European Heart Journal (2013) **34**, 932–941
doi:10.1093/eurheartj/ehs396



# Evidence of a synergistic association between heart rate, inflammation, and cardiovascular mortality in patients undergoing coronary angiography

**Bríain Ó Hartaigh[1†], Jos A. Bosch[2,3,4*†], Douglas Carroll[2], Karla Hemming[5], Stefan Pilz[6], Adrian Loerbroks[4], Marcus E. Kleber[4], Tanja B. Grammer[4], Joachim E. Fischer[4], Bernhard O. Boehm[7], Winfried März[4,8,9], and G. Neil Thomas[4,5*]**

[1]Department of Internal Medicine/Geriatrics, Yale School of Medicine, Yale University, New Haven, CT 06510, USA; [2]School of Sport and Exercise Sciences, University of Birmingham, Birmingham, UK; [3]Department of Clinical Psychology, University of Amsterdam, Amsterdam, The Netherlands; [4]Institute of Public Health, Social and Preventive Medicine, Mannheim Medical Faculty, Heidelberg University, Mannheim 68135, Germany; [5]Department of Public Health, Epidemiology and Biostatistics, University of Birmingham, Birmingham, UK; [6]Department of Internal Medicine, Division of Endocrinology and Metabolism, Medical University of Graz, Graz 8036, Austria; [7]Department of Internal Medicine I, Division of Endocrinology and Diabetes, Ulm University, Ulm 89070, Germany; [8]Clinical Institute of Medical and Chemical Laboratory Diagnostics, Medical University of Graz, Graz 8010, Austria; and [9]Synlab Academy, Synlab Services GmbH, Mannheim 68165, Germany

*Received 1 June 2012; revised 1 October 2012; accepted 31 October 2012; online publish-ahead-of-print 23 November 2012*

**Aims** Both elevated inflammatory activity and sustained tachycardia reflect unfavourable cardiovascular risk profiles, and there is evidence to suggest the deleterious effects of inflammation are amplified by increased heart rate. The purpose of this study was to assess the interaction between resting heart rate and inflammation in cardiovascular mortality.

**Methods and results** A total of 3267 patients (2283 men), aged 18–95 years, scheduled for coronary angiography, were followed prospectively. By principle component analysis, we developed an overall multi-marker index of inflammation weighting the respective coefficients of five inflammatory markers including: interleukin-6, C-reactive protein, serum amyloid A, neutrophils, and fibrinogen. Cox proportional hazard regression models were employed to evaluate the relationship between inflammation and heart rate with cardiovascular mortality. Across 29 940 person years of follow-up, there were 546 (17%) deaths due to cardiovascular disease (CVD). Significantly, we observed a strong synergistic effect of inflammatory activity and concurrent elevated heart rate. For CVD mortality, patients in the highest quartile of inflammation had an adjusted hazard ratio (95% confidence interval) of 1.84 (1.31–2.57), $P < 0.0001$ if their resting heart rate was $<75$ b.p.m. Substantially, patients had a greater adjusted HR of 7.50 (3.21–17.50), $P < 0.0001$ if their resting heart rate was $\geq 75$ b.p.m.

**Conclusion** The present analyses underline elevated inflammation as a risk factor for cardiovascular mortality. The effects of inflammation appeared to be strongly amplified by a faster resting heart rate. If confirmed by additional studies, this association may prove a useful adjunct for therapeutic approaches to alleviate symptoms and prolong survival.

**Keywords** Resting heart rate • Inflammation • Cardiovascular mortality • Angiography



† B.Ó.H. and J.A.B. contributed equally as first authors.

* Corresponding author. Tel: +44 121 414 8696, Fax: +44 121 414 7878, Email: gneilthomas@yahoo.co.uk (G.N.T.)/Tel: +31 20 525 6819, Fax: +31 20 525 6506, Email: jos.bosch@medma.uniheidelberg.de (J.A.B.)

# Introduction

Coronary artery disease (CAD) represents an important public health burden. In particular, lifetime risk of coronary heart disease (CHD) is around one in two for men and one in three for women over the age of 40.[1] Aside from the more recognized parameters of CAD risk such as smoking, diabetes, hypertension, and dyslipidaemia, compelling evidence has now accumulated in support of chronic inflammation as another risk factor, contributing towards all stages of atherosclerosis from endothelial dysfunction and plaque formation to plaque disruption and thrombosis.[2,3]

Recent evidence indicates that inflammation and sustained tachycardia interact at several levels of the cardiovascular continuum,[4] and may hereby exert a synergistic effect on cardiovascular morbidity and mortality. For example, elevated heart rate increases tensile stress which apart from inducing endothelial injury also increases endothelial permeability to circulating inflammatory mediators.[5] In addition, dysfunctional autonomic nervous activity may underlie both progression of inflammation as well as elevated resting heart rate.[6] Stimulation of efferent vagus nerve activity has been associated classically with normalizing tachycardia, whereas experimental evidence has shown that this process also inhibits inflammation via stimulation of the cholinergic anti-inflammatory pathway.[7,8] Similarly, there is ample evidence that elevated sympathetic activity modifies the inflammatory process and thereby promotes endothelial dysfunction and subsequent atheroprogression.[9,10] Though, clearly, further studies are required to test the notion that a high resting heart rate will exacerbate the effects of inflammation on cardiovascular disease (CVD).

In light of the preceding discussion, this study was undertaken to evaluate the inter-relationship between resting heart rate and inflammation with CVD mortality in a large cohort of German patients enrolled in the LUdwigshafen RIsk and Cardiovascular Health (LURIC) study.

**Table 1 Description of variables measured in the present study**

| Variable | Definition |
|---|---|
| Age | Taken at time of enrolment |
| Sex | Male/female |
| Waist | Measured in cm, horizontally around the smallest circumference between the ribs and iliac crest |
| Body mass index | weight (kg) divided by height ($m^2$) |
| Smoking | Never/ever smoker |
| Physical activity | Recorded using an 11-point scale ranging from bedridden to extremely active, and categorized into 'below average' (not very active), 'average' (usual office work), and 'above average' (heavy work or sports) |
| Arterial hypertension | Systolic $\geq$140 and/or diastolic $\geq$90 mmHg, or anti-hypertensive medication: confirmed by a physician |
| Diabetes mellitus | Fasting glucose $\geq$7.0 mmol/L, or patients receiving anti-diabetic medication: confirmed by a physician |
| Dyslipidaemia | Total cholesterol $\geq$6.2 mmol/L, HDL-cholesterol $<$1.03/1.29 mmol/L (male/female), triglycerides $\geq$1.7 mmol/L, or use of lipid-lowering medication: confirmed by a physician |
| Angina pectoris | Stable (graded severity classes I–IV described by the Canadian Cardiovascular Society); unstable (subdivided into different classes by subjective assessment of clinical symptoms using Braunwald's classification) |
| Family history | Family questionnaire regarding the history of cardiovascular disease in first and second degree relatives |
| Cardiovascular medication use | Use of ACE-inhibitors, β-blockers, or statins |

HDL, high density lipoprotein; ACE, angiotensin converting enzyme.

