Natriuretic peptides and cyclic guanosine 3',5'-monophosphate in asymptomatic and symptomatic left ventricular dysfunction

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Abstract

Background—Screening for patients with asymptomatic left ventricular dysfunction is of considerable importance because they may benefit from early treatment with angiotensin converting enzyme inhibitors. It has been suggested that atrial natriuretic peptide (ANP), brain natriuretic peptide (BNP), and cyclic guanosine 3',5'-monophosphate (cGMP) might be useful markers for screening.

Objective—To compare directly the power of the three immunoreactive forms of ANP (CT-ANP, β-ANP, NT-ANP) and BNP and cGMP to detect asymptomatic left ventricular dysfunction.

Methods and results—Radionuclide ventriculography was used to study left ventricular ejection fraction in 37 patients with asymptomatic left ventricular dysfunction, 32 patients with mild to moderate congestive heart failure, and 38 controls. CT-ANP, NT-ANP, β-ANP, BNP, and cGMP were measured at rest and 3 minutes after exercise. Plasma BNP was the most sensitive marker for patients with asymptomatic left ventricular dysfunction but it reached only a sensitivity of 58% and a specificity of 76% at rest and a sensitivity of 65% and a specificity of 84% after exercise. Combined measurements of all natriuretic peptides and cGMP did not improve the power to detect asymptomatic left ventricular function above that of a single BNP measurement.

Conclusions—Although natriuretic peptides and cGMP measured at rest and three minutes after ergometry may be useful for monitoring left ventricular dysfunction they are unlikely to be suitable for more general routine screening for completely asymptomatic left ventricular dysfunction.

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Atrial natriuretic peptide (ANP) is the prototype of a family of peptides that are activated under different physiological and pathological conditions.[1-3] ANP maintains sodium homoeostasis by natriuresis and diuresis. ANP also acts as a vasodilator and as an antagonist of the renin angiotensin system under normal conditions. It is stored in the atrial wall as a 126 amino acid pro-ANP molecule. In response to atrial stretch immunoreactive products from the N-terminal part (NT-ANP, amino acid 1–98) and from the C-terminal part (CT-ANP, amino acid 99–126) appear in the plasma.[4 5] Additionally a dimer form of CT-ANP (β-ANP) can be found.[6] Brain natriuretic peptide (BNP) is a 32 amino acid peptide with a high degree of structural homology with ANP. BNP has the same main biological actions as ANP but it is mainly secreted from the ventricle.[7 8] Cyclic guanosine 3'5'-monophosphate (cGMP) acts as a second messenger for ANP and BNP in the target cells. In response to increased secretion of ANP and BNP from the heart, cGMP is released from the target cells and this raises plasma cGMP concentrations.[9 11]

Heart failure can be the end point of various diseases such as myocardial damage caused by myocardial infarction, primary or secondary dilated cardiomyopathy, or valvar disease.[12 13] It has become evident that haemodynamic and neuroendocrine factors participate in the pathophysiology of chronic heart failure. Immunoreactive C-terminal and N-terminal cleavage products of pro-ANP as well as β-ANP are raised in the plasma of patients with congestive heart failure because of enhanced production and release from the atria, additional secretion from the ventricle, and reduced clearance in the liver and the kidneys. Circulating BNP and cGMP are also raised in patients with congestive heart failure.[14-16] The extent of the increase in natriuretic peptides is of prognostic value in congestive heart failure.[17-19] There is also some evidence that natriuretic peptides are of independent prognostic significance after myocardial infarction.[20]

Natriuretic peptides may also serve as markers for asymptomatic left ventricular (LV) dysfunction. The importance of early identification of patients with asymptomatic left ventricular dysfunction is evident from the results of the SOLVD study.[21] Early treatment with the angiotensin converting enzyme (ACE) inhibitor enalapril prevented or retarded the development of heart failure in patients with left ventricular ejection fraction (LVEF) < 35 % and no clinical signs of heart failure.

Recent studies have focused on the role of natriuretic peptides as markers for asymptomatic left ventricular dysfunction.[22-24] These studies were carried out for different natriuretic peptides, measured in different study populations and the results were partly con-

flicting. Our study adds important information to these data and may explain some of the existing discrepancies. The main purpose of our study was to compare the power of CT-ANP, β-ANP, NT-ANP, BNP, and cGMP in detecting asymptomatic left ventricular dysfunction. We also studied whether post-stress measurements or combined measurements improved the detection of patients with asymptomatic left ventricular dysfunction.

Patients and methods
STUDY POPULATION
We prospectively studied consecutive subjects between September 1993 and June 1994, who were referred for rest and exercise radionuclide ventriculography at the Centre for Cardiac Rehabilitation Grossgmain. Only patients in sinus rhythm and without frequent supraventricular and ventricular beats were included in the study. They all had normal renal (serum creatinine < 106 μmol/l) and normal hepatic function. Patients with hypertension who responded poorly to treatment, those with haemodynamically relevant valvar disease, and patients with peripheral artery disease were excluded from the study. A total of 107 subjects entered the study (83 men and 24 women; mean age 58·9 (range 25–78). Coronary artery disease had been diagnosed in more than half those in each study group. In total 78 (72·9%) participants had confirmed coronary artery disease and 64 (60%) had had at least one myocardial infarction, which had been diagnosed by electrocardiographic criteria and an increase in serum enzymes. Patients entered the study no earlier than 10 weeks after myocardial infarction. Five patients had idiopathic dilated cardiomyopathy; 27 participants were referred to rule out coronary artery disease; and 19 (17·8%) had a history of treated hypertension. Patients with systolic left ventricular dysfunction were categorised according to New York Heart Association (NYHA) classification on the basis of physical examination, detailed medical history, and LVEF determined by radionuclide ventriculography: 37 patients were NYHA class I and 32 were NYHA class II. The control group contained 38 subjects who had normal systolic left ventricular function at rest and during exercise (rest LVEF > 55% and exercise LVEF > 65%). None of the controls had signs of left ventricular diastolic dysfunction on Doppler echocardiography. Informed consent was obtained from all study participants.

