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Improvements in maximal oxygen uptake after sprint-interval training coincide with increases in central hemodynamic factors

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Short title: Central hemodynamic adaptations after SIT

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Abstract

Introduction: Sprint-interval training has been shown to improve maximal oxygen uptake, in part through peripheral muscle adaptations that increase oxygen utilization. In contrast, the adaptations of central hemodynamic factors in this context remain unexplored. **Purpose:** The aim of the current study was to explore the effects of sprint-interval training on maximal oxygen uptake and central hemodynamic factors. **Methods:** Healthy men and women (n=29, mean age 27 ± 5, height 175 ± 8 cm, body mass 72.5 ± 12.0 kg) performed 6 weeks of sprint-interval training consisting of 3 weekly sessions of 10-min low-intensity cycling interspersed with 3 x 30-s all-out sprints. Maximal oxygen uptake, total blood volume, and maximal cardiac output were measured before and after the intervention. **Results:** Maximal oxygen uptake increased by 10.3% ($p<0.001$). Simultaneously, plasma volume, blood volume, total hemoglobin mass, and cardiac output increased by 8.1% (276 ± 234 mL; $p<0.001$), 6.8% (382 ± 325 mL; $p<0.001$), 5.7% (42 ± 41 g; $p<0.001$), and 8.5% (1.0 ± 0.9 L · min^{-1}; $p<0.001$), respectively. Increased total hemoglobin mass along with measures of body surface area had significant impact on the improvements in maximal oxygen uptake. **Conclusion:** Six weeks of sprint-interval training results in significant increases in hemoglobin mass, blood volume, and cardiac output. As these changes were associated with marked improvements in maximal oxygen uptake, we conclude that central hemodynamic adaptations contribute to the improvement in maximal oxygen uptake during sprint-interval training.

Keywords: blood volume, cardiac output, HIIT, VO_{2max}, SIT

Introduction

As reported already 70 years ago (1), there is a tight relationship between total blood volume (BV) and maximal oxygen uptake (VO_{2max}) (2–4). Both blood and plasma volume (PV) respond rapidly to training, with PV expansion accounting for almost all changes in BV during the first 2-3 weeks after training onset (5,6). Later, red blood cell volume also increases until it reaches equilibrium with PV, resulting in a restored hematocrit (Htc) equivalent to pre-training levels (4). Proof-of-concept studies have shown that when BV is manipulated by phlebotomy or artificial BV expansion, VO_{2max} is directly affected by altered cardiac diastolic filling, stroke volume (SV), and cardiac output (Q) (2,7–9). Thus, collectively, there is strong evidence that improvements in VO_{2max} from traditional endurance training (TET) are strongly, but not exclusively, mediated by changes in central hemodynamic factors.

In contrast to the well-characterized adaptations contributing to the improvements in VO_{2max} following TET, and despite the fact that both (sub-)maximal high-intensity interval training (HIIT) and supramaximal sprint-interval training (SIT) elicit similar or greater improvements in VO_{2max} compared with TET (10,11), little is known about the mechanisms behind the observed increases in VO_{2max} following interval training. It has been hypothesized that following TET, an acute reduction in PV during exercise stimulates mechanisms that include an increase in albumin synthesis and albumin content, which in turn leads to expanded PV and decreased Htc, i.e., increased BV (12). We have recently reported that SIT leads to a pronounced acute reduction in PV (13), suggesting that a similar mechanism may contribute to the SIT-induced increase

in VO_{2max}. Nevertheless, central hemodynamic factors such as BV and total hemoglobin mass (tHb) have been somewhat overlooked in previous SIT-studies.

