Elevated NT-proBNP and coronary calcium score in relation to coronary artery disease in asymptomatic type 2 diabetic patients with elevated urinary albumin excretion rate

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Abstract

Background. Elevated plasma N-terminal (NT)-proBNP levels and coronary calcium score (CCS) not only predicts myocardial ischaemia and coronary artery stenosis but also adverse cardiovascular events and mortality in type 2 diabetic patients with an increased urinary albumin excretion rate (UAER), whereas low levels are associated with low frequency of coronary artery disease (CAD) and good prognosis. The underlying causes of poor prognosis in patients with elevated NT-proBNP are not known; thus, we investigated the role of putative asymptomatic CAD in type 2 diabetic patients with UAER >30 mg/24 h and elevated P-NT-proBNP and/or CCS.

Methods. We identified 200 type 2 diabetic patients without known CAD and with normal creatinine levels. Patients with P-NT-proBNP >45.2 ng/L (the median P-NT-proBNP value in this cohort and in accordance with our previous findings) and/or CCS >400 were stratified as high-risk patients for CAD (n = 133) and all other patients as low-risk patients (n = 67). High-risk patients were examined by myocardial perfusion imaging (MPI; n = 109) and/or computer tomography angiography (n = 20) and/or coronary angiography (CAG; n = 86).

Results. All patients received intensive multifactorial intervention. In 70 of 133 (53%) high-risk patients, significant
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CAD was demonstrated by MPI and/or CAG, corresponding to 35% (70/200) of the total cohort. Among high-risk patients, CCS but not P-NT-proBNP was paralleled by increased prevalence of significant CAD and in the 86 patients where CAG was performed, a CCS < 100 had a negative predictive value for coronary artery stenosis of 94% (P = 0.04).

Conclusions. Our study revealed that >50% of asymptomatic type 2 diabetic patients with UAER > 30 mg/24 h had significant CAD based on risk stratification with P-NT-proBNP and CCS. This provides some explanation to the previously reported poor prognosis in these asymptomatic patients. Optimized cardio protective treatment in these patients is warranted.

Keywords: asymptomatic coronary artery disease; coronary calcium score; micro/macrolumubinuria; plasma NT-proBNP; type 2 diabetes

Introduction

Patients with type 2 diabetes and an elevated urinary albumin excretion rate (UAER) have a poor prognosis and the most important determinant of their excessive morbidity and mortality is cardiovascular disease (CVD). Brain natriuretic peptide (BNP) and its cleavage product N-terminal (NT)-proBNP are released from the heart in response to augmented myocardial wall stress. Elevated plasma BNP and NT-proBNP (P-NT-proBNP) levels are established risk factors in patients with heart failure [1]. In addition, minor increases in P-NT-proBNP, below levels seen in heart failure, have been associated with poor outcome in the general population [2] and in patients with stable coronary artery disease (CAD) [3]. In a recently published prospective study with 15 years of follow-up, we identified P-NT-proBNP as a powerful predictor of mortality in type 2 diabetic patients, independent of UAER. In that study, 80% of patients in the upper P-NT-proBNP tertile (P-NT-proBNP > 103 ng/L) died compared to 30% of patients in the lower tertile (P-NT-proBNP < 41 ng/L; P < 0.001), and patients in the lower P-NT-proBNP tertile had a cardiovascular event rate of only 1% per year during the first 10 years of follow-up compared to 6% per year in the upper P-NT-proBNP tertile [4]. Consequently, a P-NT-proBNP < 41 ng/L identified patients with a good prognosis [4]. The underlying causes of poor prognosis in patients with elevated P-NT-proBNP are not known and particularly the role of putative asymptomatic CAD in these patients has never been ascertained. Recent studies suggest that cardiac ischaemia as defined by myocardial perfusion scintigraphy imaging (MPI) may directly influence natriuretic peptide release, independent of changes in left ventricular (LV) function [5–7]. CAD is frequent and often asymptomatic in diabetic subjects, e.g. in part or due to autonomic neuropathy. Recent recommendations suggest that the combination of functional cardiac ischaemia and anatomic coronary artery stenosis identifies the patients who can potentially benefit more from myocardial revascularization [8]. Coronary calcium score (CCS) is established as a non-invasive screening tool to assess the general coronary calcification burden and the cardiovascular risk in several populations [9], including patients with type 2 diabetes and proteinuria [10, 11]. High CCS has been reported to identify symptomatic patients with high risk of anatomic coronary artery stenosis as determined by coronary angiography (CAG), whereas low (CCS < 10) was very accurate for exclusion of patients with significant coronary artery stenosis (<1%) [12]. In addition, patients with CCS < 100 had a very low cardiac adverse event rate (0.4%) during and up to 5 years of observation, while CCS > 1000 was associated with an event rate of 20% per year [10]. The combination of P-NT-proBNP and CCS has never been examined as a marker of the presence of asymptomatic myocardial ischemia and coronary stenosis in type 2 diabetic patients with elevated UAER and elevated CVD risk. In a prognostic setting of mainly non-diabetic patients (87% without diabetes), P-NT-proBNP and CCS were related and had independent and synergistically predictive effect for CVD events in asymptomatic patients without history or signs of CAD followed for 3.9 years [13]. We hypothesized that CAD might already be subclinically present in diabetic patients when these markers are elevated and that the combination of the two, associated with different aspects of CAD, i.e. myocardial ischaemia and coronary stenosis, respectively, therefore was particularly useful in detecting CAD in asymptomatic diabetic patients. The yield of MPI defects was 22% in the Detection of Ischaemia in Asymptomatic Diabetics (DIA)D study of uncomplicated diabetes. We have previously shown that exercise electrocardiography (ECG) yielded inconclusive results in 47% of high-risk type 1 diabetic patients with albuminuria, and therefore, we did not use this method in the present study [14, 15].