# Methods

## LURIC Study Population

The LURIC study is a prospective cohort study designed to investigate environmental and genetic risk factors for CVD.[11] A total of 3316 patients (2309 men and 1007 women), aged 18–95 years were enrolled in the LURIC study. A baseline examination was performed between July 1997 and January 2000 in a single tertiary care medical centre in South-West Germany (Herzzentrum, Ludwigshafen). Inclusion criteria were availability of a coronary angiogram, Caucasians of German ancestry to limit genetic heterogeneity, and clinical stability, with the exception of acute coronary syndromes (ACS). An ACS was diagnosed if patients presented within 7 days of onset of symptoms of unstable angina pectoris or acute myocardial infarction (MI), comprising non-ST-elevation MI (troponin T $>$0.1 μg/L) and ST-elevation MI (troponin T $>$0.1 μg/L). Participants with a history of malignancy within the past 5 years, or any predominant non-cardiac disease were excluded from the study. The LURIC study was approved by the institutional review board of the ethics committee at the 'Landesärztekammer Rheinland-Pfalz' (Mainz, Germany) and written informed consent was obtained from all study participants.

## Patient examination

An overall summary of the LURIC study objectives and baseline examination procedures have been published elsewhere.[11] Briefly, all body measures were recorded by trained study nurses on the same day, during early morning, between the hours of 06:00–09:00 a.m. Resting heart rate was obtained by electrocardiography. Five measures were taken 30 s apart, following a 10 min rest in the supine position in a quiet room, with the average derived from the last two measures. Other variables included in the study were selected on the basis of previous literature, data availability, and the possibility of confounding. Details of these variables are provided in *Table 1*.

## Laboratory analysis

Samples of fasted venous blood were collected during baseline examination, with patients in the supine position. Routine laboratory parameters were immediately measured on a daily basis as previously mentioned.[11] Remaining blood samples were snap-frozen and stored in −80 C for further analyses. All markers were processed at the

Institute of Haemostaseology and Transfusion Medicine of the Ludwigshafen General Hospital. Interleukin-6 was measured using a high sensitivity enzyme immunoassay (R&D Systems, Wiesbaden, Germany). High-sensitivity C-reactive protein concentrations were measured by immunoturbidimetric assay (Roche Mannheim, Germany). Serum amyloid A was determined by immunonephelometry (Dade Behring, Marburg, Germany). Fibrinogen was measured using the Clauss method (Roche Mannheim, Germany). Neutrophil count was determined using EDTA whole blood and was later quantified using an automated analyser (Technicon H-1, Bad Vilbel, Germany until December 1998; Advia 120, Siemens Healthcare Diagnostics, Eschborn, Germany since January 1999).

## Outcome measure

The primary endpoint was death due to CVD. Information on vital status was obtained from local community registries. Death certificates were reviewed to classify the deceased into those who died from CVD and non-CVD causes. Death from CVD causes included sudden cardiac death, fatal MI, death due to heart failure, death after intervention to treat CAD, stroke, and other deaths due to heart disease. Two experienced physicians who were masked to any of the study data independently classified the causes of death. In the case of a disagreement concerning the classification, it was discussed and the final decision was made by one of the principal investigators of LURIC (W.M.), who was also masked to the key study variables.

## Statistical methods

For the purpose of this investigation resting heart rate was dichotomized into low (<75 b.p.m., $n = 2391$) and high (≥75 b.p.m., $n = 876$) according to the 75 percentile. The study sample was then summarized by comparing those with a low and a high resting heart rate, reporting means and standard deviations [or medians and interquartile ranges for those variables which were skewed], or by numbers and percentages for categorical variables. The two groups were compared using *t*-tests for continuous data, and the Mann–Whitney *U*-test for those continuous variables which were skewed, as well as $\chi^2$-tests for categorical variables. All inflammatory markers and continuous covariates were checked for normality and those which visually deviated on inspection of the frequency distribution were transformed into the natural (base e) logs. In this study, only the single markers of inflammation required logarithmic transformation. Inflammatory activity can be assessed by a number of correlated markers, such as Interleukin 6, high sensitivity C-reactive protein, serum amyloid A, fibrinogen, and the neutrophil count (*Table 2*). We employed a principle component analysis to extract from the individual markers of inflammation a single weighted multi-marker index of inflammation. In this study, the first principle component accounted for 61.3% of the explained variance and no additional significant principal components were identified. Accordingly, we developed the overall multi-marker index of inflammation by weighting the respective coefficients of each of the five inflammatory markers that contributed towards the primary underlying factor (i.e. inflammation). Kaplan–Meier survival curves with the log-rank test for equality were used to illustrate the predictive ability of inflammation (quartiles) with CVD mortality, stratified by low and high resting heart rate. The association between inflammation quartiles and CVD mortality for those with a low and a high resting heart rate was further tested using Cox proportional hazard survival models, reporting hazard ratios (HR) with 95% confidence intervals (95% CI) and *P* for trend. Three models were employed to test these relationships. The first model was unadjusted; the second model corrected for age and sex; the final model additionally controlled for body mass index, smoking, physical activity, arterial hypertension, type 2 diabetes, dyslipidaemia, angina pectoris, family history of CVD, and cardiovascular medications. To further understand the relationship between inflammation and CVD mortality, the latter analyses were also performed examining the single markers of inflammation in those with a low and a high resting heart rate, respectively. Due to the large heterogeneity of LURIC patients, we re-performed the survival models according to a number of sensitivity checks, which were computed separately. First, we examined the initial findings among those who were clinically stable at baseline. Second, patients with signs of heart failure were removed in order to examine whether the initial findings would be affected. In the current study, heart failure was determined according to a classification developed by the New York Heart Association (i.e. whereby/in whom slight physical activity caused symptoms; inability to carry on any physical activity without discomfort; symptoms of cardiac insufficiency present even at rest).[12] Third, patients accompanied with an ongoing infection who may have spuriously contributed towards elevated inflammation were excluded. Fourth, to minimize potential bias due to subclinical and undetected pre-existing disease at baseline which is associated with increased mortality, we also carried out the analyses after removing patients who died within 12 months from the time of enrolment. Fifth, we excluded those who reported the use of β-blockers, as these agents are known to artificially lower heart rate, which may have biased our initial assignment of patients to the heart rate categories. We then assessed the discriminatory value of the multi-marker index of inflammation by computing the c-statistic and integrated discrimination improvement (IDI), as well as the net reclassification improvement (NRI). For the latter, we chose *a priori* meaningful risk category of <6, 6–20, and >20% 10-year risk of CHD based on the Third Adult Treatment Panel (ATP III) risk classification.[13] Model calibration was evaluated using

**Table 2 Pairwise Pearson correlation coefficients between the individual markers of inflammation**

| | Interleukin-6 | C-reactive protein | Serum amyloid A | Fibrinogen | Neutrophils |
|---|---|---|---|---|---|
| Interleukin-6 | 1.00 | | | | |
| C-reactive protein | 0.58*** | 1.00 | | | |
| Serum amyloid A | 0.52*** | 0.79*** | 1.00 | | |
| Fibrinogen | 0.50*** | 0.73*** | 0.62*** | 1.00 | |
| Neutrophils | 0.33*** | 0.33*** | 0.30*** | 0.33*** | 1.00 |

For the single markers of inflammation, natural log transformations were applied to normalize distributions.
***$P < 0.0001$.