STUDY PROTOCOL
All medication was stopped at least 12 hours before the study except for antihypertensive agents. Patients remained supine for at least 20 minutes before the first blood samples for peptide and cGMP measurements were drawn from a cannula in a cubital or forearm vein. The second blood sample was taken 3 minutes after exercise ended.

Rest and exercise LVEFs were determined by ECG-gated equilibrium radionuclide ventriculography in supine subjects. An Elscint Apex SR camera and an Elscint Apex 1 computer system (Elscint Medical Technology, Austria) were used for data acquisition. Red blood cells were labelled by an in vitro labelling method with 700–900 MBq technetium-99m as described before.[25] Time-activity curves were obtained in phase mode using 16 frames per cycle. LVEF was determined (after background correction) independently by two experienced technicians using a semiautomatic operator-interactive computer program. As a measure of diastolic function the peak filling rate (PFR) was calculated. It is defined as the ratio between the maximum of activity curve during diastole and end diastole and is expressed as end diastolic volume/s. It was calculated by an Elscint Apex computer package, which uses an 5 point algorithm from the activity curve. In all groups PFR closely correlated with LVEF. Rest and stress imaging was carried out from the left anterior oblique (LAO) camera position.

Stress testing was carried out with the subject supine on a bicycle ergometer table starting with a workload of 25 W. This was increased by 25 W every 3 min until the maximum workload was reached. Blood pressure was measured by a standard manometer cuff technique. In most cases exercise was stopped by the study participants because of exhaustion or dyspnoea, in some patients it was stopped by the physician because of angina pectoris and significant signs of ischaemia in the ECG.

MEASUREMENT OF NATRIURETIC PEPTIDES AND CGMP
Blood was collected in prechilled tubes containing EDTA (ethylendiamine-tetraacetic acid 1·5 g/l) and immediately centrifuged at 4°C at 3000 rpm for 10 minutes before the plasma was frozen at −30°C until measurement, which was carried out within one month after blood collection.

Plasma concentrations of CT-ANP, β-ANP, NT-ANP, and BNP were measured by commercially available radioimmunoassays (RIK 8798, 9105, 9129, and 9086, Peninsula Laboratories, Belmont, CA, USA).[23 26] In brief, 1 ml (ANPs) and 2 ml (BNP) of plasma were acidified with equal volumes of 0·1% trifluoroacetic acid and the acidified plasma was applied to a C_{18} cartridge (Sep column 1, Peninsula) for extraction of peptides from plasma after cartridges were pre-activated by washing with 60% acetonitrile in 0·1% trifluoroacetic acid. After washing with of 0·1% trifluoroacetic acid, peptides were eluted with 60% acetonitrile in 0·1% trifluoroacetic acid. The eluant was evaporated to dryness in a centrifugal vacuum concentrator. The residue was resuspended in assay buffer and peptides were assayed according to the Peninsula Laboratories protocol. The mean recoveries through the extraction process were 82% (CT-ANP), 71% (β-ANP), 67% (NT-ANP), and 84% (BNP). Natriuretic peptide concentrations given in the text were not corrected for recovery. The detection limits were 1pg/tube for all four assays. The intra-assay coefficients

Table 1   Clinical characteristics of normal controls and patients with symptomless and symptomatic left ventricular dysfunction

Variable	Controls (n = 38)	NYHA I (n = 37)	NYHA II (n = 32)
Age (years)	57·5 (8·6)	57·6 (10·3)	62·1 (10·8)*
Body mass index	27·4 (4·1)	26·6 (3·6)	25·6 (2·6)
Males (%)	78·9	81·1	71·8
CAD (%)	60·5	72·9	87·5
Myocardial infarction (%)	55·3	59·5	65·6
Hypertension (%)	18·4	16·2	18·7
Drug treatment:			
β Blockers	18·4	24·3	18·7
Calcium channel blockers	13·2	16·2	15·6
ACE inhibitors	15·7	35·1	56·2
Diuretics	0	8·2	31·2
Digitalis	0	0	21·8
Nitrates	7·9	24·3	46·9

*Mean age of NYHA II patients was significantly different from controls, (P = 0·015); CAD, coronary artery disease; Body mass index was calculated with the formula body weight / height in m².
For age and body mass index, mean (SD) is shown; for the other data, percentages of the number of individuals within the groups are given.
Patients with more than one disease or taking more than one drug were listed in each group separately.

of variation were 7·3% (CT-ANP), 6·1% (β-ANP), 4·3% (NT-ANP), and 8·5% (BNP). The inter-assay coefficients of variation were 11·2% (CT-ANP), 12·5% (β-ANP), 10·3% (NT-ANP)l, and 10·5% (BNP).

Plasma cGMP was measured after plasma ethanol extraction. We added 1 ml ethanol to 250 μl of plasma, mixed it, and centrifuged it for 15 minutes and collected the supernatant. The precipitate was washed with 500 μl of ethanol and centrifuged for 15 minutes again. The supernatants were combined and evaporated to dryness at 56°C under a stream of nitrogen. The average recovery of the extraction procedure was 99·8%. The residues were redissolved in 1 ml assay buffer and 500 μl was then used in the assay for acetylation. Immunoreactive cGMP was measured with a ¹²⁵I-labelled radioimmunoassay (cGMP-assay RPA 525; Amersham International, Amersham, Buckinghamshire). The upper limit of the reference interval using this proce-

dure is 6·6 nmol/l and the intra and inter assay coefficients of variation were 6·4% and 9·2%, respectively. [11]

Plasma creatinine concentrations were determined enzymatically with reagents from Boehringer Mannheim (Mannheim, Germany).

