The impact of short duration, high intensity exercise on cardiac troponin release

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Abstract

Whilst there is substantial evidence of cardiac troponin (cTnI) appearance during or subsequent to endurance exercise the database with short duration, high intensity exercise is extremely limited. The reproducibility of any exercise-related cTnI response is unclear at this point. Consequently, we recruited 18 healthy young male adults who undertook two all-out 30 s cycle sprints separated by 7 days. cTnI, blood lactate and catecholamine concentrations were measured before, immediately after and 24 hr after each bout. Cycle performance, heart rate and blood pressure responses to exercise were also recorded. Whilst cycle performance was modestly elevated in the second trial (6.5% increase in peak power output) there was no difference in the cardiovascular, lactate or catecholamine response to the two cycle trials. cTnI was not significantly elevated from baseline through recovery (Trial 1: 0.06±0.04 ng.ml^{-1}, 0.05±0.04 ng.ml^{-1}, 0.03±0.02 ng.ml^{-1} Trial 2: 0.02±0.04 ng.ml^{-1}, 0.04±0.03 ng.ml^{-1}, 0.05±0.06 ng.ml^{-1}) in either trial. Very small within subject changes were not significantly correlated between the two trials ($r=0.06$; $p>0.05$). We can conclude that short duration, high intensity exercise does not elicit a clinically relevant response in cTnI and any small alterations likely reflect the underlying biological variability of cTnI measurement within the participants.

Introduction

Recent scientific literature is replete with observations of elevated cardiac biomarkers during and after prolonged endurance exercise [1, 2, 3, 4]. Different cardiac biomarkers reflect different pathological and/or physiological processes but most attention in the sports medicine literature has been paid to the exercise-related appearance of cardiac troponins (cTnI and/or cTnT). cTnI/T are recognised as highly tissue-specific biomarkers of myocyte cell insult [5] and are important in clinical decision making when acute cardiac events are suspected [6]. An elevation in cTn either during or after exercise has, consequently, generated both interest and concern in athletes, scientists and clinicians [4].

An exercise-associated elevation in cTnI and/or cTnI has been reported after endurance events lasting a couple of hours [7] through to multiple days [8]. The mechanism(s) associated with exercise-related appearance of cTnI or T have not been proven but likely relate to the unremitting cardiovascular work and associated changes in metabolic milieu of the cardiomycocytes [4]. Most available data related to exercise-associated increases in cTnI or cTnT have been produced from field-based studies with simple pre-post blood draws [4]. This has limited our ability to interpret this data and the exact role that exercise plays. A unique study was completed by Middleton et al. (2008) [9] whereby 9 marathon runners completed a treadmill-based marathon run with blood taken every 30 min. This study demonstrated an elevation in cTnT in every single runner with some changes noted as early as 30 min after the start of exercise. In another study by the same group, cTnI was elevated in most runners completing a high intensity, steady state 30 min run [10]. Taken together these data would suggest that exercise-associated elevations of cTn are likely very common and may occur very quickly with exercise. Despite this we still require additional data to determine if a cTn-positive response is produced in response to exercise *per se*. Neither is it

clear whether very high exercise intensity (with concomitant shortened durations) may mediate cTn release [4]. Finally, to date there is limited evidence as to whether the cTn response to any exercise bout is a reproducible phenomenon.

Consequently, the aim of the current study was to assess; a) the appearance of cTnI after a short bout (30 s) of "all-out" intense exercise, and b) to determine the stability of any exercise related cTnI release in response to repeated bouts of high intensity exercise separated by 7 days recovery.

Materials and methods

Subjects and design

Eighteen apparently healthy, physically active, male university students volunteered to participate (mean±SD age: 23±2.0 yr; body mass: 75.3±11 kg; stature 175.8±5.7 cm. The study was approved by the University Ethics Committee and all participants provided written informed consent form. All participants were full familiarised to the exercise test. The study design was a repeated measures approach to biomarker assessment before and after 2 high intensity exercise trials, separated by 7 days. For six weeks prior to data collection, and throughout the study, participants maintained normal physical activity and dietary habits and refrained from macro and micro-nutrient supplementation and/or the use of pharmaceutical agents. Repeated assessments of cardiac biomarkers were made before, immediately after and 24 h after each exercise bout.

