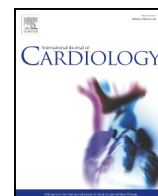


Contents lists available at [ScienceDirect](http://ScienceDirect.com)

International Journal of Cardiology

journal homepage: www.elsevier.com/locate/ijcard

Endurance exercise-induced changes in BNP concentrations in cardiovascular patients versus healthy controls[☆]

Vincent L. Aengevaeren^{a,b,1}, Maria T.E. Hopman^{a,1}, Dick H.J. Thijssen^{a,c,1}, Roland R. van Kimmenade^{b,1}, Menko-Jan de Boer^{b,1}, Thijs M.H. Eijsvogels^{a,c,*}

^a Radboud Institute of Health Sciences, Department of Physiology, Radboud University Medical Center, Nijmegen, The Netherlands

^b Radboud Institute of Health Sciences, Department of Cardiology, Radboud University Medical Center, Nijmegen, The Netherlands

^c Research Institute for Sport and Exercise Sciences, Liverpool John Moores University, UK

ARTICLE INFO

Article history:

Received 14 July 2016

Received in revised form 1 November 2016

Accepted 4 November 2016

Available online xxx

Keywords:

Walking

Biomarkers

Cardiovascular diseases

Risk factors

ABSTRACT

Background: Healthy athletes demonstrated increased B-type natriuretic peptide (BNP) concentrations following exercise, but it is unknown whether these responses are exaggerated in individuals with cardiovascular risk factors (CVRF) or disease (CVD). We compared exercise-induced increases in BNP between healthy controls (CON) and individuals with CVRF or CVD. Furthermore, we aimed to identify predictors for BNP responses.

Methods: Serum BNP concentrations were measured in 191 participants (60 ± 12 yrs) of the Nijmegen Marches before (baseline) and immediately after 4 consecutive days of walking exercise (30–50 km/day). CVRF ($n = 54$) was defined as hypertension, hypercholesterolemia, obesity or smoking and CVD ($n = 55$) was defined as a history of myocardial infarction, heart failure, atrial fibrillation or angina pectoris.

Results: Individuals walked 487 ± 79 min/day at 65 ± 10% of their maximum heart rate. Baseline BNP concentrations were higher for CVD (median: 28.1 pg/ml; interquartile range: 13–50, $p < 0.001$) compared to CVRF (3.9 pg/ml; 0–14) and CON (5.5 pg/ml; 0–14). Post-exercise BNP concentrations were elevated in CVD (35.7 pg/ml, 17–67, $p = 0.01$), but not in CVRF participants ($p = 0.11$) or CON ($p = 0.07$). No cumulative effect in BNP concentrations was observed across the consecutive walking days ($p > 0.05$). Predictors for post-exercise BNP ($R^2 = 0.77$) were baseline BNP, beta-blocker use and age.

Conclusion: Prolonged moderate-intensity walking exercise increases BNP concentrations in CVD participants, but not in CVRF and CON. BNP increases were small, and did not accumulate across consecutive days of exercise. These findings suggest that prolonged walking exercise for multiple consecutive days is feasible with minimal effect on myocardial stretch, even for participants with CVD.

© 2016 The Authors. Published by Elsevier Ireland Ltd. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).

1. Introduction

B-type natriuretic peptide (BNP) is a hormone that promotes natriuresis, diuresis and arterial vasodilation and is secreted by cardiomyocytes in response to myocardial stretch [1]. Since BNP concentrations are elevated in subjects with myocardial stress, BNP has been intensively studied as a circulating biomarker in cardiovascular diseases. BNP and its cleavage equivalent NT-proBNP have nowadays become established diagnostic and prognostic biomarkers in cardiovascular medicine [2].

BNP concentrations are known to acutely increase in cardiac patients undergoing a symptom-limiting exercise test [3,4], with the magnitude of rise proportional to the degree of myocardial ischemia [5,6]. Other factors associated with BNP increase include baseline BNP, age, area of scarred myocardium, change in wall-motion score, workload, lower resting heart rate and higher percentage of predicted heart rate achieved [7–9]. Subjects with hypertension [10,11] as well as healthy controls [12] also showed an increase in BNP after a short bout of high intensity exercise, with hypertensive subjects showing higher concentrations of exercise-induced BNP than controls. Elevations of BNP after short-duration high-intensity exercise appear to peak immediately post-exercise and return to baseline within 1 h post-exercise [3,12].

Previous studies revealed that BNP was also elevated after prolonged high-intensity exercise (>3–4 h, >80% of maximum heart rate) in assumingly healthy endurance athletes [13,14]. Age was the main predictor of the magnitude of the BNP increase [15,16]. The kinetics of post-exercise BNP concentrations following prolonged exercise show an increase immediately after intense exercise [15,17–19], with peak

[☆] Acknowledgement of grant support: V.L.A is financially supported by a grant from the Radboud Institute for Health Sciences (RIHS-2014). T.M.H.E is supported by a Horizon 2020 grant from the European Commission (Marie Skłodowska-Curie Fellowship 655502).

