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Title

Reliability of near-infrared spectroscopy for measuring intermittent handgrip contractions in sport climbers

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Reliability of NIRS

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ABSTRACT

The use of near infra-red spectroscopy (NIRS) to investigate muscle oxygenation changes during physical tasks such as rock climbing has rapidly increased within recent years; yet there is no known measure of reliability. The current study aimed to determine inter-session reliability and minimal detectable change (MDC) of continuous wave NIRS parameters during intermittent handgrip contractions in rock climbers. Thirty-two **s**port climbers were tested for exhaustive intermittent handgrip exercise (8s contraction – 2s relief) at 60 % of maximal voluntary contraction on three separate days. During each visit, continuous wave NIRS was used to determine tissue saturation index (TSI) as the measure of tissue oxygenation in the flexor digitorum profundus. To assess the inter-session reliability, the intra-class correlation (ICC), standard error of measurement (SEM), coefficient of variation (CV), and minimal (MDC) were used. Mean de-oxygenation during the contractions provided reliable results (Δ TSI; first trial -8.9 ± 2.9%, second trial -8.8 ± 2.7%, and third trial -8.4 ± 2.6%; ICC = 0.692; SEM = 1.5%; CV=17.2%; MDC = 4.2%). Mean muscle re-oxygenation during the relief periods was similarly reliable (Δ TSI; first trial 9.0 ± 3.1%, second trial 8.8 ± 2.9%, and third trial 8.5 ± 2.7%; ICC = 0.672; SEM = 1.7%; CV=19.0%, MDC = 4.7%). As such, continuous wave NIRS provides a reliable measure of de-oxygenation and re-oxygenation during intermittent contractions to failure in the forearm flexors of rock climbers. Differences exceeding ~4.5% for ΔTSI during contraction and relief periods should be considered meaningful.

KEYWORDS. NIRS, tissue saturation, haemoglobin, forearm flexors, oxygenation

INTRODUCTION

The use of near infra-red spectroscopy (NIRS) to investigate muscle oxygenation changes during physical activity has rapidly increased within recent years (8). The technique relies on assessing the relative transparency of near-infrared light through tissue, and oxygen dependent absorption characteristics of haemoglobin and myoglobin (9, 15). As the light spectrum cannot distinguish haemoglobin from myoglobin, for convenience (6, 20), we will refer to haemoglobin in the manuscript. Using continuous wave NIRS (cw-NIRS), relative or absolute concentrations of oxy-haemoglobin (O_2Hb), de-oxyhaemoglobin (HHb) and total haemoglobin (tHb) can be estimated (8). Cw-NIRS assumes homogeneity of the detected tissue and due its simplicity, the devices are suitable for field measurement.

Previously, cw-NIRS derived parameters during exercise have been shown to be a valid tool for determination of local muscle oxygen consumption, oxygenation threshold, tissue de-oxygenation and O_2 recovery rate (3, 27, 29). The use of cw-NIRS is, therefore, promising as a diagnostic tool in sports where muscular hypoxia is common such as local isometric contractions or high intensity exercise seen in rock climbing. Although this technique has been extensively used in this population to assess muscle oxygenation (1, 11-14, 18, 19, 22), the reliability of cw-NIRS is not known. As such, it is not known whether reported changes are of physiological significance or due to variability with the data collection methods. Previously, reliability studies assessing muscle oxygenation have been limited to laboratory conditions and/or using frequency domain spectroscopy (5, 21, 24, 26). There is no known study which has aimed to determine reliability within an ecologically valid setting. This may be in part due to the complex nature of determining reliability in sport specific settings where controlling confounding factors is inherently difficult. However, given the growing use of this technique in both climbing and clinical exercise populations (1, 11-14, 18, 19, 22), determining the reliability of cw-NIRS derived parameters would provide important information for clinicians and couches.

Therefore, the aim of the current study was to determine inter-session reliability and minimal detectable change of cw-NIRS parameters during intermittent handgrip contractions in rock climbers.

