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ORIGINAL ARTICLE

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Acute exercise is not cardioprotective and may induce apoptotic signalling in heart surgery: a randomized controlled trial†

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Abstract

OBJECTIVES: During open-heart surgery, the myocardium experiences ischaemia–reperfusion injury. A single bout of moderate, 30-min exercise induces preconditioning and protects the heart from ischaemia–reperfusion injury in rats, but this has never been investigated in humans. We aimed to investigate whether 1 bout of moderate exercise 24 h prior to surgery protects against mitochondrial and cardiac damage.

METHODS: Patients scheduled for elective coronary artery bypass were eligible for this pilot study. Twenty were included and randomized to the treadmill exercise group (the EX group, $n = 10$) 24 h preoperatively or to standard presurgical procedures (control $n = 10$). Right atrial (RA) and left ventricular (LV) biopsies were collected immediately before and as long as possible after aortic cross-clamping to assess the primary outcome of mitochondrial respiration by respirometry, in addition to reactive oxygen species production by fluorometry and apoptotic transcripts. Cardiac troponin T and creatine kinase myocardial brain were measured in plasma at arrival, before surgery and 6 and 24 h postoperatively.

RESULTS: Mitochondrial respiration was lower in the EX group after surgery in the LV (Complex I -22%, $P < 0.05$ and maximal -23%, $P < 0.05$) and the right atrium (Complex I -25%, $P < 0.05$). Transcript level of the apoptosis-related marker caspase 3 was increased 1.5-fold in the LV prior to surgery in the EX group when compared with the control group, $P < 0.05$. Cardiac troponin T was 45% higher in the EX group than in the control group 6 h postoperatively ($P = 0.03$), although not significant when corrected for aortic cross-clamping time.

CONCLUSIONS: Results indicate that exercise did not precondition the heart against surgery-related damage. Exercise may render the myocardium and mitochondria more vulnerable to perioperative damage.

Clinical trials registration number: NCT00218985 (https://clinicaltrials.gov/ct2/show/NCT00218985)

Keywords: Preconditioning • Ischaemia–reperfusion • Mitochondria • Troponin • Caspase-3

INTRODUCTION

Ischaemic heart disease is the leading cause of mortality worldwide [1]. Myocardial ischaemia–reperfusion (I/R) is important in the pathophysiology of ischaemic heart disease and occurs when the heart is subjected to ischaemia and subsequent reperfusion, such as during salvage of an acute myocardial infarction [2]. Considerable effort has been made to find therapeutic strategies to combat I/R injury, and several interventions have been suggested to protect the heart from damage. Exercise was previously proposed as a strategy to protect the heart, and studies have shown that in addition to reducing cardiovascular risk factors, exercise also exerts a direct cardioprotective effect [3, 4]. Exercise as preconditioning has shown promising results [5, 6], and 2 studies have shown that 30 min [7] and 100 min [8] of treadmill running at $\approx$25 m·min^{-1} at 0° and 6° inclination, respectively, is sufficient to reduce I/R injury in rats [7, 8]. From our experience with the use of exercise in rats in our laboratory, this should correspond

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to a moderate intensity. The protection appears immediately after exercise and then disappears only to reappear 24 h [8] or 36 h [5] later, referred to as the second window of preconditioning. The effects of exercise prior to I/R injury have, however, not been investigated in a clinical setting. During conventional on-pump open-heart surgery, the myocardium experiences I/R injury despite perioperative cardioprotective initiatives such as hypothermia and cardioplegia [9]. The most apparent sign of this cardiac damage is elevated levels of Troponin T (cTnT) and creatine kinase myocardial brain (CK-MB) postoperatively, which is associated with a proportionally increased risk of adverse clinical outcomes [10]. As conventional on-pump heart surgery involves a planned ischaemic event, this opens for studying potential measures to reduce I/R injury during surgery.

The mitochondria have been shown to play an important role in I/R injury [11], and effects on the mitochondrial respiratory capacity and production of reactive oxygen species (ROS) have been suggested as central mechanisms of I/R injury [12]. Previous studies have also implicated that the cardioprotective effects of exercise could be caused by biochemical adaptations in the mitochondria [13].

The aim of this study was to investigate whether a single bout of moderate intensity exercise 24 h prior to coronary artery bypass graft (CABG) surgery was sufficient to reduce the I/R injury imposed during aortic cross-clamping (ACC). We hypothesized that 1 bout of exercise would preserve mitochondrial respiration during surgery and reduce postoperative levels of cardiac cTnT and CK-MB and be an easy implementable cardioprotective initiative in clinical practice.