* Corresponding author at: Department of Physiology (392), Radboud University Medical Center, P.O. Box 9101, 6500 HB Nijmegen, The Netherlands.

E-mail address: Thijs.Eijsvogels@radboudumc.nl (T.M.H. Eijsvogels).

¹ All authors take responsibility for all aspects of the reliability and freedom from bias of the data presented and their discussed interpretation.

concentrations at approximately 1 day post-exercise [20–22] and a return to baseline concentrations within 3 days [20].

Individuals with cardiovascular diseases (CVD) or risk factors (CVRF) participate in endurance exercise events more often nowadays [23,24]. Prolonged moderate-intensity exercise such as walking is typically performed at 60–70% of the maximum heart rate and appears to be feasible and attractive for these patients [23,25]. Whether exercise-induced changes in BNP concentrations after prolonged exercise differ between CVD or CVRF compared to healthy controls (CON) is unknown. Therefore, the primary aim of the present study was to compare BNP concentrations at rest and after a single bout of prolonged moderate-intensity walking exercise between individuals with CVD, CVRF and CON. Secondly, we sought to compare post-exercise BNP concentrations across 4 consecutive days of walking exercise. Thirdly, we aimed to identify predictors of the exercise-induced increase in BNP concentrations. We hypothesized that individuals with cardiovascular risk factors and/or disease would show greater changes in BNP after exercise compared to healthy peers due to greater myocardial stress for a similar bout of exercise. Furthermore, we speculated that BNP concentrations show a cumulative effect over the four subsequent walking days, because previous studies demonstrated peak BNP concentrations at 1 day post-exercise [20–22].

2. Methods

2.1. Study population

This was an observational cohort study. A selection of participants of the 2014 and 2015 edition of the Nijmegen Four Days Marches were invited to participate in our study. The Four Days Marches is the largest multiple day walking event in the world (<http://www.4daagse.nl/en/>). Participants of the Nijmegen Marches walked 30, 40 or 50 km per day for 4 consecutive days. Participants could self-select their distance, pace and timing of breaks during walking. CVD, CVRF and CON participants were recruited via social media and the website of the Nijmegen Marches. CVRF was defined as hypertension, hypercholesterolemia, obesity or smoking. CVD was defined as a history of myocardial infarction, heart failure, atrial fibrillation or angina pectoris. Participants were allowed to have multiple diagnoses, but could only be part of a single group. Therefore, if a participant with CVRF also had CVD, one would be allocated to the CVD group. Healthy controls were free from CVRF and CVD. The medical ethical committee of the Radboud University Medical Center approved the study and all participants provided written informed consent before participation. The study was conducted in line with the Declaration of Helsinki.

2.2. Study procedures

Participants completed an extensive questionnaire including general demographic data, walking distance and detailed questions on their (cardiovascular) health status. One or two days before the start of the event (i.e. baseline), body weight (Seca 888 Scale, Seca, Hamburg, Germany) and height were measured. In addition, heart rate and blood pressure at rest were measured using an automated sphygmomanometer (M5-1 Intellisense, Omron Health Care, Hoofddorp, The Netherlands) after 5 min of supine rest. Furthermore, at baseline and ~10 min after the finish on each walking day (day 1–4), a venous blood sample (10 mL) was drawn from an antecubital vein. Blood samples were collected in serum-gel Vacutainer tubes and allowed to clot for ~45 min. After centrifugation, serum was aliquoted, frozen, and stored at -80°C for later analysis. BNP concentrations were analyzed using a high sensitive BNP assay (Centaur BNP, Siemens Healthcare Diagnostics, The Hague, The Netherlands), with a detection limit of 2 pg/mL and a coefficient of variation of 20% at 2.5 pg/mL, 4.7% at 30 pg/mL, and 2.3% at 1500 pg/mL. Start and finish times were used to determine exercise duration and walking speed. Heart rate was

recorded during the first exercise day with a 2-channel electrocardiographic chest band system (Polar Electro Oy, Kempele, Finland) and measured with a data recorder every 5 km along the route. Exercise intensity was calculated as average heart rate during exercise divided by estimated maximum heart rate ($208 - 0.7 \times \text{age}$) [26]. An overview of all measurements is presented in Fig. 1. Safety of the participants was secured by multiple medical assistance places along the walking route.

2.3. Data analysis

Statistical analyses were performed using SPSS Statistics 21 (SPSS, Inc., Chicago, Illinois). Continuous variables were reported as mean \pm SD or median (interquartile range) and categorical variables as proportions. BNP concentrations at baseline and post-exercise were transformed with the natural logarithm as $\text{Ln}(\text{BNP concentration} + 2)$ prior to statistical analysis. Change in BNP (ΔBNP), was defined as $\text{Ln}(\text{BNP post-exercise} + 2) - \text{Ln}(\text{baseline BNP} + 2)$. One-way ANOVA was used to compare baseline characteristics across groups. Pearson Chi-Square tests were used to compare categorical variables and a post-hoc z-test comparison of columns with Bonferroni correction was done in case of statistical significance. To assess aim 1 and 2, one-way repeated measures ANOVA was used to compare the acute and prolonged effect of exercise on BNP concentrations between CON, CVRF and CVD participants. Bonferroni was used as post-hoc test in case of statistical significance. To assess aim 3, linear regression analysis was used to identify predictors of post-exercise BNP and ΔBNP . First, univariate linear regression analysis of variables was used to select potential predictors for multivariate analysis. Variables that were significantly associated with BNP concentrations using univariate analysis were then included in a multivariate analysis. We performed a backward multivariate linear regression analysis to gain insight in which variables could independently predict post-exercise BNP and ΔBNP . Statistical significance was assumed at $p < 0.05$.