METHODS

Experimental Approach to the Problem

A repeated measures design (test-retest method) was used to assess inter-session reliability of cw-NIRS derived parameters. Subjects visited an environmentally controlled laboratory (20.0 ± 0.7 °C) on four separate occasions during a three-week period (Figure 1). Prior to each visit subjects were asked to restrain from heavy physical activity and alcohol for 24 hours, caffeine for 12 hours and food 3 hours. The first visit was used to determine anthropometric markers and finger flexor maximal voluntary contraction (MVC) using a climbing specific handgrip dynamometer (Figure 2). Additionally, subjects familiarized themselves with the intermittent handgrip test. During the second, third and fourth visit (three separate sessions), subjects performed the intermittent handgrip endurance task until volitional failure occurred (contracting for 8 s at 60% MVC and relaxing for 2s). Each of these three visits was separated by 3-6 days and completed in the afternoon between 14.00 and 18.00. All testing was conducted during early spring when climbing training is focused on high volume with submaximal intensity.

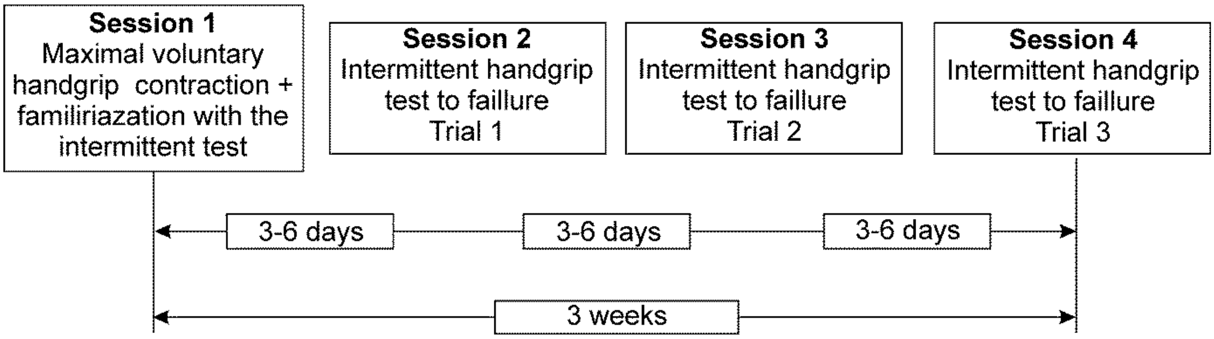

Figure 1. Design of the study indicating four visits of all subjects (N=32). Inter-session reliability of the intermittent handgrip endurance test to failure was assessed from three trials.

Subjects

Sport climbers were chosen as they are accustomed to the repeated nature of high intensity contractions. As such, 32 sport climbers (age range 20-44 yrs) volunteered (15 males: age 27.7 ± 10.2 yrs; body mass 71.0 ± 9.3 kg, height 178.3 ± 9.7 cm; 17 females: age 26.3 ± 4.6, body mass 57.7 ± 5.6 kg; 166.4 ± 5.7 cm). Self-reported climbing ability ranged from 11 to 23 on the IRCRA scale which corresponds to intermediate to advanced ability level defined by Draper et al. (7). The mean climbing experience was 9.3 ± 6.2 years and the volume of climbing training 5.8 ± 2.3 hours per week. Subjects were healthy non-smokers who were not taking any vascular acting medications. The study conformed to the recommendations of the local Ethics Committee in accordance with the Declaration of Helsinki. All subjects were informed of the experimental risks and signed an informed consent document prior to data collection.

Anthropometric and skinfold measurement

Body mass was measured on an electronic scale (Soehnle 7730.01.001, Backnang, Germany) with the subjects wearing underwear only. Height was measured using a stadiometer (SECA, Vogel & Halke GmbH, Hamburg, Germany). Forearm skinfold thickness (under the cw-NIRS optode) was measured using a skinfold calliper (Harpenden, Baty, West Sussex, UK). Adipose fat was not considered to effect the NIR signal as skinfold measures were below the limits set by Van Beekvelt et al. (25) (1.7 ± 0.5 and 2.0 ± 0.5 mm for male and females respectively).

Finger strength and endurance tests

Each session began with the same warm-up, which consisted of 5-min stair walking, 5-min traversing on a climbing wall and 5-min individual one-arm intermittent hanging on a 30- and 23-mm-wide rung.

To test for finger flexor strength and endurance, a custom-made dynamometer (3D-SAC, Spacelab, Sofia, Bulgaria) (Figure 2) was used. The dynamometer enabled the subjects to control the intensity and duration of each contraction. Further, the device provided real time feedback using both visual and acoustic signals. The dynamometer was calibrated for a wooden hold 23 mm deep which maximized activation of the flexor digitorum profundus (FDP) (23).

