: aspirin 100 mg/day (n = 93, 76%), nitrates (n = 41, 34%), angiotensin converting enzyme (ACE) inhibitors (n = 26, 21%), β blockers (n = 69, 57%), calcium channel antagonists (n = 26, 21%), diuretics (n = 16, 13%), and statins (n = 22, 18%).

The study was performed in accordance with the principles of the Declaration of Helsinki and was approved by the local ethics com-

mittee. Written informed consent was obtained from all participants.

STUDY PROTOCOL

One day before coronary angiography the patients were asked to refrain from smoking, drinking coffee or tea, and carrying out any exhausting physical activity six hours before the blood sampling. All blood samples were drawn two hours before the ultrasound measurements. The determination of serum total cholesterol, low density lipoprotein (LDL), high density lipoprotein (HDL), triglycerides, and blood glucose was achieved using routine laboratory methods. The next day, after at least 10 hours fasting, all patients underwent coronary angiography.

PROCEDURES

The ultrasound procedures were performed with modifications to the methods of Celermajer *et al* and Wendelhag *et al*,^{9,16} as previously reported.^{17,18} Briefly, the measurements were conducted in supine position at a stable room temperature of 25°C \pm 2°C. The lumen diameter of the brachial artery was sought in a cross sectional view and then measured blind in longitudinal section, using B mode stand-by. Two focus zones were set to the depth of the transducer for near wall and far wall, respectively. In addition to individual optimisation of the depth and gain settings, a preset vascular imaging program was used to standardise the measurements. All scans were electrocardiographically triggered (at end diastole). Blood pressure was documented simultaneously in the contralateral arm at two minute intervals.

Table 2 Concurrent drug treatment in the five patient groups

	No CAD	CAD-0 Stenosis < 50%	CAD-1 Stenosis > 50%	CAD-2 Stenosis > 50%	CAD-3 Stenosis > 50%	p value
Aspirin	17/21 (81%)	24/30 (80%)	22/26 (85%)	16/21 (76%)	22/24 (92%)	0.448
Nitrates	9/21 (43%)	12/30 (40%)	7/26 (27%)	7/21 (33%)	6/24 (25%)	0.156
Ca antagonists	4/21 (19%)	3/30 (10%)	6/26 (23%)	4/21 (19%)	9/24 (38%)	0.066
β Blockers	10/21 (48%)	16/30 (53%)	17/26 (65%)	11/21 (52%)	15/29 (63%)	0.363
ACE inhibitors	6/21 (29%)	4/30 (13%)	5/26 (19%)	7/21 (33%)	4/24 (17%)	0.967
Diuretics	2/21 (10%)	2/30 (7%)	3/26 (12%)	2/21 (10%)	7/24 (29%)	0.049
Statins	0/21 (0%)	5/30 (17%)	7/26 (27%)	4/21 (19%)	6/24 (25%)	0.044

Coronary artery disease classification: CAD-0, vessel alterations but lesion < 50%; CAD-1, lesion > 50% in one vessel; CAD-2, lesion > 50% in two vessels; CAD-3, lesion > 50% in three vessels.

ACE inhibitors, angiotensin converting enzyme inhibitors; Ca antagonists, calcium channel antagonists.