Baseline measures

At baseline body mass, stature and body composition was determined using a calibrated balanced weighing scales (Seca, UK), stadiometer (Seca, UK) and underwater

weighing procedures, respectively. Body density was assessed as described previously [11]. Relative body fat was estimated from body density [12]. Residual lung volume was measured using the simplified oxygen re-breathing method [13]. Fat free mass (FFM) was determined by subtracting fat mass from total body mass.

The Acute Exercise Test

Participants performed 2 all-out 30 s cycle ergometer tests (Monark, 864, Monark-Crescent AB, Varberg, Sweden). A force velocity test was performed one week prior to the 30s cycle ergometer test to determine optimal resistive forces based on total body mass (TBM) and fat free mass (FFM). Briefly, the test consisted of six short maximal sprints (6-8 s) against randomly assigned resistive forces (70, 75, 80, 85, 90 and 95 $g.kg^{-1}$). Successive exercise bouts were separated by a 5 min rest period. The resistive force that produced the highest PPO value for both TBM and FFM protocol was used in the 30s test. Reliability of the optimal resistive force obtained for both TBM and FFM protocol was determined using test retest methods. The order of the two 30s tests were randomised and separated by 7 days. The cycle ergometer was set-up and calibrated in the same way for all tests [14]. Saddle heights were adjusted to accommodate partial knee flexion of between 170° to 175° (with 180° denoting a straight leg position) during the down stroke and were consistent between repeated tests. Feet were firmly supported by toe clips and straps. All subjects were instructed to remain seated during the test and were verbally encouraged to perform maximally. All participants performed a standardised 5 min warm up prior to experimental data collection [15]. Participants were given a rolling start at 60 rpm for a 5 s period prior to resistive force application. On the command 'go', the subjects began to pedal maximally, the resistive force applied simultaneously, and data capture initiated. Indices of performance were calculated from flywheel revolutions using an inertia corrected computer program [14]. Data transfer was made possible using a mounted sensor unit and power supply attached to the fork of the ergometer located opposite the flywheel. The

sampling frequency of the sensor was 18.2 Hz. Validity of the cycle ergometer as a test of muscle power has been reported as r = 0.93 [16]. Heart rate was recorded throughout exercise using a short range telemetry system (Sport Tester 3000, Polar Electro Finland). Peak power output (PPO), the highest l-s value of power attained during each 30-s sprint as well as mean power output (MPO), the average power output for the 30-s period, were recorded for each trial. A fatigue index, the drop in power from a maximal to a minimal value over the 30 s was expressed as a percentage and total work done was estimated across the 30 s sprint.

Blood Sampling and analysis

Duplicate blood samples were collected at the same time of day and by the same investigator in all trials in an attempt to control for biological and between subject variation [17]. In an attempt to control for plasma volume changes, all resting samples were taken following 30 min of supine rest. The immediate post exercise samples were taken with subjects placed in a supine position on a clinical couch to minimise the risk of fainting. Capillary blood samples were collected from the right ear lobe and were analysed immediately for Haematocrit (Hawksley Micro haematocrit reader, Sussex, UK), haemoglobin (Haemocue, Sussex, UK) and blood lactate concentration (Analox P-LM5, London, UK). Changes in plasma volume were calculated from haematocrit and haemoglobin using the equations of Dill and Costill (1974) [18]. This data was used to adjust absolute data of biomarkers for any alterations in plasma volume.

At the same time venous blood samples were collected from an antecubital forearm vein using the Vacutainer System (Becton Dickinson, Rutherford, NJ, USA) and placed immediately on ice. Samples were centrifuged at 3,500 rpm for 10 min. Serum was then extracted and placed into plastic storage containers and stored at -80°C.