**Table 3 Baseline characteristics of patients according to low and high resting heart rate**

| | Low RHR (<75 b.p.m.) | High RHR (≥75 b.p.m.) | *P*-value |
|---|---|---|---|
| Characteristics | ($n = 2391$) | ($n = 876$) | |
| Cardiovascular mortality, *n* (%) | 366 (15.3) | 180 (21.0) | <0.0001 |
| Age, mean ± SD, years | 62.7 ± 10.5 | 62.3 ± 10.7 | 0.32 |
| Male sex, *n* (%) | 1682 (70.4) | 601 (68.6) | 0.34 |
| Waist circumference, mean ± SD, cm | 98.8 ± 11.6 | 99.5 ± 12.6 | 0.11 |
| Body mass index, mean ± SD, kg/m$^2$ | 27.4 ± 3.9 | 27.7 ± 4.5 | 0.04 |
| Resting heart rate, mean ± SD | 63 ± 7 | 83 ± 8 | <0.0001 |
| Smoking status, *n* (%) | | | |
| Never | 858 (35.9) | 312 (35.6) | 0.89 |
| Ever | 1533 (64.1) | 564 (64.4) | |
| Physical activity, *n* (%) | | | |
| Low | 564 (23.6) | 278 (31.7) | <0.0001 |
| Average | 1277 (53.4) | 443 (50.6) | |
| High | 550 (23.0) | 155 (17.7) | |
| Arterial hypertension, *n* (%) | 1208 (50.5) | 528 (60.3) | <0.0001 |
| Type 2 diabetes mellitus, *n* (%) | 363 (15.2) | 217 (24.8) | <0.0001 |
| Dyslipidaemia, *n* (%) | 1642 (68.7) | 612 (69.9) | 0.52 |
| Angina pectoris, *n* (%) | | | |
| Stable | 893 (37.7) | 339 (38.9) | 0.01 |
| Unstable | 776 (32.8) | 241 (27.7) | |
| Family history of CVD, *n* (%) | 1266 (53.0) | 440 (50.2) | 0.17 |
| Cardiovascular medication use, *n* (%) | | | |
| ACE-inhibitors | 1268 (53.0) | 483 (55.0) | 0.29 |
| β-blockers | 1617 (67.6) | 465 (53.1) | <0.0001 |
| Statins | 1189 (49.7) | 354 (40.4) | <0.0001 |
| Inflammatory markers | | | |
| Interleukin-6, median (IQR), pg/mL[a] | 3.00 (1.74–5.69) | 3.89 (2.11–7.29) | <0.0001 |
| C-reactive protein, median (IQR), mg/L[a] | 3.02 (1.21–7.69) | 4.69 (1.63–10.7) | <0.0001 |
| Serum amyloid A, median (IQR), mg/L[a] | 4.80 (2.70–10.80) | 6.15 (3.30–18.60) | <0.0001 |
| Neutrophils, median (IQR), $10^3/\mu L$[a] | 3.94 (3.11–4.88) | 4.39 (3.52–5.76) | <0.0001 |
| Fibrinogen, median (IQR), g/L[a] | 3.71 (3.16–4.43) | 3.96 (3.34–4.76) | <0.0001 |

RHR, resting heart rate ; CVD, cardiovascular disease; ACE, angiotensin converting enzyme; IQR, interquartile range. Categorical variables are shown as percentages, and continuous data are presented as mean ± SD, or median (IQR).
[a]*t*-test was computed for continuous parameters (with Mann–Whitney *U*-test for skewed data), and $\chi^2$-test for categorical variables.

the Hosmer–Lemeshow goodness-of-fit test according to 10 risk categories. All calculations were performed using STATA version 11.2 (StataCorp, Texas).

# Results

## Baseline characteristics

Of the 3316 patients enrolled in the LURIC study, 49 (1.5%) were excluded due to missing data on a number of parameters examined. Hence, the analytic sample included a total of 3267 patients (2283 men and 984 women). Preliminary analyses revealed that there were no sex-specific differences present. Baseline characteristics of the overall population are reported in *Table 3*. The median duration of follow-up was 9.9 years. Across the 29940 person years of follow-up, there were 546 (17%) deaths due to CVD. There was a trend towards higher body mass index, lower physical activity, arterial hypertension, and type 2 diabetes in patients with a high resting heart rate. In contrast, there was a higher incidence of unstable angina pectoris in patients with a low resting heart rate. Similarly, use of β-blockers and statins was significantly higher among patients with a low resting heart rate (*Table 3*). Of the individual inflammatory markers, interleukin-6, high sensitivity C-reactive protein, serum amyloid A, fibrinogen, and neutrophils were all significantly higher in patients with a high resting heart rate. Patients with a low resting heart rate and who were in the highest quartile on the inflammation index had an increased risk of CVD mortality compared with those with lower levels of inflammation (*Figure 1A*). For those with a high resting heart rate, there was an even stronger increase in CVD mortality with increasing levels of inflammation. That is, patients in the highest quartile of inflammation had a greater risk of CVD mortality when resting