STATISTICAL ANALYSIS

For statistical analysis we used the SPSS for Windows 6·0 (SPSS, USA, 1993) computer package. Variables shown in tables 1 and 2 were compared using the Wilcoxon rank sum W test. The Tukey HSD (honest significance difference) with unequal sample size was applied to compare peptide concentrations in table 3. Comparisons were carried out on log transformed values for all peptides. The Wilcoxon matched-pairs signed-ranks test for dependent samples was used to compare rest and post-stress measurements. For description of the diagnostic performance (correctly and falsely classified patients) of each variable a receiver-operating characteristic (ROC) plot analysis was carried out.[27] ROC plot curves and areas under the curves were calculated by maximum likelihood estimation of binormal receiver operating characteristic curves and the areas under the curves were compared using the software CLABROC and LABROC1.[28] CHAID (χ^2 automatic interaction detector) analysis is part of the SPSS software package and was used for testing the usefulness of combined natriuretic peptide and cGMP measurements.

Results

Demographic data on the patients with asymptomatic and symptomatic left ventricular dysfunction are compared with the control

Table 2   Left ventricular systolic and diastolic function, blood pressure, and maximal workload among controls and patients with asymptomatic and symptomatic left ventricular dysfunction (means (SD))

| Variable	Controls (n = 38)	NYHA I (n = 37)	Controls v NYHA I	NYHA II (n = 32)	Controls v NYHA II
LVEF rest (%)	60·2 (6)	47·5 (8)	P < 0·0001	36 (13)	P < 0·0001
LVEF stress (%)	61·6 (8)	46·1 (8)	P < 0·0001	34·5 (13·8)	P < 0·0001
PFR rest	2·01 (0·6)	1·66 (0·6)	P < 0·001	1·42 (0·5)	P < 0·001
PFR stress	4·11 (1·2)	3·49 (1·0)	P = 0·0301	2·31 (1·0)	P = 0·0301
BP systolic rest	142 (18)	139 (20)	P = 0·418	142 (27)	P = 0·813
BP diastolic rest	83 (11)	84 (9)	P = 0·688	83 (16)	P = 0·971
BP systolic stress	202 (28)	191 (29)	P = 0·632	172 (27)	P = 0·0002
BP diastolic stress	85 (15)	87 (13)	P = 0·124	86 (17)	P = 0·947
Maximal workload	93·4 (35)	86·5 (28)	P = 0·7591	49·4 (20)	P < 0·0001

LVEF, left ventricular ejection fraction; PFR, peak filling rate (end diastolic volume/s); BP, blood pressure (mm Hg).

Table 3   Plasma peptide measurements in controls compared with patients with symptomless and symptomatic left ventricular dysfunction (means (SD)) at rest and post-stress

Comparison	C-ANP (ng/l)	β-ANP (ng/l)	N-ANP (ng/l)	BNP (ng/l)	cGMP (pmol/l)
			Rest measurements		
Controls	54·3 (22·6)	71·5 (60·9)	902 (408)	22·8 (17·7)	3·5 (0·9)
NYHA I	77·9 (72·0)	119 (155·4)	1071 (760)	42·3 (38·9)	4·4 (2·4)
C v NYHA I	P = 0·264	P = 0·501	P = 0·630	P = 0·004	P = 0·739
NYHA II	128·9 (88·1)	141·4 (117·1)	1632 (880)	72·6 (74·2)	6·3 (3·2)
C v NYHA II	P = 0·0004	P = 0·089	P = 0·0005	P = 0·0001	P = 0·003
			Post-stress measurements		
Controls	164 (92)	167 (139)	1306 (593)	30·8 (24)	8·2 (3·3)
NYHA I	215 (151)	285 (382)	1869 (2343)	61·2 (51)	8·9 (4·2)
C v NYHA I	P = 0·227	P = 0·162	P = 0·749	P = 0·004	P = 0·888
NYHA II	240 (136)	258 (224)	2068 (1081)	92·1 (82)	10·2 (4·9)
C v NYHA II	P = 0·052	P = 0·225	P = 0·006	P = 0·0001	P = 0·212

C v NYHA I, controls compared with patients in NYHA class I; C v NYHA II, controls compared with patients in NYHA class II (age was used as a covariate).

Figure 1 Correlation between left ventricular ejection fraction at rest and plasma BNP concentrations among patients with (A) asymptomatic LV dysfunction (NYHA I, n = 37), (r = −0·0086, P = 963) and (B) symptomatic patients (NYHA II, n = 32), (r = −0·61, P < 0·001).
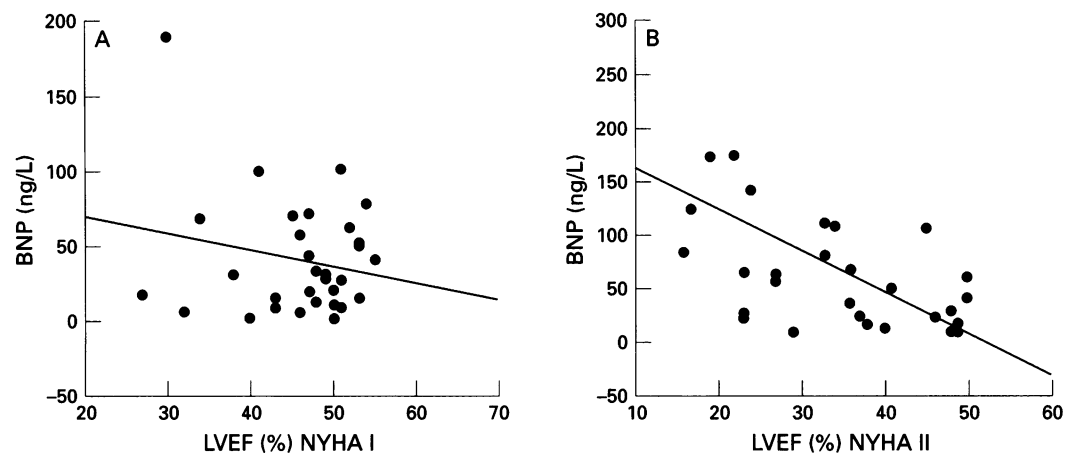
group in table 1. There was no significant difference between the three groups in mean body mass index. Mean age was similar in the NYHA I group (57·6 years) and the controls (57·5 years). The mean age of the NYHA II patients (62·1 years) was significantly higher than that of the controls. Prevalence of coronary artery diseases (CAD), myocardial infarction (MI), and treated hypertension among patients and controls as well as drug treatments are shown in table 1. There was no statistically significant difference between NYHA I patients and controls in the prevalence of CAD, MI, and hypertension.