Accordingly, we investigated the presence of CAD in subjects with elevated P-NT-proBNP and/or CCS in a cohort of asymptomatic type 2 diabetic subjects with elevated UAER using MPI and CT angiography (CTA) or CAG.

Materials and methods

Patient cohort and clinical measurements

In a cross-sectional study at Steno Diabetes Centre, we identified from January 2007 to February 2008 a cohort of 200 type 2 diabetic patients without known CAD or other cardiac diseases. All patients had persistent UAER >30 mg/24 h and received intensive multifactorial intervention, constituting improved glycaemic, lipid and blood pressure control, as well as antithrombotic therapy and lifestyle modification according to international guidelines [16]. A written invitation to participate in the study was sent to all patients aged 20–70 years from the outpatient clinic [n = 613, 69% males and a mean (standard deviation) age of 47 (8) years]. A total of 72 patients refused to participate. Furthermore, patients were excluded (n = 341) if one or more of the following characteristics were present: normal UAER or non-persistent elevated UAER, significant Q-waves in the 12-lead ECG and relative contra-indications to CTA or CAG, including abnormal plasma creatinine.

P-NT-proBNP was measured in all patients and analysed by an immunoassay as previously described [4]. UAER was measured in 24 h urine collections by an enzyme immunoassay [17]. Diabetic retinopathy was assessed in all patients by fundus photography. Systolic blood pressures in the ankle and big toe were measured by strain gage technique and worst ankle and big toe pressures were recorded and calculated as ankle-brachial and toe-brachial blood pressure index [18]. Peripheral artery disease was defined as ankle-brachial index <0.90 and/or toe-brachial index <0.64 and/or prior surgery for peripheral artery disease [19]. Tests for autonomic neuropathy, heart rate variability assessed by the expiration-inspiration variation of the heart rate and orthostatic blood pressure measurements were performed [20, 21]. In addition, somatic nerve function (vibratory perception threshold) was evaluated by biothesiometry.

Coronary calcium score

Coronary calcium scanning was performed in all patients during a single breath hold using a 16 multidetector-row CT scanner with 3-mm-slice...
thicknes (Philips Precedence MX 8000 IDT 16 slice; Philips Medical Systems, Best, The Netherlands). Quantification of Agatston CCS [22], including intimal and medial calcification in the left main, left anterior descending artery, circumflex artery and right coronary artery, was performed on a separate workstation with dedicated software (Heartbeat-CS, EBW; Philips Medical Systems).

**CAD screening algorithm and study end points**

Patients were stratified into high-risk (the group of primary interest) and low-risk groups: (i) P-NT-proBNP >452 ng/L = high-risk patients (n = 104); this P-NT-proBNP cutoff value was selected for the following reasons. An early separation of a group of high- and low-risk patients was necessary in order to decide individual examinations. We decided to use the median P-NT-proBNP of the first 50 patients examined in the study, all without acute illness or symptomatic heart failure. We have previously demonstrated good prognosis in an unselected cohort of patients with P-NT-proBNP below 41 ng/L, but as the previous study was performed retrospectively on frozen samples, we decided to establish a new cutoff value based on fresh samples, but the new cutoff turned out to be in accordance with our previous study [4]; (ii) P-NT-proBNP ≤452 ng/L and CCS >400 = low-risk patients (n = 29) and (iii) P-NT-proBNP ≤452 ng/L and CCS <400 = low-risk patients (n = 67).