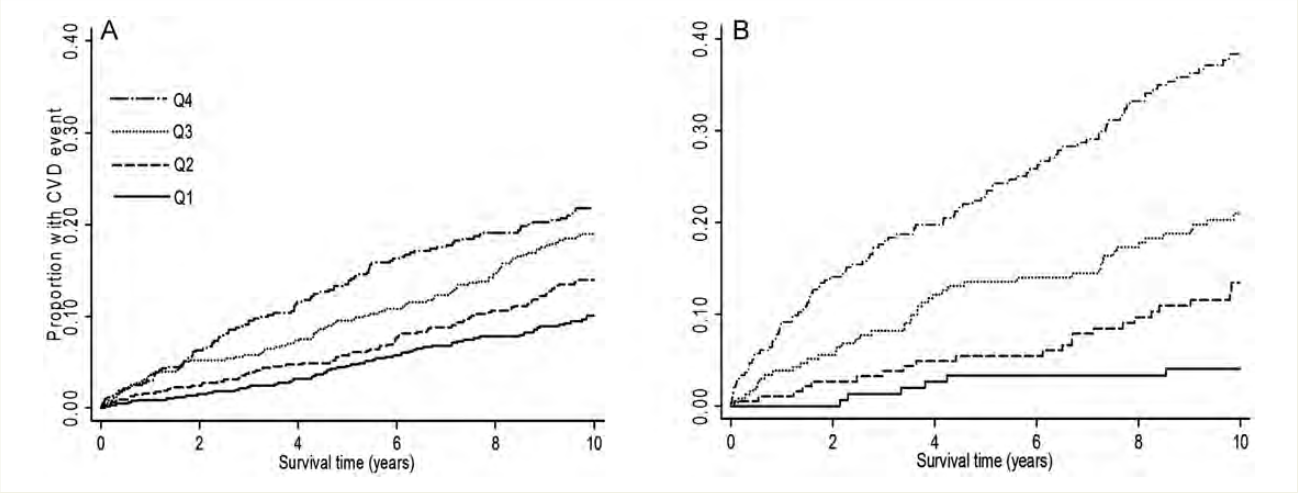



**Figure 1** Kaplan–Meier plots indicating the proportion of cardiovascular deaths according to quartiles of inflammation by (*A*) low (<75 b.p.m.) and (*B*) high (≥75 b.p.m.) resting heart rate. The log-rank test for equality found quartiles of inflammation was significantly different in both models ($P < 0.0001$, respectively).

**Table 4 Relationship between inflammation and cardiovascular mortality by low and high resting heart rate**

| Variable | Inflammation (quartiles) | | | | $P_{trend}$ |
|---|---|---|---|---|---|
| | Q1 | Q2 | Q3 | Q4 | |
| Resting heart rate (<75 b.p.m.) | | | | | |
| No. of alive/CVD event | 593/64 | 539/85 | 471/109 | 407/108 | |
| Median follow-up time, years | 9.7 | 9.1 | 8.8 | 8.3 | |
| Model 1, HR (95% CI) | 1 (reference) | 1.49 (1.07–2.06) | 2.13 (1.56–2.90) | 2.52 (1.85–3.44) | <0.0001 |
| Model 2, HR (95% CI) | 1 (reference) | 1.39 (1.01–1.93) | 1.95 (1.43–2.66) | 2.09 (1.53–2.86) | <0.0001 |
| Model 3, HR (95% CI) | 1 (reference) | 1.28 (0.91–1.79) | 1.69 (1.22–2.34) | 1.84 (1.31–2.57) | <0.0001 |
| Resting heart rate (≥75 b.p.m.) | | | | | |
| No. of alive/CVD event | 146/6 | 162/24 | 188/46 | 197/103 | |
| Median follow-up time, years | 9.7 | 9.0 | 8.5 | 7.0 | |
| Model 1, HR (95% CI) | 1 (reference) | 3.49 (1.43–8.53) | 5.61 (2.40–13.13) | 11.72 (5.15–26.72) | <0.0001 |
| Model 2, HR (95% CI) | 1 (reference) | 3.09 (1.26–7.57) | 4.67 (1.99–10.96) | 9.21 (4.03–21.03) | <0.0001 |
| Model 3, HR (95% CI) | 1 (reference) | 2.40 (0.97–5.92) | 4.08 (1.72–9.64) | 7.50 (3.21–17.50) | <0.0001 |

Values were obtained from Cox proportional hazard models. Model 1 was unadjusted. Model 2 corrected for age and sex. Model 3 additionally corrected for body mass index, smoking, physical activity, arterial hypertension, type 2 diabetes, dyslipidaemia, angina pectoris, family history of cardiovascular disease, and cardiovascular medication. Inflammation index included: interleukin-6, high-sensitivity C-reactive protein, serum amyloid A, neutrophil numbers, and fibrinogen. B.p.m., beats per minute; HR, hazard ratio; CI, confidence interval.

heart rate was ≥75 b.p.m. (*Figure 1B*). The log-rank test for equality revealed that there was a significant difference between quartiles ($P < 0.001$).

## Relationship of inflammatory index with cardiovascular mortality according to resting heart rate

Cox proportional hazard regression reported a significant interaction effect between heart rate and inflammatory activity for the primary endpoint ($P = 0.04$). Separate Cox proportional hazard regression models for those with a low and a high resting heart rate are presented in *Table 4*. There was a survival reduction of 1.4 and 2.7 years due to the primary endpoint in those with a low and a high resting heart rate, respectively, when the highest quartile of inflammation was compared with the lowest quartile. For those with a low resting heart rate, the fully adjusted HR (95% CI) for CVD mortality among patients in the upper quartile of inflammation was 1.84 (1.31–2.57), $P_{trend} < 0.0001$. However, for patients with a high resting heart rate, the fully adjusted risk increased by more than seven-fold, HR (95% CI) = 7.50 (3.21–17.50), $P_{trend} < 0.0001$, for those in the highest inflammatory quartile (*Table 4*). Since patients involved in this study were scheduled

for coronary angiography, we subsequently explored if the degree of CAD severity could explain, in part, the interaction between resting heart rate and inflammatory status in predicting risk of CVD mortality. Interestingly, we observed that the adjusted risk of death due to CVD increased further depending on the magnitude of CAD severity (*Figure 2*). Patients with ≥20% stenosis of at least one main luminal vessel and who were in the upper-most quartile of inflammation with a high resting heart rate had a comparable risk, [HR (95% CI) = 6.68 (2.66–16.78)], $P_{trend} < 0.0001$ for CVD mortality compared with the overall cohort (*Table 4*). On the other hand, the magnitude of cardiovascular risk increased further to approximately 10-fold HR [(95% CI) = 9.83 (3.05–31.65), $P_{trend} < 0.0001$] among those with ≥50% narrowing of at least one main coronary artery, who were in the highest quartile of inflammation on the background of a high resting heart rate. In *Figure 3*, the fully adjusted HR estimates for the single markers of inflammation are presented. For each marker, we compared the upper-most quartile to the lowest and found the risk of CVD mortality was greater among patients who had a high resting heart rate (*Figure 3*). Of all these markers, the greatest prediction was provided by fibrinogen, HR (95% CI) = 3.68 (2.02–6.70), $P < 0.0001$. Though each individual marker was predictive of CVD mortality among those with a high resting heart rate, the strongest prediction for CVD mortality was afforded by the overall marker of inflammation (*Figure 3*).

To better understand the relationship between inflammation and CVD mortality, we repeated the latter analyses including only patients with clinical stability ($n = 2234$) at baseline. After

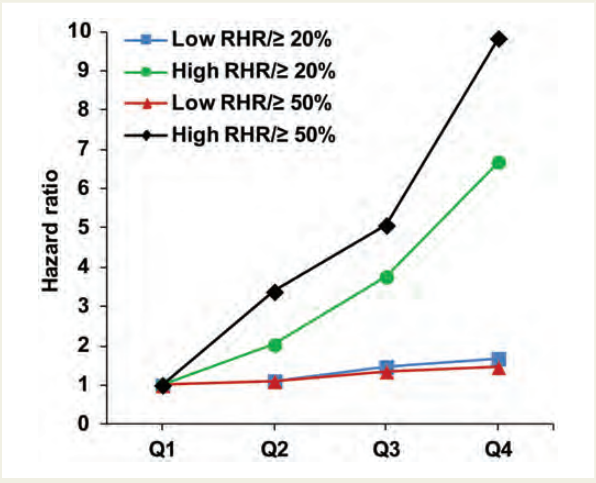



**Figure 2** Hazard ratios for incident cardiovascular mortality according to quartiles (Q) of inflammation by low (<75 b.p.m.) and high (≥75 b.p.m.) resting heart rate (RHR) with either ≥20% or ≥50% stenosis of at least one main coronary artery. Model fully adjusted as outlined in *Table 4*.