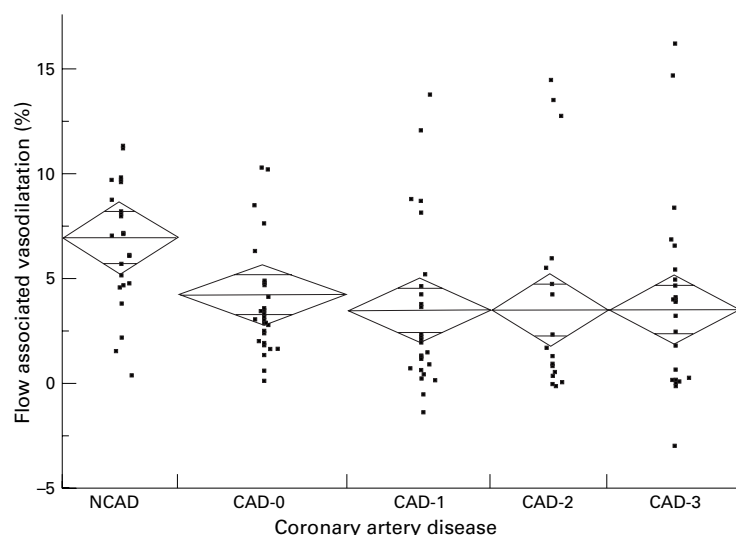


Figure 1 Flow associated vasodilatation (FAD%) related to the extent of coronary artery disease (CAD). NCAD, no vessel alterations; CAD-0, vessel alterations but lesion < 50%; CAD-1, lesion > 50% in one vessel; CAD-2, lesion > 50% in two vessels; CAD-3, lesion > 50% in three vessels; FAD%, flow associated (vaso)dilatation derived as percentage change during reactive hyperaemia relative to the baseline scan at rest (100%).

Ultrasound measurements were taken as follows: (1) after 10 minutes of rest; (2) 45–60 seconds after sudden deflation of a tourniquet placed on the ipsilateral forearm, after which—during a 20 minute period for vessel recovery—the intimal media thickness of the common carotid artery on both sides was determined^{19–21}; (3) a third measurement of the brachial artery was then made, after which 400 µg of glyceryl trinitrate was given sublingually; (4) four minutes later the brachial artery was scanned a fourth time to assess endothelium independent vasodilatation.

Flow associated dilatation

FAD% comprises the change in arterial diameter in response to reactive hyperaemia as an endothelium dependent stimulus to vasodilatation, caused by an increase in shear stress with the release of nitric oxide. FAD% is defined as

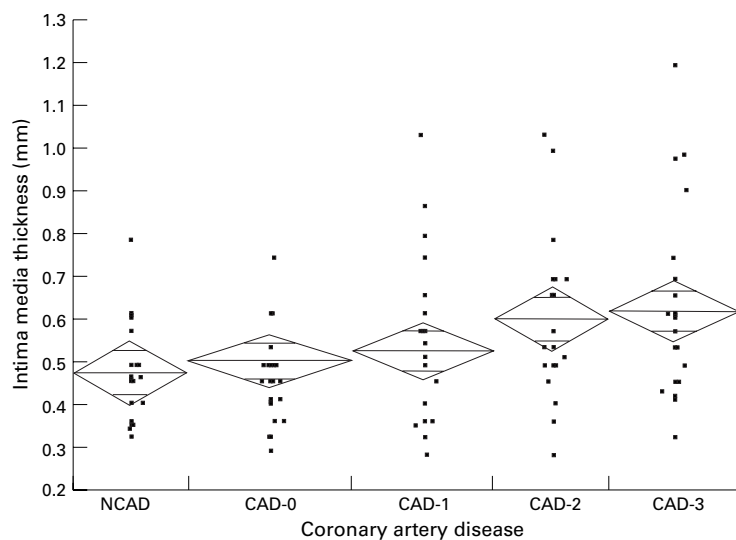


Figure 2 Intimal media thickness related to the extent of coronary artery disease (CAD). NCAD, no vessel alterations; CAD-0, vessel alterations, but lesion < 50%; CAD-1, lesion > 50% in one vessel; CAD-2, lesion > 50% in two vessels; CAD-3, lesion > 50% in three vessels.

flow associated (vaso)dilatation, derived as percentage change relative to the baseline scan at rest (= 100%).

Change in arterial diameter after glyceryl trinitrate administration

The change in arterial diameter after glyceryl trinitrate administration (GTN%) was defined as the change in diameter in response to the application of 400 µg of glyceryl trinitrate, resulting in endothelium independent vasodilatation caused by relaxation of the smooth muscle cell and expressed as percentage change relative to the baseline scan (= 100%).

Intimal media thickness

The intimal media thickness of the common carotid artery was determined by methods described previously.^{22–23} Briefly, we scanned the common carotid artery bilaterally and performed six different blinded measurements in longitudinal projections approximately 2 cm below the bifurcation, at the start of the bulbous.