During the first visit, MVC of the dominant finger flexors was completed twice (separated by two minutes). On presentation of an acoustic signal, subjects were asked to progressively pull on the rock climbing hold as hard as possible for 5 s. Subjects were verbally encouraged to achieve maximal effort throughout. The highest value from the two trials was used to determine their relative workloads (60% MVC) in the following endurance trials. MVC was determined whilst standing with the shoulder flexed to 180° and the elbow fully extended to simulate sport specific conditions (2), and to prevent blood pooling (25).

The intermittent handgrip endurance test was performed in the same anatomical position as the MVC trials. Each contraction was performed at 60% MVC with a ratio of 8 s contraction to 2 s relief. The test started with an acoustic signal and subjects were provided with visual feedback to ensure the correct force was applied. If this force dropped by more than 10% for more than 1 s, the test was automatically terminated. An acoustic signal as well as the visual display marked the start and end of each contraction/relief period. Subjects were required to achieve the desired target force as quickly as possible. Only the time in the target zone was used in the analysis.

Near-infrared spectroscopy

NIRS optodes were placed in 1/3 of proximal distance between medial epicondyle of humerus and carpus. The FDP was located by a chartered physiotherapist using the technique suggested by Schweizer and Hudek (23), where the thumb and the first finger were squeezed together. The flexor was then palpated to locate the middle of the muscle belly. Optodes were placed in a secure holder on the skin using bi-adhesive tape. The holder was fixed using dark tape to prevent contamination from ambient light (Figure 2). To ensure no variability in optode placement between sessions, pen marks were used on the skin.

Cw-NIRS (Oxymon, Artinis Medical System, BV, The Netherlands) was used to assess the oxygenation and blood volume changes. OxyMon uses two lengths of NIR light at 765 and 855nm. The absorption changes of the wavelengths used are converted into concentration changes of O_2Hb and HHb using a modified Lambert–Beer law in which a path-length factor is incorporated to correct for scattering of light in the tissue (9). Two emitting optodes were spaced 4 cm and 3.6 cm from one receiver, respectively. This configuration enables the calculation of absolute ratios of O_2Hb, HHb and TSI using the spatially resolved spectroscopy approach. Absolute concentration of tHb were estimated from the path-length factor, which was fixed to 4.0 as suggested by van Beckvelt et al. (26) for forearm measurements. Sampling frequency was set to 10 Hz and data were sent via optical cables in personal computer, where online recording and all parameters were undertaken using the Oxysoft software (Artinis Medical System, BV, The Netherlands). Only TSI and tHb from spatially resolved spectroscopy were included in the analysis as the measures of tissue oxygenation and blood volume (15). TSI is determined as O_2Hb/(HHb+O_2Hb) and, it is, therefore, independent of actual volume of tHb under the optodes. Resting tHb and TSI were determined using the last minute of 10 min seated rest. Changes of tissue oxygenation and blood volume during exercise (Δ TSI, Δ tHb) were calculated from all 8 s contractions and 2 s relief periods, respectively. Moreover, the minimum and mean minimum concentrations of tHb (tHb_{min}, $tHb_{mean\ min}$) and TSI (TSI_{min}, $TSI_{mean\ min}$) from the contraction periods were used in the analysis (Figure 3). To enable comparisons with other studies (12, 22), de-oxygenation (Δ TSI during contraction) and re-oxygenation (Δ TSI during relief) from the first and last three contraction and relief periods were calculated.

