Title

The reliability of a pre-loaded treadmill time-trial in moderate normobaric hypoxia

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Abstract

The purpose of this study was to assess the reliability of a pre-loaded 1500 m treadmill time-trial, conducted in moderate normobaric hypoxia. Eight trained runners/ triathletes (24 ± 3 years, 73.2 ± 8.1 kg, 182.5 ± 6.5 cm, altitude specific $\dot{V}O_{2max}$: 52.9 ± 5.5 $ml \cdot kg^{-1} \cdot min^{-1}$) completed three trials (the first as a familiarisation), involving two, 15 minute running bouts at 45 % and 65 % $\dot{V}O_{2max}$, respectively, and a 1500 m time-trial in moderate normobaric hypoxia equivalent to a simulated altitude of 2500 m (F_iO_2 ~ 15 %). Heart rate, arterial oxygen saturation, skeletal muscle and cerebral tissue oxygenation (StO_2), expired gas ($\dot{V}O_2$ and $\dot{V}CO_2$), and ratings of perceived exertion were monitored. Running performance (Trial 1: 352.7 ± 40; Trial 2: 353.9 ± 38.2 s) demonstrated a low CV (0.9 %) and high ICC (1). All physiological variables demonstrated a global CV ≤ 4.2 %, and ICC ≥ 0.87, with the exception of muscle (CV 10.4 %; ICC 0.70) and cerebral (CV 4.1 %; ICC 0.82) StO_2. These data demonstrate good reliability of the majority of physiological variables, and indicate that a pre-loaded 1500 m time-trial conducted in moderate normobaric hypoxia is a highly reliable test of performance.

Introduction

The assessment of exercise performance is central to many investigations in the field of Sport and Exercise Science, allowing researchers to monitor the efficacy of a treatment or training schedule, compare different populations, and track performance over a particular time period. It is important to know the reliability of the performance test being applied, alongside accompanying physiological parameters, for similar conditions and participant cohorts, to allow accurate interpretation of results. This may include estimating the magnitude of a treatment effect, exploring individual differences, and calculating the smallest worthwhile effect [14]. Likewise, knowledge of protocol reliability can help inform sample size estimations for future investigations [14].

Typically, exercise performance is assessed using a time to exhaustion (TTE) or a time trial (TT) exercise test. The assessment of TTE involves exercise to volitional fatigue at a fixed intensity such as a percentage maximal oxygen uptake ($\dot{V}O_{2max}$), and affords the opportunity to monitor steady-state physiology whilst simultaneously providing an indication of performance response. However, TTE tests have been criticised for lacking face validity, inflicting participant boredom, and demonstrating poor reproducibility [18], with coefficient of variation (CV) values often > 25 % [18]. In contrast, TTs, which involve the completion of a set distance or amount of work (W) in the shortest possible time, more closely represent 'real world' competition, and typically demonstrate a coefficient of variation (CV) < 5 % [8].

Exercise testing in a hypoxic (i.e. low oxygen) environment is common across a range of research areas. Notably, exercise physiologists explore the effects of hypoxia on physiological functioning and exercise performance, or else evaluate the efficacy of interventions designed to mitigate the ergolytic effect of hypoxia. This has relevance for the thousands of individuals ascending to altitude (i.e. hypobaric hypoxia) each year for recreational and sporting purposes. However, it is unclear whether protocols predominantly designed for application in normoxia (i.e. sea-level) are appropriate for testing unacclimatised individuals exercising in hypoxia. Indeed, exposure to even mild hypoxia (≥ 580 m simulated altitude) reduces $\dot{V}O_{2max}$ [11,22,24,28] and has a deleterious effect on exercise performance [10,29,35] relative to normoxia. Likewise, it remains to be established whether hypoxic exercise demonstrates similar reliability to sea-level tests, given, for example, the hypoxic exercise environment will likely be unfamiliar to most participants, and it is possible that individuals may struggle to correctly self-regulate their pacing in this environment [13].