3. Results

Our cohort consisted of $n = 83$ healthy individuals, $n = 53$ individuals with CVRF, and $n = 55$ individuals with CVD. Participant characteristics are summarized in Table 1. One CON participant dropped out after day 1, whereas no BNP concentration could be determined in 6 samples of participants on one of the walking days due to insufficient sample volume. On average, participants walked 487 ± 79 min/day at $65 \pm 10\%$ of their predicted maximum heart rate (Table 1).

Compared to CVRF participants and CON, participants with CVD were older, more often men, had lower diastolic blood pressure, walked at a lower exercise intensity and more frequently used medication. Compared to CVRF participants, the CVD group had lower mean and systolic blood pressures. Furthermore, compared with CON, the CVD group had a larger height, higher weight, higher BMI and slower walking speed. Whereas compared to CON, CVRF participants were more often men and had a higher mean weight and BMI (Table 1).

3.1. Effects of exercise on BNP concentrations.

Baseline BNP concentrations were significantly higher in CVD (28.1 pg/mL; 13–50, $p < 0.001$) compared to CVRF participants (3.9 pg/mL; 0–14) and CON (5.5 pg/mL; 0–14) (Fig. 2). At baseline, six participants (3%, all CVD) had a BNP concentration > 100 pg/mL. Post-exercise BNP concentrations (day 1: 35.7 pg/mL, 17–67) were significantly ($p = 0.01$) elevated in CVD participants, but not in CVRF participants (8.2 pg/mL, 0–14, $p = 0.11$) or CON (5.2 pg/mL, 0–14, $p = 0.07$). Seven (4%, all CVD) participants had a BNP concentration > 100 pg/mL after day 1 of walking exercise. Post-exercise BNP concentrations at day 2 to 4 were not different compared to post-exercise levels found on day 1 for any of the groups ($p > 0.05$).

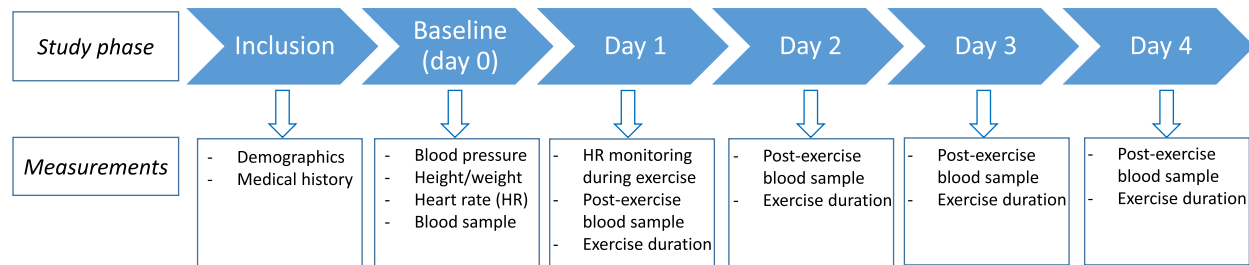


Fig. 1. Flowchart indicating study phase and accompanying measurements.

3.2. Predictors of BNP

Univariate analysis revealed that age, sex, walking distance, walking duration, walking speed, exercise intensity, baseline BNP, study group (CON, CVRF, CVD), the use of beta-blockers, angiotensin-converting enzyme/angiotensin II/renin inhibitors, mineralocorticoid antagonist, diuretics, statins and anticoagulants were significantly positively associated with post-exercise BNP concentrations (Table 2). Because of multicollinearity, walking duration was excluded from multivariate analysis based on its lowest R^2 with univariate analysis. Multivariate analysis revealed that baseline BNP, beta-blocker use and age significantly predict post-exercise BNP ($R^2 = 0.77$) (Table 3). Baseline BNP and beta-blocker use were significantly associated with Δ BNP in both univariate and multivariate analysis ($R^2 = 0.18$; Tables 2 & 3).

4. Discussion

Our results indicate that ~8 h of moderate intensity walking exercise cause a mild but significant increase in post-exercise BNP concentrations in CVD participants, but not in CVRF participants or CON. In addition, bouts of prolonged walking on subsequent days did not result in a further increase in BNP concentrations. We also found that baseline BNP concentrations, beta-blocker use and age independently predicted post-exercise concentrations, whereas baseline BNP concentrations and beta-blocker use predicted Δ BNP concentrations.

4.1. Acute effects of exercise

Participants with CVD demonstrated higher baseline BNP concentrations compared to CON and CVRF, which is in line with literature [27].