METHODS

Patients and study design

This study was designed as a single-centre, single-blinded, prospective, randomized, controlled trial. In this pilot study, 20 patients undergoing elective primary isolated on-pump CABG surgery at St. Olav's University Hospital, Trondheim, Norway, were included. Patients with unstable angina, recent myocardial infarction (within 4 weeks), diabetes mellitus, poorly controlled hypertension, severe pulmonary disease, orthopaedic and/or neurological limitations or in need of heart valve surgery were excluded. Patients willing to participate in this study signed written consent forms prior to randomization. The patients were randomized to 1 bout of incremental treadmill exercise 24 h prior to surgery (the EX group, $n = 10$) or were prepared for surgery according to standard protocols [control (the CTR group) $n = 10$]. All subjects were analysed unless otherwise stated. Block randomization was performed by the Unit for Applied Clinical Research of St. Olav's University Hospital with 1 block consisting of 10 + 10 patients. This study was approved by the Regional Ethics Committee and conducted in conformance with the Declaration of Helsinki. Personnel performing patient care, data collection and analyses were blinded to group allocation until all data analyses were completed. A.W. enrolled and K.H.S. assigned participants to interventions (see Supplementary Material, Methods section).

Exercise protocol

The exercise protocol consisted of incremental treadmill walking for 30 min, divided into 3 intensity intervals of 10 min. Intensity was increased stepwise (1 $km \cdot h^{-1}$, 2% inclination or a composite of 0.5 $km \cdot h^{-1}$ and 1% inclination), based on the subjects perceived rating of exhaustion using the Borg scale, which ranges from 6 to 20. This corresponds to heart rates between resting state and maximal exertion (60–200 bpm), meaning a rating of 15 should correspond to approximately 150 bpm and 75% of maximal heart rate [14]. The aim of the first interval was to warm up, and intensity was increased at 3 and 6 min if Borg $\leq$10. The second interval aimed to strain the patients to a Borg level of between 10 and 12. Third interval targeted a workload corresponding to 15 Borg, which was achieved by increasing by 1 step every second minute if Borg $\leq$15 and by 2 steps if Borg $\leq$12. Oxygen consumption was monitored by indirect ergospirometry (Metamax II, Cortex, Germany). Blood pressure, ECG and heart rate were continuously monitored by a medical doctor, and the exercise intervention was terminated if any of the stop criteria were reached (see Supplementary Material, Methods section).

Blood and tissue sampling

Blood samples were collected at 4 different time points (Fig. 1): upon admission (T1), immediately before surgery (T2) and 6 (T3) and 24 h after surgery (T4). The secondary outcomes cTnT and CK-MB were analysed by the laboratory of St. Olav's University Hospital, Norway, using chemiluminescence and electrochemiluminescence immunoassay, respectively.

Biopsies from the right atrial appendage and the left ventricle (LV) were collected before ACC (prior to ischaemia) and after the removal of ACC (after reperfusion). Right atrial biopsies were collected in connection with cannulation and decannulation of the right atrium, and ventricular samples were harvested using an Argon Biopince 16G biopsy needle. All biopsies were separated into 2 parts immediately after excision. One part was placed in ice-cold relaxation medium (BIOPS, see Supplementary Material, Methods section) for mitochondrial analyses, and the remaining tissue was snap frozen in liquid nitrogen for RNA analyses.

Mitochondrial respiration

Mitochondrial respiration, which was the primary outcome, and hydrogen peroxide (H_2O_2) production were measured simultaneously in permeabilized fibres from the LV and the right atrium using the Oxygraph-2k (Oxygraph-2k; Oroboros Instruments, Innsbruck, Austria). The titration protocol will be described here, and for a full description, see Supplementary Material, Methods section.

To assess respiration from fat, as well as Complex I (CI), Complex II (CII) and Complex IV (CIV) of the electron transport chain, subsequent additions of substrates, inhibitors and uncouplers were performed. We obtained respiratory states in this order: leak respiration (L) from fat (L_{fat}); oxidative phosphorylation (P) from fat (P_{fat}), thereafter supplementing with CI (P_{fat+CI}) and CI + II ($P_{fat+CI\&II}$); leak respiration from CI&II ($L_{fat+CI\&II}$) and uncoupled respiration (E) from CI + II ($E_{fat+CI\&II}$), CII (E_{fat+II}) and CIV (E_{CIV}) (for details, see Supplementary Material, Methods section).