At present, the reproducibility of the hypoxic exercise response is poorly understood. MacNutt and others [23] reported good reliability (≤ 10 %) of physiological and metabolic responses across five testing sessions involving cycling in hypoxia (F_iO_2 13.0 %). However, exercise performance was not evaluated. The reliability of other physiological parameter may also be relevant during hypoxic exercise testing, yet is presently unclear. Notably, near-infrared spectroscopy (NIRS) has become a popular non-invasive method of assessing tissue oxygenation, and has been applied to distinguish between local versus systemic effects of hypoxia [30,32]; identify disparate responses to hypoxia between trained and untrained individuals [7]; and provide mechanistic information regarding the effect of interventions

designed to offset the decline in exercise performance in hypoxia [25]. Amann and colleagues [1] observed a CV of 1.0 % for a 5 km cycle ergometry TT in hypoxia (F_iO_2 15.0 %), and Arnold et al. [2] reported a CV of 3.9 % for a 10 km run TT in hypoxia (F_iO_2 15.4 %). The CV values suggest similar reliability to sea-level performance tests (i.e. < 5 %) [8]. However, in both studies, reliability data was reported secondary to a wider investigation, and only involved a sub-group of participants (four and six, respectively). Likewise, reliability of physiological variables was not reported, and limited information was provided on participant characteristics, making replication difficult. To the best of the authors' knowledge, no studies have assessed the reliability of a pre-loaded treadmill TT in normobaric hypoxia.

Therefore, the aim of this study was to investigate the reliability of a short-duration high-intensity (1500 m) treadmill TT performed in moderate normobaric hypoxia equivalent to 2500 m altitude (F_iO_2 ~15.0 %), and pre-loaded with two, 15 minute bouts of steady-state exercise corresponding to low (45 % $\dot{V}O_{2max}$) and moderate (65 % $\dot{V}O_{2max}$) exercise intensities. The TT was designed to provide a measure of high-intensity continuous running performance at a moderate altitude equivalent to that experienced by athletes on training camps or competing at altitude. The pre-load affords the opportunity to monitor important steady-state variables, which may provide useful mechanistic information to inform the performance measure. Such a protocol, if sufficiently reproducible, could be used to determine the effect of acclimatisation, training, nutritional or other interventions on physiological response and exercise performance in moderate normobaric hypoxia.

Methods

Subjects

Eight trained male runners / triathletes aged 24 ± 3 years, with a body mass of 73.2 ± 8.1 kg, height of 182.5 ± 6.5 cm, and maximal oxygen consumption ($\dot{V}O_{2max}$) (assessed at a simulated altitude of 2500 m) of 52.9 ± 5.5 $ml \cdot kg^{-1} \cdot min^{-1}$ volunteered to take part in this study. The study received institutional ethical approval and was conducted in line with the journals ethical standards [12].

Study overview

Subjects attended the laboratory on four separate occasions, all of which involved exercise in a normobaric hypoxic facility, equivalent to 2500 m altitude (F_iO_2 ~ 15.0 %). F_iO_2 was adjusted daily to account for fluctuations in barometric pressure. The first visit to the laboratory involved an incremental running test to volitional exhaustion to determine $\dot{V}O_{2max}$, whilst the second visit involved a familiarisation trial. The third and fourth visits (Trial 1 and Trial 2) constituted the experimental trials, comprising a 1500 m treadmill TT preceded by two, 15 minute steady-state exercise bouts at 45 % and 65 % of altitude $\dot{V}O_{2max}$, respectively. All exercise trials were performed at the same time of day to avoid any influence of circadian variance, and were separated by 3 – 10 days, to ensure adequate recovery. Participants completed a 24 hour food diary prior to the first exercise trial, and used this to replicate their diet as closely as possible for all subsequent visits. During this time period, participants avoided caffeine, alcohol, and strenuous exercise.