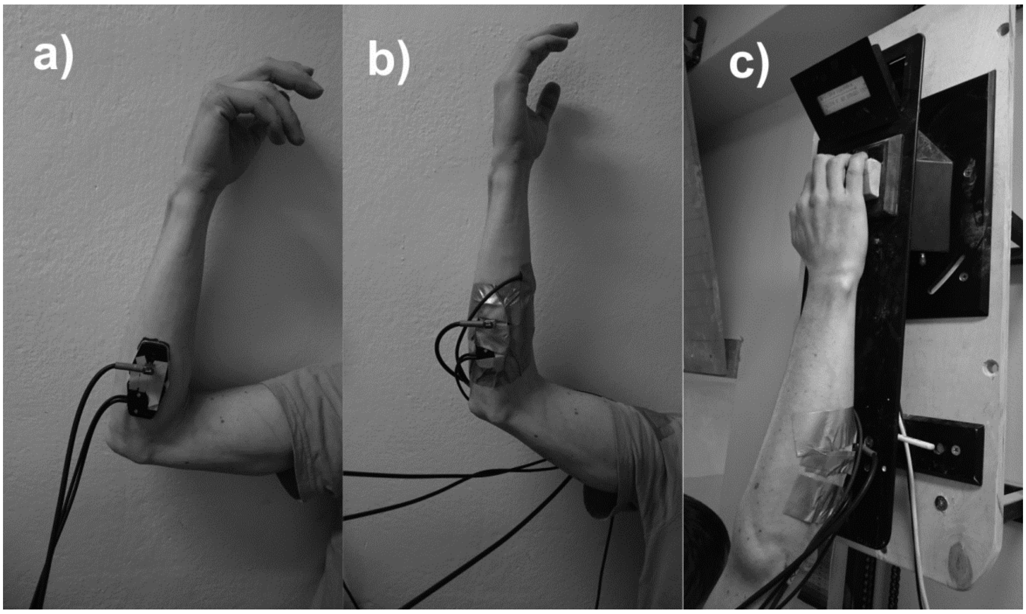

Figure 2. a) Placement of optodes over the belly of flexor digitorum profundus; b) fixing the optodes by dark tape; c) arm and finger position on the custom made dynamometer

Statistical analysis

Descriptive statistics (mean ± *s*) were used to characterize time of the test, TSI, and tHb during rest and exercise. The differences between trials were assessed using repeated measures ANOVA. To assess the inter-session reliability, several coefficients were used as recommended by Hopkins (17) and Weir (28).

ICC was calculated from the equation:

$$ICC = \frac{MSB - MSW}{MSB + (k-1)MSW},$$

where MSB and MSW correspond to mean squares between and within subjects from repeated measure ANOVA, respectively, and k is the number of trials (3 in this case). This equation encompasses both the variability due to systematic changes between trials and error variability. ICC was expressed with 95% confidence interval.

Coefficient of variation (CV) was calculated as:

$$CV = \frac{\sqrt{MSW}}{\mu},$$

where μ is the mean of the three trials.

Standard error of measurement (SEM) was computed from the equation $SEM = \sqrt{MSW}$. This equation is to be preferred as it is not dependent on the type of ICC (28).

The minimal detectable change (MDC) often referred to as the "smallest detectable difference" was calculated from the SEM as $MDC = SEM \times 1.96 \times \sqrt{2}$ (28). MDC determines the magnitude of change that must be observed before the change can be considered to exceed the measurement error and variability at the 95% confidence level (16).

Statistical significance was set to $P < 0.05$. Partial eta squared (η_p^2) was calculated to evaluate the effect size of differences between trials. As Weir (28) showed that the ICC values are influenced by between subject variability and, therefore, higher ICC values can be found in heterogeneous samples even if the reliability is poor and vice versa, the reliability was not scaled as high, moderate or low. Additionally, CV is often criticized as it determines the error variability related to a common mean. When the common mean approaches zero the CV increases even for low standard deviations. Due to these delimitations, MDC was introduced.

MDC is an absolute measure of reliability (measurement error) and is being increasingly used to assist in interpreting results and determining whether a change between repeated tests is random variation or a true change (16). Statistical computations were performed using IBM SPSS for Windows (version 22, Chicago, Il., USA).