High-risk patients were examined according to the following algorithm (Figure 1): (i) patients with P-NT-proBNP >452 ng/L underwent MPI. Patients with abnormal MPI (n = 55) or CCS >100 (n = 29) were referred for CAG; (ii) patients with P-NT-proBNP ≤452 ng/L and CCS 400–1000 underwent CTA [n = 20; CTA was only used in patients with CCS 400–1000 since severe coronary artery calcifications (CCS >1000) compromise the validity of CTA] [23]. Patients with abnormal CTA were referred for CAG (n = 15) and (iii) patients with P-NT-proBNP ≤452 ng/L and CCS >1000 underwent MPI (n = 9). Patients with abnormal MPI (n = 6) were referred for CAG.

**Fig. 1.** Algorithm used for determination of the prevalence of CAD in 200 asymptomatic type 2 diabetic patients with the use of plasma NT-proBNP, CCS and flow chart showing the selective referral of patients with abnormal test results for further examination. *Patients with plasma NT-proBNP >452 ng/L; †patients with plasma NT-proBNP ≤452 ng/L and CCS ≤400; ‡patients with plasma NT-proBNP >452 ng/L and CCS >400; §significant CAD, defined as the presence of one or more significant myocardial perfusion defects on MPI, and/or one or more significant major epicardial coronary artery stenosis at CAG. In addition, we report the prevalence of coronary artery stenosis alone, including the clinical indication for myocardial revascularization.**

Study end point in the high-risk patients was ‘significant CAD’ defined as the presence of significant myocardial perfusion defects on MPI and/or one or more significant major epicardial coronary artery stenosis at CAG as defined below. In addition, we determined the number of patients with clinical indication for myocardial revascularization (see below).

**CTA, MPI and echocardiography**

CTA was performed with a 64-slice CT scanner (Philips Brilliance CT; Philips Medical Systems), using routine angiographic scanning parameters. MPI was performed using a standard 2-day protocol with 99mTc-Tetrofosmin (600–900 MBq) injected at rest and at peak stress with intravenous dipyridamole (0.6 mg/kg over 4 min, Philips Precedence MX 8000 IDT 16 slice; Philips Medical Systems) [24]. New or worsening defects after stress were considered to represent reversible myocardial ischaemia caused by CAD. The perfusion defects were coded as minor, moderate or severe. All CTA and MPI data were analysed by specialists in nuclear medicine in a blinded manner and disagreements were solved by consensus. Echocardiography was performed using a Philips IE 33 machine (Philips Medical Systems). All patients were examined with conventional two-dimensional and Doppler modalities.

**CAG and invasive myocardial revascularization**

CAG was performed according to standard techniques. Significant major epicardial coronary artery stenosis was defined as left main luminal diameter stenosis >50% or luminal stenosis >70% in the left anterior descending artery, circumflex artery and/or right coronary artery. Stenosis detection was based on visual estimation according to standard clinical practice with evaluation by quantitative CAG analysis (Philips Medical Systems) in indeterminate cases (n = 6). Experienced interventional cardiologists evaluated the CAG and determined the indication for invasive myocardial revascularization. Significant major epicardial coronary artery stenosis was categorized as 1-, 2- and 3-vessel disease and/or left main stenosis and the indication for revascularization was determined according to established guidelines, i.e. coronary artery bypass surgery (CABG) for subjects with left main disease and/or 2- to 3-vessel CAD including proximal left anterior descending artery and with LV ejection fraction <45% and percutaneous coronary intervention (PCI) for subjects with 1- to 2-vessel CAD suitable for PCI, with corresponding reversible myocardial perfusion defect(s) [8, 25]. The study was approved by the local ethics committee and all patients gave written informed consent.

**Statistical analysis**

Data were expressed as means and standard deviation, except for non-normal distribution variables, which were log10 transformed before analysis and were given as medians and interquartile range. As CCS was highly skewed with values of zero, log10 (CCS + 1) was used for analysis. Our primary objective was to examine patients with the highest risk of CAD with the use of P-NT-proBNP and/or CCS for risk stratification, as these markers identify patients with a poor prognosis. In addition, we also evaluated P-NT-proBNP in relation to cardiac ischaemia in patients where MPI was performed and CCS in relation to coronary stenosis in patients where CAG was performed. These associations were assessed by logistic regression models and expressed as unadjusted and adjusted odds ratios (ORs) per 10-fold increase in P-NT-proBNP or CCS, respectively. Covariate adjustments were made for sex, age and variables significantly (P < 0.10) associated with end points in univariate analyses, including P-total cholesterol, P-creatinine, peripheral systolic blood pressure, vibratory perception threshold and heart rate variability unless otherwise stated. ORs and 95% confidence intervals for risk factors and significance level for χ² (likelihood ratio test) were calculated. In addition, the predictive accuracy of the covariate-adjusted models with and without inclusion of P-NT-proBNP or CCS were compared by
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Results

Patient characteristics and risk groups

The clinical characteristics of all patients and the high-risk group compared with the low-risk group are summarized in Table 1. In addition to the use of oral antidiabetic medication and insulin, cardiovascular medical treatment included statins (95% of all patients), aspirin (90%), renin-angiotensin-aldosterone system blockade (98%), diuretics (64%), calcium channel blockers (40%) and beta-blockers (14%).