LVEF, PFR, blood pressure, and maximal work loads are compared in table 2. All these variables were significantly different in symptomatic patients and controls except for blood pressure during rest. The comparison of asymptomatic patients with LV dysfunction with controls showed significantly lower LVEF during rest (P < 0·001) and stress (P < 0·001) and significantly lower PFR during rest (P < 0·001) and stress (P = 0·030) in asymptomatic patients. In contrast to these highly significant differences there was no significant difference in maximal workload.

Within the control group and the group of patients with symptomless left ventricular dysfunction no significant correlation between left ventricular ejection fraction and circulating natriuretic peptides and cGMP was found. However, in the group of patients with symptomatic heart failure (n = 32), LVEF was significantly inversely correlated with CT-ANP (r = −0·54, P = 0·002), NT-ANP (r =

−0·52, P = 0·002) and BNP (r = −0·61, P < 0·001). The lack of correlation of natriuretic peptides among patients with asymptomatic LV dysfunction and significant for BNP correlation among symptomatic patients is shown in fig 1.

Mean plasma concentrations for all natriuretic peptides and for cGMP significantly increased during exercise among controls, NYHA class I patients, and NYHA class II patients. In table 3 mean values (SD) for circulating natriuretic peptides and cGMP of the control group are compared with those in patients with NYHA class I and NYHA class II disease. The group of patients with symptomatic left ventricular dysfunction (NYHA class II) had significantly higher concentrations for all natriuretic peptides and cGMP than the control group. These significant differences were observed at rest as well as post-stress. For patients with symptomless left ventricular dysfunction (NYHA class I) plasma concentrations of all peptides and cGMP were higher than in the controls, again both at rest and post-stress. However, the difference was statistically significant only for BNP (rest measurement P = 0·004 and post-stress measurement P = 0·004).
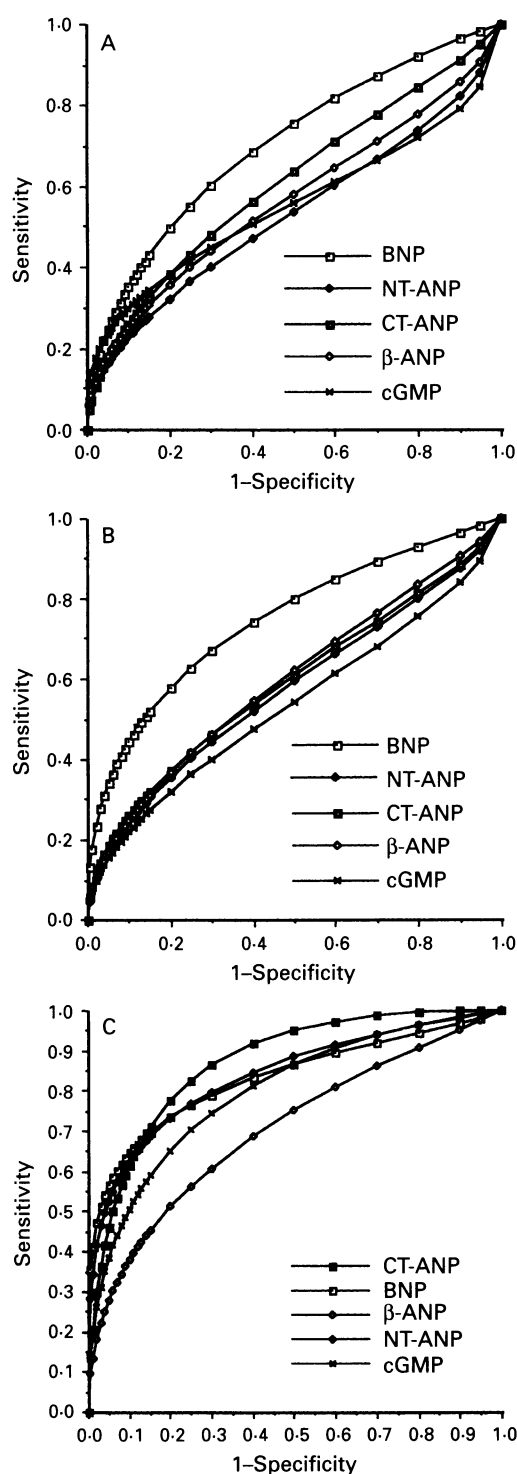
We looked for correlations between the natriuretic peptides and cGMP in the controls and in NYHA class I and NYHA class II patients at rest and after exercise. For each peptide and for cGMP, plasma concentrations at rest correlated highly significantly with post-stress measurements with correlation coefficients ranging from r = 0·61 for CT-ANP in the control group to r = 0·89 for BNP in the NYHA II group (data not shown). Within the three groups significant correlations of CT-ANP, β-ANP, and NT-ANP with each other were found at rest and after exercise. Differences in BNP between controls and NYHA I patients were sought by comparing correlation coefficients between BNP, NT-ANP, and cGMP in controls and symptomless patients (table 4). In the control group we found no correlation between BNP and NT-ANP either at rest or after exercise. Among NYHA class I patients (and NYHA II patients, data not shown) BNP correlated highly significantly with N-ANP at rest as well as after exercise.