H_2O_2 production in the tissue samples was assessed using the O2k-Fluorescence LED2-Module, thoroughly described by other authors [15]. In brief, 10-µM Amplex Ultrared and 4-µ/ml horseradish peroxidase were added to the chamber at the beginning of the experiment, and detector probes recorded emitted light from the chamber as a product of H_2O_2 production.

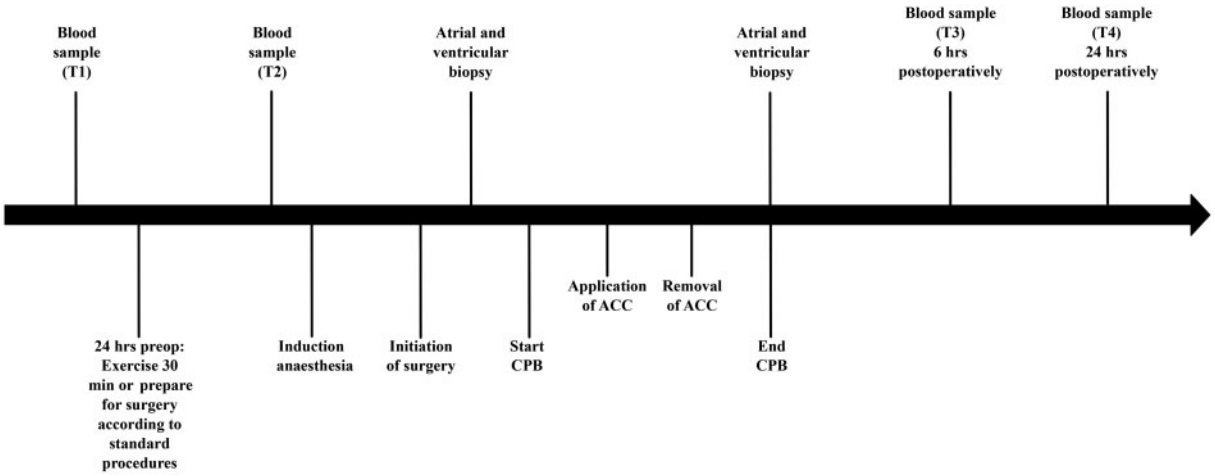

Figure 1: Timeline of blood and biopsy sampling. ACC: aortic cross-clamp; CPB: cardiopulmonary bypass.

After the protocol, the tissue samples were removed from the chambers and weighed (Sartorius ME235P-SD). Respiration rates were expressed as pmol·mg^{-1}·min^{-1}.

Messenger RNA analysis

Quantitative real-time reverse-transcriptase polymerase chain reaction was performed to determine messenger RNA (mRNA) levels of the target genes *BNIP3* (BCL2 interacting protein 3), *BAX* (BCL2 associated X), *BCL2* (apoptosis regulator), *CASP3* (caspase 3), *CYPD* (peptidylprolyl isomerase D), *B2M* (B2 microglobulin). For a detailed description, see Supplementary Material, Methods section.

Statistical analyses

In this pilot study, 20 patients were included. This sample size was chosen, because meaningful results were reported in other studies using the same methods of mitochondrial respiration [16]. All results were presented as mean ± standard error of the mean unless otherwise specified. The effect of exercise preconditioning on postsurgical variables was determined by analysis of covariance (ANCOVA). The post-value (6 h postoperatively for cTnT and CK-MB) was used as the dependent variable with group as the fixed factor, while controlling for the pre-value as a covariate as previously recommended for randomized trials [17]. In additional analysis, we added ACC time (ischaemic time) as a covariate as this could have influenced the results. Unpaired *t*-tests were used to assess pre-ACC differences in mitochondrial respiration. Because of non-normal distribution of preoperative cTnT, CK-MB, Pro-BNP, ROS production, bypass time and ACC time, preoperative between-group differences were analysed using the Mann–Whitney *U*-test and reported as median ± interquartile range. The Fisher's exact test was used on categorical variables. For mRNA data, 1-way ANOVA with Newman–Keuls *post hoc* test was used. We considered a *P*-value of <0.05 as statistically significant. The SPSS version 22 was used for statistics.