Table 1
Baseline cohort characteristics.

| Cohort characteristics | Total | Controls | CVRF | CVD | p-Value |
|---|------------|------------|-------------------------|-------------------------|---------|
| N | 191 | 82 | 54 | 55 | |
| Age, years | 60 ± 12 | 56 ± 13 | 60 ± 10 | 67 ± 7 ^{†*} | 0.000 |
| Sex, male, n (%) | 123 (64) | 36 (44) | 36 (67) [†] | 51 (93) ^{†*} | |
| Female, n (%) | 68 (36) | 46 (56) | 18 (33) | 4 (7) | |
| Height, cm | 174 ± 8 | 172 ± 8 | 175 ± 9 | 177 ± 6 [†] | 0.007 |
| Weight, kg | 79 ± 15 | 73 ± 12 | 83 ± 18 [†] | 84 ± 11 [†] | 0.000 |
| BMI, kg/m ² | 25.8 ± 3.5 | 24.4 ± 2.8 | 26.7 ± 4.0 [†] | 26.9 ± 3.3 [†] | 0.000 |
| Mean BP, mm Hg | 101 ± 12 | 100 ± 12 | 105 ± 13 | 97 ± 11 [†] | 0.001 |
| Systolic BP, mm Hg | 136 ± 17 | 135 ± 17 | 142 ± 19 | 133 ± 16 [*] | 0.02 |
| Diastolic BP, mm Hg | 83 ± 10 | 83 ± 10 | 87 ± 11 | 79 ± 10 ^{†*} | 0.000 |
| Walking distance | | | | | 0.000 |
| 30 km, n (%) | 79 (41) | 24 (29) | 18 (33) | 37 (67) ^{†*} | |
| 40 km, n (%) | 93 (49) | 48 (59) | 28 (52) | 17 (31) [†] | |
| 50 km, n (%) | 19 (10) | 10 (12) | 8 (15) | 1 (2) [*] | |
| Walking duration, min | 487 ± 79 | 491 ± 75 | 503 ± 98 | 465 ± 57 [*] | 0.04 |
| Walking speed, km/h | 4.6 ± 0.7 | 4.7 ± 0.7 | 4.6 ± 0.7 | 4.3 ± 0.6 [†] | 0.01 |
| Rest HR | 65 ± 9 | 65 ± 8 | 66 ± 9 | 63 ± 10 | 0.38 |
| Exercise intensity, (%HR _{max}) | 65 ± 10 | 67 ± 9 | 68 ± 10 | 60 ± 9 ^{†*} | 0.000 |
| CVRF, n (%) | 92 (48) | – | 54 (100) | 39 (71) | – |
| Hypertension, n (%) | 43 (23) | – | 21 (40) | 22 (40) | – |
| Hypercholesterolemia, n (%) | 43 (23) | – | 16 (30) | 27 (49) | – |
| Obesity (BMI > 30), n (%) | 22 (12) | – | 13 (24) | 9 (16) | – |
| Smoking (current), n (%) | 9 (5) | – | 8 (15) | 1 (2) | – |
| CVD, n (%) | 55 (29) | – | – | 55 (100) | – |
| Myocardial infarction, n (%) | 31 (16) | – | – | 31 (56) | – |
| Angina pectoris, n (%) | 24 (13) | – | – | 24 (44) | – |
| Heart Failure, n (%) | 20 (11) | – | – | 20 (36) | – |
| Atrial fibrillation, n (%) | 19 (10) | – | – | 19 (35) | – |
| Cardiovascular medication use, n (%) | 76 (40) | 0 (0) | 23 (43) [†] | 53 (96) ^{†*} | 0.000 |
| Beta-blocker, n (%) | 45 (24) | 0 (0) | 6 (11) [†] | 39 (71) ^{†*} | 0.000 |
| Calcium antagonist, n (%) | 10 (5) | 0 (0) | 3 (6) | 7 (13) [†] | 0.01 |
| ACE/AT2/Renin inhibitor, n (%) | 46 (24) | 0 (0) | 5 (9) [†] | 41 (75) ^{†*} | 0.000 |
| Anticoagulants, n (%) | 54 (28) | 0 (0) | 6 (11) [†] | 48 (87) ^{†*} | 0.000 |
| Statin, n (%) | 54 (28) | 0 (0) | 11 (20) [†] | 43 (78) ^{†*} | 0.000 |
| Diuretic, n (%) | 21 (11) | 0 (0) | 7 (13) [†] | 14 (26) [†] | 0.000 |
| Antimineralocorticoid, n (%) | 3 (2) | 0 (0) | 0 (0) | 3 (6) [†] | 0.03 |

Data are presented as mean ± standard deviation or frequency (%). Continuous variables were compared using one-way ANOVA; categorical variables were compared using Pearson Chi-squared tests. CVD = cardiovascular disease, BMI = body mass index, HR = heart rate. * = significantly different from CVRF group, [†] = significantly different from controls. ACE/AT2 = angiotensin-converting enzyme/angiotensin 2.