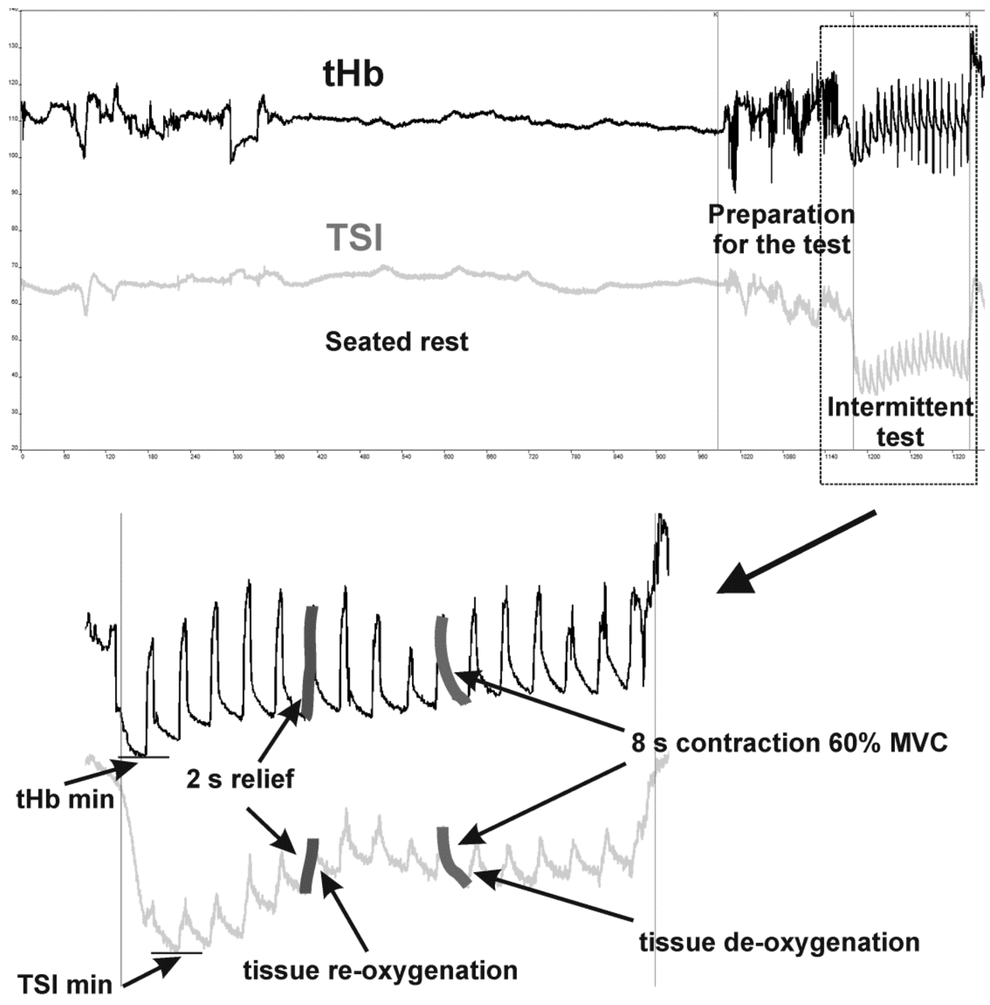

Figure 3. Typical example of the testing protocol with seated rest, preparation for the test and the intermittent test. Total haemoglobin (tHb) and tissue saturation index (TSI) are depicted on the lines. TSI min and tHb min indicate the lowest value during contraction in the intermittent test. Tissue re-oxygenation and de-oxygenation designate % changes in TSI during relief and contraction period of the test.

RESULTS

There were no differences between trials for time to failure or cw-NIRS derived parameters, which indicates no systematic error between trials (Table 1). The ICC for cw-NIRS derived parameters ranged from 0.294 to 0.692 and CV from 8.3% to 41.8%. Mean ΔTSI from all contractions and relief periods (Table 2) provided the highest inter-session reliability with respect to other parameters (ICCs = ~0.7, CV <20%, and MDC = ~ 4.5%). ΔTSI from the first and last three contractions showed higher error variability and measurement error (ICCs = 0.48-0.65, CV = 21-24%, MDC = 5.3-6.1%) than mean ΔTSI from all contractions. Scores of ΔtHb, tHb_{min} and TSI_{min} provided the lowest inter-session reliability, and high measurement error (Table 2). Individual responses are depicted in Figure 4 where dark lines show climbers with the variability between trials exceeding the MDC.

Table 1. Mean (± *s*) time to failure in the intermittent handgrip test with tissue saturation index (TSI) and total haemoglobin (tHb) changes during rest, contraction and relief periods in three trials. Statistical differences (*P*) between trials were assessed by RM-ANOVA.