Coronary angiography

Coronary angiography was conducted using the standard Judkins technique or through the right brachial artery using the Sones technique.^{24–25} Interpretation of the angiograms was performed by two different observers who were blinded both for the interpretation of the angiograms and the ultrasound scores. Coronary artery disease was defined as any viewable lesion. According to the severity and extent of atherosclerosis patients were divided into five subgroups: (1) NCAD, no vessel alterations; (2) CAD-0, vessel alterations, but lesion < 50%; (3) CAD-1, lesion > 50% in one vessel; (4) CAD-2, lesions > 50% in two vessels; (5) CAD-3, lesion > 50% in three vessels.

High resolution ultrasound

FAD%, GTN%, and intimal media thickness were assessed using a high resolution ultrasound device (AU4 Idea, ESAOTE Biomedica, Munich, Germany), equipped with an integrated electrocardiography package and a 13 MHz linear array transducer with an axial resolution of 0.12 mm and a penetration depth of 1.0–4.5 cm.

A continuous measurement of blood pressure by cuff inflation in two minute intervals was performed using an automatic device (Bosatron 2, Bosch und Sohn GmbH, Jungingen, Germany). Documentation of data was accomplished by a Sony videographic printer UP-890CE (Sony Corporation, Tokyo, Japan).

REPRODUCIBILITY

Intraobserver variability was determined for FAD% and GTN% on the basis of 15 measurements each. A coefficient of variation of 1.2% to 4.2% was found. The interobserver variability was evaluated as a mean (SD) difference of maximum 0.18 (0.10) mm, representing a difference of < 0.21 mm in 95% of all measurements. The intra- and interobserver variability for the measurement of intimal media thickness was comparable.^{22–23}

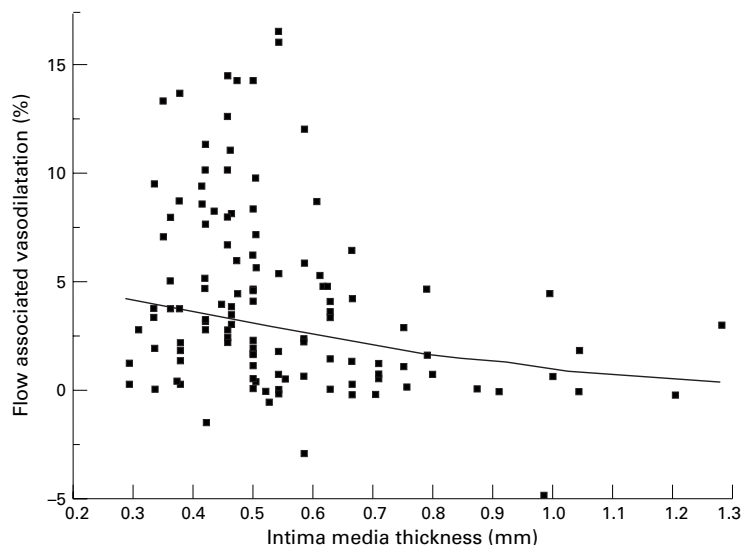


Figure 3 Inverse relation of flow associated vasodilatation (FAD%) and intimal media thickness ($R = -0.317$, $p = 0.0004$, FAD% log transformed). FAD% derived as percentage change during reactive hyperaemia relative to the baseline scan at rest (100%).

STATISTICAL ANALYSIS

Continuous variables were described using means and standard deviations (mean (SD)). Discrete variables were described as counts and percentages. The distributions of discrete variables (risk factors and clinical characteristics) by coronary artery disease group were compared using χ^2 tests. We used analysis of variance to compare continuous variables by coronary artery disease severity group if normally distributed, and the Kruskal-Wallis test if not normally distributed. Spearman rank correlations were used to describe correlations between variables that were not normally distributed. Test results are presented as nominal two tailed p values. Significance was inferred at $p \leq 0.05$. Because of the explorative nature of the study, all tests were interpreted in a descriptive sense.