Preliminary testing

A two-part incremental running test was conducted on a motorised treadmill (Woodway, Cranlea, Birmingham, UK) [19]. Participants completed five to eight sub-maximal stages of three minutes duration, interspersed with one minute recovery periods during which time finger-tip blood samples were obtained to determine blood lactate concentrations (YSI 2300 STAT plus, Yellow Springs, Ohio). Running speed was increased by 1 $km \cdot h^{-1}$ each stage. The treadmill gradient was set to 1 %, to approximate the energetic demands of outdoor running [20]. Exercise was continued until blood lactate concentrations exceeded 4 mM or ratings of perceived exertion (RPE) reached 18. Following approximately 5 minutes recovery, the second phase of the test commenced. Running speed was fixed at the final speed obtained during the first part of the test, minus 2 $km \cdot h^{-1}$. Gradient was increased by 1 % every minute, until volitional exhaustion. Expired gas was monitored continuously throughout exercise using an online gas analysis system calibrated before each trial according to the manufacturer's instructions (MedGraphics Ultima CPX, MGC Diagnostics, MN, USA). Gas data was used to determine $\dot{V}O_{2max}$ (highest 30 second average in $\dot{V}O_2$), and the required sub-maximal running speeds for the experimental trials, via regression analysis of the $\dot{V}O_2$-speed relationship. Participants were deemed to have obtained $\dot{V}O_{2max}$ when at least two of the following criteria were met: A plateau in $\dot{V}O_2$ observed in the last stage [34], RER $\geq$ 1.15 [17], heart rate (HR) within 10 $b \cdot min^{-1}$ of age-predicted maximum (220 – age), RPE $\geq$ 19, and blood lactate concentrations $\geq$ 8 mmol [26].

Experimental protocol

Participants completed three separate experimental testing sessions, with the first serving as a familiarisation trial and the following two sessions used to calculate reliability of the exercise protocol.

Participants sat comfortably in a chair in normoxia for 15 minutes on arrival, before entering the normobaric hypoxic chamber, where they rested for a further 30 minutes. The exercise protocol then began. Participants ran on a treadmill for 15 minutes at 45 % $\dot{V}O_{2max}$. This was followed by a 5 minute rest period before a second 15 minute run commenced at 65 % $\dot{V}O_{2max}$. Participants rested for a further 5 minutes, after which they ran a 1500 m TT. Participants ran at speed approximating 80 % $\dot{V}O_{2max}$ for 30 seconds, before the TT commenced. This served as a rolling start, to limit the time taken to reach running velocity [36]. Participants manipulated speed manually using the control buttons on the treadmill unit. Running speed and time were not visible during the TT, although feedback on distance covered was given at 200 m intervals. A fixed gradient of 1 % was applied throughout the exercise protocol [20].

Measurements

After entering the normobaric hypoxic chamber, and sitting quietly for 10 minutes, a resting expired gas sample was collected. Expired gas was then continuously monitored throughout steady-state exercise via on-line gas-analysis as previously described. Breath by breath data obtained during the final 5 minutes of pre-exercise rest and final five minutes of each 15 minute steady-state exercise stage was averaged and used for data analysis. Arterial oxygen saturation (SpO_2) (Nellcor, Medtronic, Minneapolis, MN) and HR (Polar Electro, Oy, Finland) were

recorded during the final 2 minutes of pre-hypoxic exposure rest and pre-exercise rest, during the final 2 minutes of each 15 minute steady-state exercise bout, and immediately post-TT.

Muscle and cerebral tissue oxygenation was monitored continuously throughout the testing session using near-infrared spectroscopy (NIRS) (INVOS 5100C, Medtronic, Minneapolis, MN). Prior to analysis, NIRS data was averaged over the final 5 minutes of pre-hypoxic exposure rest, pre-exercise rest, and each 15 minute steady-state exercise bout, throughout the entire TT, and the first 5 minutes of post-TT recovery. The INVOS 5100C measures the reflection of NIR light to determine the ratio of oxygenated and deoxygenated haemoglobin at the measurement site. Data is expressed as a percentage of regional oxygen saturation (StO_2), with the measurable range between 15 and 95 %. Little is known about the reproducibility of StO_2 measures obtained via this device during running. A probe was placed in the middle portion of the lateral gastrocnemius on the left leg, parallel to the longitudinal axis of the lower leg. A second probe was positioned horizontally on the forehead, approximately 2 cm above the left eyebrow to measure oxygenation of the pre-frontal cortex [25]. Probes were held in place via elastic non-compressive bandages to prevent displacement and shield from external light. Hair underneath the probes was removed prior to placement via electric clippers, to avoid attenuation of the NIR light.