In contrast to the sparse literature investigating hemodynamic adaptations to interval training, peripheral processes involved in oxygen utilization, such as mitochondrial biogenesis (14), promotion of skeletal muscle mitochondrial content (15–17), function (18,19), and capillarization (20), have been more thoroughly examined. Consequently, SIT-induced improvements in VO_{2max} have been suggested to be mediated by peripheral adaptations, with little or no contribution from central hemodynamic factors (21–23). While the ability of working skeletal muscle to extract oxygen is undoubtedly important (24–26), we consider it unlikely that improvements in VO_{2max} are solely due to peripheral adaptations, given that the respiratory capacity of skeletal muscle exceeds maximal oxygen delivery (27) and central hemodynamic factors such as BV and Q play a central role in explaining exercise-induced improvements in VO_{2max} after TET (1,4,7,8,28). A more comprehensive investigation of the underlying biological processes responsible for the improvement in VO_{2max} after interval training therefore seems warranted. This would improve our understanding of the key stimuli and mechanisms behind the overall central hemodynamic adaptations and aid in the design of time-efficient training protocols to maximize increases in VO_{2max}.

Accordingly, the aim of the current study was to explore the effects of 6 weeks of low volume SIT on VO_{2max} and canonical central hemodynamic factors. We hypothesized that 6 weeks of SIT would results in a marked improvement in VO_{2max} in tandem with

improved oxygen delivery capacity, through expanded BV, increased tHb, and improved Q_{max}.

Method

Ethical approval

All subjects were informed verbally and in writing about the study before providing written informed consent to participate. The study was approved by the Swedish Ethical Review Authority (ref 2019-0449, approved 2019-09-26).

Subjects

None of the subjects regularly participated in any form of interval training or other structured training programs. During the study intervention, all subjects were asked to refrain from any other training that might affect the outcome of the study. Of the 33 participants recruited, 29 completed the study protocol (13 women and 16 men (baseline VO_{2max} 41.0±7.8 mL · kg^{-1} · min^{-1}, age 27±5 years, height 175.3±8.1 cm, body mass 72.5±12.0 kg, BMI 23.5±2.5 kg · m^{-2}). Three subjects dropped out due to illness (cold or flu-like symptoms) and one subject dropped out due to lack of motivation. Of the 29 subjects included, one subject was unable to perform the VO_{2max} and Q_{max} test after the intervention because of flu-like symptoms, one subject did not perform the Q_{max} test after the intervention because he became ill before the test, and we were unable to place a peripheral venous catheter in one subject at the time of total hemoglobin mass measurement.

Training intervention

The training intervention consisted of 3 training sessions per week for 6 weeks. Each session lasted 10 minutes and consisted of three 30-second all-out sprints on a mechanically braked cycle ergometer (Monark 894E, Varberg, Sweden) against a braking force equivalent to 7.5% of body weight (Figure 1). Sprints were separated by 2 minutes of unloaded cycling, referred to as “rest” in Figure 1. Before the first interval, subjects completed a short warm-up (2.5 minutes of unloaded cycling). The third interval was followed by 2 minutes of unloaded cycling, which served as a cool-down. Subjects were instructed to pedal as fast as possible against the inertial resistance of the cycle ergometer. When maximal cadence was reached, braking force was applied manually. Subjects were strongly encouraged to pedal as fast as possible during each 30-second interval. Power output was measured during each training session. Peak power corresponded to the highest 1-second average during each bout, and average power included the entire 30-second bout.

Pre- and post-intervention measurements

Pre- and post-intervention tests were administered the week before and the week after the intervention. The test battery included: (i) determination of tHb (ii) an incremental cycling test to determine VO_{2max} (iii) an incremental cycling test to determine Q_{max} (Figure 1).

The optimized carbon monoxide rebreathing method was used to determine tHb. Briefly, subjects rested for 15 minutes before a venous blood sample was drawn from the median cubital vein. The sample was immediately analyzed for baseline carboxyhemoglobin (%HbCO), hemoglobin concentration [Hb], and Hct. End-tidal carbon monoxide (CO) was measured at baseline and after the rebreathing procedure

using a CO gas analyzer (Dräger, PAC 700, Lübeck, Germany). Throughout rebreathing, the same gas analyzer was used to check for CO leaks. Subjects breathed a gas mixture of pure CO (0.8 ml x kg^{-1}) and medical oxygen (AGA, Stockholm, Sweden) for 2 minutes before disconnecting from the spirometer (Blood tec, GmbH, Germany). Two blood samples (1 mL) were collected, one before rebreathing and one 7 minutes after administration of CO. The samples were then analyzed for %HbCO (ABL 800, Radiometer A/S, Copenhagen). tHb was calculated as described previously (29). The coefficient of variation for this method in our laboratory is 1.6%, which is in agreement with previous publications (30,31).