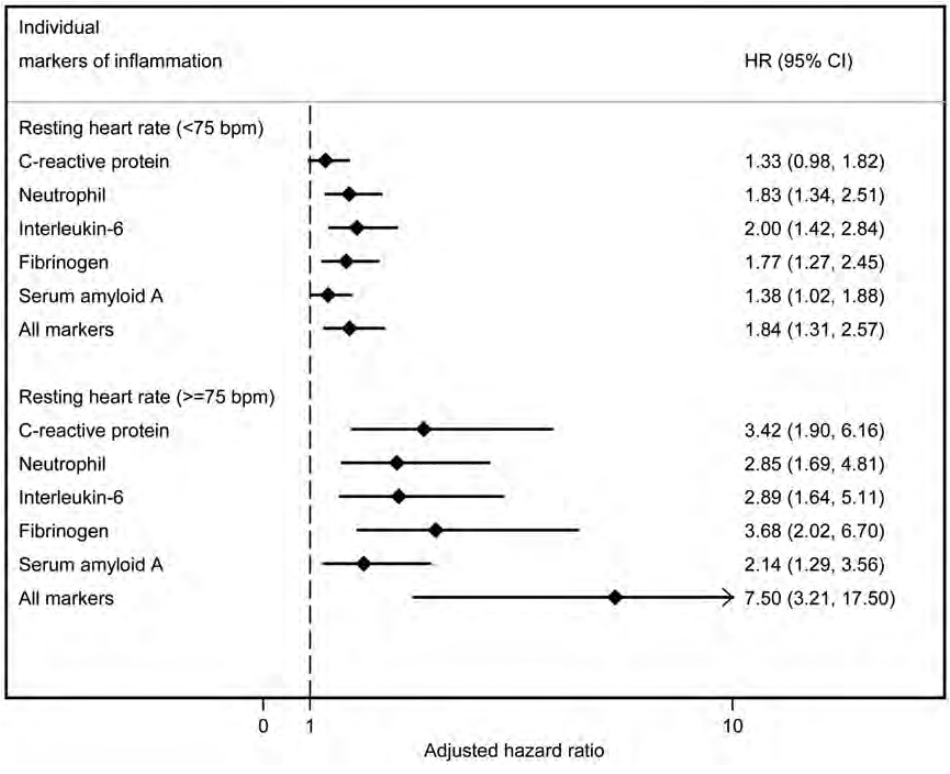



**Figure 3** Hazard ratios (quartile 4 vs. quartile 1) for incident cardiovascular mortality according to the individual markers of inflammation and the composite score, by low (<75 b.p.m.) and high (≥75 b.p.m.) resting heart rate. Model was fully adjusted as abbreviated in *Table 4*.

comparing the upper-most quartile to the lowest, we found the relationship between inflammation and CVD mortality increased HR (95% CI) = 2.37 (1.56–3.60), $P < 0.0001$ in patients with a low resting heart rate, as well as for those HR (95% CI) = 9.18 (3.23–26.00), $P < 0.0001$ with a high resting heart rate. The analyses were then carried out after excluding patients with suspected heart failure ($n = 619$). The fully adjusted association between inflammation and CVD mortality remained unchanged, HR (95% CI) = 1.87 (1.26–2.78), $P = 0.001$ for those with a low resting heart rate. However, there was a slight attenuation in the risk HR (95% CI) = 6.81 (2.63–17.63), $P < 0.0001$ for those with a high resting heart rate. We also removed patients accompanied by an ongoing infection ($n = 315$) which may have contributed towards a heightened inflammatory status at baseline. Likewise, we found the adjusted association between inflammation and CVD mortality for the highest compared with the lowest quartile did not materially change HR (95% CI) = 1.86 (1.31–2.64), $P < 0.0001$ in patients with a low resting heart rate, while again, there was a slight attenuation for individuals HR (95% CI) = 6.19 (2.63–14.60), $P < 0.0001$ with a high resting heart rate. To minimize potential bias due to subclinical and undetected pre-existing disease at baseline which is associated with increased mortality, we removed patients who died within 12 months ($n = 130$) of enrolment. The results were virtually identical, HR (95% CI) = 1.85 (1.29–2.65), $P = 0.001$ for those with a low resting heart rate, and HR (95% CI) = 6.23 (2.64–14.72), $P < 0.0001$ for patients with a high resting heart rate. Finally, after excluding those who reported the use of β-blockers, the adjusted association between inflammation and CVD death among those with a low resting heart rate was somewhat attenuated HR (95% CI) = 1.78 (1.03–3.08), $P = 0.04$, albeit the association remained significant. Conversely, the strong independent relationship between inflammation and CVD mortality HR (95% CI) = 5.35 (2.05–14.00), $P = 0.001$ persisted among patients with a high resting heart rate, even after removing those who reported the use of these agents. Further exploration of the data, using instead, the median cut-off value (median = 68 b.p.m.) for resting heart rate revealed similar findings (data not shown).

## Measures of discrimination for the multi-marker inflammatory index (c-statistic, integrated discrimination improvement, net reclassification improvement)

We assessed the discriminatory value of the multi-marker index of inflammation by computing the c-statistic and area under the receiver operating characteristic (AUC) curve; comparing a model including the conventional risk factors with a model based on a combination of these risk factors and the multi-marker index. Initially, the conventional risk factor model achieved reasonable discrimination, obtaining a c-statistic of 0.751 (*Figure 4*). After including the multi-marker index of inflammation, the discriminatory value marginally improved by 0.010 (0.761, $P = 0.015$) (*Figure 4*). In addition, we employed the reclassification statistics of the IDI and NRI. According to the IDI, inclusion of the multi-marker index significantly improved model discrimination by 0.010 ($P <$ 0.0001). Moreover, the addition of the multi-marker index to the conventional risk factors significantly improved reclassification of patients to a different risk category by 6% ($P = 0.002$) (*Table 5*).