	1st trial	**2nd trial**	**3rd trial**	***P* value**	η_p^2
Time till failure (s)	91.3 ± 23.7	91.3± 23.3	92.9 ± 25.2	0.667	0.011
TSI rest (%)	59.1 ± 5.5	58.9 ± 6.6	58.6 ± 7.3	0.914	0.003
tHb rest (μmol)	109.4 ± 19.1	112.8 ± 27.8	110.2 ± 25.5	0.626	0.015
Contractions					
Δ TSI - all contractions (%)	-8.9 ± 2.9	-8.8 ± 2.7	-8.4 ± 2.6	0.487	0.009
Δ TSI - first three contractions (%)	-9.2 ± 2.7	-9.1 ± 2.9	-8.6 ± 2.3	0.383	0.001
Δ TSI - last three contractions (%)	-9.2 ± 3.9	-8.9 ± 3.2	-8.8 ± 3.5	0.746	0.022
Δ tHb - all contractions (μmol)	-17.4 ± 9.8	-15.0 ± 7.0	-15.4 ± 6.5	0.307	0.006
TSI min (%)	32.5 ± 9.7	31.5 ± 10.9	33.4 ± 10.0	0.605	0.016
TSI mean min (%)	36.9 ± 8.3	36.0 ± 10.6	37.6 ± 9.8	0.676	0.019
tHb min (μmol)	89.5 ± 14.4	84.9 ± 14.1	88.8 ± 15.6	0.153	0.028
tHb mean min (μmol)	95.8 ± 15.7	90.9 ± 14.2	94.6 ± 15.9	0.163	0.026
Reliefs					

∆ TSI - all reliefs (%)	9.0 ± 3.1	8.8 ± 2.9	8.5 ± 2.7	0.547	0.012
∆ TSI - first three reliefs (%)	8.2 ± 2.9	8.4 ± 3.1	7.8 ± 2.4	0.398	0.002
∆ TSI - last three reliefs (%)	9.6 ± 3.9	9.1 ± 3.4	9.1 ± 3.5	0.620	0.016
∆ tHb – all reliefs (µmol)	18.7 ± 10.1	16.3 ± 7.3	16.8 ± 6.6	0.337	0.003

Table 2. Intra-class coefficient of correlation (ICC), coefficient of variation (CV), standard error of measurement (SEM) and minimal detectable change (MDC) for time to failure, tissue saturation index (TSI) and total haemoglobin (tHb) changes during rest, contraction and relief periods of the intermittent handgrip test.

	ICC	**95 % ICC**	**CV**	**SEM**	**MDC**
Time (s)	0.855	0.759-0.921	10.0 %	9.2	25.5
TSI rest (%)	0.420	0.205-0.629	8.3 %	4.9	13.6
tHb rest (µmol)	0.656	0.478-0.798	12.9 %	14.3	39.6
Contractions					0.0
∆ TSI - all contractions (%)	0.692	0.526-0.822	17.2 %	1.5	4.2
∆ TSI - first three contractions (%)	0.480	0.269-0.674	21.5 %	1.9	5.3
∆ TSI - last three contractions (%)	0.652	0.473-0.795	23.5 %	2.1	5.8
∆ tHb - all contractions (µmol)	0.294	0.078-0.524	41.8 %	6.6	18.3
TSI min (%)	0.437	0.223-0.642	23.5 %	7.6	21.1
TSI mean min (%)	0.432	0.217-0.638	19.6 %	7.2	20.0
tHb min (µmol)	0.522	0.316-0.705	11.6 %	10.2	28.3
tHb mean min (µmol)	0.502	0.293-0.690	11.5 %	10.8	29.9
Reliefs					
∆ TSI - all reliefs (%)	0.672	0.499-0.808	19.0 %	1.7	4.7
∆ TSI - first three reliefs (%)	0.545	0.342-0.722	23.6 %	1.9	5.3
∆ TSI - last three reliefs (%)	0.612	0.423-0.769	24.3 %	2.2	6.1
∆ tHb – all reliefs (µmol)	0.304	0.088-0.533	39.4 %	6.8	18.8

DISCUSSION

To date there is no known research determining reliability of cw-NIRS during high intensity intermittent contractions during an ecologically valid sport performance setting. As such the main aim of the study was to determine the inter-session reliability and MDC of cw-NIRS parameters during intermittent handgrip contractions to failure in rock climbers. The main finding of the study was that 1) cw-NIRS provides a reliable measure of forearm muscle oxygenation, and 2) during both rest and exercise, MDC was determined for indicators of muscle oxygenation and blood volume.