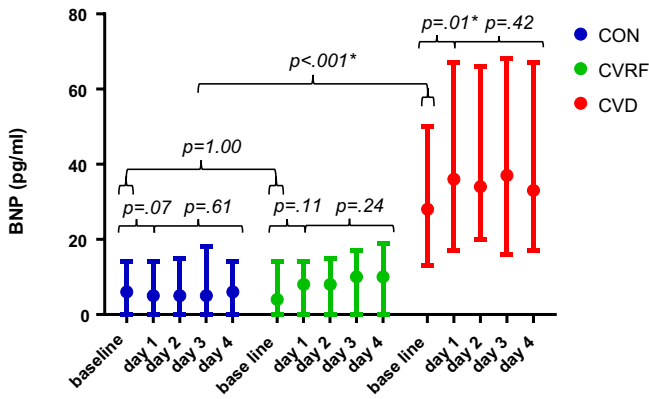


Fig. 2. B-type Natriuretic Peptide (BNP) concentrations at baseline and after prolonged walking exercise on days 1–4 for controls (CON), participants with risk factors (CVRF) and participants with cardiovascular diseases (CVD); BNP concentrations displayed as median with interquartile range. * = $p < 0.05$.

More importantly, we found that prolonged walking exercise produced a subtle but significant increase in BNP concentrations in CVD participants only. The lack of BNP increase in CON and CVRF is in contrast with previous studies that assessed exercise-induced changes in BNP concentrations in athletic populations [15,17,18]. A potential explanation for these discrepant findings may relate to the intensity of the exercise bout. Whereas previous studies performed exercise at a high-intensity (>80% HRmax) [8,15,17], our participants performed moderate-intensity prolonged walking exercise at $65 \pm 10\%$ of their predicted maximum heart rate. Indeed, exercise intensity was previously established as a contributor to the magnitude of BNP release [8]. The CVD group showed an increase in BNP, although their exercise intensity was lower ($60 \pm 9\%$) compared to the other groups. Use of beta-

Table 3
Multivariate linear regression analysis.

| Prediction model | B (CI) | p-Value |
|-------------------|---------------------|---------|
| Post-exercise BNP | | |
| | $R^2 = 0.77$ | |
| Baseline BNP | 0.71 (0.62–0.80) | 0.00 |
| Beta-blocker use | 0.56 (0.33–0.78) | 0.00 |
| Age, years | 0.01 (0.00–0.02) | 0.02 |
| Δ BNP | | |
| | $R^2 = 0.18$ | |
| Baseline BNP | –0.23 (–0.31––0.15) | 0.00 |
| Beta-blocker use | 0.56 (0.33–0.79) | 0.00 |

blockers is a likely explanation for this observation. Besides lowering the maximum attained heart rate, beta-blocker use may independently increase exercise-induced BNP concentrations [9]. For CON and CVRF, prolonged moderate-intensity walking exercise may induce a lower, or absent, increase in BNP concentration following exercise.

4.2. Effect of repetitive bouts of exercise

BNP concentrations did not further increase after the first day of prolonged walking exercise. This finding was unexpected considering previous observations in marathon runners, who demonstrated peak BNP concentrations > 24 h post-exercise [20–22]. It was hypothesized that the delayed BNP peak previously found could point towards transient myocardial dysfunction [20]. However, we previously demonstrated that CVD participants demonstrate no signs of (transient) myocardial dysfunction after prolonged walking exercise [28]. An alternative explanation for the lack of accumulation may relate to the short half-life time of BNP (~20 min), which would cause BNP concentrations to drop back to baseline before initiation of exercise on day 2–4.

Table 2
Univariate linear regression analysis of cohort characteristics.