RESULTS

Twenty patients electively referred for CABG surgery were included in this study between October 2014 and December 2015. There was no 30-day mortality. Baseline characteristics are presented in Table 1. More individuals in the EX group had a diagnosis of hypertension and were on antihypertensive medication. However, difference in blood pressure levels or left ventricular hypertrophy at admission between the groups was not observed. Trial was completed when we reached 20 participants. We only obtained 6-h postoperative blood samples from 6 in the EX group and 9 in the CTR group due to practical matters. There were no dropouts.

Table 1: Baseline characteristics

	Control (n = 10)	Exercise (n = 10)
Sex, female (%)	0 (0)	1 (10)
Age (years)	65 ± 7.6	62.6 ± 8.5
BMI (kg/m^2)	26.5 ± 2.8	27.4 ± 3.6
ASA score	3.3 ± 0.48	3.4 ± 0.52
EuroSCORE II	0.88 ± 0.37	0.92 ± 0.38
Creatinine clearance, (ml·min^{-1})	105.1 ± 31.0	103.2 ± 42.8
LVEF (%)		
>50	9 (90)	9 (90)
31–50	1 (10)	1 (10)
<30	0 (0)	0 (0)
Current smoker (%)	1 (10)	1 (10)
Previous smoker (%)	5 (50)	3 (30)
Hypertension (%)	1 (10)	7 (70)
Previous cerebral insult (%)	1 (10)	0 (0)
Previous myocardial infarction (%)	2 (20)	2 (20)
Previous PCI (%)	2 (20)	1 (10)
ACE inhibitor or ARB (%)	1 (10)	5 (50)
Aspirin (%)	10 (100)	10 (100)
Beta blockers (%)	7 (70)	7 (70)
Calcium channel blockers (%)	3 (30)	1 (10)
Diuretics (%)	1 (10)	4 (40)
Lipid-lowering agent (%)	10 (100)	8 (80)
Prednisone (%)	1 (10)	0 (0)
Warfarin (%)	0 (0)	1 (10)

Data are presented as mean ± standard deviation.

ACE: angiotensin converting enzyme; ARB: angiotensin receptor blocker; BMI: body mass index; PCI: percutaneous coronary intervention; LVEF: left ventricular ejection fraction.

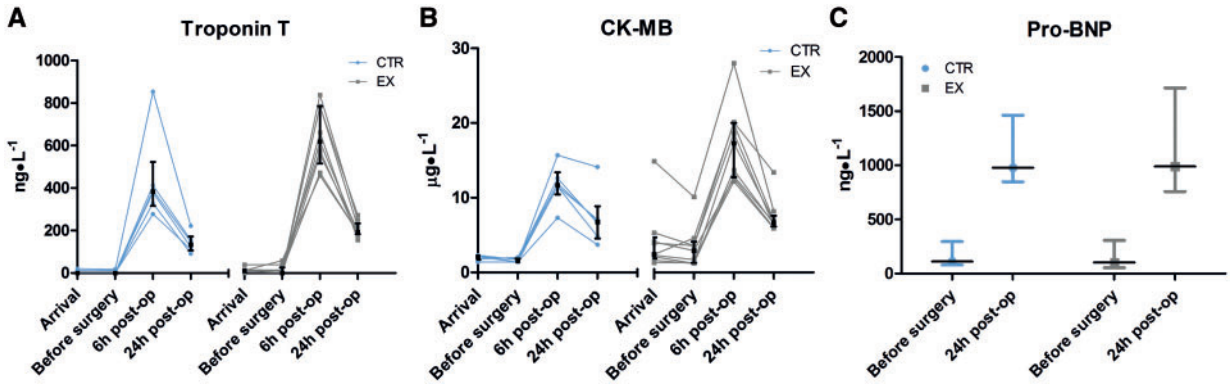

Figure 2: Circulating markers of cardiac damage. All values are presented as median ± interquartile range. (**A**) Cardiac troponin T and (**B**) CK-MB values at arrival, before surgery (24 h after exercise), 6 h after surgery ($n = 6$ in the EX group and $n = 9$ in the CTR group) and 24 h after surgery. (**C**) Pro-BNP before surgery and 24 h after surgery. CK-MB: creatine kinase myocardial brain; CTR: control; cTnT: cardiac troponin T; EX: exercised.

Table 2: Surgical data

	Control ($n = 10$)	Exercise ($n = 10$)	P-value
Aortic cross-clamp time (min:s)	31:30 (15:15)	39:00 (11:00)	0.023*
Cardiopulmonary bypass time (h:min:s)	00:58:00 (00:21:30)	01:15:00 (00:18:00)	0.015*
Cardioplegia (ml)	970 ± 95	955 ± 44	0.66
Distal coronary graft anastomoses (n)	3.1 ± 0.74	3.6 ± 0.52	0.096

Data are presented as mean ± standard deviation except aortic cross-clamp time and cardiopulmonary bypass time, which are median (interquartile range).
*indicates a statistical significant difference.