To evaluate FAD% and intimal media thickness as a screening test for coronary artery disease the optimal cut off point was determined by receiver operating characteristic (ROC) analysis, using the coronary angiography result as the gold standard for the diagnosis of coron-

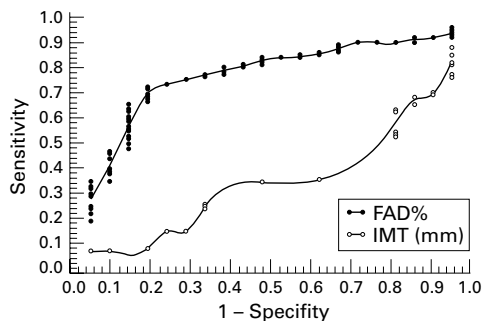


Figure 4 Receiver operator characteristic (ROC) analysis of flow associated vasodilatation (FAD%). Determination of a cut off point of FAD% $\leq 4.5\%$ as a predictor for coronary artery disease, showing a positive linear relation between intimal media thickness and coronary artery disease ($R = 0.324$, $p = 0.0003$). True positive rates (sensitivity) for the different criteria are plotted against false positive rates (1-specificity). FAD% derived as percentage change during reactive hyperaemia relative to the baseline scan at rest (100%).

ary artery disease. ROC analysis applies to tests that report clinical data as a continuous range of variables. The ROC is the relation between sensitivity and specificity for different test cut off values used as criteria for detecting disease.²⁶ The sensitivity and specificity are presented with 95% confidence intervals.

Results

PATIENT DATA

The 122 patients were examined both by peripheral high resolution ultrasound and coronary angiography. In 21 patients coronary angiography was normal (the NCAD group). Table 1 gives the baseline characteristics and the cardiovascular profile of the five subgroups. Only triglycerides showed differences between the groups ($p = 0.042$); otherwise all five groups were comparable with respect to numbers, age, sex, body mass index, blood pressure, family history, the distribution of smoking, diabetes, and hypertension. One patient in the CAD-0 group had a plasma triglyceride concentration of 17.3 mmol/l. This resulted in the difference among the groups.

ULTRASOUND STUDIES

The lumen diameter at rest did not differ between the five groups (table 2). FAD% was decreased in the four coronary artery disease groups compared with the NCAD group (3.7 (4.1)% *v* 7.0 (3.5)%, $p < 0.001$). No difference was found for FAD% between the different coronary artery disease groups: CAD-0, 4.2 (3.5)%; CAD-1, 3.5 (3.9)%; CAD-2, 3.5 (4.6)%; CAD-3, 3.5 (4.8)% (fig 1). After sublingual application of 400 μ g glyceryl trinitrate, vessel dilatation in patients without angiographically detectable coronary artery disease (NCAD group) was increased in comparison with the patients with coronary artery disease (14.9 (7.4)% *v* 10.2 (6.8)%, $p = 0.005$), but without further differences between the coronary artery disease groups: CAD-0, 9.5 (6.1)%; CAD-1, 9.9 (8.1)%; CAD-2, 10.8 (6.0)%; CAD-3, 11.0 (7.0)%.

The intimal media thickness of the common carotid artery tended to be increased in the coronary artery disease groups compared with the NCAD group (0.59 (0.35) *v* 0.47 (0.11) mm, $p = 0.053$). Intimal media thickness increased continuously with the extent of coronary artery disease (fig 2). We found an inverse relation between intimal media thickness and FAD% (fig 3). ROC analysis was calculated for FAD% and intimal media thickness to characterise the ability to predict coronary artery disease from these non-invasive sonographic indices. A cut off point of FAD% of $\leq 4.5\%$ with a sensitivity of 0.71 (95% confidence interval 0.61 to 0.80), a specificity of 0.81 (0.61 to 0.80), and a positive predictive value of 0.95 was determined (fig 4). In contrast to FAD%, intimal media thickness showed a positively linear relation with coronary artery disease (fig 4), and a cut off point could not be calculated by ROC analysis.

Discussion

An association between sonographic variables (such as intimal media thickness and FAD%) and coronary artery disease has recently been described.^{2 3 13} The aim of this study was to evaluate and to compare the ability of the two non-invasive indices to predict coronary artery disease. The two indices differ with regard to the pathophysiological information they provide, since FAD% is a functional variable and intimal media thickness a morphological variable. Both were correlated with the presence and extent of coronary artery disease assessed by coronary angiography.

