Capacidad de difusión pulmonar bajo diferentes modalidades de ejercicio a nivel del mar y en hipoxia hipobárica simulada de 4.000 m

Lung diffusing capacity after different modalities of exercise at sea level and hypobaric simulated altitude of 4,000 m

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Resumen

Introducción: La difusión pulmonar para el monóxido de carbono (DL_{CO}) proporciona una medida de la transferencia de gas en los pulmones, que aumenta con relación al ejercicio y disminuye en presencia de una lesión intersticial pulmonar. El objetivo de este estudio ~~es~~ fue evaluar los cambios en la difusión pulmonar después de un ejercicio aeróbico y anaeróbico en cicloergómetro.

Material y método: Los participantes fueron 9 sujetos físicamente activos, incluyendo seis mujeres (edad: 24.6 ± 3.6 años) y tres hombres (edad: 23.7 ± 1,5 años). La DL_{CO} se estudió bajo dos protocolos diferentes: El primer día, la DL_{CO} fue medida a nivel del mar en reposo (SL-R), después de un esfuerzo máximo de 30 segundos (SL-ANA), y después de un ejercicio moderado continuo de 15-min (SL-AER). El segundo día, la DL_{CO} fue evaluada a nivel del mar en reposo (SL-R, y luego en altitud (4,000 m) en reposo (HA-R) y después de un ejercicio interválico de 30 minutos (HA-AER).

Resultados: Se produjo un aumento de la DL_{CO} de la SL-R a la SL-ANA (32,5 ± 6,4 a 40,3 ± 11,6 $mL \cdot min^{-1} \cdot mmHg^{-1}$, P = 0,027). En el segundo día, la DL_{CO} no se modificó después de la exposición en altitud, ya sea en reposo a 4.000 m (HA-R) o después del ejercicio interválico moderado a dicha intensidad (HA-AER).

Conclusiones: La difusión pulmonar aumentó ampliamente después de un esfuerzo máximo de 30 segundos en cicloergómetro, aunque la dependencia del oxígeno en este tipo de esfuerzos es pequeña. La intensidad del esfuerzo es un modulador determinante en las modificaciones de la difusión pulmonar con relación al ejercicio.

Palabras clave: Capacidad de difusión pulmonar. Ejercicio intermitente en hipoxia. Edema pulmonar de altura. Hipoxia hipobárica. Capacidad de difusión pulmonar para el monóxido de carbono.

Abstract

Introduction: Lung diffusion capacity of carbon monoxide (DL_{CO}) provides a measure of gas transfer in the lungs, which increase in relation to exercise and decrease in the presence of lung interstitial disease. The aim of this study is to evaluate the changes in lung diffusion after anaerobic and aerobic exercise in a cycle ergometer.

Material and method: The participants were 9 healthy active subjects, including 6 females and 3 males (age: 24.3 ± 3.1 years). Lung diffusion capacity for carbon monoxide (DL_{CO}) was studied under two different protocols: In the first day, DL_{CO} was measured at SL at rest (SL-R), after 30-s maximal exercise (SL-ANA), and after 15-min moderate continuous exercise (SL-AER). In the second day, DL_{CO} was evaluated at rest at SL, and then at HA (4,000 m) at rest (HA-R) and after 30-min of moderate interval exercise (HA-AER).

Results: There was an increase in DL_{CO} from rest to after SL-ANA (32.5 ± 6.4 to 40.3 ± 11.6 $mL \cdot min^{-1} \cdot mmHg^{-1}$, P = 0.027). In the second day, DL_{CO} was evaluated at rest at SL, and then at HA (4,000 m) at rest (HA-R) and after 30-min of moderate interval exercise (HA-AER). During HA exposure, there was no changes in DL_{CO}, either at HA-R, or after HA-AER.

Conclusions: Lung diffusion capacity largely increased after 30-s maximal exercise in a cycle ergometer, although the O_2-dependence is small during this type of anaerobic exercise. Thus, exercise intensity may be a key modulator of the changes in lung diffusing capacity in relation to exercise.

Keywords: Diffusing capacity. Intermittent hypoxic exercise. High-altitude pulmonary edema. Hypobaric hypoxia. Lung diffusing capacity for carbon monoxide.