Exercise training

Average VO_2 reached 19.5 ± 1.4 ml·kg^{-1}·min^{-1} at the end of the 30-min session, corresponding to 5.6 ± 0.4 METS. Heart rate reached 110 ± 8 bpm, and the average total caloric cost of the session was 197.2 ± 9.1 kcal. For 2 of the subjects, the exercise was aborted with approximately 5 min remaining due to ECG changes suggesting ischaemia. They were still included in the analysis.

Circulating biomarkers of cardiac damage

To determine the degree of I/R injury to the myocardium, we measured markers of cardiac damage. Results of the biomarkers CK-MB, cTnT and pro-BNP are depicted in Fig. 2. ANCOVA, using the preoperative value as a covariate, revealed that cTnT 6 h after surgery was higher in the EX group than the CTR group (adjusted means 651.1 ± 58.2 ng·l^{-1} vs 422.5 ± 72.0 ng·l^{-1}, $P = 0.034$) indicating a higher degree of cardiac injury. Significant differences between groups at any other time point were not observed. No significant differences were observed for CK-MB 6 h postoperatively (adjusted means 16.1 ± 0.9 µg·l^{-1} vs 13.5 ± 1.1 µg·l^{-1}, $P = 0.089$). Patients in the EX group had significantly longer CPB and ACC times (Table 2), which may have influenced the results [18], and adding ACC duration as a covariate in the ANCOVA model, differences between the groups were no longer significant for cTnT (adjusted means 630.3 ± 61.0 ng·l^{-1} vs 453.7 ± 77.4 ng·l^{-1}, $P = 0.13$ in the EX group and the CTR group, respectively) and still not significant for CK-MB (adjusted means 16.2 ± 0.9 µg·l^{-1} vs 13.3 ± 1.2 µg·l^{-1}, $P = 0.10$ in the EX group and the CTR group, respectively).

Pro-BNP increased 24 h after surgery when compared with that before surgery, but differences between the groups were not observed (Fig. 2C).

A residual analysis of the biomarkers with natural logarithmic transformation is presented in the Supplementary Material. The results were similar.

Mitochondrial function

Respiration rates in permeabilized left ventricular and atrial muscle fibres were analysed before and after exposure to I/R by ACC. Respiration did not differ before ACC (Fig. 3A and B). In contrast to our hypothesis, we observed a lower respiration in the left ventricular muscle fibres following ACC in the EX group when compared with the CTR group during P_{fat} ($P = 0.025$), P_{CI} ($P = 0.018$) and $P_{fat+CI\&II}$ ($P = 0.011$, Fig. 3A). Even following correction for ACC duration, the 2 latter remained significant (-22% $P = 0.024$ and -23% $P = 0.019$, respectively). In the right atrial muscle fibres (the right atrium), P_{fat+CI} was lower in the EX group than that in the CTR group after ACC ($P = 0.03$), and this remained significant when adjusting for ACC duration (-25%, $P = 0.04$). Together this suggests that preoperative exercise may lead to a reduction in mitochondrial respiration after surgery.

After ACC, ROS production was 28% lower in the EX group than that in the CTR group in the left ventricular muscle fibres during $L_{fat+CI\&II}$ ($P = 0.04$), and this persisted after controlling for ACC duration ($P = 0.003$). In the right atrial muscle fibres, ROS production during $L_{fat+CI\&II}$ was 33% lower in the EX group than that in the CTR group ($P = 0.05$ and 0.02 after correction for ACC duration). ROS production was otherwise unchanged.