To our knowledge, the reliability of NIRS in forearm muscles during exercise was only assessed using frequency domain NIRS and/or whilst completing protocols without exhaustion in very standardized laboratory settings (4, 5, 26). Frequency domain spectroscopy provides absolute concentrations of HHb + O_2Hb and accounts for the variability of many biological factors which cw-NIRS does not (8). Using incremental handgrip exercise, Celie et al. (4) reported ICCs between 0.025 and 0.873 for HHb, and 0.22 to 0.774 for TSI. From similar handgrip exercise, Van Beekvelt et al. (26) found CVs for muscle oxygen consumption ranging from 13.3% to 23.2%. No other reliability parameters were reported and we can only estimate that the high variability in the ICCs might be due to low reproducibility of the measurements, or low between-subject variability. Nevertheless, the authors suggested that their protocol was highly reliable for determination of HHb and TSI. These findings cannot be compared to our protocol due to different methodology and parameters used. Parameters in the current study were chosen due to the extensive amount of sport rock climbing literature which has focused on them over the past decade, with no known measure of reliability (1, 11, 12, 19, 22).

Previous rock climbing literature has shown that Δ TSI (rate of de-oxygenation) during intermittent handgrip contractions at 40 - 60% MVC was greater in experienced climbers than in non- or lower grade climbers (group differences for Δ TSI ranging from 3% to 9.4%) (1, 12, 19, 22). As such, it has been suggested that rock climbers may have vascular adaptions occur during training. The current study found that MDC for Δ TSI during contractions is 4.2%; this exceeds the previously reported differences between less and more experienced athletes in numerous previous studies (1, 12, 22). As such, all between group differences, which are less than 4.2% maybe due to error variability or low reproducibility of measurement, and so caution should be used when interpreting such data. It seems plausible that climbing ability level is not solely related to hemodynamic changes in forearms, and other factors such as muscle fiber types, training preferences (endurance or strength) or general fitness may also play a substantial role in TSI responses. Data from the current study is in agreement with recent research into the population which suggested that the unknown variability in NIRS methods should mean that data should be interpreted with caution (1, 10).

TSI_{min} during handgrip exercise has previously been associated with a greater endurance performance and oxygen utilization in the muscle (1, 13). In the current study, the MDC for $TSI_{mean\ min}$ was 20%. This high variability may be caused by the high variability in TSI_{rest} ($TSI_{mean\ min}$ depends on the baseline value of TSI_{rest}). At rest, TSI had poor inter-session reliability (ICC = 0.420; SEM = 4.9 %; CV = 8.3 %). It is likely that this was affected by the previous warm-up, which was needed for safety reasons. To increase inter-session reliability, it seems advisable that future research provides 1) a prolonged rest time following a warm-up, and 2) as previously suggested by Celie et al. (4); use the relative decrease in TSI (from baseline/rest).

There were a similar number of men and women used; separate reliability analyses for each sex were calculated. No substantial changes between males and females were found and consequently the results were not reported. We are in agreement with Crenshaw et al. (5) who did not find any systematic sex differences in forearm and shoulder oxygenation after isometric contractions and blood occlusion.

Individual responses in three trials (Figure 4) suggest that the inter-session reliability was decreased by the high intra-individual variability of few subjects. Several reasons may have led to these individual responses such as small shift of optode placements, tissue movement during contractions, different dietary uptake and hydration statuses and the state of fatigue and/or recovery. Although the climbers were instructed to abstain from previous heavy exercise, caffeine and food intake (see Methods); hemodynamic responses are very complex and the control of all factors during testing in an ecologically valid setting is difficult.

In order to fully interpret data from the current study it is important to highlight a number of strengths and weaknesses. This was the first study to determine reliability of high intensity contractions in a sport specific setting, thus increasing ecological validity whilst potentially compromising reliability. The current study had a relatively large sample size; however, data is limited to athletic populations. The use of this experienced group and the inclusion of a familiarization session would have meant the learning effect was likely negligible. Due to the inherent interpretation problems of ICC's and CV's, several reliability coefficients were calculated to better describe the reproducibility of the measurements.