| Cohort characteristics | Post-exercise BNP | | | Δ BNP | | |
|---------------------------------|------------------------|----------------|---------|----------------------|----------------|---------|
| | B (CI) | R ² | p-Value | B (CI) | R ² | p-Value |
| Age, years | 0.06 (0.04–0.07) | 0.28 | 0.000* | 0.001 (–0.007–0.009) | 0.00 | 0.76 |
| Sex, male | –0.71 (–1.07––0.35) | 0.08 | 0.000* | –0.19 (–0.38–0.01) | 0.02 | 0.07 |
| Height, cm | 0.02 (–0.004–0.04) | 0.01 | 0.12 | 0.01 (–0.003–0.02) | 0.01 | 0.14 |
| Weight, kg | 0.01 (–0.003–0.02) | 0.01 | 0.13 | 0.01 (–0.001–0.01) | 0.02 | 0.09 |
| BMI, kg/m ² | 0.03 (–0.02–0.08) | 0.01 | 0.23 | 0.02 (–0.01–0.05) | 0.01 | 0.12 |
| Mean BP, mm Hg | –0.01 (–0.03–0.01) | 0.01 | 0.18 | –0.001 (–0.01–0.01) | 0.001 | 0.76 |
| Systolic BP, mm Hg | –0.003 (–0.01–0.01) | 0.002 | 0.55 | –0.002 (–0.01–0.003) | 0.004 | 0.39 |
| Diastolic BP, mm Hg | –0.02 (–0.03–0.001) | 0.02 | 0.07 | 0.001 (–0.01–0.01) | 0.00 | 0.83 |
| Walking distance, 30 km ref. | 0 | 0.19 | 0.000* | 0 | 0.01 | 0.42 |
| 40 km | –0.96 (–1.30––0.62) | – | 0.000* | –0.05 (–0.25–0.16) | – | 0.65 |
| 50 km | –1.52 (–2.08––0.95) | – | 0.000* | 0.18 (–0.16–0.51) | – | 0.31 |
| Walking duration, min | –0.003 (–0.005––0.001) | 0.04 | 0.01* | 0.001 (0.00–0.002) | 0.02 | 0.09 |
| Walking speed, km/h | –0.51 (–0.75––0.26) | 0.08 | 0.000* | –0.09 (–0.22–0.05) | 0.01 | 0.22 |
| Rest HR | –0.01 (–0.03–0.01) | 0.01 | 0.25 | 0.002 (–0.01–0.01) | 0.001 | 0.71 |
| Heart rate/max heart rate, % | –0.04 (–0.06––0.03) | 0.13 | 0.000* | –0.004 (–0.01–0.01) | 0.003 | 0.48 |
| Ln baseline BNP | 0.85 (0.78–0.93) | 0.73 | 0.000* | –0.15 (–0.22––0.07) | 0.07 | 0.000* |
| Cardiovascular health, CON ref. | – | 0.25 | 0.000* | – | 0.002 | 0.50 |
| CVRF | –0.57 (–0.95 ––0.18) | – | 0.004 | 0.06 (–0.16–0.27) | – | 0.60 |
| CVD | 1.55 (1.23–1.87) | – | 0.000* | 0.04 (–0.17–0.25) | – | 0.72 |
| Medication use | 0.89 (0.56–1.22) | 0.13 | 0.000* | 0.08 (–0.11–0.27) | 0.004 | 0.42 |
| Beta-blockers | 1.56 (1.21–1.91) | 0.30 | 0.000* | 0.26 (0.04–0.48) | 0.03 | 0.02* |
| Calcium antagonist | 0.74 (–0.04–1.53) | 0.02 | 0.06 | 0.02 (–0.40–0.44) | 0.00 | 0.93 |
| ACE/AT2/renin inhibitor | 1.50 (1.15–1.85) | 0.28 | 0.000* | 0.11 (–0.11–0.33) | 0.005 | 0.34 |
| Antimicrobialocorticoid | 1.61 (0.21–3.01) | 0.03 | 0.02* | 0.36 (–0.40–1.11) | 0.005 | 0.35 |
| Diuretics | 0.69 (0.12–1.26) | 0.03 | 0.02* | –0.17 (–0.48–0.13) | 0.007 | 0.27 |
| Statin | 1.16 (0.80–1.52) | 0.18 | 0.000* | 0.11 (–0.10–0.32) | 0.006 | 0.30 |
| Anticoagulants | 1.32 (0.98–1.67) | 0.24 | 0.000* | 0.09 (–0.13–0.30) | 0.003 | 0.43 |

* = $p < 0.05$, significant univariate predictor. CI = Confidence Interval; BMI = body mass index (weight/length²); BP = blood pressure; HR = heart rate; BNP = B-type natriuretic peptide; CON = controls; CVRF = Cardiovascular risk factors; CVD = cardiovascular disease; ref. = reference group.; ACE/AT2 = Angiotensin-converting enzyme/Angiotensin 2.

Subsequently, a similar BNP rise is expected after each walking day, leading to comparable post-exercise BNP concentrations. This hypothesis is in contrast with previous literature on BNP kinetics after prolonged high intensity exercise, suggesting that the exercise intensity may change BNP kinetics. However, it aligns with previous observations on BNP kinetics after short-term high-intensity exercise [3,12]. This suggests BNP kinetics may be influenced by a combination of both exercise intensity and duration, and that exercise bouts may need to be both long-term and of high intensity to induce prolonged elevations in BNP concentrations.

4.3. Predictors of BNP

Baseline BNP, beta-blocker use and age predicted post-exercise BNP concentrations in a positively correlated fashion. These findings are consistent with previous studies on predictors of post-exercise BNP concentrations [7–9,15,16]. The exercise-induced increase in BNP (Δ BNP) was predicted by baseline BNP and beta-blocker use in our multivariate linear regression model. Interestingly, we found baseline BNP to be negatively associated with Δ BNP. This was an unexpected finding [8] and may be explained by the heterogeneous fitness levels of CVD participants that were included in the present study. Our data suggest that lower baseline BNP concentrations may represent fitter CVD participants who are able to exercise at a higher intensity and therefore have greater Δ BNP compared to less fit CVD participants with higher baseline BNP concentrations. Nonetheless, the contribution of baseline BNP to predict Δ BNP was low (7%) and appears to be of little clinical significance. More importantly, baseline BNP concentrations were tightly correlated to post-exercise BNP concentrations ($R^2 = 0.73$, $p < 0.001$), indicating that the relative change in BNP was rather small. However, this resulted in the excellent predictability of post-exercise BNP using our multivariate model.