Adrenaline and noradrenaline concentrations were determined using the Gilson ASTED.XL (Anachem; Luton, Beds, UK), a fully automated sample processing system, with an improved sample handling system that included dialysis and sample clean-up on a strong cation trace-enrichment cartridge. The catecholamines were separated by reverse phase ion-pair chromatography. Calibrations were run every tenth and controls, every fifth sample. Coefficients of variation ranged between 1.1% and 9.3%.

Cardiac Troponin I (cTnI) concentrations were determined using the Chiron Diagnostics ACS: 180 ® Automated Chemiluminescence Systems (Medfield, MA, U.S.A.). The detection limit for cTnI was set at 0.06 ug.L^{-1}. The coefficient of variation was established at 3.5%.

Statistical Analysis

Data were analysed using a computerised statistical package (SPSS Version 2, Chigaco, USA) using parametric statistics. Significance was set at the $P < 0.05$ level. Confirmation that all dependent variables were normally distributed was assessed via repeated Kolmogorov-Smirnov tests. Changes in exercise performance and peak HR data between trials were compared using repeated measures ANOVA. Blood borne parameters and cTnI data were compared via repeated measures 2-way ANOVA (TIME: pre; immediately; and 24 h post exercise; TRIAL: TBM, FFM). Following simple main and interaction effects, Bonferroni-corrected paired samples *t*-tests were applied to make *posteriori* comparisons of the effect of time at each level of the trial factor. The delta change in cTnI from baseline to immediately post-exercise was compared between trials using a intra-class correlation test.

Results

Values for performance data and peak heart rate generated during the study for the two 30-s all out cycling trials are presented in Table 1. Although there was a small but significantly higher PPO with the FFM trial (c. 6.5%, $P < 0.05$), all other performance test data were not different between trials ($P > 0.05$). Resting HR (68 beats.min^{-1}) was the same prior to both trials and peak HR attained was not different between trials. There was a significant PVL between baseline and end-exercise ($P < 0.05$) that was not different between trials ($P > 0.05$; Table 2), with a return to baseline at 24 h post-test in both trials. This data was used to correct data for blood borne parameters and biomarkers. Of note the blood lactate concentrations rose with exercise ($P < 0.05$) and declined to baseline at 24 h post trial but these changes were similar between trials ($P > 0.05$; Table 2). Biomarkers of sympathetic neural activation, adrenaline and noradrenaline, were elevated post-exercise in both trials ($P < 0.05$; Table 2) and returned to baseline at 24 h post exercise. Again the kinetics of adrenaline and noradrenaline change with all-out 30-s exercise was consistent between trials ($P < 0.05$).

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188 Table 1. Exercise performance and HR data for the cohort over both 30 s trials.

Parameter	**Trial**	**Mean ± SD**
PPO (W)	1-TBM	953 ± 111
	2-FFM	1020 ± 130 *
Time to PPO (s)	1-TBM	4.1 ± 3.1
	2-FFM	3.5 ± 1.5
MPO (W)	1-TBM	535 ± 81
	2-FFM	512 ± 86
FI (%)	1-TBM	42 ± 8
	2-FFM	38 ±10
Total Work (J)	1-TBM	16050 ± 1828
	2-FFM	15369 ± 1975
Peak HR (beats.min^{-1})	1-TBM	177 ± 8
	2-FFM	175 ± 5

189 TBM-total body mass, FFM-fat free mass, PPO-peak power output, MPO-mean power output, FI-fatigue index,

190 HR-heart rate, * significantly different from the other trial ($P < 0.05$).

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198 Table 2. Blood borne biomarker data for both short duration high intensity cycle trials