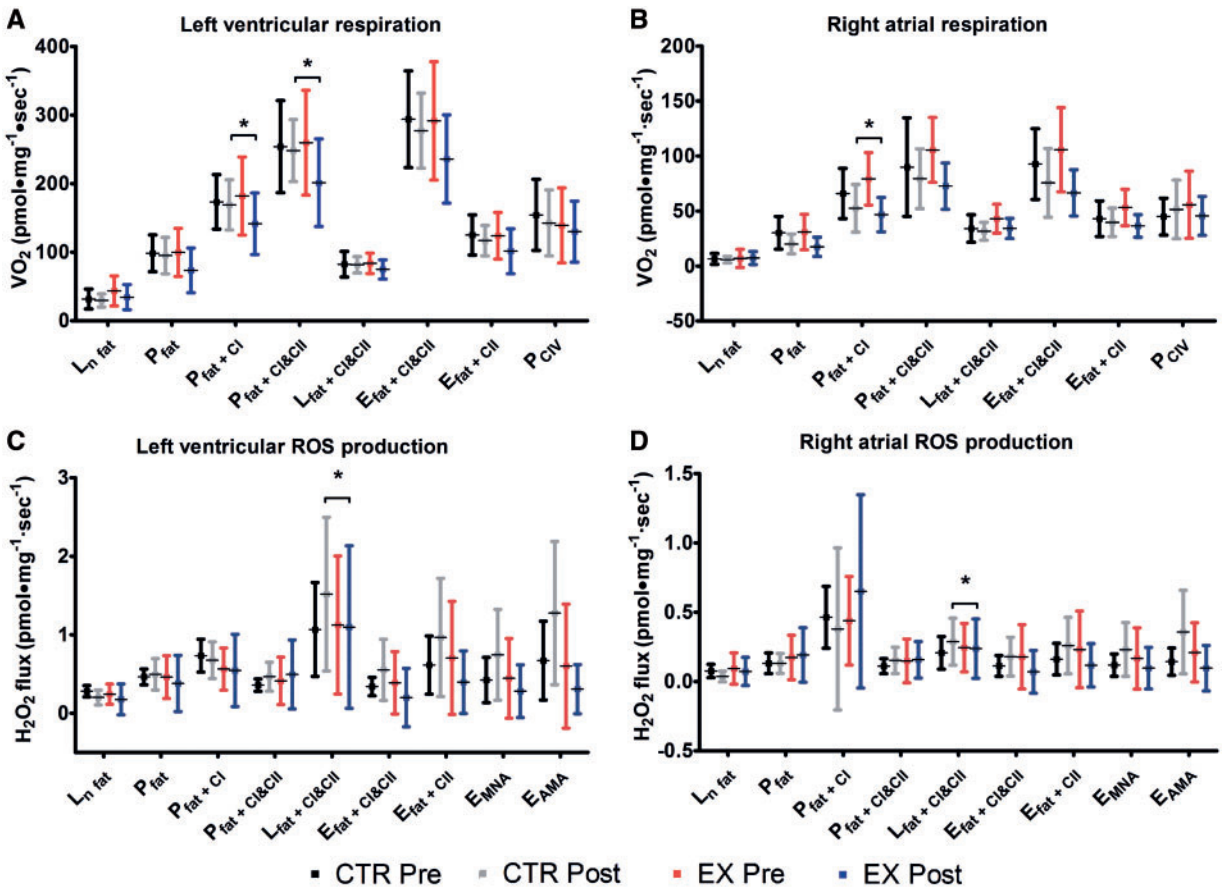

Figure 3: Effect of exercise prior to CABG surgery on mitochondrial function. Mitochondrial respiration and reactive oxygen species (H_2O_2) production in all respiratory states after subsequent addition of substrates, uncoupler and inhibitors; mean ± standard deviation. (**A**) Left ventricular and (**B**) right atrial respiration in the CTR group and the EX group before (pre) and after (post) induction of ischaemia by aortic cross-clamp and (**C**) left ventricular and (**D**) right atrial H_2O_2 production in the CTR group and the EX group before (pre) and after (post) induction of ischaemia by aortic cross-clamp. AMA: inhibition of CIII by antimycin A; CI: complex I with pyruvate and glutamate; CII: complex II with succinate, CI blocked by rotenone; CIV: complex IV with ascorbate and TMPD; CTR: control; E: uncoupled respiration; EX: exercised; Fat: palmitoyl-carnitine and octanoyl-carnitine; L: leak respiration; L_n: leak respiration in the absence of adenylates; MNA: inhibition of complex II by malonic acid; P: oxidative phosphorylation respiration. *P-value <0.05 vs control group, corrected for prevalue and aortic cross-clamp time.

Activation of apoptotic signalling

We analysed left ventricular samples before ACC to determine the transcript levels of key mediators of cell death. The mRNA level of proapoptotic *CASP3* was increased 1.5-fold in the EX group when compared with the CTR group ($P = 0.047$, Fig. 4A). However, transcript levels of other proapoptotic genes, such as *BNIP3* and *BAX*, and the anti-apoptotic *BCL-2* did not change significantly between groups (Fig. 4C–F). Additionally, difference was not observed in the expression of the necrosis marker *CYPD* between the groups (Fig. 4B).