4.4. Limitations

The strengths of the present study are its large sample size and the inclusion of a cardiovascular patient population that performed endurance exercise. The limitations include that we allowed self-selected distance and pace and did not match the groups for age and other baseline characteristics. Nevertheless, these parameters were included in the univariate and multivariable adjusted regression analyses, to establish predictors of exercise-induced BNP release. Moreover, we did not match or select people based on level of physical fitness. Another potential limitation may be the timing of our blood drawing. We obtained blood samples typically <10 min post-exercise. Although the half-life time of BNP is considered ~20 min, the time of blood sampling was similar across all days and study groups and aligns with a previous field based study [18]. Finally, we only measured BNP and not its cleavage equivalent NT-proBNP. Given the different exercise-induced kinetics [20] of both biomarkers, it is unfortunately not possible to compare our findings with previous research on NT-proBNP.

4.5. Conclusion

In conclusion, prolonged moderate-intensity walking exercise induces a mild but significant increase in BNP concentrations in individuals with CVD, but not in individuals with CVRF and CON. Walking for consecutive days did not further increase BNP concentrations after the first day of prolonged walking. Post-exercise BNP concentrations can be well predicted as baseline BNP, beta-blocker use and age can explain 77% of its variance. Δ BNP is hard to predict, as baseline BNP and beta-blocker use only explain 18% of Δ BNP variance. Overall, these findings suggest that prolonged walking exercise for multiple consecutive days is feasible with minimal effect on myocardial stretch, even for participants with CVD.

Conflicts of interest

The authors report no relationships that could be construed as a conflict of interest.

Acknowledgements

V.L.A. is supported by a grant from the Radboud Institute for Health Sciences. T.M.H.E. is supported by a Horizon 2020 grant from the European Commission (Marie Skłodowska-Curie Fellowship 655502). We recognize the excellent help of Gilbert van de Tillaart, Ernst Lindhout and Marloes Geurts from Future Diagnostics with performing the analyses of our samples. Furthermore, we thank Lucas Kasper, Inge Bakker and Stan van Dijk from Siemens Healthcare Diagnostics for provision of the BNP assays.