Table 1. The clinical characteristics of all patients, low- versus high-risk patients and patients with significant CAD (abnormal MPI and/or stenosis on CAG)

<table>
<thead>
<tr>
<th></th>
<th>All patients (n = 200)</th>
<th>Low-risk patients (n = 67)</th>
<th>High-risk patients (n = 133)</th>
<th>P-values</th>
<th>Patients without significant CAD (n = 70)</th>
<th>Patients with significant CAD (n = 70)</th>
<th>P-values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex (male%)</td>
<td>152 (76)</td>
<td>50 (75)</td>
<td>102 (77)</td>
<td>0.747</td>
<td>14 (20)</td>
<td>56 (80)</td>
<td>0.33</td>
</tr>
<tr>
<td>Age (years)</td>
<td>59 (9)</td>
<td>53 (10)</td>
<td>61 (6)</td>
<td>0.001</td>
<td>57 (9)</td>
<td>62 (6)</td>
<td>0.001</td>
</tr>
<tr>
<td>Duration of diabetes (years)</td>
<td>13 (7)</td>
<td>10 (7)</td>
<td>14 (7)</td>
<td>0.001</td>
<td>12 (7)</td>
<td>15 (7)</td>
<td>0.006</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>32.6 (5.8)</td>
<td>32.9 (6.0)</td>
<td>32.4 (5.7)</td>
<td>0.592</td>
<td>32.3 (5.8)</td>
<td>33.1 (5.7)</td>
<td>0.32</td>
</tr>
<tr>
<td>HbA₁c (%)</td>
<td>7.9 (1.3)</td>
<td>8.0 (1.3)</td>
<td>7.8 (1.4)</td>
<td>0.194</td>
<td>7.8 (1.3)</td>
<td>7.8 (1.5)</td>
<td>0.80</td>
</tr>
<tr>
<td>UAR (mg/24 h)</td>
<td>103 (39–230)</td>
<td>105 (44–194)</td>
<td>97 (38–97)</td>
<td>0.814</td>
<td>97 (43–194)</td>
<td>138 (38–491)</td>
<td>0.22</td>
</tr>
<tr>
<td>P-creatinine (μmol/L)</td>
<td>76 (18)</td>
<td>72 (17)</td>
<td>79 (19)</td>
<td>0.007</td>
<td>73 (17)</td>
<td>82 (20)</td>
<td>0.001</td>
</tr>
<tr>
<td>Systolic blood pressure (mmHg)</td>
<td>130 (17)</td>
<td>130 (16)</td>
<td>130 (18)</td>
<td>0.953</td>
<td>129 (17)</td>
<td>132 (18)</td>
<td>0.38</td>
</tr>
<tr>
<td>Total cholesterol (mmol/L)</td>
<td>3.9 (0.9)</td>
<td>4.1 (1.0)</td>
<td>3.8 (0.9)</td>
<td>0.041</td>
<td>4.0 (1.0)</td>
<td>3.8 (0.9)</td>
<td>0.08</td>
</tr>
<tr>
<td>Vibratory perception threshold (mV—mean of both sides)</td>
<td>33 (15)</td>
<td>28 (14)</td>
<td>36 (14)</td>
<td>0.001</td>
<td>30 (14)</td>
<td>39 (13)</td>
<td>0.001</td>
</tr>
<tr>
<td>Heart rate variation during deep breathing (b.p.m.)</td>
<td>7 (4.5–11.5)</td>
<td>9 (7–14)</td>
<td>6 (4–10)</td>
<td>0.001</td>
<td>8 (5–12)</td>
<td>6 (4–10)</td>
<td>0.013</td>
</tr>
<tr>
<td>Retinopathy, n (%)</td>
<td>120 (60)</td>
<td>28 (42)</td>
<td>92 (69)</td>
<td>0.001</td>
<td>71 (55)</td>
<td>49 (70)</td>
<td>0.034</td>
</tr>
<tr>
<td>Periphery artery disease, n (%)</td>
<td>40 (20)</td>
<td>8 (12)</td>
<td>32 (24)</td>
<td>0.025</td>
<td>19 (15)</td>
<td>21 (30)</td>
<td>0.003</td>
</tr>
<tr>
<td>Abnormal heart rate variability, n (%)</td>
<td>130 (65)</td>
<td>38 (57)</td>
<td>92 (69)</td>
<td>0.022</td>
<td>80 (62)</td>
<td>50 (71)</td>
<td>0.12</td>
</tr>
<tr>
<td>Orthostatic hypotension, n (%)</td>
<td>17 (9)</td>
<td>4 (6)</td>
<td>13 (10)</td>
<td>0.40</td>
<td>11 (8)</td>
<td>6 (9)</td>
<td>0.99</td>
</tr>
<tr>
<td>Oral antidiabetic medication, n (%)</td>
<td>170 (85)</td>
<td>57 (85)</td>
<td>113 (85)</td>
<td>0.98</td>
<td>111 (85)</td>
<td>59 (84)</td>
<td>0.83</td>
</tr>
<tr>
<td>Insulin treatment, n (%)</td>
<td>124 (62)</td>
<td>38 (57)</td>
<td>86 (65)</td>
<td>0.28</td>
<td>77 (59)</td>
<td>47 (67)</td>
<td>0.27</td>
</tr>
<tr>
<td>RAAS blockade, n (%)</td>
<td>196 (98)</td>
<td>65 (97)</td>
<td>131 (98)</td>
<td>0.48</td>
<td>127 (98)</td>
<td>69 (99)</td>
<td>0.67</td>
</tr>
<tr>
<td>Statin therapy, n (%)</td>
<td>189 (95)</td>
<td>62 (93)</td>
<td>127 (95)</td>
<td>0.39</td>
<td>122 (94)</td>
<td>67 (96)</td>
<td>0.58</td>
</tr>
<tr>
<td>Aspirin therapy, n (%)</td>
<td>183 (92)</td>
<td>58 (87)</td>
<td>125 (94)</td>
<td>0.076</td>
<td>119 (92)</td>
<td>69 (99)</td>
<td>0.98</td>
</tr>
<tr>
<td>Beta-blocker therapy, n (%)</td>
<td>27 (14)</td>
<td>2 (3)</td>
<td>25 (19)</td>
<td>0.002</td>
<td>12 (9)</td>
<td>15 (21)</td>
<td>0.016</td>
</tr>
<tr>
<td>Calcium channel blockers, n (%)</td>
<td>80 (40)</td>
<td>21 (31)</td>
<td>59 (44)</td>
<td>0.076</td>
<td>47 (36)</td>
<td>33 (47)</td>
<td>0.13</td>
</tr>
<tr>
<td>Use of diuretics, n (%)</td>
<td>128 (64)</td>
<td>35 (52)</td>
<td>93 (70)</td>
<td>0.014</td>
<td>82 (63)</td>
<td>46 (66)</td>
<td>0.71</td>
</tr>
<tr>
<td>Current smoker, n (%)</td>
<td>59 (30)</td>
<td>18 (27)</td>
<td>41 (31)</td>
<td>0.56</td>
<td>36 (28)</td>
<td>23 (33)</td>
<td>0.45</td>
</tr>
<tr>
<td>LV ejection fraction (%)</td>
<td>59.4 (4.9)</td>
<td>59.2 (4.2)</td>
<td>59.5 (5.2)</td>
<td>0.562</td>
<td>59.9 (4.3)</td>
<td>58.6 (5.7)</td>
<td>0.084</td>
</tr>
</tbody>
</table>