Introduction

The physiological benefits of exercise training have long been studied, including cardiac remodelling, increase in capillary density, and improvement of muscle oxidative capacity among others with continuous and interval exercise training (1-3). However, pulmonary structural and functional capabilities seem to do not significantly change in response to anaerobic nor aerobic training in healthy subjects (4), except in aquatic sports such as swimming (5) or artistic swimming (6).

Different exercise modalities have been largely utilized to improve exercise performance and health. During last years, evidence is amounting regarding the positive effect of exercise, from sport high performance to clinical rehabilitation, both in elite athletes and subjects with chronic pathologies (7,8). However, it remains unknown whether there are acute changes in the structural or functional properties of the lungs in response to anaerobic and aerobic exercise.

Measures of carbon monoxide diffusing capacity of the lungs (DL_{CO}) are widely utilized to evaluate the gas conductance from the alveoli to the blood (9). Acute changes in DL_{CO} have been already described in relation to exercise. Lung diffusion capacity increase with exercise to meet the demand of O_2 by means of an expansion of the pulmonary capillary network due to the increase in cardiac output and pulmonary perfusion pressure at sea level (10, 11). Then, from rest to peak exercise, DL_{CO} may increase up to 150% (12). Consequently, aerobic performance (13), maximal oxygen uptake (VO_2max) (14), and quality of life (15) has been correlated with DL_{CO} values. However, in some cases the permeability of the alveolar-capillary barrier has been impaired after exercise (16), possibly due to pulmonary hypertension and capillary wall stress failure in the lungs (17).

High-altitude exposure also provokes changes in DL_{CO} although there is no consensus about the conditions needed to provoke changes in DL_{CO} in relation to exercise at high-altitude, with some studies describing slightly decrease or increase and other studies finding no changes in DL_{CO} (18-21). Although intermittent hypoxic exercise is becoming popular, to the best of our knowledge it remains unclear how lung function cope with this exercise modality.

In this study, we aimed to evaluate the acute changes in DL_{CO} after different modalities of exercise, at SL and simulated HA under artificial hypobaric conditions. We evaluate DL_{CO} at SL, after a 30-s maximal intensity exercise (SL-ANA) and after moderate intensity continuous exercise (SL-AER). An additional aim is to analyse whether changes in DL_{CO} are correlated to power output (watts) performed in the (SL-ANA). Later, we evaluate DL_{CO} at 4,000 m of HA, at rest (HA-R) and after moderate intensity interval exercise (HA-AER).

Material and method

Participants

The participants were 9 healthy non-smoker subjects, including 6 females and 3 males (age: 24.3 ± 3.1 years, height: 167.9 ± 9.8 cm, body mass: 60.3 ± 8.7 kg) with no history of cardiovascular or respiratory abnormalities. All of them were physically active university students who performed on average 3 sessions of moderate exercise per week. None had asthma, recent upper respiratory tract infections or other respiratory conditions.

Experimental design

The participants performed two DL_{CO} measurements before the start of the study to become familiar with the procedure. A cycle ergometer (in aerobic test: Corival Lode BV, Groningen, Netherlands; in Wingate test: Excalibur Lode BV, Groningen, Netherlands) was used to do the exercise protocols, and a computerized spirometer (Easy One Pro, ndd Medical Technologies, Zurich Switzerland) was used to evaluate DL_{CO} and other pulmonary parameters.

The participants reported to the laboratory on two occasions. Figure 1 shows a schematic representation of the experimental design. The first day, measurements of lung diffusion capacity were performed at rest at sea level (SL), after 30-seconds maximal intensity exercise (Wingate test) (SL-ANA), and after 15-minutes of continuous aerobic exercise at 30% Watts (W) of the maximal W performed in the Wingate test (SL-AER). The second day, lung diffusing capacity was evaluated in relation to exercise during a short-term exposure to hypobaric HA at 4,000 m. The participants performed another basal measurement in resting condition at sea level (SL-R). Then, they reached the target barometric pressure of 462 torr (equivalent to 4,000 m of altitude) in the hypobaric chamber. After at least 30 minutes of reaching target barometric pressure, measurements were performed again in a resting condition (HA-R), and immediately after 30 minutes of moderate interval exercise at the same artificial high-altitude (HA-AER).