References

- [1] E. Braunwald, Biomarkers in heart failure, *N. Engl. J. Med.* 358 (2008) 2148–2159.
- [2] P. Ponikowski, A.A. Voors, S.D. Anker, H. Bueno, J.G. Cleland, A.J. Coats, et al., 2016 ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure: the task force for the diagnosis and treatment of acute and chronic heart failure of the European Society of Cardiology (ESC). Developed with the special contribution of the Heart Failure Association (HFA) of the ESC, *Eur. J. Heart Fail.* 18 (2016) 891–975.
- [3] H.K. Win, S.M. Chang, M. Raizner, G. Shah, F. Al Basky, U. Desai, et al., Percent change in B-type natriuretic peptide levels during treadmill exercise as a screening test for exercise-induced myocardial ischemia, *Am. Heart J.* 150 (2005) 695–700.
- [4] M. Maeder, T. Wolber, H. Rickli, J. Myers, D. Hack, W. Riesen, et al., B-type natriuretic peptide kinetics and cardiopulmonary exercise testing in heart failure, *Int. J. Cardiol.* 120 (2007) 391–398.
- [5] R.S. Foote, J.D. Pearlman, A.H. Siegel, K.T. Yeo, Detection of exercise-induced ischemia by changes in B-type natriuretic peptides, *J. Am. Coll. Cardiol.* 44 (2004) 1980–1987.
- [6] M.S. Sabatine, D.A. Morrow, J.A. de Lemos, T. Omland, M.Y. Desai, M. Tanasijevic, et al., Acute changes in circulating natriuretic peptide levels in relation to myocardial ischemia, *J. Am. Coll. Cardiol.* 44 (2004) 1988–1995.
- [7] S. Bergeron, J.E. Moller, K.R. Bailey, H.H. Chen, J.C. Burnett, P.A. Pellikka, Exertional changes in circulating cardiac natriuretic peptides in patients with suggested coronary artery disease, *J. Am. Soc. Echocardiogr.* 19 (2006) 772–776.
- [8] M.T. Maeder, D. Staub, Y. Surnier, T. Reichlin, M. Noveanu, T. Breidhardt, et al., Determinants of absolute and relative exercise-induced changes in B-type natriuretic peptides, *Int. J. Cardiol.* 147 (2011) 409–415.
- [9] P.Y. Marie, P.M. Mertes, N. Hassan-Sebbag, N. de Talence, K. Djaballah, W. Djaballah, et al., Exercise release of cardiac natriuretic peptide is markedly enhanced when patients with coronary artery disease are treated medically by beta-blockers, *J. Am. Coll. Cardiol.* 43 (2004) 353–359.
- [10] M. Tanaka, Y. Ishizaka, Y. Ishiyama, J. Kato, O. Kida, K. Kitamura, et al., Exercise-induced secretion of brain natriuretic peptide in essential hypertension and normal subjects, *Hypertens. Res.* 18 (1995) 159–166.
- [11] M. Kohno, K. Yasunari, K. Yokokawa, T. Horio, H. Kano, M. Minami, et al., Plasma brain natriuretic peptide during ergometric exercise in hypertensive patients with left ventricular hypertrophy, *Metabolism* 45 (1996) 1326–1329.
- [12] W.S. Huang, M.S. Lee, H.W. Perng, S.P. Yang, S.W. Kuo, H.D. Chang, Circulating brain natriuretic peptide values in healthy men before and after exercise, *Metabolism* 51 (2002) 1423–1426.
- [13] J. Scharhag, K. George, R. Shave, A. Urhausen, W. Kindermann, Exercise-associated increases in cardiac biomarkers, *Med. Sci. Sports Exerc.* 40 (2008) 1408–1415.
- [14] F. Sedaghat-Hamedani, E. Kayvanpour, L. Frankenstein, D. Mereles, A. Amr, S. Buss, et al., Biomarker changes after strenuous exercise can mimic pulmonary embolism and cardiac injury – a metaanalysis of 45 studies, *Clin. Chem.* 61 (2015) 1246–1255.
- [15] D. Konig, Y.O. Schumacher, L. Heinrich, A. Schmid, A. Berg, H.H. Dickhuth, Myocardial stress after competitive exercise in professional road cyclists, *Med. Sci. Sports Exerc.* 35 (2003) 1679–1683.
- [16] M.P. Leers, R. Schepers, R. Baumgarten, Effects of a long-distance run on cardiac markers in healthy athletes, *Clin. Chem. Lab. Med.* 44 (2006) 999–1003.
- [17] H. Ohba, H. Takada, H. Musha, J. Nagashima, N. Mori, T. Awaya, et al., Effects of prolonged strenuous exercise on plasma levels of atrial natriuretic peptide and brain natriuretic peptide in healthy men, *Am. Heart J.* 141 (2001) 751–758.
- [18] A. La Gerche, W.J. Inder, T.J. Roberts, M.J. Brosnan, H. Heidebuchel, D.L. Prior, Relationship between inflammatory cytokines and indices of cardiac dysfunction following intense endurance exercise, *PLoS One* 10 (2015), e0130031.
- [19] A. La Gerche, K.A. Connelly, D.J. Mooney, A.I. MacIsaac, D.L. Prior, Biochemical and functional abnormalities of left and right ventricular function after ultra-endurance exercise, *Heart* 94 (2008) 860–866.
- [20] W. Frassi, R. Kowoll, N. Katz, M. Speth, A. Stangl, L. Brechtel, et al., Cardiac markers (BNP, NT-pro-BNP, troponin I, troponin T) in female amateur runners before and up until three days after a marathon, *Clin. Lab.* 54 (2008) 81–87.
- [21] A.J. Siegel, E.L. Lewandrowski, K.Y. Chun, M.B. Sholar, A.J. Fischman, K.B. Lewandrowski, Changes in cardiac markers including B-natriuretic peptide in runners after the Boston marathon, *Am. J. Cardiol.* 88 (2001) 920–923.

- [22] J.E. Trivax, B.A. Franklin, J.A. Goldstein, K.M. Chinnaiyan, M.J. Gallagher, A.T. deJong, et al., Acute cardiac effects of marathon running, *J. Appl. Physiol.* (1985) 108 (2010) 1148–1153.
- [23] T. Eijssvogels, K. George, R. Shave, D. Gaze, B.D. Levine, M.T. Hopman, et al., Effect of prolonged walking on cardiac troponin levels, *Am. J. Cardiol.* 105 (2010) 267–272.
- [24] S. Mohlenkamp, N. Lehmann, F. Breuckmann, M. Brocker-Preuss, K. Nassenstein, M. Halle, et al., Running: the risk of coronary events: prevalence and prognostic relevance of coronary atherosclerosis in marathon runners, *Eur. Heart J.* 29 (2008) 1903–1910.
- [25] T.M. Eijssvogels, S. Molossi, D.C. Lee, M.S. Emery, P.D. Thompson, Exercise at the extremes: the amount of exercise to reduce cardiovascular events, *J. Am. Coll. Cardiol.* 67 (2016) 316–329.
- [26] H. Tanaka, K.D. Monahan, D.R. Seals, Age-predicted maximal heart rate revisited, *J. Am. Coll. Cardiol.* 37 (2001) 153–156.
- [27] M. Yoshimura, H. Yasue, K. Okumura, H. Ogawa, M. Jougasaki, M. Mukoyama, et al., Different secretion patterns of atrial natriuretic peptide and brain natriuretic peptide in patients with congestive heart failure, *Circulation* 87 (1993) 464–469.
- [28] N.M. Benda, M.T. Hopman, A.P. van Dijk, D. Oxborough, K.P. George, D.H. Thijssen, et al., Impact of prolonged walking exercise on cardiac structure and function in cardiac patients versus healthy controls, *Eur. J. Prev. Cardiol.* (2016).