**CAD determined by MPI and CAG**

Stratification of patients according to P-NT-proBNP and CCS and subsequent examinations for significant CAD by MPI, CTA and/or CAG are shown in Figure 1. MPI was performed in 96% (105/109) of patients referred for this examination. Three patients refused MPI and one died before MPI in a traffic accident. All patients referred for CTA (n = 20) completed the scan, and 82% (86/105) of cases referred for CAG completed the invasive examination. Of the 19 patients who did not, 9 did not want the examination, 3 died (1 cardiovascular and 2 non-cardiovascular deaths), 4 developed severe non-CVD and 3 were cancelled due to development of contraindications for CAG.

In total, 70 high-risk patients had significant CAD defined by abnormal MPI and/or CAG. Among these patients, 23 patients had coronary artery stenosis, including 12 patients fulfilling established criteria for coronary revascularization. According to our study design and the previously reported good prognosis in these patients and lower prevalence of CAD reported in other patient groups, 67 low-risk patients (P-NT-proBNP ≤ 45.2 ng/L and CCS < 400) were not examined for CAD [4, 7, 10, 12]. Our model therefore

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*aHigh-risk patients = patients with plasma NT-proBNP levels > 45.2 ng/L or plasma NT-proBNP levels ≤ 45.2 ng/L and CCS ≥ 400, all other low-risk patients; nr, not relevant.