To determine VO_{2max} and maximal workload (W_{max}), subjects performed an incremental cycling test to volitional fatigue on an electronically braked ergometer (Lode, Groningen, The Netherlands) with identical protocols before and after the intervention. Using an online gas collection system (Vmax Encore, Illinois, USA), O_2 and CO_2 concentrations were measured continuously and recorded as breath-by-breath values. Subjects began the test at 50 W for 5 minutes as a warm-up. Subsequently, resistance was increased by 1 W every 3 s, corresponding to an increase of 20 W per minute, until subjects reached volitional fatigue. VO_{2max} was considered to be the highest 20-second value achieved during the test. Criteria for the test were either a plateau in VO_2 or RER > 1.15, a heart rate within 10 beats of age-predicted maximum, and volitional exhaustion.

Q_{max} was determined using a rebreathing technique in which Q_{max} is derived from pulmonary uptake of the inert gas nitrous oxide (N_2O). This assumes that pulmonary uptake of blood-soluble gas is proportional to pulmonary blood flow (32). This method

has been shown to be reliable and is widely used (33); however, it is also known to underestimate absolute Q_{max} values (34). During the maximal incremental test, subjects inhaled a gas mixture containing 5% blood-soluble N_2O, 1% insoluble sulfur hexafluoride (SF_6), and 94% O_2. The gas mixture was mixed with ambient air before the start of the rebreathing protocol. When subjects reached maximal workload (as determined by the VO_{2max} assessment), they were switched from ambient air breathing to a closed-circuit system in which they inhaled the gas mixture while photoacoustic gas analyzers continuously quantified closed-circuit gas concentrations (Innovision, Odense, Denmark). Pulmonary N_2O uptake was determined by the decrease in N_2O concentration over three consecutive exhalations after complete mixing between the remaining pulmonary air and the gas in the rebreathing bag, as determined by a stable gas fraction of the blood insoluble gas (SF_6). Subjects were instructed on how to perform rebreathing, and each subject had two trial runs of the rebreathing protocol before the actual measurement.

Estimation of O_2 delivery (QaO_2) was calculated as the product of Q_{max} times arterial O_2 content (CaO_2), which in turn was calculated as the product of hemoglobin concentration, arterial oxygen saturation (SaO_2) and the physiological O_2 binding coefficient of hemoglobin (1.34 ml $\cdot$ g^{-1}). As SaO_2 was not measured during the VO_{2max} tests, we have assumed that there was no change in SaO_2 between pre- and post-intervention.

Statistics

All data are presented as mean and standard deviation unless otherwise stated. Training effects, i.e., comparison of pre- and post-intervention data, were derived using mixed linear models with time as a fixed effect and subject/time as a random effect to

account for the repeated measures design. The Linear and Nonlinear Mixed Effects Models (nlme) library in R version 3.5.5 was used for this purpose. Power output variables from the training sessions were analyzed using a mixed linear model with training session x bout as the fixed effect and subject/session as the random effect. Here, session denotes the training sessions from 1 (first) to 18 (last training session) and bout denotes the sprint intervals 1, 2, and 3 in each session. All dependent variables were treated as independent hypothesis tests and the correction for multiple hypothesis tests was calculated using the Benjamini-Hochberg false discovery rate, where a false discovery rate of 1% was considered significant.

The study was primarily designed to detect changes in VO_{2max} and PV based on the following power and effect-size calculations: To detect a change in VO_{2max} of 300 ml or 8% with an interindividual variance of 10% in response and a power of 0.8 and $p<0.01$, 22 independent observations were sufficient. For plasma volume, with a variance of 18%, 25 subjects were required to detect a change of >7%, and 30 subjects for a change of 9% with a power of 0.8 and $p<0.05$. Based on this, 30 subjects were included.