Table 4 Correlations between N-ANP, BNP, and cGMP among controls and patients with symptomless left ventricular dysfunction (NYHA I) under rest (A) and stress conditions (B)

| | Controls | | NYHA I | |
	N-ANP	cGMP	N-ANP	cGMP
(A) Rest				
BNP	r = 0·20 P = 0·224	r = 0·15 P = 0·377	r = 0·56 P < 0·001	r = 0·31 P = 0·63
cGMP	r = 0·61 P < 0·001		r = 0·65 P < 0·001	
(B) Post-stress				
BNP	r = 0·16 P = 0·339	r = 0·01 P = 0·963	r = 0·52 P = 0·001	r = 0·17 P = 0·312
cGMP	r = 0·68 P < 0·001		r = 0·68 P < 0·001	

Spearman correlation coefficients r are given in the upper line and significance levels P at 95% confidence intervals are given below.

Figure 2 Diagnostic performance of natriuretic peptides and cGMP in identifying patients with asymptomatic left ventricular dysfunction (A) at rest and (B) three minutes after exercise ended. (C) Diagnostic power to detect patients with a resting ejection fraction below 35%, most of which were symptomatic. Receiver-operating-characteristic (ROC) plots curves for CT-ANP, β-ANP, NT-ANP, BNP, and cGMP for measurements are shown. A diagonal line represents a worthless test (0·5). The larger the area under the ROC curve the better the discriminative power of the test.

Table 5 BNP as a predictor of symptomless left ventricular dysfunction at a cutoff value of 30 ng/l at rest and 40 ng/l post stress

	Rest	Post-stress
Sensitivity	0·58	0·65
Specificity	0·76	0·84
Positive predictive value*	0·71	0·80
Negative predictive value†	0·64	0·71
Efficiency‡	0·67	0·75
Positive likelihood ratio§	2·4	2·2

*True positive test results of all positive test results.
†True negative test results of all negative test results.
‡True positive and negative test results of all positive and negative test results.
§Sensitivity/(1 − Specificity).

nificant differences. As examples, sensitivity, specificity, positive predictive value, negative predictive value, efficiency, and positive likelihood ratio for rest and post-stress BNP are summarised in table 5. Optimal cutoff values according to ROC analysis (discriminator with the largest deviation from the diagonal line in the plot) were used to calculate the variables mentioned above.

The power of natriuretic peptides to detect asymptomatic patients with left ventricular dysfunction might potentially be improved by combining peptide measurements. We investigated this posibility by applying a CHAID model. However, combined measurement of the natriuretic peptides under investigation and cGMP did not improve the efficiency of single measurement of BNP at rest or after exercise (data not shown).

We also studied the ability of natriuretic peptide and cGMP concentrations to correctly identify patients with a resting ejection fraction below 35%, regardless of their NYHA classification. Sixteen of 107 study participants had a LVEF < 35%. ROC blot analysis showed that CT-ANP had the best discriminative power for with a sensitivity of 0·81, a specificity of 0·88, a positive predictive value of 0·54, a negative predictive value of 0·96, and an efficiency of 0·87. The efficiencies of NT-ANP, BNP, and cGMP were not significantly different from CT-ANP. Only β-ANP showed a significantly lower predictive value (fig 2C).

To compare the ability of natriuretic peptides and cGMP to correctly classify subjects into groups with normal and reduced left ventricular function (NYHA class I) we constructed ROC plots[27] for rest (fig 2A) and stress measurements (fig 2B). The area under the curve is an excellent variable for comparing different predictive tests under investigation. For BNP it was 0·70 (0·06) at rest and 0·75 (0·06) post stress. For CT-ANP, β-ANP, NT-ANP, and cGMP it ranged from 0·53 (0·07) for rest NT-ANP to 0·61 (0·06) for rest CT-ANP. At rest as well as after exercise BNP discriminated significantly better between NYHA I patients and controls than the other peptides and cGMP. Among NT-ANP, β-ANP, CT-ANP, and cGMP there were no sig-

Discussion

Several studies have shown that natriuretic peptides are significantly raised in symptomatic congestive heart failure. This was confirmed by the present study for CT-ANP, β-ANP, NT-ANP, BNP, and cGMP by comparing patients in NYHA class II with a control group. We also found significant correlations between left ventricular ejection fraction and plasma concentrations of BNP, NT-ANP, and CT-ANP in patients with symptomatic LV dysfunction, with the correlation being strongest for BNP. This accords with recent reports by Choy et al[29] and Motwani et al.[24] Both groups found better correlations between LVEF and BNP than between LVEF and ANP.

Our study concentrated on the power of natriuretic peptides to identify patients with symptomless left ventricular dysfunction,

because these patients could benefit from early treatment with ACE inhibitors. Comparatively few and partly conflicting data are available regarding this question. Winters *et al*[5] showed that immunoreactive parts of the N-terminal ANP could discriminate a group of five patients classified as NYHA I from normal controls. In a substudy of the SOLVD investigation Francis *et al*[22] found significantly raised concentrations of ANP in patients with an LVEF of 35% or less but without overt congestive heart failure. In a recent publication Lerman *et al*[23] reported that NT-ANP is an excellent marker for early identification of patients with symptomless heart failure, with high sensitivity (90%) and specificity (92%). Choy *et al* studied 75 survivors of a recent myocardial infarction to see whether patients with a LVEF < 40% could be identified by clinical assessment, echocardiography, or measurement of ANP and BNP. Measurement of plasma BNP concentrations reached a sensitivity (84%), similar to that of qualitative visual assessment echocardiography (82%). Plasma ANP concentration was less sensitive (64%).[29]