Statistical analysis

A combined approach to assessing test-retest reliability was applied [14]. Firstly, prior to analysis, data was naturally log-transformed to minimise heteroscedasticity [14]. Mean values were compared between Trial 1 and Trial 2 to assess systematic changes using a paired t-test (TT data) and two-way repeated measures analysis of variance (ANOVA) (all other variables) using IMB SPSS Statistics version 21.0. The degrees of freedom were adjusted in cases of asphericity, with the Greenhouse Geisser correction applied for $\varepsilon < 0.75$, and the Huynh-Feldt correction was adopted for $\varepsilon > 0.75$. A significant difference was accepted at an alpha level of $p < 0.05$. A custom made spreadsheet was then used to calculate typical error as a CV (%) and intraclass correlation coefficient (ICC) [Hopkins, W.G. Precision of measurement (2011) in internet: newstats.org/precision.html; (01/06/2015)]. An ICC > 0.90 was considered as high, 0.80 - 0.90 as moderate, and < 0.80 as low. Global data (i.e. reliability of data averaged throughout the trial) was calculated alongside time point specific data, which is relevant for analyses comparing mean values across an entire trial (e.g. ANOVA treatment effect). The smallest worthwhile change (SWC) (0.3 x within subject SD) was also computed for performance data, to determine the minimum effect necessary to represent a 'real world' change [31]. As the effects of familiarisation on TT reliability are well established [3,8], only data from Trial 1 and Trial 2 (i.e. the test-retest) is reported.

Results

1500 m TT performance

There was no systematic change ($p > 0.05$) in 1500 m run time between Trial 1 (352 ± 40 s) and Trial 2 (353.9 ± 38.2 s). The mean within-subject CV was 0.9 % (95 % CI: 0.6 – 1.9 %), and the ICC was 1.0 (95 % CI: 0.98 – 1.0). The SWC for performance was calculated as 0.8

s. Fig. 1 shows a spaghetti plot of individual 1500 m TT times and their pattern of change between trials.

Physiological variables

There was no significant difference between any of the physiological variables between trials overall or at specific measurement points (Table 1.) ($p > 0.05$). Mean HR and SpO_2 had the lowest CV (≤ 1 %) and ICC (0.98) of measured physiological variables when averaged across the different time points (Table 2.). StO_2 measured at the gastrocnemius muscle showed poor reliability across the trial (Table 1.). Further, CV increased and ICC decreased for gastrocnemius StO_2 with running speed (Table 2). Descriptive data of physiological variables are presented in Table 1. The reliability statistics for each measurement time point and global data are displayed in Table 2.

Discussion

Performance

The main novel finding of the present study was that a pre-loaded 1500 m treadmill TT conducted in moderate normobaric hypoxia ($F_iO_2 \sim 15$ %) is a highly reliable test of running performance in healthy males. The CV for 1500 m TT performance (0.9 %) is superior to values reported for an isolated (i.e. not preceded by a pre-load) 1500 m treadmill TT in well-trained runners conducted in normoxia (CV: 3.3 %) [21]. Likewise, these results compare favourably against other preloaded treadmill TTs conducted in normoxia (CV: 1.0 – 5.0 %) [8]. When viewed alongside the results of Amann and colleagues [1] who reported a CV of 1.0 % for a 5 km treadmill TT in four male runners in hypoxia (F_iO_2 15 %) and Arnold et al. [2] who observed a CV of 3.9 % for a 10 km treadmill TT in six male runners in hypoxia (F_iO_2 15.4 %), it is apparent that the addition of a moderate hypoxic stress does not necessarily affect the reproducibility of an endurance running performance test. The high ICC (1.0) and low SWC (0.8 s) also suggest this is an appropriate performance test for intervention studies conducted in hypoxia, allowing the detection of small but potentially meaningful performance changes.