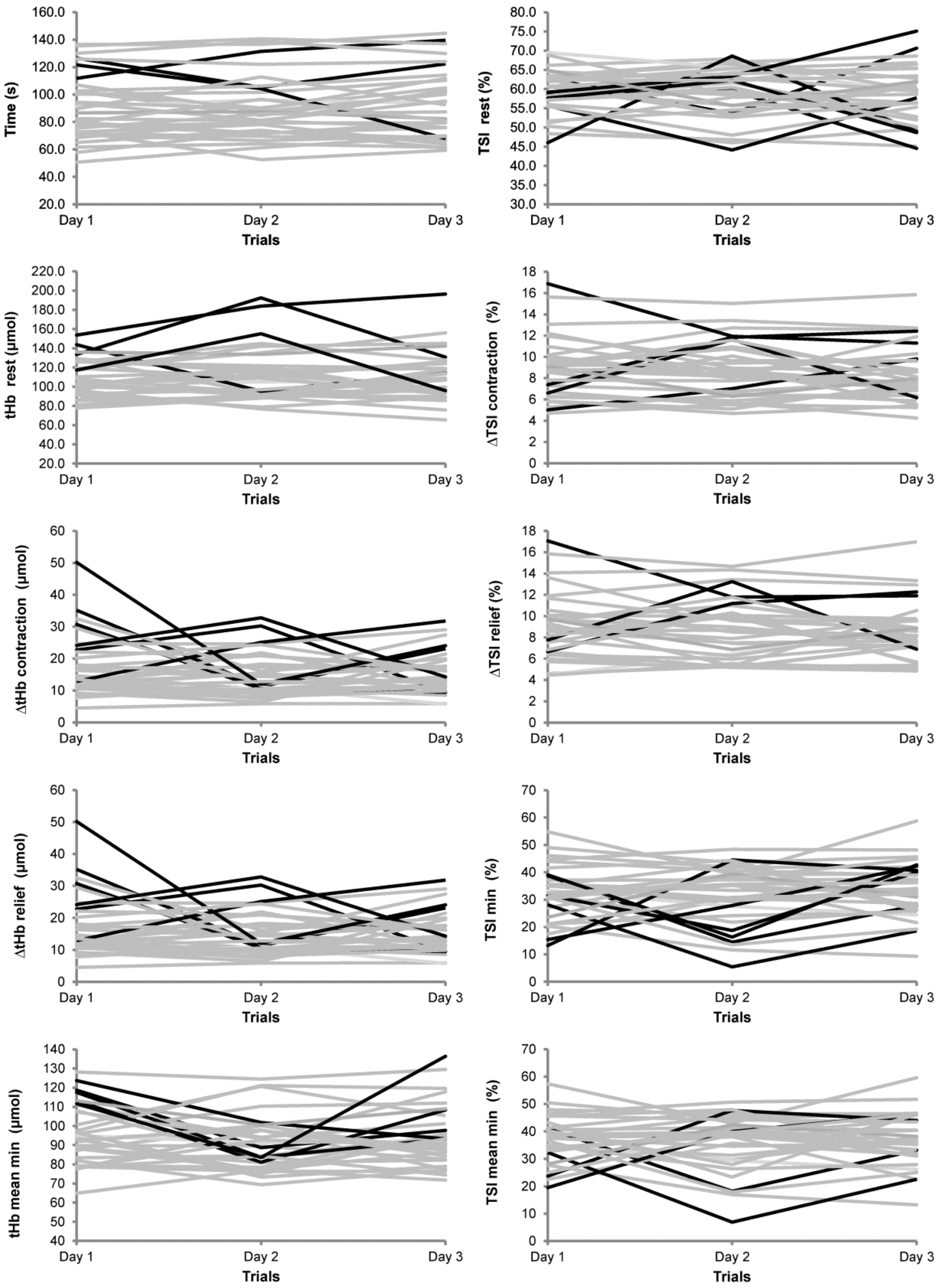

Figure 4. Individual responses in three trials (days) for time to failure, tissue saturation index (TSI) and total haemoglobin (tHb) changes during rest, contraction and relief periods of the intermittent handgrip test. Dark lines represent subjects with the variability between trials exceeding the minimal detectable change.

PRACTICAL APPLICATIONS

Continuous wave NIRS provided a reliable measure of tissue oxygenation changes in rock climbing specific conditions. The most reliable parameters were mean TSI changes during the contraction and relief periods. Mean changes in TSI from all contractions and relief periods were more reliable than those from the first and last three contractions. ΔtHb, tHb_{min} and TSI_{min} provided the lowest reliability and should be interpreted with caution. Consequently, we recommend that differences during contraction and relief periods should exceed ~4.5% for ΔTSI and ~18.5 μmol for ΔtHb to be considered meaningful (based on MDS). Moreover, previous studies which investigated the oxygenation responses in rock climbers may have not been accurate and should be interpreted with caution.

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