One of the main goals of our study was to define a control group and a group of symptomless patients as accurately as possible. In contrast to the studies mentioned above our study gives a more precise definition of symptomless patients with left ventricular dysfunction. As well as reporting no signs of congestive heart failure in response to detailed questioning, our NYHA I patients showed no significant difference in the mean maximal work load compared with controls. Our definition of normal LVEF was above 55% at rest or above 65% at peak exercise. Although the definition of normal left ventricular ejection fraction measured by radionuclide ventriculography shows considerable variation between different laboratories, we can assume that our cutoff value for normal LVEF lies above those chosen in the other reports. On the basis of our definition of left ventricular dysfunction, patients in NYHA class I had higher plasma concentrations of CT-ANP, β-ANP, NT-ANP, BNP, and cGMP than the controls. However, only BNP was significantly higher in patients in NYHA class I than in the controls and therefore was a better discriminator for asymptomatic LV dysfunction than the immunoreactive C-terminal and N-terminal parts of ANP. This finding accords with that of Choy *et al*.[29] Our findings do not confirm the results of Lerman *et al*,[23] who found that NT-ANP was a powerful marker for patients with symptomless LV dysfunction. In our study neither CT-ANP nor NT-ANP were suitable for detecting asymptomatic LV dysfunction. A possible explanation for the differing results might be differences in selection criteria used for NYHA classification. Lerman *et al* did not compare maximal work load in the NYHA I patients and controls. Furthermore the predictive power of ANP and BNP for asymptomatic LV dysfunction cannot be compared because BNP was not measured by Lerman *et al*. In our study, although cGMP is a second messenger for BNP, it could not discriminate between NYHA I patients and controls at rest and 3 minutes after exercise. This confirms our previous results with cGMP. However, in an earlier study we showed that cGMP was a marker for symptomless left ventricular dysfunction when measured 30 minutes after ergometric exercise.[26] Because natriuretic peptides are known to have very short half lives we did not carry out late measurements in the present study.

For correlations between natriuretic peptides and cGMP there was a clear difference between NYHA class I patients and controls in terms of BNP relations. At rest as well as post-stress BNP did not correlate with NT-ANP in the control group. However, BNP and NT-ANP were highly correlated in the group containing NYHA I patients. This observation might reflect a relation between atrial and ventricular dysfunction in mild heart failure. This difference and the fact that BNP was the best marker for symptomless left ventricular dysfunction in our study may reflect the different sites of secretion of BNP and ANP. BNP is mainly secreted from the ventricle, whereas ANP is primarily secreted from the atrium. An additional secretion of ANP from the left ventricle in addition to the atrium has been reported in severe congestive heart failure.[30] Secretion of ANP from the ventricle might not have influenced the results of our study, because our study population did not contain patients in NYHA classes III and IV. Post-stress measurements and combined measurements based on an CHAID algorithm, which was carried out in our study, extended the usefulness of previously available data. However, this approach did not add important information to rest measurement and might not justify cost and effort in routine diagnosis.

We also studied the ability of natriuretic peptides and cGMP to identify patients with markedly impaired LVEF. By doing so we did not apply our criteria for asymptomatic left ventricular dysfunction because most of these patients (LVEF < 35%) were symptomatic. As we did for NYHA I patients we used ROC plot analysis to study the efficiency of these factors to correctly pick up patients with a LVEF < 35% in our sample. With this approach we obtained similar results for NT-ANP, CT-ANP, BNP, and cGMP, the sensitivity (0.81) and the specificity (0.88) tended to be highest for CT-ANP (fig 2C). The predictive performance of β-ANP was significantly lower. Wei *et al*[6] found that β-ANP was highly specific for the diagnosis of heart failure. Our results differ from their results. The reason for this discrepancy might be that the study of Wei *et al*[6] mainly contained patients with severe congestive heart failure and this might greatly have influenced the result.

We suggest that natriuretic peptides and cGMP measured at rest and post-stress have a low diagnostic yield for screening purposes. Our observations are supported by the work of Chaidhal *et al*[31] and Wallen *et al*.[32] These two studies investigated atrial natriuretic pep-

tide concentrations in samples drawn from the general population. Both studies accord with our suggestion that the plasma concentrations of natriuretic peptides in an individual do not have sufficient predictive value for asymptomatic heart failure to justify their use in population screening.

Limitations of our study

The control group was primarily defined on the basis of LVEF. Because radionuclide ventriculography studies in normal volunteers were not possible for ethical reasons we studied patients with coronary artery disease with preserved LV function. This was also the case in the study by Lerman et al.[23] We used Doppler echocardiography to exclude LV diastolic dysfunction in the control group. To compare diastolic function in controls, NYHA I, and NYHA II patients we used the PFR derived from equlibrium ventriculography. We must stress that PFR was calculated from 16 frame aquisition data which might not be sufficient to obtain an accurate *absolute* measure of diastolic function. In our study it was used to rule out major differences in diastolic function between the study groups. In all groups, PFR correlated with LVEF.

More study participants in the NYHA I and NYHA II group were taking β blockers, calcium channel blockers, ACE inhibitors, diuretics, digitalis, and nitrates than in the control group. Because of the variety of drugs that were taken at different doses in comparatively small numbers of patients and controls we did not carry out multivariate analysis using treatment as a covariate. Potential confounding effects of treatment cannot be excluded.