Physiological measurements

Reliability statistics were also calculated for physiological parameters commonly monitored during exercise in hypoxia. There were no systematic changes in physiological variables between Trial 1 and Trial 2. This is perhaps not surprising, as significant adaptation/ acclimatisation is unlikely to occur following such a brief period of exercise in moderate normobaric hypoxia [23]. Reliability statistics averaged across the whole experimental trial for HR, RPE, $\dot{V}O_2$, $\dot{V}CO_2$ and SpO_2 are good (CV: ≤ 4.2, ICC: ≥ 0.87), and comparable to those reported during cycling (CV: ≤ 6.0 %, ICC: 0.89) [37] and running (CV: < 5.0 %, ICC: >0.9) [33] TTs conducted in normoxia.

This is the first study to investigate the reliability of StO_2 measures obtained from the INVOS 5100C during running. The assessment of StO_2 may of value to the researcher or practitioner during hypoxic exercise, by helping distinguish between local and systemic oxygen availability [30,32]. This parameter may also be useful for comparing tissue oxygenation between different populations [7], and evaluating the efficacy of interventions designed to minimise the decline

in exercise performance in hypoxia [25]. The INVOS 5100C was primarily designed for use in a clinical environment [9], but has been used to monitor recovery from a mixed exercise protocol [6] and during arterial occlusion, with and without the addition of dorsiflexion exercise [27]. Previous investigations conducted in normoxia have reported good reliability across a range of resting StO_2 values using this device [15,16].

Average CV for cerebral tissue StO_2 across the experimental trial was good (4.5 %) and at specific time-points (< 7.0 %), although ICC ranged from low to moderate (0.66 – 0.85). There was no apparent decrement in reliability statistics for cerebral tissue StO_2 measurements during exercise (Table 2.). In contrast, the reliability of StO_2 at the gastrocnemius was highly variable. During low intensity exercise (45 % $\dot{V}O_{2max}$) (CV: 5.0 %, ICC: 0.96), gastrocnemius StO_2 showed superior reliability to that previously reported for running at lactate threshold (ICC: 0.87) and $\dot{V}O_{2max}$ (ICC: 0.88) [4], and comparable to during a 5 km TT (CV: 3 %, ICC: 0.94) [33] in other NIRS devices. However, variability increased during moderate intensity exercise (CV: 10.9 %, ICC: 0.88) and TT (CV: 28.8 %, ICC: -0.18), suggesting decreasing reliability at faster running speeds. Considerable muscle deformation during high-intensity running likely changed the pathlength of NIR light through the muscle, leading to motion artefacts which may have been misinterpreted as alterations in the relevant chromophore (i.e. oxyhaemoglobin and deoxyhaemoglobin) concentrations [5]. Anecdotally, there was also regular loss of signal during high-intensity running, probably due to separation of the NIRS probe and skin, which may have introduced further variability into the NIRS signal. This suggests an unsuitability of this device during high-intensity running, when trying to measure StO_2 at the gastrocnemius, as considerable measurement variability would likely mask any changes in StO_2 subsequent to an intervention. Whether there would be improved reliability at a different muscle group or non-exercising muscle is yet to be established. There may be a viable role for this device in monitoring changes in cerebral tissue StO_2, and gastrocnemius StO_2 during resting/ low-intensity exercise.

Interestingly, reliability appeared to be better for both resting cerebral and gastrocnemius tissue StO_2 in hypoxia relative to normoxia (Table 2). This is puzzling, but may be related to the duration of rest prior to measurement. Pre-exposure measurements were taken after 10 minute of rest, which was assumed to be sufficient to provide a consistent baseline value. Conversely, pre-exercise measurements were preceded by 25 minutes of rest in hypoxia, deemed necessary to elicit a physiological response to the low oxygen environment. The longer period of rest may have allowed a more consistent resting value to be obtained.