FLOW ASSOCIATED VASODILATATION

A reduced FAD% was found in all patients with haemodynamically significant lesions by angiography compared with patients without angiographically visible lesions. This is in accordance with previously published data.^{3 27} In contrast to Neunteufl *et al*,²⁷ we did not find any difference in FAD% between the various coronary disease patient groups. One explanation for this difference might be that besides age, sex, and lumen diameter other possible confounding factors for FAD% were present, such as diabetes mellitus,²⁸ hyperlipidaemia,^{9 29} arterial hypertension,³⁰ smoking,³¹ and a positive family history.³² We corrected FAD% for body mass index, blood pressure, cholesterol, family history, smoking history, diabetes, and hypertension within the different patient groups, which was not done by Neunteufl *et al*.

The ROC analysis revealed a cut off point of FAD% of $\leq 4.5\%$ with a sensitivity of 0.71, a specificity of 0.81, and a positive predictive value of 0.95 for the presence of coronary artery disease. This cut off value is similar to the value of 3% found by Anderson *et al* in predicting coronary endothelial dysfunction. The calculated cut off value for FAD% in the study by Neunteufl *et al* was 10% with a sensitivity of about 85% and a specificity comparable to our results.²⁷

The most probable reason for a small cut off point and generally smaller FAD% and GTN% values in our study is that our measurements were performed without discontinuing the patients' drug treatment, which may have influenced endothelial dependent and independent vasodilatation. It is, however, of interest that this did not result in significant differences in mean FAD% within the different patient groups. Another reason may be that a different definition of the lumen diameter was used in our study. According to the definition of Wendelhag *et al*,¹⁶ the A line (intima/lumen interface at the near wall) was visualised and the actual morphological lumen diameter determined by measuring the distance between the A line and the B line. This was different from the study by Neunteufl *et al*,²⁷ where the lumen diameter was measured as the difference between the two M lines.

INTIMAL MEDIA THICKNESS

We found a trend towards increased intimal media thickness in patients with coronary artery disease compared with the group

without angiographic coronary artery disease. There was a positive correlation between the intimal media thickness of the common carotid artery and the extent of coronary artery disease. These findings are in agreement with other recent studies.^{33 34} In addition, our results showed an inverse relation between intimal media thickness and FAD% within the same cohort of patients with coronary artery disease. In the patients with coronary artery disease we found a continuous linear correlation between intimal media thickness of the common carotid artery and coronary artery disease, and thus—in contrast to Visona and colleagues³⁴—we were unable to calculate a cut off point for intimal media thickness to predict the presence of coronary artery disease. Our data support the hypothesis of Adams *et al*,³³ that the measurement of intimal media thickness is not sufficient for screening patients with suspected coronary artery disease.

CHANGE IN ARTERIAL DIAMETER AFTER GLYCERYL TRINITRATE

It has been shown previously that determination of GTN% is an index of reduced nitric oxide sensitivity in the smooth muscle cell of the arterial wall. We found a reduced GTN% in patients with coronary artery disease compared with patients with angiographically normal coronary arteries. This is in agreement with the study by Neunteufl *et al*,²⁷ who also described a reduced GTN% in coronary artery disease patients. These findings suggest that more advanced coronary atherosclerosis is associated with systemic atherosclerosis presenting with dysfunction of the smooth muscle cells of the peripheral arterial system.²⁴

STUDY LIMITATIONS

A certain preselection bias needs to be taken into account in interpreting these data, since all patients were scheduled for coronary angiography by expert cardiologists as a result of symptoms typical of coronary artery disease.³⁵ Thus all the ultrasound measurements were performed without discontinuing concurrent drug treatment (table 3). However, the different patient groups were quite well matched for drug treatment distribution, and concurrent medication did not significantly influence the results of FAD% and intimal media thickness (table 3).