**P-values reflect comparison between high- and low-risk patients.

*bP-values reflect comparison of patients with or without significant CAD.

^Data are expressed as means (SD) or medians (interquartile range).

**Heart rate variability ≤ 10 b.p.m. and an orthostatic blood pressure fall > 30 mmHg were considered abnormal.
identified 67% (133/200) high-risk patients in a group of asymptomatic type 2 diabetes patients with elevated UAER and significant CAD in 53% (70/133) of high-risk patients.

MPI showed myocardial perfusion defects in 56% (61/109) of high-risk patients, including 23 patients with large reversible or permanent myocardial perfusion defects. CAG showed at least one significant major epicardial coronary artery stenosis in 27% (23/86) of patients with 1-, 2- and 3-vessel disease in 7, 4 and 12 patients, respectively. Myocardial revascularization was performed in 12 patients according to the predefined treatment protocol and 9 patients underwent CABG (1 with left main stenosis and 8 with 3-vessel disease) and 2 patients underwent PCI. One patient declined CABG. Another patient with colon cancer died 6 days after CABG. All other revascularization procedures were performed without complications. The clinical characteristics of patients with significant CAD are shown in Table 1.

NT-proBNP levels and CCS

As discussed above, the P-NT-proBNP cutoff value (45.2 ng/L) was selected to identify increased risk and was determined as the median P-NT-proBNP in the first 50 patients examined in the study. P-NT-proBNP, CCS and heart rate variation during deep were normalized by log10 transformation and thus the corresponding ORs refer to a 10-fold rise in the variables.

Table 2. The relations between conventional risk factors, plasma NT-proBNP or CCS and the prevalence of significant CAD in 133 high-risk patients assessed by a multivariable logistic regression enter model and displayed as adjusted ORs

<table>
<thead>
<tr>
<th>Predictors of significant CAD (n = 133)</th>
<th>OR_unadj (95% CI)</th>
<th>OR_adj (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>1.03 (0.98–1.09)</td>
<td>0.92 (0.85–1.00)</td>
</tr>
<tr>
<td>Sex (male versus female)</td>
<td>1.48 (0.66–3.32)</td>
<td>2.08 (0.67–6.46)</td>
</tr>
<tr>
<td>Cholesterol per 1 mmol/L increase</td>
<td>0.84 (0.57–1.25)</td>
<td>0.77 (0.47–1.24)</td>
</tr>
<tr>
<td>Plasma creatinine per 10 mmol/L increase</td>
<td>1.25 (1.03–1.53)</td>
<td>1.30 (0.99–1.70)</td>
</tr>
<tr>
<td>Vibratory perception threshold per 10 mV increase</td>
<td>1.35 (1.05–1.75)</td>
<td>1.44 (1.01–2.05)</td>
</tr>
<tr>
<td>Heart rate variation during deep breathing per 10-fold increase</td>
<td>0.87 (0.28–0.73)</td>
<td>2.07 (0.46–9.33)</td>
</tr>
<tr>
<td>Peripheral toe systolic pressures per 10 mmHg decrease</td>
<td>1.07 (0.97–1.18)</td>
<td>1.01 (0.88–1.15)</td>
</tr>
<tr>
<td>P-NT-proBNP per 10-fold increase</td>
<td>1.26 (0.55–2.89)</td>
<td>2.04 (0.65–6.46)</td>
</tr>
<tr>
<td>CCS per 10-fold increase</td>
<td>1.83 (1.25–2.69)</td>
<td>2.57 (1.47–4.50)</td>
</tr>
</tbody>
</table>

*Adj., adjusted for the other variables in the column. Column 2 show unadjusted ORs and confidence intervals (CIs) and column 3 show adjusted ORs. NT-proBNP, CCS and heart rate variation during deep were normalized by log10 transformation and thus the corresponding ORs refer to a 10-fold rise in the variables.

Fig. 2. CCS and the frequency (%) of significant CAD, coronary artery stenosis and patients with clinical indication for myocardial revascularization among high-risk patients.

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median CCS was 251 (interquartile values 24–872) in patients with P-NT-proBNP above cutoff value compared to 87 (1–426) in patients with P-NT-proBNP below cutoff value (P = 0.003).