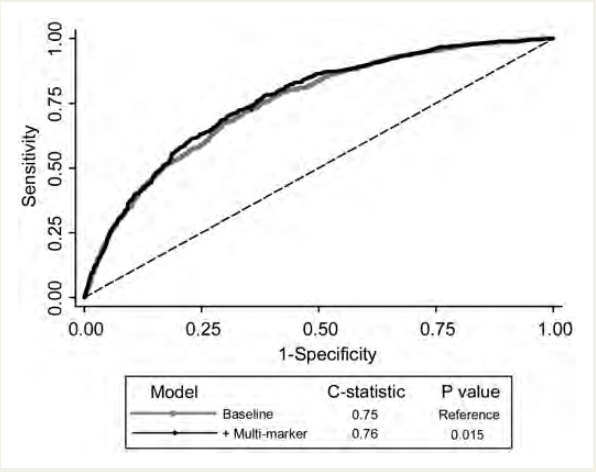



**Figure 4** C-statistic estimates for the prediction of cardiovascular mortality according to the baseline conventional risk factors and multi-marker index of inflammation. Baseline model (grey) included age, sex, waist circumference, smoking, physical activity, arterial hypertension, Type 2 diabetes, dyslipidaemia, family history of cardiovascular disease, angina pectoris, and cardiovascular therapy.

**Table 5 Reclassification of intermediate (defined as 6–20%) risk patients with and without the cardiovascular endpoint**

| Classification according to conventional risk factors | Reclassification accounting for multi-marker | | | |
|---|---|---|---|---|
| | Low | Intermediate | High | Total |
| Patients with CVD endpoint | | | | |
| Low <6% in 10 years | 16 | 6 | 0 | 22 |
| Intermediate 6–20% in 10 years | 5 | 157 | 34 | 196 |
| High >20% in 10 years | 0 | 17 | 301 | 318 |
| Total no. with event | 21 | 180 | 335 | 536 |
| Patients without CVD endpoint | | | | |
| Low <6% in 10 years | 587 | 58 | 0 | 645 |
| Intermediate 6–20% in 10 years | 95 | 1165 | 79 | 1339 |
| High >20% in 10 years | 0 | 88 | 571 | 659 |
| Total no. without event | 682 | 1311 | 650 | 2643 |
| Net reclassification improvement | 6.0% ($P = 0.002$) | | | |

Conventional risk factor model included age, sex, body mass index, smoking, physical activity, arterial hypertension, type 2 diabetes, dyslipidaemia, angina pectoris, family history of cardiovascular disease, and cardiovascular medication.

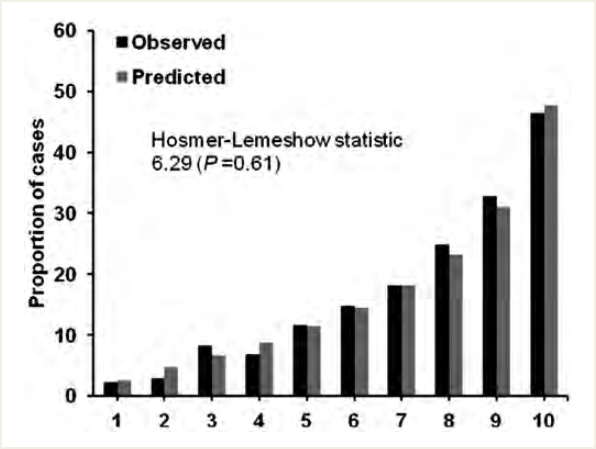



**Figure 5** Proportion of observed and predicted cases according to the risk model that included the multi-marker index of inflammation. The Hosmer–Lemeshow goodness-of-fit test yielded no significant deviation between observed and predicted risk ($P = 0.61$).

Model calibration using the Hosmer–Lemeshow goodness-of-fit test also yielded a Chi-square of 6.29 ($P = 0.61$), indicating no significant deviation between observed and predicted risk (*Figure 5*).

# Discussion

The present study assessed the inter-relationship between inflammation and heart rate with CVD mortality. We found that the risk associated with elevated inflammation was amplified four-fold in patients with a high resting heart rate (≥75 b.p.m.), compared with those with a low resting heart rate (HR 7.50 vs. 1.84). This synergistic effect remained unaltered after adjusting for an extensive number of established risk factors. Further, these observations appeared somewhat dependent on the degree of CAD, since in the current investigation the risk of experiencing the study endpoint increased by almost 10-fold among those with severe CAD (≥50% luminal stenosis of at least one major coronary artery). Our study adds to the current literature because, it is, to our knowledge, the only investigation to address resting heart rate as an effect modifier of inflammation for predicting future CVD outcome.

The European Society of Hypertension/European Society of Cardiology guidelines recently proposed the inclusion of elevated heart rate when evaluating the cardiovascular risk profile of an individual.[14] The present results indicate that the predictive utility of resting heart rate may be substantially enhanced when considered alongside other risk factors, specifically inflammation. Consequently, we may speculate that slowing the heart rate may prove a useful adjunct for interrupting the patho-physiological process initiated by inflammation within the local pro-atherosclerotic vascular environment. If this conjecture is correct, then it may be anticipated that the protective effects of selective heart rate-lowering agents (i.e. β-blockers, calcium antagonists, and the more novel specific heart rate-lowering agent ivabradine) may differ as a function of lower and higher inflammatory activity.

At this point, the exact mechanisms by which tachycardia modulates the effects of inflammation remain to be elucidated, although the extant literature provides compelling evidence for several candidate processes. Among these is autonomic nervous damage. Sympathetic overactivity plays a key role in atheroprogression by inducing tachycardia[5] as well as by causing immune dysregulation. For example, the sympathetic mediator's adrenaline and noradrenaline have potent effects on cytokine release by activated T cells and macrophages and also regulate the migratory behaviour of these cells. Likewise, stimulation of parasympathetic outflow, which normalizes tachycardia, also short circuits the inflammatory cascade via the cholinergic anti-inflammatory pathway,[6–8,15] which involves down regulation of macrophage activity via direct neural stimulation of macrophage cholinergic receptors.

Following this above logic, a raised heart rate could be considered a marker of autonomic nervous system dysfunction rather than a risk factor *per se*.[5,16–18] Nonetheless, several lines of research provide evidence to suggest that heart rate is also a primary risk factor for CVD,[9,19,20] and therefore other interactive mechanisms could be considered as well. For instance, disturbed flow-generated shear stress as a consequence of increased heart rate stimulates specific mechanosensors located on the surface of endothelial cells,[4,5] subsequently upregulating pro-atherogenic genes and downregulating athero-protective genes.[5,21,22] Many of these genes also control inflammatory processes. These include, but are unlikely limited to, reduced nitric oxide activity, which has potent immunomodulatory properties, increased vascular permeability, which is a hallmark of the inflammatory process, as well as increased expression of inflammatory molecules such as adhesion molecules (e.g. vascular cell adhesion molecule-1, intercellular adhesion molecule-1), chemoattractants (monocyte chemoattractant protein-1), and cytokines (interleukin-6, tumour necrosis factor-α).[5,9,23] Thus, an elevated heart rate may enhance the magnitude of an unfavourable cardiovascular risk profile by amplifying the effects of ongoing inflammatory processes.