Conclusion

The current research suggests that a pre-loaded 1500 m treadmill TT conducted in moderate normobaric hypoxia is a highly reliable test of running performance in trained men. The majority of physiological variables demonstrated good reliability. Caution is advised when applying the INVOS 5100C to monitor StO_2 at the gastrocnemius during higher-intensity running, given the large measurement variability observed in this study. Together with the data of other investigations, this study suggests that the addition of a hypoxic stimulus does not necessarily compromise the reliability of physiological and performance measures obtained

from a pre-loaded TT. Therefore, this protocol is suitable for use in studies evaluating the effects of acclimatisation, training, nutritional or other similar interventions on physiological responses and exercise performance conducted in trained men exercising in moderate normobaric hypoxia.

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Table 1. Descriptive statistics of physiological variables calculated at individual time points and globally throughout the experimental trials. Data is presented as Mean ± SD.

	Pre-exposure		Pre-exercise		45 % $\dot{V}O_{2max}$		65 % $\dot{V}O_{2max}$		TT		Post-exercise		Global data	
	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
HR ($b \cdot min^{-1}$)	62 ± 8	62 ± 7	63 ± 8	65 ± 8	105 ± 14	104 ± 13	147 ± 17	148 ± 15	180 ± 11	182 ± 9	-	-	112 ± 49	112 ± 49
RPE (AU)	-	-	-	-	10 ± 2	10 ± 1	13 ± 2	12 ± 1	-	-	19 ± 2	19 ± 2	14 ± 4	14 ± 4
$\dot{V}O_2$ ($ml \cdot kg^{-1} \cdot min^{-1}$)	-	-	5.0 ± 0.5	5.0 ± 0.8	19.4 ± 5.8	19.4 ± 5.7	34.8 ± 2.6	35.2 ± 2.9	-	-	-	-	19.7 ± 12.9	19.9 ± 13.1
$\dot{V}CO_2$ ($ml \cdot kg^{-1} \cdot min^{-1}$)	-	-	5.0 ± 0.7	5.0 ± 0.7	19.0 ± 6.6	18.8 ± 7.0	35.0 ± 3.1	34.6 ± 2.7	-	-	-	-	19.7 ± 13.2	19.4 ± 13.0
SpO_2 (%)	97 ± 1	98 ± 1	91 ± 2	91 ± 2	85 ± 5	85 ± 5	83 ± 3	82 ± 4	-	-	78 ± 3	78 ± 3	87 ± 7	87 ± 8
Gastroc StO_2 (%)	68 ± 8	66 ± 10	65 ± 8	65 ± 9	63 ± 12	63 ± 12	54 ± 13	52 ± 12	51 ± 8	53 ± 16	72 ± 10	73 ± 10	62 ± 11	60 ± 12
Cerebral StO_2 (%)	72 ± 7	71 ± 6	63 ± 5	66 ± 6	59 ± 4	62 ± 5	59 ± 5	60 ± 5	52 ± 7	54 ± 6	63 ± 7	63 ± 7	61 ± 8	63 ± 8

s = seconds, $b \cdot min^{-1}$ = beats per minute, AU = arbitrary units, SpO_2 = arterial oxygen saturation, RPE = ratings of perceived exertion, $\dot{V}O_2$ = oxygen consumption, $\dot{V}CO_2$ = expired carbon dioxide, Gastroc StO_2 = regional oxygen saturation of the gastrocnemius, Cerebral StO_2 = regional oxygen saturation of the pre-frontal cortex, CV = coefficient of variation, ICC = intraclass correlation coefficient, TT = time-trial.

Table 2. Reliability statistics of physiological variables calculated at individual time points and globally throughout the experimental trials.