---Insert Figure 1--

Due to the inability to sustain 15 minutes of continuous exercise at the intensity proposed at SL-AER, the exercise duration was separated in interval sets during HA-AER. The exercise interval protocol consisted of 5 sets with 3 minutes at moderate intensity (30% W of the maximal W performed in the Wingate test) interspersed with 3 minutes of active recovery (25 W). The computerized spirometer utilized to measure DL_{CO} was placed inside the hypobaric chamber during the HA measurements. Measurements in the HA-AER condition was taken between 1 to 2 h after hypoxic exposure. Exercise at HA was monitored by pulse oximeter oxygen saturation (S_pO_2) and heart rate (HR) to ensure an optimal health status during exercise. To ensure a safe HA exposure in the unacclimated subjects, there was no Wingate test at 4,000 m.

All the measures considered were "grade A" manoeuvres (> 90% of VC_{IN} and VA within 0.2 L or 5% of largest VA from another acceptable manoeuvre) (22). In addition, the haemoglobin (Hb) concentration was determined from a small blood sample obtained by venepuncture to adjust DL_{CO} to individual parameters before the beginning of the study.

Pulmonary function measurements

The procedure used to obtain diffusion lung capacity parameters was the single-breath method, for which a computerized spirometer was attached to a gas mixture cylinder. This method involves measuring the uptake of CO from the lungs over a short breath-holding period. The recommendations made in a recent joint statement by the American Thoracic

Society (ATS) and the European Respiratory Society (ERS) were followed (22). The participants were placed in a seated position, with a mouthpiece and nose-clip in place throughout the test procedure. The test started with tidal breathing for 2–4 breaths until the subject felt comfortable with the mouthpiece. Then the DL_{CO} manoeuvre began with an unforced exhalation to residual volume (RV). At residual volume (RV) the subject's mouthpiece was connected to the source of test gas, and the subject inhaled rapidly to maximal inspiration. After that, the participant was asked to hold their breath for 10 s and then exhale completely without interruption in fewer than 4 s and to continue with a tidal breath to finish the test. The test gases mixture used to calculate pulmonary function and diffusion capacity was composed of 0.3% of carbon monoxide (CO), 11% of a tracer inert gas (He) used to measure VA and the initial alveolar CO, a mixture of 20.9% of oxygen (O_2) and the remainder was nitrogen (N_2). In addition to DL_{CO} and VA, transfer coefficient of the lung for carbon monoxide (K_{CO}), total lung capacity (TLC), vital capacity inspired (VC_{IN}) and residual volume (RV) were calculated.

Ethics approval and consent to participate

The study was developed in accordance with the Helsinki Declaration concerning the ethical principles of human experimentation and approved by the Institutional Ethical Committee from the University of Barcelona (Institutional Review Board number IRB00003099), in accordance with current Spanish legislation. The participants were informed and familiarized with all the experimental procedures, as well as the risks and benefits of the study. They signed an informed consent form and were free to withdraw from the experimental protocol at any time.

Statistical analysis

Data are reported as mean values ± standard deviation (SD). Differences in pulmonary functional and structural parameters between conditions were analysed using a one-way repeated measures analysis of variance (ANOVA) respectively, and in case of detecting statistical effects ($p<0.05$), Bonferroni corrections were performed. Effect sizes as partial eta squared (η^2_p) values were employed to present the magnitude of differences and statistical power (sp) was also described. The analyses were performed using the SPSS v. 20 (IBM SPSS Statistics, Armonk, New York, USA).

Results

Table 1 shows the response in pulmonary functional and structural parameters to SL conditions. There was a significant interaction between changes in DL_{CO} and exercise conditions at SL (F = 7.82, P = 0.004, η^2_p = 0.49, sp = 0.905; Figure 2), including an increase in DL_{CO} from SL-R to SL-ANA (32.5 ± 6.4 to 40.3 ± 11.6 $mL \cdot min^{-1} \cdot mmHg^{-1}$, P = 0.027). However, there was no differences from SL-R to SL-AER (P = 0.873) or from SL-ANA to SL-AER (P = 0.058). In the case of K_{CO}, there was also a significant interaction between conditions at sea level and K_{CO} (F = 8.32, P = 0.003, η^2_p = 0.51, sp = 0.992), presenting a significant increase from SL-R to SL-ANA (P = 0.003).

---Insert Table 1---

--Insert Figure 2--

Regarding lung volumes, there were no significant differences in structural parameters along the SL conditions such as VA (P = 0.115), TLC (P = 0.115) or RV (P = 0.095).