DISCUSSION

This is the first study to examine whether exercise preconditioning prior to I/R injury in human hearts can reduce cardiac damage. One bout of exercise 24 h before CABG surgery was not protective. In contrast, exercise prior to I/R injury may render the myocardium more susceptible to damage during I/R. This was indicated by a decline in mitochondrial function during surgery and increased levels of apoptotic transcripts before the initiation of ischaemia in the EX group when compared with the CTR group.

Cardiac injury

The EX group patients had higher levels of cTnT 6 h after CABG surgery. However, patients in the EX group had a longer ACC duration. Most likely, this represents a random occurrence due to a low number of patients. In an attempt to control for the group difference in ACC duration, this was added as a covariate to the ANCOVA model. The adjusted mean difference in cTnT was reduced by 23% and lost statistical significance. The loss of significance could be due to low power and should be interpreted with caution because adjustment for ACC time did not completely remove the group difference. We did not obtain blood samples from 4 subjects in the EX group and 1 in the CTR group. This was due to a miscommunication with hospital staff. Subjects with and without missing samples had similar ACC duration making a selection bias unlikely.

Mitochondrial respiration

Preserving mitochondrial respiration can protect the heart during I/R injury [19, 20]. In our study, mitochondrial respiration was lower after I/R in the EX group, indicating increased injury to the mitochondria. Despite only 2 respiratory states being significantly

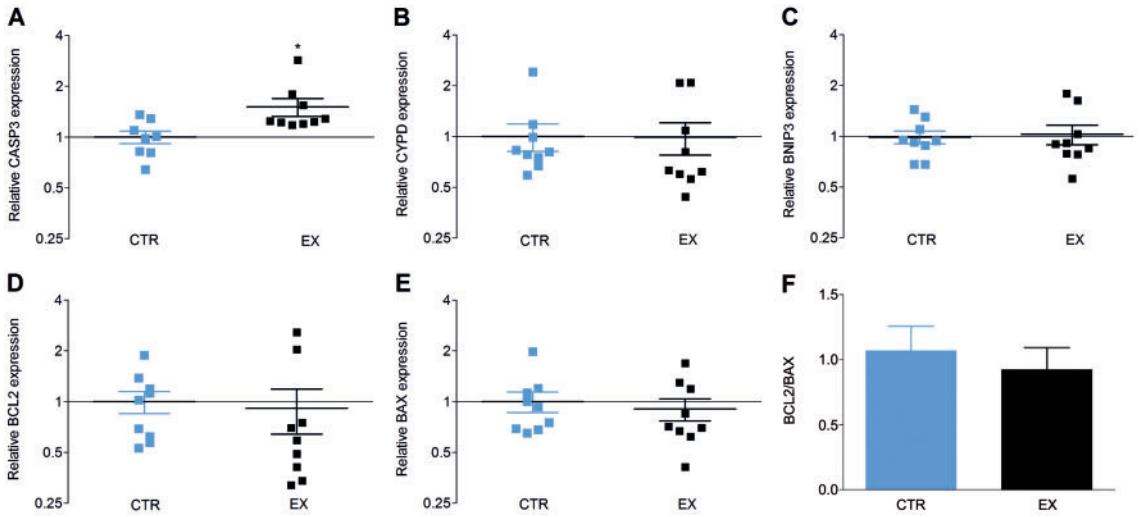

Figure 4: Exercise increases expression of the apoptotic marker CASP3 before the induction of ischaemia. Relative expression data (quantitative real-time reverse-transcriptase polymerase chain reaction; mean ± standard error of the mean) of CASP3 (**A**), CYPD (**B**), BNIP3 (**C**), BCL2 (**D**), BAX (**E**) and BCL-2/BAX ratio (**F**) in the the CTR group and the EX group. Normalization of data was performed with B2M and validated among other (c1orf and TBP) reference genes. BAX: BCL2 associated X; BCL2: apoptosis regulator; BNIP3: BCL2 interacting protein 3; CASP3: caspase 3; CTR: control; CYPD: peptidylprolyl isomerase D; EX: exercised. *P <0.05.

impaired in the left ventricle, all states showed the same tendency (Fig. 3). Together with the arguably higher cTnT values following surgery in the EX group, this shows that exercise is not inducing preconditioning as we have hypothesized. In contrast, signs of mitochondrial dysfunction, a common mechanism in I/R injury [21] became apparent. Electron transport chain complexes damaged during ischaemia are possible sources of increased ROS production during reperfusion [12], an important signal in mitochondria-initiated apoptotic pathways [22]. ROS production occurs continuously during IR, with a spike seen in early reperfusion [23]. We only saw changes in ROS production during maximal leak respiration, where it was higher in the CTR group than that in the EX group. However, as our measurements of ROS were taken after reperfusion, we cannot rule out an increased ROS production during ischaemia and early reperfusion in the EX group.