Table 3 No significant influence of current drug treatment on flow associated vasodilatation* (FAD%) and intimal media thickness (IMT)

Variable	FAD% (p value)	IMT (p value)
Aspirin	0.765	0.594
Nitrates	0.131	0.166
Ca antagonists	0.608	0.955
β Blockers	0.077	0.495
ACE inhibitors	0.942	0.481
Diuretics	0.948	0.997
Statins	0.273	0.777

*Derived as percentage change during reactive hyperaemia relative to the baseline scan at rest (100%).

ACE inhibitors, angiotensin converting enzyme inhibitors; Ca antagonists, calcium channel antagonists.

CONCLUSION

Our data indicate that an impaired FAD% may be an useful non-invasive marker of the presence of coronary artery disease in patients with clinically suspect coronary artery disease. In contrast, determination of increased intimal media thickness is not useful in discriminating between presence or absence of coronary artery disease, though in patients with coronary artery disease, it appears to be associated with the extent of the disease. The role of these ultrasound procedures in the management of patients with chest pain in comparison with established non-invasive diagnostic tools (for example, electrocardiography, stress electrocardiography, nuclear myocardial scintigraphy) remains to be evaluated.

This study was supported by a grant from the Fortuene Programme (379-F, 1284053.2), Tübingen.

- 1 Goldner J, Wisnant JP, Taylor WF. Long-term prognosis of cerebral ischemic attacks. *Stroke* 1971;2:160-7.
- 2 Uehata A, Gerhard MD, Meredith IT, et al. Close relationship of endothelial dysfunction in coronary and brachial artery [abstract]. *Circulation* 1993;88:1-618.
- 3 Anderson TJ, Uehata A, Gerhard MD, et al. Close relation of endothelial function in the human coronary and peripheral circulations. *J Am Coll Cardiol* 1995;26:1235-41.
- 4 Ludmer PL, Selwyn AP, Shook TL, et al. Paradoxical vasoconstriction induced by acetylcholine in atherosclerotic coronary arteries. *N Engl J Med* 1986;315:1046-51.
- 5 Werns SW, Walton JA, Hsia HH, et al. Evidence of endothelial dysfunction in angiographically normal coronary arteries of patients with coronary artery disease. *Circulation* 1989;79:287-91.
- 6 Saenz de Tejada I, Goldstein I, Azadzi K, et al. Impaired neurogenic and endothelium-mediated relaxation of penile smooth muscle from diabetic men with impotence. *N Engl J Med* 1989;320:1025-30.
- 7 Smits P, Kapma JA, Jacobs MC, et al. Endothelium-dependent vascular relaxation in patients with type I diabetes. *Diabetes* 1993;42:148-53.
- 8 Calver A, Collier J, Vallance P. Inhibition and stimulation of nitric oxide synthesis in the human forearm arterial bed patients with insulin-dependent diabetes. *J Clin Invest* 1992;90:2548-54.
- 9 Celermajer DS, Sorensen KE, Gooch VM, et al. Non-invasive detection of endothelial dysfunction in children and adults at risk of atherosclerosis. *Lancet* 1992;340:1111-15.
- 10 Sorensen KE, Celermajer DS, Spiegelhalter DJ, et al. Non-invasive measurement of human endothelium dependent arterial responses: accuracy and reproducibility. *Br Heart J* 1995;74:247-53.
- 11 Wikstrand J, Wendelhag I. Quantitative ultrasonography of carotid and femoral arteries. In: *Vascular diagnostics*. Berlin: Springer, 1995:129-38.
- 12 Salonen JT, Salonen R. Ultrasound B-mode imaging in observational studies of atherosclerotic progression. *Circulation* 1993;87:II56-65.
- 13 Geroulakos G, O'Gorman D, Nicolaides A, et al. Carotid intima-media thickness: correlation with the British Regional Heart Study risk score. *J Intern Med* 1994;235:431-3.
- 14 Bots ML, Hofman A, Grobbee DE. Common carotid intima-media thickness and lower extremity arterial atherosclerosis. The Rotterdam Study. *Arterioscler Thromb* 1994;14:1885-91.