Correlations between change-scores in the main outcome variable (VO_{2max}), potential moderators (tHb, BV, W_{max}, and Q_{max}), and baseline characteristics (sex, body surface area, and baseline VO_{2max}) were explored using principal component analysis and then modelled with mixed linear models, where the change-score of VO_{2max} served as the dependent variable and was first analyzed with the potential moderators in univariate models and then controlled for baseline characteristics as fixed effects. Principal

component analysis was performed using Factominer and mixed linear models was performed using LME.

Results

At baseline, VO_{2max} was 3.0±0.8 L · min^{-1} and increased by 10.3% to 3.3±0.9 L · min^{-1} ($p<0.001$) following 6 weeks of training (Figure 2). W_{max} was 235.6±45.0 W at baseline and increased by 11.3% (26.6±12.2 W; $p<0.001$). SV and Q_{max} were 65±13 mL and 12.5±2.5 L · min^{-1} at baseline (Figure 2) and increased by 9.0% (5.3±5.1 mL) and 8.5% (1.0±0.9 L · min^{-1}), respectively ($p<0.001$, Figure 2). QaO_2 increased by 5.5% from 2.4±0.7 L · min^{-1} at baseline to 2.5±0.6 L · min^{-1} after the intervention ($p=0.018$). Maximum heart rate (HR_{max}) did not change significantly from baseline values of 193±7 bpm. PV increased by 8.1% (276±234 mL) from 3374±396 mL at baseline ($p<0.001$). BV and tHb were 5512±788 mL and 706±165 g, respectively, at baseline and increased by 6.8% (382±325 mL) and 5.7% (42±41 g; $p<0.001$). [Hb] and Htc decreased by 2.6% ($p<0.001$) and 2.3% ($p=0.002$), respectively (Figure 2). Body mass did not change from baseline (72.5±12.0 vs 73.1±11.0 kg). Mean and standard deviations presented in a table format can be found in supplementary table 1.

At baseline, the average power output was 5.79 · 0.92 W · kg^{-1} during bout 1. In bouts 2 and 3, it decreased to 4.49±0.86 W · kg^{-1} and 3.94±0.78 W · kg^{-1}, representing a decrease of 22% and 31%, respectively. Average power in bout 1 remained stable throughout the training sessions and was 5.82±0.99 W · kg^{-1} in the last training session ($P>0.05$). The average power in bout 2 and 3 increased by 0.013±0.002 W · kg^{-1} · $session^{-1}$ ($p<0.001$) and was 4.79±0.89 and 4.42±0.79 W · kg^{-1} during the last training session, respectively (Figure 3).

Potential modifiers of changes in VO_{2max} were first explored using a principal component analysis (Figure 4), in which the change score of VO_2max was examined along with changes in measures of tHb, BV and PV, Q_{max}, and W_{max}, as well as baseline body surface area and baseline VO_{2max}. This yielded a covariance of ~48% using the first two principal components, with baseline VO_{2max}, change in tHb, and baseline body surface area being the variables with highest covariance with changes in VO_{2max}. Based on the results of the PCA, it was decided to test the potential modifiers of Δ VO_{2max} while controlling for baseline VO_{2max} and sex. A mixed linear model analysis was then performed testing the potential moderators as independent variables with the change in VO_{2max} as the dependent outcome variable. To allow for effect-size comparisons between variables, the independent variables were scaled to Z-scores (Table 1).

Based on a low model contribution and the fact that there was no significant correlation between sex and change in VO_{2max} (p=0.074 univariate and p=0.82 in the model with BSA and baseline VO_{2max}), sex was removed and the final model was based on baseline VO_{2max}, body surface area, and Δ tHb. This model had better performance than a model with sex, baseline VO_{2max}, and Δ tHb (AIC 2.9 and adjusted R^2 0.44 vs. AIC 9.8 and R^2 0.29) and was also superior to a model with sex added in addition to body surface area, baseline VO_{2max}, and Δ tHb (AIC 4.9 R^2 0.44, Table 2).