	Pre-exposure		Pre-exercise		45 % $\dot{V}O_{2max}$		65 % $\dot{V}O_{2max}$		TT		Post-exercise		Global data	
	CV (%)	ICC	CV (%)	ICC	CV (%)	ICC	CV (%)	ICC	CV (%)	ICC	CV (%)	ICC	CV (%)	ICC
HR ($b \cdot min^{-1}$)	3.5	0.95	3.8	0.95	1.5	0.99	1.4	0.99	-	-	1.2	0.97	1.0	0.98
RPE (AU)	-	-	-	-	12.9	0.58	5.0	0.89	-	-	3.6	0.91	4.2	0.87
$\dot{V}O_2$ ($ml \cdot kg^{-1} \cdot min^{-1}$)	-	-	9.2	0.70	6.0	0.97	4.1	0.80	-	-	-	-	4.0	0.93
$\dot{V}CO_2$ ($ml \cdot kg^{-1} \cdot min^{-1}$)	-	-	10.2	0.62	4.4	0.99	4.4	0.80	-	-	-	-	4.0	0.96
SpO_2 (%)	0.8	0.51	0.7	0.95	0.6	1.0	2.2	0.81	-	-	1.3	0.95	0.5	0.98
Gastroc StO_2 (%)	9.4	0.58	5.3	0.89	5.0	0.96	10.9	0.88	28.8	-0.18	3.3	0.97	10.1	0.71
Cerebral StO_2 (%)	6.2	0.66	4.2	0.85	4.3	0.75	5.2	0.71	6.6	0.79	6.9	0.74	4.5	0.80

s = seconds, $b \cdot min^{-1}$ = beats per minute, AU = arbitrary units, SpO_2 = arterial oxygen saturation, RPE = ratings of perceived exertion, $\dot{V}O_2$ = oxygen consumption, $\dot{V}CO_2$ = expired carbon dioxide, Gastroc StO_2 = regional oxygen saturation of the gastrocnemius, Cerebral StO_2 = regional oxygen saturation of the pre-frontal cortex, CV = coefficient of variation, ICC = intraclass correlation coefficient, TT = time-trial.

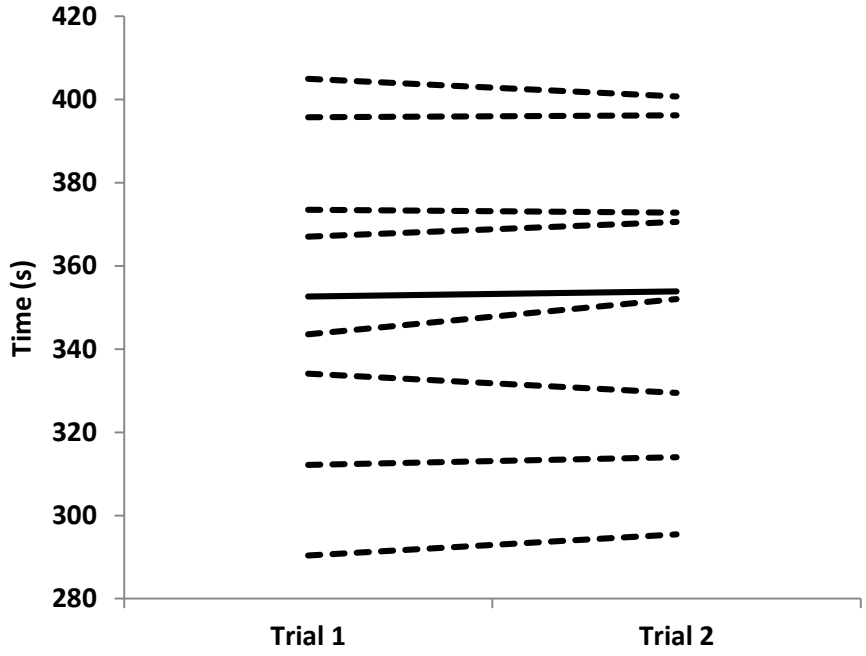

Figure 1. Spaghetti plot of individual performance times (dashed lines) and mean TT time (solid line) for 2 repeat pre-loaded 1500 m running TTs.