CONCLUSIONS

Our study confirmed that natriuretic peptide secretion was increased in patients with symptomatic and symptomless left ventricular dysfunction. BNP was a better marker for symptomless LV dysfunction than CT-ANP, β-ANP, NT-ANP, and cGMP. In our sample natriuretic peptides and cGMP identified patients with markedly impaired LVEF (< 35%), but we must stress that most of these patients had symptoms. However, even with BNP the sensitivity and specificity were not sufficiently high for screening large populations for symptomless left ventricular dysfunction. Post-stress measurements and combined measurements did not add substantially to the information provided by a single measurement at rest. Our data suggest that of all the natriuretic peptides and cGMP, BNP is the best marker for symptomless LV dysfunction when measured at rest or 3 minutes after exercise. But its relatively low sensitivity and specificity for the detection of completely symptomless patients might limit the use of BNP measurements for a wide routine screening for symptomless left ventricular dysfunction. It could, however, be helpful in monitoring treatment in patients with poor echocardiographic images.

1 Rosenzweig A, Seidmann CE. Atrial natriuretic factor and related peptide hormones. *Annu Rev Biochem* 1991;**60**: 229–55.
2 Wei CM, Heublein DM, Perrella MA, Lerman A, Rodeheffer RJ, McGregor GY, et al. Natriuretic petide system in human heart failure. *Circulation* 1993;**88**: 1004–9.
3 Brandt RR, Wright RS, Redfield MM, Burnett JC. Atrial natriuretic peptide in heart failure. *J Am Coll Cardiol* 1993;**22** suppl A:86A-92A.
4 Edwards BS, Zimmerman RS, Schwab TR, Heublein DM, Burnett JC. Atrial stretch, not pressure, is the principal determinant controlling the acute release of atrial natriuretic factor. *Circulation* 1988;**62**:191–5.
5 Winters CJ, Sallman AL, Baker BJ, Meadows J, Rico DM, Vesely DL. The N-terminus and a 4,000-MW peptide from the midportion of the N-terminus of the atrial natriuretic factor prohormone each circulate in humans and increase in congestive heart failure. *Circulation* 1989;**80**: 438–49.
6 Wei CM, Kao PC, Lin JT, Heublein DM, Schaff HV, Burnett JC. Circulating β-atrial natriuretic factor in congestive heart failure in humans. *Circulation* 1993;**88**: 1016–20.
7 Mukoyama M, Nakao K, Hosoda K, Suga S, Saito Y, Ogawa Y, et al. Brain natriuretic peptide as a novel cardiac hormone in humans. *J Clin Invest* 1991;**87**:1402–12.
8 Lang CC, Choy AM, Struthers AD. Atrial and brain natriuretic peptides—a dual natriuretic peptide system potentially involved in circulatory homeostasis. *Clin Sci* 1992; **83**:519–27.
9 Song DL, Kohse KP, Murad F. Brain natriuretic factor. Augmentation of cellular cyclic GMP, activation of particulate guanylate cyclase and receptor binding. *FEBS Lett* 1988;**232/1**:125–9.
10 Hirata Y, Ishii M, Matsuoka H, Sugimoto T, Iizuka M, Uchida Y, et al. Plasma concentrations of α-human atrial natriuretic polypeptide and cyclic GMP in patients with heart disease. *Am Heart J* 1987;**113**:1463–69.
11 Vorderwinkler KP, Artner-Dworzak E, Jakob G, Mair J, Dienstl F, Pichler M, et al. Release of cyclic guanosine monophosphate evaluated as a diagnostic tool in cardiac disease. *Clin Chem* 1991,**37**:186–90.
12 Tsutamoto T, Kanamori T, Morigami N, Sugimoto Y, Yamaoka O, Kinoshita M. Possibility of downregulation of atrial natriuretic peptide receptor coupled to guanylate cyclase in peripheral vascular beds of patients with chronic severe heart failure. *Circulation* 1993;**87**:70–5.
13 Packer M. Pathophysiology of chronic heart failure. *Lancet* 1992;**340**:88–91.
14 Burnett JC, Kao PC, Hu DC, et al Atrial natriuretic peptide elevation in congestive heart failure in human. *Science* 1986;**231**:1145–7.
15 Haug C, Metzele A, Koch M, Hombach V, Grüner A. Plasma brain natriuretic peptide and atrial natriuretic peptide concentrations correlate with left ventricular end-diastolic pressure. *Clin Cardiol* 1993;**16**:553–7.
16 Jakob G, Mair J, Vorderwinkler KP, Judmaier G, König P, Zwierzina H, Pichler M, Puschendorf B. Clinical significance of urinary cyclic guanosine monophosphate in diagnosis of heart failure. *Clin Chem* 1994;**40/1**:96–100.
17 Gottlieb SS, Kukin ML, Ahern D, Packer M. Prognostic value of atrial natriuretic peptide in patients with chronic heart failure. *J Am Coll Cardiol* 1989;**13**:1534–9.
18 Hall C, Kjekshus J, Eneroth P, Snapinn S. The plasma concentration of N-terminal proatrial natriuretic factor (ANF(1–98) is related to prognosis in severe heart failure. *Clin Cardiol* 1994;**17**:191–5.
19 Swedberg K, Eneroth P, Kjekshus J, Wilhelmsen L, for the CONSENSUS Trial Study Group. Hormones regulating cardiovascular function in patients with severe congestive heart failure and their relation to mortality. *Circulation* 1990;**82**:1730–6.
20 Hall C, Rouleau JL, Moyè L, de Champlain J, Bichet D, Klein M, Sussex B, Packer M. N-terminal proatrial natriuretic factor. An independent predictor of long-term prognosis after myocardial infarction. *Cirulation* 1994;**89**: 1934–42.
21 SOLVD Investigators. Effect of enalapril on mortality and the development of heart failure in asymptomatic patients with reduced left ventricular ejection fraction. *N Engl J Med* 1992;**327**:685–91.
22 Francis GS, Benedict C, Johnstone DE, Kirlin PC, Nicklas J, Liang C, Kubo SH, for the SOLVD Investigators. Comparison of neuroendocrine activation in patients with left ventricular dysfunction with and without congestive heart failure. A substudy of the Studies of Left Ventricular Dysfunction. *Circulation* 1990;**82**:1724–9.
23 Lerman A. Gibbons RJ, Rodeheffer RJ, Bailey KR, McKinley LJ, Heublein DM, et al. Circulating N-terminal atrial natriuretic peptide as a marker for symtomless left-ventricular dysfunction. *Lancet* 1993;**341**:1105–9.
24 Motwani JG, McAlpine H, Kennedy N, Struthers AD. Plasma brain natriuretic peptide as an indicator for angiotensin-converting-enzyme inhibition after myocardial infarction. *Lancet* 1993;**341**:1109–13.
25 Pichler M, Mostbeck G, Friedrich L, Eghbalian F, Gassner A. Scintigraphic left ventricular function during exercise in elderly patients with coronary artery disease. *Eur Heart*

J 1984;5:55–7.