Of additional importance, we derived a composite marker of inflammation from a principle component analysis. In doing so, we were able to combine each of the individual markers into a single component, thereby reflecting a general marker of inflammation, permitting us to retain most of the information attributed to each marker. From a clinical perspective, the inclusion of a multi-marker strategy in this study based on C-reactive protein, neutrophils, interleukin-6, fibrinogen, and serum amyloid A was more informative for improving risk estimation, compared with single marker determination. Foremost, the clinical utility of individual biomarkers towards predicting subsequent cardiovascular events has recently been questioned. Indeed, the prognostic reliability of C-reactive protein, the most commonly measured inflammatory marker in cardiovascular epidemiology, has been challenged by other authors.[24–26] Thus, we postulate that simultaneous assessment of multiple markers of inflammation may provide additional prognostic information beyond single marker determination, at least among secondary care patients.

The present study is not without limitations. Patients in the present investigation were Caucasian of German ancestry. Thus, caution is warranted in generalizing our findings to the understanding of CVD in other populations. Although prospective in patient enrolment, resting heart rate was only assessed at a single time point and therefore may not reflect heart rate over the longer term and allow us to identify sustained tachycardia. Further, heart rate shows significant diurnal variation which presents additional problems for a one-off measure. Nevertheless, we would counter that resting heart rate was measured in a sufficiently standardized manner to yield reliable data, permitting comparison among patients, while minimizing methodological bias as much as possible.[27] For the purpose of this investigation, resting heart rate was dichotomized according to the 75 percentile cut-off value. Thus, subsequent studies may wish to include other metrics of heart rate when evaluating its relationship with inflammation and adverse cardiovascular outcome. Similarly, the individual markers of inflammation were determined at a single time point. We also cannot exclude the possibility that other functionally distinct markers of inflammation may be implicated in CVD risks that were not examined here. Although we performed multivariable adjustments, we cannot rule out that our results may be influenced by unmeasured or residual confounding. However, due to the large sample size and extensive range of measurements taken, we were able to adjust for a range of important covariates including arterial hypertension, type 2 diabetes, dyslipidaemia, and family history of CVD. In this study, the observed association between resting heart rate and inflammation with CVD mortality appeared somewhat stronger with more severe CAD. However, due to the lack of non-fatal CVD endpoints in LURIC, we were unable to explore this relationship further. In light of this, we hope these findings spur future investigations to evaluate the putative link between resting heart rate and inflammatory activity with non-fatal CVD events.

In conclusion, the present data highlight a potential role for heart rate as a risk factor for CVD mortality by amplifying the effect of inflammation, which may explain, in part, the poor cardiovascular prognosis in individuals with elevated heart rate. Should these observations be confirmed, slowing heart rate may prove a useful adjunct in prolonging survival among CVD patients that exhibit an elevated inflammatory activity.

## Acknowledgements

We thank the LURIC study team either temporarily or permanently involved in patient recruitment, sample, and data handling; the laboratory staff at the Ludwigshafen General Hospital and the Universities of Freiburg, Ulm, and Graz; the German registration offices and local public health departments for their assistance.

## Funding

This work was supported by the 6th Framework Program (integrated project Bloodomics, grant LSHM-CT-2004–503485) and the 7th Framework Program (integrated project Atheroremo, Grant Agreement number 201668) of the European Union.

**Conflict of interest:** none declared.

## References

1. Lloyd-Jones DM, Larson MG, Beiser A, Levy D. Lifetime risk of developing coronary heart disease. *Lancet* 1999;**353**:89–92.
2. Biasillo G, Leo M, Della Bona R, Biasucci LM. Inflammatory biomarkers and coronary heart disease: from bench to bedside and back. *Intern Emerg Med* 2010;**5**:225–233.
3. Hansson GK. Inflammation, atherosclerosis, and coronary artery disease. *N Engl J Med* 2005;**352**:1685–1695.
4. Zamorano JL. Heart rate management: a therapeutic goal throughout the cardiovascular continuum. *Eur Heart J. Suppl* 2008;**10**:F17–F21.
5. Giannoglou GD, Chatzizisis YS, Zamboulis C, Parcharidis GE, Mikhailidis DP, Louridas GE. Elevated heart rate and atherosclerosis: an overview of the pathogenetic mechanisms. *Int J Cardiol* 2008;**126**:302–312.
6. Tracey KJ. The inflammatory reflex. *Nature* 2002;**420**:853–859.
7. Borovikova LV, Ivanova S, Zhang M, Yang H, Botchkina GI, Watkins LR, Wang H, Abumrad N, Eaton JW, Tracey KJ. Vagus nerve stimulation attenuates the systemic inflammatory response to endotoxin. *Nature* 2000;**405**:458–462.
8. Bernik TR, Friedman SG, Ochani M, DiRaimo R, Ulloa L, Yang H, Sudan S, Czura CJ, Ivanova SM, Tracey KJ. Pharmacological stimulation of the cholinergic antiinflammatory pathway. *J Exp Med* 2002;**195**:781–788.
9. Custodis F, Schirmer SH, Baumhakel M, Heusch G, Bohm M, Laufs U. Vascular pathophysiology in response to increased heart rate. *J Am Coll Cardiol* 2010;**56**:1973–1983.
10. Redwine LS, Wirtz PH, Hong S, Bosch JA, Ziegler MG, Greenberg B, Mills PJ. Depression as a potential modulator of Beta-adrenergic-associated leukocyte mobilization in heart failure patients. *J Am Coll Cardiol* 2010;**56**:1720–1727.
11. Winkelmann BR, Marz W, Boehm BO, Zotz R, Hager J, Hellstern P, Senges J. Rationale and design of the LURIC study—a resource for functional genomics, pharmacogenomics and long-term prognosis of cardiovascular disease. *Pharmacogenomics* 2001;**2**:S1–S73.
12. McMurray JJ, Adamopoulos S, Anker SD, Auricchio A, Bohm M, Dickstein K, Falk V, Filippatos G, Fonseca C, Gomez-Sanchez MA, Jaarsma T, Kober L, Lip GY, Maggioni AP, Parkhomenko A, Pieske BM, Popescu BA, Ronnevik PK, Rutten FH, Schwitter J, Seferovic P, Stepinska J, Trindade PT, Voors AA, Zannad F, Zeiher A, ESC Committee for Practice Guidelines, Bax JJ, Baumgartner H, Ceconi C, Dean V, Deaton C, Fagard R, Funck-Brentano C, Hasdai D, Hoes A, Kirchhof P, Knuuti J, Kolh P, McDonagh T, Moulin C, Reiner Z, Sechtem U, Sirnes PA, Tendera M, Torbicki A, Vahanian A, Windecker S, Document Reviewers, Bonet LA, Avraamides P, Ben Lamin HA, Brignole M, Coca A, Cowburn P, Dargie H, Elliott P, Flachskampf FA, Guida GF, Hardman S, Iung B, Merkely B, Mueller C, Nanas JN, Nielsen OW, Orn S, Parissis JT, Ponikowski P. ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure 2012: The Task Force for the Diagnosis and Treatment of Acute and Chronic Heart Failure 2012 of the European Society of Cardiology. Developed in collaboration with the Heart Failure Association (HFA) of the ESC. *Eur Heart J* 2012;**33**:1787–1847.
13. Executive Summary of the Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III). *JAMA* 2001;**285**:2486–2497.
14. Mancia G, De Backer G, Dominiczak A, Cifkova R, Fagard R, Germano G, Grassi G, Heagerty AM, Kjeldsen SE, Laurent S, Narkiewicz K, Ruilope L, Rynkiewicz A, Schmieder RE, Boudier HA, Zanchetti A. 2007 ESH-ESC Practice Guidelines for the Management of Arterial Hypertension: ESH-ESC Task Force on the Management of Arterial Hypertension. *J Hypertens* 2007;**25**:1751–1762.
15. Thayer JF. Vagal tone and the inflammatory reflex. *Cleve Clin J Med* 2009;**76**:S23–S26.
16. Tardif JC. Heart rate as a treatable cardiovascular risk factor. *Br Med Bull* 2009;**90**:71–84.
17. Dominguez-Rodriguez A, Blanco-Palacios G, Abreu-Gonzalez P. Increased heart rate and atherosclerosis: potential implications of ivabradine therapy. *World J Cardiol* 2011;**3**:101–104.
18. Palatini P. Elevated heart rate: a 'new' cardiovascular risk factor? *Prog Cardiovasc Dis* 2009;**52**:1–5.
19. Swedberg K, Komajda M, Bohm M, Borer JS, Ford I, Dubost-Brama A, Lerebours G, Tavazzi L. Ivabradine and outcomes in chronic heart failure (SHIFT): a randomised placebo-controlled study. *Lancet* 2010;**376**:875–885.
20. Borer JS. Heart rate: from risk marker to risk factor. *Eur Heart J Suppl* 2008;**10**:F2–F6.
21. Chiu JJ, Usami S, Chien S. Vascular endothelial responses to altered shear stress: pathologic implications for atherosclerosis. *Ann Med* 2009;**41**:19–28.
22. Brooks AR, Lelkes PI, Rubanyi GM. Gene expression profiling of human aortic endothelial cells exposed to disturbed flow and steady laminar flow. *Physiol Genomics* 2002;**9**:27–41.