Parameter	Trial	Baseline	Immediate Post-Ex	24 h Post-Ex
PVL (%)	1-TBM	-	-12.2 ± 5.8	4.3 ± 10.0
	2-FFM	-	-12.6 ± 6.9	5.1 ± 6.2
BLa ($mmol.l^{-1}$)*	1-TBM	0.5 ± 0.7	9.0 ± 1.2	0.6 ± 0.6
	2-FFM	1.1 ± 0.9	9.3 ± 1.4	0.7 ± 0.8
A ($nmol.l^{-1}$)*	1-TBM	0.3 ± 0.1	2.8 ± 1.6	0.3 ± 0.1
	2-FFM	0.2 ± 0.1	3.3 ± 1.9	0.3 ± 0.1
NA ($nmol.l^{-1}$)*	1-TBM	1.3 ± 0.4	19.1 ± 7.9	1.5 ± 0.5
	2-FFM	1.7 ± 0.4	20.0 ± 9.6	1.3 ± 0.4

199 PVL-plasma volume loss, BLa-blood lactate concentration, A-adrenaline, NA-noradrenaline, Ex-exercise, *-
200 significant main effect of time, but no significant main effect of trial or interaction.

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202 Mean cohort data for cTnI are presented in Figure 1. In both trials there was no
203 significant main effect of sample time, trial or interaction effect. Cohort data can mask small
204 individual changes and consequently we plotted delta cTnI from baseline to immediately
205 post-exercise from both trials in Figure 2. This Figure demonstrates that any individual
206 change is very small and largely unpredictable from trial to trial (r=-0.02; $P > 0.05$).

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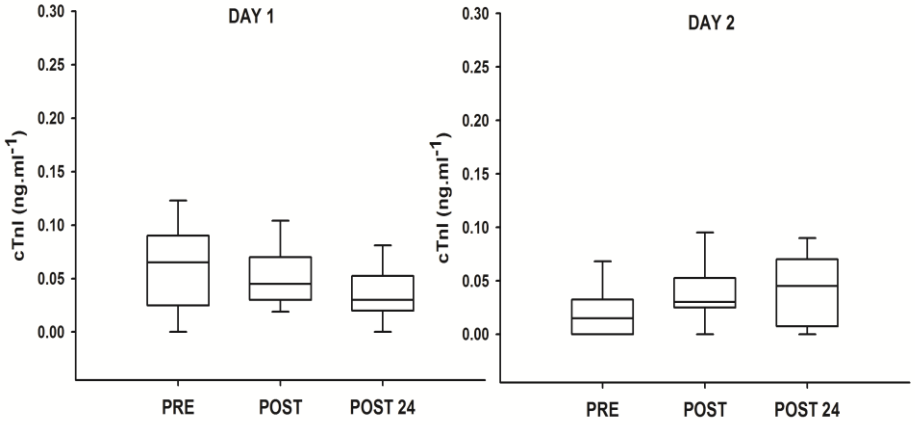

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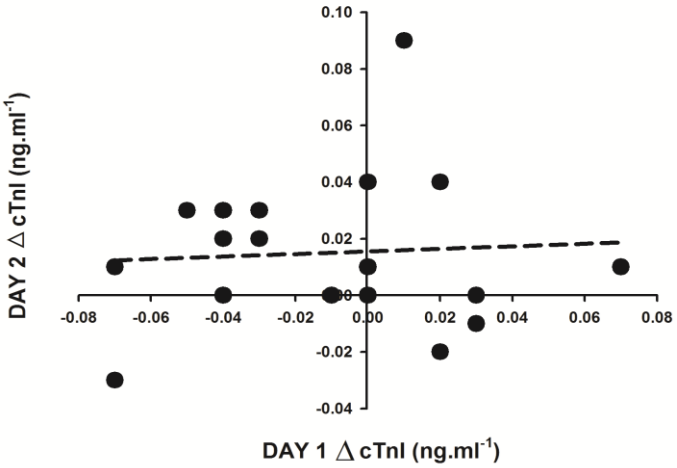
210 Figure 1. Group mean data for cTnI in Trial 1 and 2 at baseline, immediately post-exercise

211 and 24 h post-exercise.



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213 Figure 2. Scatterplot of delta cTnI responses from pre-exercise to post-exercise for trial 1 and

214 2.

215 **Discussion**

216 The key novel finding from this study is that short duration, high intensity cycling bouts (30

217 s) does not elicit a statistically or clinically significant increase in circulating cTnI during

218 immediate or late (24 hr post) recovery. Any small individual changes in response to exercise

219 are not consistent between trials and likely reflects the small biological variance in the cTnI

220 assay within a young, healthy cohort.