- 15 World Health Organisation. *Diabetes mellitus*. WHO Technical Report Series 1985. Geneva: WHO.
- 16 Wendelhag I, Gustavsson T, Suurkula M, et al. Ultrasound measurement of wall thickness in the carotid artery: fundamental principles and description of a computerized analysing system. *Clin Physiol* 1991;11:565-77.
- 17 Enderle MD, Benda N, Schmuelling RM, et al. Preserved endothelial function compared with healthy subjects in IDDM patients, but not in NIDDM patients. *Diabetes Care* 1998;21:271-7.
- 18 Enderle MD, Balletshofer BM, Schmuelling RM, et al. Early detection of atherosclerosis in type 2 diabetic patients using high resolution ultrasound. *Ultraschall Med* 1998;19:16-21.
- 19 Pignoli P. Ultrasound B-mode imaging for arterial wall thickness measurement. *Atheroscler Rev* 1984;12:177-84.
- 20 Pignoli P, Tremoli E, Poli A, et al. Intimal plus medial thickness of the arterial wall: a direct measurement with ultrasound imaging. *Circulation* 1986;74:1399-406.
- 21 Furchgott RF, Zawadzki JV. The obligatory role of endothelial cells in the relaxation of arterial smooth muscle by acetylcholine. *Nature* 1980;288:373-6.
- 22 Kawamori R, Yamasaki Y, Matsushima H, et al. Prevalence of carotid atherosclerosis in diabetic patients. Ultrasound high-resolution B-mode imaging on carotid arteries. *Diabetes Care* 1992;15:1290-4.
- 23 Yamasaki Y, Kawamori R, Matsushima H, et al. Atherosclerosis in carotid artery of young IDDM patients monitored by ultrasound high-resolution B-mode imaging. *Diabetes* 1994;43:634-9.
- 24 Judkins MP. Selective coronary arteriography. Part I. A percutaneous transfemoral technic. *Radiology* 1967;89:815-24.
- 25 Sewell WH. Coronary arteriography by the Sones technique. Technical considerations. *Am J Roentgenol Radium Ther Nucl Med* 1965;95:673-83.
- 26 McNeil B, Keeler E, Adelstein S. Primer on certain elements of medical decision making. *N Engl J Med* 1975;293:211-15.
- 27 Neunteufl T, Katzschlager R, Hassan A, et al. Systemic endothelial dysfunction is related to the extent and severity of coronary artery disease. *Atherosclerosis* 1997;129:111-18.
- 28 Clarkson P, Celermajer DS, Donald AE, et al. Impaired vascular reactivity in insulin-dependent diabetes mellitus is related to disease duration and low density lipoprotein cholesterol levels. *J Am Coll Cardiol* 1996;28:573-9.
- 29 Sorensen KE, Celermajer DS, Georgakopoulos D, et al. Impairment of endothelium-dependent dilation is an early event in children with familial hypercholesterolemia and is related to the lipoprotein (a) level. *J Clin Invest* 1994;93:50-55.
- 30 Iiyama K, Nagano M, Yo Y, et al. Impaired endothelial function with essential hypertension assessed by ultrasonography. *Am Heart J* 1996;132:779-82.
- 31 Celermajer DS, Sorensen KE, Georgakopoulos D, et al. Cigarette smoking is associated with dose-related and potentially reversible impairment of endothelium-dependent dilation in healthy young adults. *Circulation* 1993;88:2149-55.
- 32 Celermajer DS, Sorensen KE, Bull C, et al. Endothelium-dependent dilation in the systemic arteries of asymptomatic subjects relates to coronary risk factors and their interaction. *J Am Coll Cardiol* 1994;24:1468-74.
- 33 Adams MR, Nakagomi A, Keech A, et al. Carotid intima-media thickness is only weakly correlated with the extent and severity of coronary artery disease. *Circulation* 1995;92:2127-34.
- 34 Visona A, Pesavento R, Lusiani L, et al. Intimal medial thickening of common carotid artery as indicator of coronary artery disease. *Angiology* 1996;47:61-6.
- 35 Douglas PS. Is noninvasive testing for coronary artery disease accurate? *Circulation* 1997;95:299-302.