26 Jakob G, Mair J, Pichler M, Puschendorf B. Ergometric exercise improves the sensitivity of cyclic guanosine 3'5'-monophosphate to diagnose asymptomatic left ventricular dysfunction. *Br Heart J* 1995;73:145–50.

27 Zweig MH, Campbell G. Receiver-operating characteristic (ROC) plots: a fundamental tool in clinical medicine. *Clin Chem* 1993;39/4:561–77.

28 Metz CE. Some practical issues of experimental design and data analysis in radiological ROC studies. *Invest Radiol* 1986;24:234–45.

29 Choy AMJ, Darbar D, Lang CC, Pringle TH, McNeill GP, Kennedy NSJ, *et al.* Detection of left ventricular dysfunction after acute myocardial infarction: comparison of

clinical, echocardiographic, and neurohormonal methods. *Br Heart J* 1994;72:16–22.

30 Yasue H, Obata K, Okumura K, Kurose M, Ogawara H, Matsuyama K, *et al.* Increased secretion of atrial natriuretic polypeptide from the left ventricle in patients with dilated cardiomyopathy. *J Clin Invest* 1989;83:46–51.

31 Chaidahl K, Hedner J, Hedner T. Atrial natriuretic peptide and catecholamines in periphereal blood as indicators of cardiac dysfunction in the general population. *Clin Sci* 1988;74:587–94.

32 Wallen T, Landahl S, Hedner T, Hedner J, Hall C. Atrial peptides, ANP(1–98 and ANP(99–126) in health and disease in an elderly population. *Eur Heart J* 1993;14:1508–13.

IMAGES IN CARDIOLOGY

Anomalous origin of the left coronary artery from the pulmonary artery
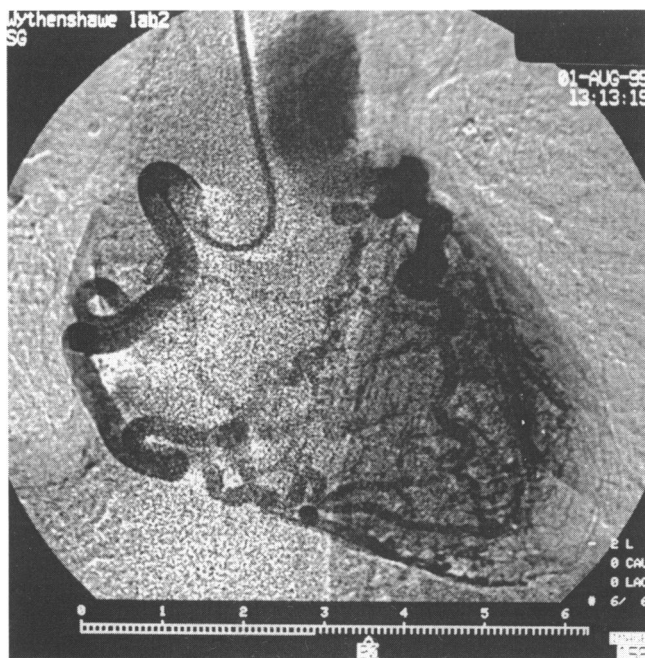
The figure shows a digitally subtracted right coronary angiogram in the posteroanterior projection. The patient was a 42 year old woman who had had exertional breathlessness for 18 months and mild angina for four months. She had a continuous murmur. Echocardiography was normal and an exercise test showed electrocardiographic features of ischaemia at low workload.

The Sones coronary catheter is seen in the ascending aorta with its tip in the right coronary ostium. The right coronary artery is enormous, with extensive tortuous collaterals filling the entire left system and draining into the pulmonary artery, which is also opacified. There was no origin of the left coronary artery from the aorta.

This case is an example of anomalous origin of the left coronary artery from the pulmonary artery. It occurs in 2·5 to 5 per 100 000 live births[1] and was first described in 1886 and then as a syndrome of neonatal angina, infarction and congestive cardiac failure in 1933.[2] Eighty five percent of patients with this anomaly die in childhood. It is rare in adults. Forty four cases surviving to adulthood were described in 1977[3] with 41% being diagnosed postmortem. The oldest age at diagnosis was 49. Treatment is by surgery, for which a variety of procedures have been described. Before operation was possible 80 to 90% of those surviving to adulthood died suddenly, at a mean age of 35 years. In this case the pulmonary origin was ligated and oversewn. Because of the very extensive collateral supply, grafting was not thought necessary.

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1 Karr SS, Giglia TM. Anomalous coronary arteries and coronary artery fistulas in infants and children. *Coronary Artery Disease* 1993;4:139–47.

2 Bland EF, White PD, Garland J. Congenital abnormalities of the coronary arteries: report of an unusual case associated with cardiac hypertrophy. *Am Heart J* 1933;8:787–801.

3 Wilson CL, Dlabal PW, Holeyfield RW, Akins CW, Knauf DG. Anomalous origin of left coronary artery from pulmonary artery. *J Thorac Cardiovasc Surg* 1977;73:887–93.