23. Walpola PL, Gotlieb AI, Cybulsky MI, Langille BL. Expression of ICAM-1 and VCAM-1 and monocyte adherence in arteries exposed to altered shear stress. *Arterioscler Thromb Vasc Biol* 1995;**15**:2–10.
24. Danesh J, Wheeler JG, Hirschfield GM, Eda S, Eiriksdottir G, Rumley A, Lowe GD, Pepys MB, Gudnason V. C-reactive protein and other circulating markers of inflammation in the prediction of coronary heart disease. *N Engl J Med* 2004; **350**:1387–1397.
25. Tall AR. C-reactive protein reassessed. *N Engl J Med* 2004;**350**:1450–1452.
26. Blankenberg S, McQueen MJ, Smieja M, Pogue J, Balion C, Lonn E, Rupprecht HJ, Bickel C, Tiret L, Cambien F, Gerstein H, Munzel T, Yusuf S. Comparative impact of multiple biomarkers and N-Terminal pro-brain natriuretic peptide in the context of conventional risk factors for the prediction of recurrent cardiovascular events in the Heart Outcomes Prevention Evaluation (HOPE) Study. *Circulation* 2006;**114**:201–208.
27. Vogel CU, Wolpert C, Wehling M. How to measure heart rate? *Eur J Clin Pharmacol* 2004;**60**:461–466.



## CARDIOVASCULAR FLASHLIGHT

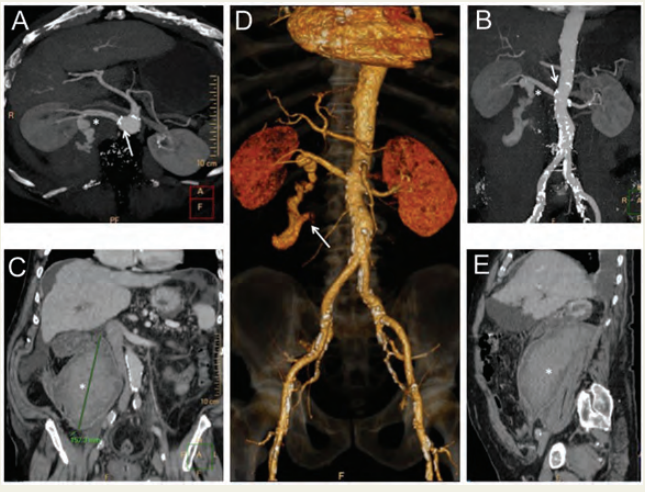
doi:10.1093/eurheartj/ehs320
Online publish-ahead-of-print 1 October 2012

# Dissection and ruptured pseudoaneurysm of a renal artery: a non-described complication during transcatheter aortic-valve implantation

**Alfonso Jurado Roman[1]*, Jose Manuel Montero Cabezas[1], Ana Robles Alonso[2], and Agustin Albarran Gonzalez-Trevilla[1]**

[1]Cardiology Department, Hospital Universitario 12 de Octubre, Avenida de Córdoba s/n, Madrid 28026, Spain and [2]Radiology Department, Hospital Universitario 12 de Octubre, Avenida de Córdoba s/n, Madrid 28026, Spain

* Corresponding author. Tel: +34 629871863, Fax: +34 914695775, Email: alfonsojuradoroman@gmail.com




A 72-year-old man with severe symptomatic aortic stenosis and multiple comorbidities (logistic Euroscore:10.29%) was rejected for conventional valvular surgery because of high surgical risk. After a thorough evaluation, a transcatheter aortic-valve implantation (TAVI) was performed by transfemoral access, and there were no immediate complications.

After 72 h, the patient developed abdominal pain radiating to right lower quadrant, associated with anaemia and hypotension. An emergent CT-scan revealed a dissection and a ruptured pseudoaneurysm of the right renal artery (RRA) (*Figure 1A* and *B*, asterisk) with a high-flow active bleeding (*Figure 1D*, arrow) resulting in a large retroperitoneal haematoma (*Figure 1C* and *E*, asterisk). Percutaneous embolization of the RRA was required to control bleeding. Six months later, the patient remains stable with mild renal dysfunction.

In the emergent CT-scan, a calcium spicule on the RRA ostium was observed (*Figure 1A* and *B*, arrow). During the procedure, the probable impact of the sheath at that level could have produced the RRA dissection, resulting in a pseudoaneurysm which rupture occurred 72 h later.

Vascular complications of TAVI are common and worsen the prognosis. Most are related to closure devices failure and some predictors are operator experience, femoral calcification, and sheath-to-femoral artery ratio. To prevent them, ultrasound guidance during the intervention and follow-up after TAVI should be considered, in particular, if difficulties occurred during the procedure. To the best of our knowledge, we describe for the first time a dissection and ruptured pseudoaneurysm of the renal artery as a rare but severe complication related to TAVI.