The effect sizes in the final model were an increase of 2.5±1.1 mL VO_2 per gram increase in tHb, 934±293 mL VO_2 per m^2 increase in body surface area at baseline, and a decrease of -150±73 mL VO_2 per liter VO_{2max} at baseline. The model had an R^2

of 0.44, which corresponds to an explanatory value of 44% of the total variance of ΔVO_{2max}. Adding the tHb change score to the baseline parameters improved model performance from AIC 6.3 and R^2 of 0.33 to AIC 3.9 and R^2 of 0.44 with a likelihood-ratio of 5.4 (p=0.06, Figure 5).

Discussion

The current study investigated whether BV and other central hemodynamic factors were affected by 6 weeks of low-volume SIT in parallel with the expected increase in VO_{2max}. Indeed, robust improvements in VO_{2max} were observed with concomitant increases in SV, Q_{max}, and total BV but the increase in VO_{2max} was larger than the changes in these central hemodynamic components, indicating that peripheral adaptations were also of importance, as has been shown in previous studies (15–17). While tHb also increased, BV expansion was driven by increased PV, which was reflected in a decrease in Htc and [Hb].

The observed increase in VO_{2max} is consistent with previous publications in which comparable training was performed. Similar increases have been demonstrated after 4 weeks of repeated 15-30-second bouts of SIT (35), after 6 weeks of combined HIIT and SIT (36), and after a variety of other SIT protocols (10,37). An interesting comparison of the elicited changes in VO_{2max} in the present study with some of the above reports is that despite the lower total number of bouts of sprints in the current training protocol, the increase in VO_{2max} between interventions was similar. This is consistent with previous studies reporting that 2-3 supramaximal intervals in a session

are sufficient to produce robust improvements in VO_{2max} in previously untrained subjects (37–39).

A key finding of the current study was that low volume SIT resulted in an increase in VO_{2max} with concomitant increases in BV, PV and tHb. This is consistent with previous studies reporting central hemodynamic adaptations associated with increased VO_{2max} following TET (4,28). The results are also corroborated by studies using a higher volume of interval training and showing that vascular volume and Q_{max} increased after 6 and 12 weeks of training in parallel with the increase in VO_{2max} (40,41). This suggests that similar central hemodynamic factors mediate the increase in VO_{2max} after SIT and TET. The observed relationship between central hemodynamic factors and VO_{2max} supports this assumption, especially given the previously verified causal relationship between Q and VO_{2max} (1,2,7).

The observed increase in BV was likely driven by the expanding PV as reflected by the decrease in [Hb] and Htc. This hemodilution reflects the typical kinetics of exercise-induced hypervolemia, in which the PV rapidly expands followed by an increase in erythrocyte volume until equilibrium is reached and Htc is restored (4,40,42,43). The decreased Htc and [Hb] in our data show that this equilibrium was not reached after 6 weeks SIT. The increase in BV, PV, and tHb is in contrast to a previous study that reported no effect on the aforementioned variables following a 2-week HIIT intervention (23). While it is not surprising that 2 weeks of training did not elicit an increase in tHb, it was somewhat unexpected that PV remained unchanged, as this has been demonstrated after a wide range of training protocols with varying intensities and durations (40,42).

The importance of increased BV relates to the resulting increased venous return, SV and Q. Consistent with the findings reported here, studies employing both short (≤4 weeks) (44) and longer (>6 weeks) training interventions have reported pronounced effects on Q and SV (36,37,41). However, in addition to the above study with unchanged PV, another study reported unchanged Q_{max} following cycling-based SIT (22,23). The reason for this discrepancy is unclear, but most likely multifactorial, including differences in methodology, exercise intensity, intervention time, baseline fitness status of subjects and outcome variables assessed. Collectively, however, it appears that interval intensity, rather than total work volume is the key modifier of VO_{2max}. This is supported by previous work reporting that adding training sessions at the expense of exercise intensity may reduce the training effect (45) and that exercise with a relatively high workload for a longer duration has a smaller effect on muscle volume expansion and plasma volume drop (46). In this regard, it should be noted that in the two studies in which no increase in central hemodynamic factors was found, the intensity in each sprint bout was lower than in the present study. Based on our recent publication (13), it appears that training with a high workload for a shorter duration actually has a greater effect on muscle volume expansion and PV drop than a lower workload for a longer duration. It is plausible that this leads to a greater hypovolemic stimulus, which has greater effects on vascular volumes. This is also supported by reports showing that glycogenolysis (and thus metabolite accumulation) is maximally activated during the first 15 seconds of the first exercise bout (47). While more detailed information is needed to explore these plausible mechanisms and how they relate to an increase in albumin synthesis, the present data support the hypothesis of a central hemodynamic adaptation through an increase in BV with SIT.