Predictors of significant CAD in high-risk patients

As shown in Table 2, among high-risk patients (n = 133), CCS was associated with significant CAD, whereas P-NT-proBNP was not. Furthermore, as shown in Figure 2, increased CCS was paralleled by increased prevalence of significant CAD, coronary artery stenosis and myocardial revascularization in high-risk patients. CCS was related with myocardial perfusion defects in patients where MPI was performed (P = 0.001). This was not seen for P-NT-proBNP (P = 0.32), which probably reflects that few patients with low levels of P-NT-proBNP were investigated (i.e. 29 patients with P-NT-proBNP below cutoff but CCS >400) were examined for significant CAD compared to 59 patients with P-NT-proBNP above cutoff or CCS <400) or could be due to lack of power. Finally, in the 86 patients where CAG was performed, a CCS <100 had a negative predictive value for coronary artery stenosis of 94% (P = 0.04) and for CCS <400, the negative predictive value for myocardial revascularization was 93% (P = 0.003). The AUC was highest (75.7%) in the covariate model that in addition also included CCS compared to 66.8% in the covariate model alone and 50.0% in the United Kingdom Prospective Diabetes Study (UKPDS) model.

Discussion

A main finding in the present study was the high prevalence of asymptomatic CAD in type 2 diabetic patients with UAER >30 mg/24 h, despite the fact that patients had no history of CAD and received multifactorial intervention aimed at CAD prevention. By the use of a simple predefined P-NT-proBNP and CCS algorithm, we were able to identify a high-risk CAD group with significant CAD in 53% of patients. Among patients with significant CAD (n = 70), 23 patients had coronary artery stenosis, including 12 patients fulfilling guidelines for myocardial revascularization. CCS but not P-NT-proBNP was related to our CAD end points in the high-risk patients.

Several studies have demonstrated that elevated urinary albumin excretion identifies a subgroup of diabetic patients with increased morbidity and mortality from CVD, but recent studies have suggested that adding additional markers such as BNP can further stratify the group into patients with high/low risk for events, but the reason for this has not been identified [4, 10, 13]. We have extended these findings by applying a simple risk model to a group of patients with the highest risk of CVD, and we demonstrated that the high-risk patients known to have a poor prognosis had a high prevalence of significant CAD despite their lack of symptoms when investigated with MPI and CAG. Average P-NT-proBNP levels in the present study were slightly lower than in the previous study which included patients with overt heart disease [4]. Our present aim was to more conclusively examine patients with the highest risk of adverse cardiovascular events and prevalence of CAD. In addition to P-NT-proBNP, we also included CCS as a screening marker based on previous work by other investigators [10]. The CCS ≥400 cutoff value was selected in accordance with a previously described threshold of all-cause mortality risk in diabetic patients with proteinuria but also according to the close association with coronary artery stenosis [11, 12]. We therefore combined P-NT-proBNP and CCS as potential markers of myocardial ischaemia or coronary stenosis and suggest a synergistically predictive CVD effect in asymptomatic patients [13]. The selective referral of patients with abnormal results of these two tests for CAD examination resulted in identification of a substantial number of asymptomatic patients with significant CAD identified by MPI and/or CAG. Performance of MPI and/or CAG in the 67 low-risk patients may have identified additional patients with significant CAD. However, 35 of the 67 low-risk patients had CCS <10, which is very accurate for exclusion of patients with significant coronary artery stenosis [12]. Furthermore, a recent study showed that low CCS is also associated with normal MPI results [27]. Indeed, in the latter study, all patients with CCS <10 had normal MPI results, and in patients with CCS 11–100 or 100–399, 97.4% or 88.7% had normal MPI, respectively [27]. In our study, CCS <10 ruled out MPI defects in 74% of patients who underwent MPI (P = 0.001; data not shown).

Our primary objective was to examine patients with the highest risk of CAD by the use of P-NT-proBNP/CCS as risk stratification. Our study, however, also allowed us to evaluate the diagnostic performances of these markers when excluding low-risk patients who did not undergo cardiac examination. To evaluate the relationship between P-NT-proBNP or CCS and CAD, we therefore performed analyses in high-risk patients and two smaller high-risk patient subgroups including patients where MPI was performed (to report any association with cardiac ischaemia) or patients where CAG was performed (to any report association with coronary artery stenosis) accepting the potential selection bias in the latter two groups. In the group with P-NT-proBNP levels above the cutoff value, the actual levels of P-NT-proBNP were not associated with CAD end points in the high-risk patients. It is assumed, but not demonstrated in the present study, that patients with low levels of P-NT-proBNP have fewer CAD end points, in accordance with the lower CCS in this group and the better prognosis. In addition, P-NT-proBNP and patient prognosis may also be related through other factors such as: atherosclerosis in other territories, echocardiography abnormalities other than ejection fraction and cardiac autonomic neuropathy. Increased CCS was paralleled by an increased prevalence of CAD end points in high-risk patients, and CCS <100 ruled out coronary artery stenosis in 94% of patients who underwent CAG. The predictive accuracy of covariate-adjusted screening models utilizing conventional risk factors or UKPDS_CAD risk engine scores were low among high-risk patients, confirming that the current risk engines are not sufficiently predictive of CAD [28]. CCS added to the predictive capability of the covariate model. The true impact of CCS, P-NT-proBNP, MPI, CAG and CTA on mortality and clinical end points in our cohort will be examined by prospective follow-up. Our results highlight the necessity to define prospectively if
baseline NT-proBNP and CCS identify better patients with the worst prognosis compared to traditional risk markers.