Apoptotic markers

Despite increased transcription levels of the known apoptotic marker *CASP3* [24] in the EX group when compared with the CTR group, the transcription of other members of the BCL-2 family upstream to *CASP3*, including *BAX* and *BCL-2*, was not affected by exercise. This suggests that other pathways of caspase activation may be involved. Indeed, besides the BCL2 family apoptotic cascade, *CASP3* also integrates signalling from the cytokine/Fas and the Ca^{2+}/ER pathways [25]. Further studies are clearly needed to assess how exercise might modulate *CASP3* activation and apoptosis through these pathways. Nevertheless, this initial apoptotic signalling may have left the heart more vulnerable and thereby led to the increased cardiac injury seen in the EX group after surgery. *CASP3* activation and apoptosis have been shown after a single bout of vigorous exercise in the skeletal muscle of rats [26], but chronic exercise training has shown no effect on *CASP3* in left ventricle of rats [27]. However, as our subjects performed a single exercise session 24 h prior to the biopsy, the heart might not have had the opportunity to recover and adapt to the exercise stress, and *CASP3* activation may indicate a transient exercise induced muscle damage. Although these results are reported exclusively based on mRNA levels, the increased *CASP3* levels indicate that exercise initiated apoptotic signalling before I/R, which might have left the heart more susceptible to injury.

Exercise preconditioning

There are several possible reasons for the findings conflicting with previous studies of exercise preconditioning in rodents. Rats and mice have higher metabolisms and protein turnover rates [6], and thus, the time course established in rodents may be different in humans. Patient heterogeneity in, for instance, age, comorbidities, medication use and lifestyle may also complicate the translation of animal studies [28]. Importantly, animal studies investigating exercise preconditioning normally use healthy animals in an experimental I/R protocol, whereas we selected patients with coronary disease in need of surgery, as well as comorbidities. After exercise, cardiac function can be temporarily impaired [29], and the ischaemic stress of CABG surgery superimposed on the stress from exercise may therefore leave an already diseased heart more susceptible to I/R injury, whereas healthy animals might tolerate this stress better. Two previous rat studies showing exercise preconditioning did not specify the intensity used other than to state the speed (≈25 m·min^{-1}) [7, 8], making it difficult to compare with this study. However, they employed continuous running for 30 and 100 min, precluding high intensity, meaning it was probably similar to ours. Also, we performed 30 min of exercise that was sufficient to induce preconditioning in one of the rat studies [7]. Upregulation of apoptotic markers 24 h after exercise supports the view that exercise may leave a diseased heart in a temporarily vulnerable state. This study supports that patients should refrain from strenuous exercise prior to cardiac surgery, although we cannot rule out potential preconditioning effects of exercise. Nor should the findings be taken as evidence to contraindicate long-term exercise training as a therapy in this patient group. We have previously exercise-trained

similar patients and observed improved health with no adverse effect [30].

Limitations

Being a pilot study, the number of patients included was low, and a larger number of patients may have contributed to a clearer result. We made an effort to adjust for the differences in ACC duration between groups. However, our study gave an indication for a harmful effect of exercise 24 h before CABG surgery, and therefore, we decided against repeating the same protocol in a larger study due to ethical concerns.

CONCLUSIONS

One bout of exercise 24 h prior to open-heart surgery leads to activation of apoptotic signalling and does not protect the heart from postoperative I/R injury. Although our study is small and differences in ACC time preclude a definitive conclusion, the results indicate that exercise may render the heart more vulnerable to cardiac damage during surgery, and patients should not be advised to exercise the day before CABG surgery. Although the low number of subjects in our study and differences in ACC time precludes a definitive conclusion that exercise the day before surgery is directly harmful, the indications are strong enough to recommend against protocols of similar nature.

SUPPLEMENTARY MATERIAL

Supplementary material is available at *ICVTS* online.

Conflict of interest: none declared.

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