The extant sports medicine literature is replete with descriptive evidence of a rise in cTnI or cTnT during or after bouts of endurance exercise [4]. This phenomenon has been reported in different participant groups from the elite athlete to relatively untrained "weekend warrior" [e.g. 7, 19], from young adolescents to those in later life [e.g. 20, 21], across a range of exercise bouts [22, 23]. What has characterised nearly all studies to date has been the employment of endurance activity as it has been assumed that any cTn release with exercise is due to some mechanism associated with the unremitting haemodymanic or metabolic stress of prolonged activity on the heart. Recently, two studies have shed new and interesting insights into exercise-related cTn appearance. Firstly, Middleton et al. (2008) [22] assayed for cTn every 30 min during a treadmill-based marathon run and reported early (at 30 min) elevation of cTn is some runners and an exercise-related cTn rise in all runners at some point during the marathon. The same group then employed a high intensity 30 min run and observed a rise in cTn in most participants [10]. Taken together it was hypothesised that a cTn rise with exercise was potentially an inevitable phenomenon that would likely be physiological in nature. The current study was conceived as an extension to the work of Middleton et al. (2008) [22] and Shave et al. (2010b) [10] to determine if any exercise (per se), even 30 s of high intensity cycling, could act as a stimulus to induce cTn changes in the circulation. Whilst exercise may be a potent stimulus for cTn appearance in the systemic circulation, the lack of a statistical or clinically significant rise in cTn after a 30s all-out cycle trial in the current study would suggest that the total volume of exercise undertaken (30 s all-out) placed insufficient stress on the heart. Despite a rapid acceleration of HR with 30 s of all out cycling, there is a subsequent quick deceleration of HR suggesting that the total myocardial work and oxygen demand is very low in comparison to previous studies that have employed endurance exercise, over many hours, days and even weeks [4].

A secondary aim from this study was to determine if the cTn response to a short duration, high intensity cycle trial was consistent and repeatable if the exposure was repeated after 7 days. The repeatability of the exercise-related cTn response has received scant attention [20] but may be insightful when addressing the potential impact or mechanisms involved in cTn appearance with exercise. At one level the cTn response to the exercise intervention employed was highly consistent as there was no significant rise in cTn after both cycling trials. This consistency in a "null" response adds to the apparent repeatability in the "cTn-positive" response to exercise in a mixed adult/adolescent running study [20]. A secondary, within subject analysis in the current study assessed the consistency of the very small changes in cTn observed post-exercise. Not surprisingly the correlation of delta responses was very low (r=0.06) given the small absolute and relative changes. This likely provides some insight into the very low levels of biological variability of this cTn assay in these participants' undertaking short bouts of activity.

The implications from this study are straightforward. Short bursts of high intensity activity do not seem to result in an elevation in cTn either immediately or during later recovery from exercise suggesting this type of exercise bout does not activate the mechanism(s) required to elicit a statistically or clinically meaningful cTn response. Within the applied sports medicine setting the appearance of large cTn increase (above clinical cut-offs 0.2 $ng.ml^{-1}$) in any participants or athletes with a recent history of exercise of less than 30 min should raise a potential red flag for further clinical investigation.

As with all studies there as some limitations of note that should be followed up in on-going studies. The data in the current study pertain only to young, healthy male participants. Given

that there may be different exercise-cTn responses in different participant populations this study should be replicated in broader groups (age, sex, fitness status). Future work may employ blood samples in recovery to provide conclusive evidence of a lack cTn response to short bouts of physical activity.

In conclusion, a short duration, high intensity cycle trial does not result in an elevation of cTnI immediately post-exercise or later during recovery (24 hr). The lack of a cTn response was consistent over two cycling trials suggesting the overall response was repeatable. Small between subject variance in cTn response to the exercise stimulus was clinically meaningless and likely reflects a low level of biological variability in this assay in the current participants.

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