Patients in the present study all had normal P-creatinine according to the study inclusion criteria. Reduced kidney function is a well-known risk factor for CAD in diabetic patients and the current CAD frequencies are therefore probably underestimated compared to the prevalence of CAD in an unselected population of type 2 diabetic patients with elevated UAER. The prevalence of significant myocardial perfusion defects in 56% of high-risk patients in our study is higher than the 22% observed in the recent large-scale DIAD type 2 diabetic screening study [14], and this difference most likely reflects different patient selection criteria, e.g. in our study, all patients had elevated UAER compared with only 25% of patients in DIAD. Furthermore, all our patients examined with MPI had elevated P-NT-proBNP and/or CCS. Although microalbuminuria in general is associated with increased cardiovascular morbidity and mortality, we have previously demonstrated that patients with low (<41 ng/L) P-NT-proBNP levels have a very favourable prognosis even in the presence of microalbuminuria, and thus, we have assumed the prevalence of CAD end points was small in the low-risk patients in the present study.

Although the combined presence of MPI perfusion defects and significant stenosis determined by CAG portends the worst adverse prognosis [8, 29, 30], this observation does not necessarily imply that risk reduction can be achieved by therapeutic interventions. Indeed, so far, no randomized controlled trial has investigated the benefit of myocardial revascularization in asymptomatic type 2 diabetic patients with CAD as determined by MPI and CAG. In diabetic patients with symptomatic CAD, a retrospective study has shown a survival benefit with myocardial revascularization compared to optimal medical therapy in patients with moderate to large MPI perfusion defects [31]. The BARI-2D trial recently compared myocardial revascularization (PCI or CABG) with aggressive medical therapy in type 2 diabetic patients with mild or moderate symptomatic CAD [32]. No difference was observed in overall mortality between the treatment groups, but CABG reduced major cardiovascular events as compared to intensive medical therapy in patients with extensive CAD. Since patients in the present study were asymptomatic, our indication for revascularization required both the presence of a proximal major coronary artery stenosis and a corresponding reversible MPI perfusion defect. Earlier studies support the benefit of CABG in asymptomatic patients with severe anatomic CAD associated with MPI defects [8, 25]. Most of our patients, however, did not have severe CAD. Presently, the recommended medical CAD primary and secondary prevention are only slightly different, i.e. the use of aspirin as primary prevention is unclear and treatment targets for low-density lipoprotein are lower in secondary compared to primary prevention. It is not known whether this is also beneficial in patients with asymptomatic CAD. Based on current guidelines, future randomized trials are needed to test whether more aggressive intensive medical treatments and/or newer myocardial revascularization strategies are superior treatments in diabetic patients with asymptomatic CAD. Finally, the present study results may be useful to consider when such primary prevention trials are designed in order to identify high-risk patients, who are most likely to benefit from intervention.

In conclusion, our study revealed that >50% of asymptomatic type 2 diabetic patients with UAER but receiving multifactorial intervention aimed at CAD prevention had significant CAD if high-risk stratification with P-NT-proBNP and/or CCS is applied. However, at present, no additional medical or interventional therapy is recommended in patients with diabetes and asymptomatic CAD, and therefore, screening is not warranted. New treatment strategies in high-risk patients identified in the present study should be investigated.

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Competing interests. P.R reports having received lecture fees from Novartis and Boehringer Ingelheim, and research grant from Novartis, has served as a consultant for Merck and having equity interest in NovoNordisk. Dr. H.-H.P. reports having served as a consultant for Novartis, Merck, Pfizer and Sanofi-Aventis, having equity interest in Merck and NovoNordisk and having received lecture fees from Novartis, Merck, Pfizer and Sanofi-Aventis. H.-H.P. has received grant support from Novartis, AstraZeneca and Sanofi-Aventis.

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