The contribution of the baseline characteristics of the subjects and the change values of the physiological parameters were analyzed in relation to the improvement of VO_{2max}. Based on the initial exploratory PCA analysis, baseline VO_{2max}, body surface area, and sex were examined as potential moderators of the training response. In the final and best performing model, BSA made a significant contribution with Δ VO_{2max} increasing by 600 ml per standard deviation increase in BSA, or 934 ml VO_2 per m^2 increase in BSA. The model with BSA outperformed the models with sex as a moderator, and a model that included sex in addition to the BSA was not significantly better at predicting Δ VO_{2max}. Based on the inherited mutual correlation of BSA and sex, the present dataset is arguably too small to infer possible independent effects but nevertheless it suggests that BSA is a more important moderator of the training effect of this type of training intervention than sex. This is consistent with earlier reports showing that there is no difference in VO_{2max} gain between men and woman when subjected to interval-training regimes (11). However, it cannot be ruled out that there is residual variance in Δ VO_{2max} related to sex that could have been quantified in a larger cohort than the present study or that scaling exaggerated the BSA to Δ VO_{2max} relationship. Nevertheless, it is tempting to speculate that the mechanism underlying the positive relationship between BSA and Δ VO_{2max} is due to greater metabolic and hemodynamic stress induced by exercise with greater muscle mass.

Baseline VO_{2max} was negatively correlated with Δ VO_{2max}, with Δ VO_{2max} being 150 ml lower for each liter of baseline VO_{2max}. In agreement with previous studies, the influence of baseline VO_{2max} on the model, and therefore the predictability of the exercise response by baseline power, is rather small. It is also possible that there is a

regression to the mean phenomena with repeated measures in this correlation, which cannot be quantified in the present study due to the absence of a non-exercising control group. It must therefore be considered uncertain whether this is a true biological variation in which individuals with lower baseline VO_{2max} are more responsive to this type of training than individuals with higher aerobic capacity. Of the physiological parameters examined as moderators of Δ VO_{2max}, the change in tHb made the strongest contribution. The final model based on baseline BSA, baseline VO_{2max}, and change in tHb had an R^2 of 0.44, meaning it explained 44% of the total variance in Δ VO_{2max}. Bivariate correlations can be found in supplementary table 2.

Initially, studies examining interval training focused on processes involved in peripheral oxygen handling, such as mitochondrial biogenesis (14) and capillarization (20). Because some previous studies reported little or no contribution from central hemodynamic factors (21–23), it was assumed that the increase in VO_{2max} with SIT was mediated mainly by peripheral adaptations. Yet, if SIT did not increase Q_{max}, a higher VO_{2max} can only be explained by improved blood flow coordination or increased oxygen extraction. This appears unlikely given the limitation in oxygen delivery during maximal work and the excess capacity of mitochondrial respiration (27). Moreover, given the already low venous Hb saturation at peak-VO_2 in the untrained state, it is questionable whether peripheral remodeling alone can explain the observed increases in VO_{2max} seen with SIT. Although Q_{max} and tHb are the major determinants of aerobic capacity during exercise using large muscle groups (4,48), the ability to achieve VO_{2max} also depends on efficient peripheral mechanisms for oxygen uptake and utilization (49), as demonstrated by unilateral training models (48). This is also supported by the improvements in sprint performance over the course of the training intervention. No

change in W_{max} was observed, but a successive increase in average power output during each sprint and an improvement in recovery between each sprint was apparent, i.e., the power output of the 2nd and 3rd sprint approached the power output of the first bout over the course of the training intervention (Figure 3). This is likely due to peripheral adaptations, as our previous work suggests that power output during bouts 2 and 3 reflects endurance capacity, i.e., the ability to sustain higher power output for longer periods of time (13). Thus, our findings, together with previously published literature, support the concept of symmorphosis, i.e., that both central and peripheral adaptations are involved in the improvements in VO_{2max} following a SIT intervention.

Because central and peripheral adaptations are regulated by different mechanisms, it is likely that the design of interval training (e.g., intensity vs. duration/volume) will affect these two processes to different degrees, as will the kinetics at which these adaptations occur. In order to fine-tune SIT programs to the desired outcome, future studies should attempt to tease out the independent and combined effects of the various program design factors on these different adaptations. Furthermore, this spurs future research to explore plausible regulatory mechanisms that may explain how contrasting exercise modalities such as TET and SIT may lead to similar improvements in central hemodynamic factors despite the large differences in exercise stimuli. As mentioned earlier, one of the limitations in the present study is the underestimation of Q_{max} in our measurements. The limitations of the Q_{max} measurements are also evident in the PCA (Figure 4), where, unlike tHb, the variables derived from this measurement were not associated with the changes in VO_{2max}, even though the variables are surrogates for oxygen transport. One proposed explanation for the measurement error is the recirculation of nitrous oxide into the pulmonary system during the rebreathing

procedure (36). However, the reproducibility and reliability of the method have shown good results (33,50). Nevertheless, one should be cautious when interpreting the absolute values generated by the method, especially if one is attempting to derive other physiological parameters from the existing data on Q_{max}.

In conclusion, we report that 6 weeks of SIT elicits robust improvements in VO_{2max} and that increases in central hemodynamic factors together with BSA explain a significant proportion of the variance in improvements in VO_{2max}. Thus, in contrast to previous reports suggesting that the training effects on VO_{2max} observed with SIT are mainly explained by peripheral adaptations, our data strongly suggest that adaptations in central hemodynamic factors also play a significant role.

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Conflict of interest

The authors have no conflict of interest to declare.

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Figure and Table legends

Figure 1. Study overview.

Figure 2.Group mean and individual data with directional changes in workload max (W_{max}), maximal oxygen uptake (VO_{2max}), stroke volume (SV), maximal cardiac output (Q_{max}), plasma volume (PV), blood volume (BV), total hemoglobin mass (tHb), hemoglobin concentration [Hb] and hematocrit (Htc). **** p<0.0001, *** p<0.001, ** p<0.01.

Figure 3. Average power output in watts per kilogram of body weight ($W \cdot kg^{-1}$) during each training session, grouped by sprint bouts throughout the 6 weeks of training. Values are presented as mean (O) and standard deviation (whiskers), Significant changes in each bout across the 18 training sessions are denoted. ** p<0.001.

Figure 4. Principal component analysis and biplot of Δ VO_{2max}, change-scores in hemodynamic parameters and baseline characteristics. The size of each individual observation denotes the magnitude of Δ VO_{2max} where a larger dot indicates a more substantial response. 52% of the total variance across all variables was retained using two principal components, indicative of a high covariance across all change-scores. The variable loadings indicate that baseline VO_2, body surface area and sex along with changes in hemoglobin mass and blood volume are the most important moderators of Δ VO_{2max}.

Figure 5. Model performance. A linear model considering body-surface area, baseline VO_{2max} and change-score in total hemoglobin mass achieved a R^2 of 0.44 corresponding to 44% of the total variance in Δ VO_{2max}. The effect sizes in the final model were an increase of 2.5±1.1 mL VO_2 per gram increase in total hemoglobin mass, 934±293 mL VO_2 per m^2 increase in body surface area at baseline, and a decrease of -150±73 mL VO_2 per liter VO_{2max} at baseline. Standardized effect-sizes were an increase of 359±161 mL VO_2 per standard-deviation increase in total hemoglobin mass, 654±206 mL VO_2 per standard-deviation increase in body surface

618 area at baseline, and a decrease of -418±201 mL VO_2 per standard-deviation increase
619 in VO_{2max} at baseline.