Figure 3 shows the correlation between DL_{CO} at SL-R and average Watts (W) performed in the Wingate test (R = 0.95), in which the studied sample developed an average of 523 ± 166 W and 8.56 ± 1.65 W/Kg in the 30-s of exercise. It is also showed the correlation between the changes in DL_{CO} (Δ DL_{CO}) from basal to after SL-ANA and the Watts performed at the Wingate test (R = 0.63).

--Insert Figure 3--

Table 2 shows the response in pulmonary functional and structural parameters to HA conditions. At the hypobaric chamber, there were no differences between SL-R, HA-R, and HA-AER in any of the main pulmonary parameters evaluated such as $DL_{CO}adj$ (DL_{CO} adjusted to barometric pressure) (Figure 4), K_{CO} and VA.

--Insert Table 2--

--Insert Figure 4--

Discussion

The main finding of this study was the high increase in DL_{CO} (+24%) after 30-s maximal intensity exercise (Wingate test) in a cycle ergometer when compared to rest. However, after 15-min of moderate intensity exercise, DL_{CO} returned to resting levels, suggesting that exercise intensity may be a key modulator of pulmonary function in healthy subjects.

During HA exposure, there were no changes in any pulmonary parameter during the exposure to 4,000 m in the hypobaric chamber (HA-R and HA-AER), suggesting that pulmonary system of healthy subjects cope well with a short-term conditional exposure to exercise and high altitude.

Changes in DL_{CO} in relation to exercise at SL

The Wingate test is considered the most common test to evaluate anaerobic (sprint) cycling performance. In our study, lung diffusing capacity (DL_{CO} and K_{CO}) increased more in this short and explosive exercise compared to 15-min of moderate intensity continuous exercise. To the best of our knowledge, this is the first study that evaluates acute changes in DL_{CO} after anaerobic exercise, although some studies have investigated the relationship between aerobic performance and DL_{CO} both in the short-term and long-term. Lalande et al. (23) showed that individuals with higher maximal aerobic capacity have a more distensible pulmonary circulation. The expansion of the pulmonary vasculature appeared not to reach a plateau during maximal aerobic exercise (14). Interestingly, the changes in DL_{CO} found by Lalande et al. (23) were similar to our results, with an increase of 27 and 24% respectively. We probably did not find a similar DL_{CO} response after 15-min of moderate intensity exercise due to the lower intensity applied compared to the Wingate test and the maximal aerobic exercise utilized by Lalande et al. (23). Therefore, exercise intensity seems to be an important factor to provoke a short-term increase in lung diffusion, ahead of oxygen requirements or exercise duration, and probably due to the increased requirements for CO_2 elimination.

During exercise, alveolar-capillary diffusion increases in proportion to the increase in metabolic rate, but there is no causal response between metabolic rate and hyperpnea, and the mechanisms involved in the increase in ventilation during exercise has not been fully elucidated (24). Volitional exercise requires activation of the central nervous system (CNS), in which neural feed-forward (central command) mediate the exercise hyperpnea (24). The rapid increase in DL_{CO} from our study probably take part of the same physiological mechanism. The entire-organism tried to adjust the cardiovascular and ventilatory systems to maximal intensity exercise (25), despite 30-s anaerobic exercise barely relying on O_2-dependent energy production. This rapid response also makes sense since lung diffusion in the first limiting step of aerobic performance along the O_2 transport cascade and the increase in cardiac output has been shown to be faster than VO_2 kinetics (26).

Correlation DL_{CO} - Wingate

Anaerobic performance measured in Watts correlated closely with DL_{CO} at SL-R (R = 0.95; Figure 2), suggesting that central command-mediated intensity rather than O_2-dependent metabolism is the key in DL_{CO} changes. Figure 2 also shows how changes in DL_{CO} (Δ DL_{CO}) respond to Wingate test anaerobic power (R = 0.63). In this regard, DL_{CO} does not only correlates with VO_2max and aerobic performance (27), but also correlates with neuromuscular anaerobic power. Muscular strength has been already correlated with lung function in some studies (28) which may explain the close relationship between DL_{CO} and neuromuscular power.

Our results also suggest that lung volume (VA and TLC) tend to increase, but this change is not statistically significant, after maximal intensity exercise (SL-ANA). Changes in lung volumes also has been suggested to participate in DL_{CO} changes during exercise periods (28), but at the best of our knowledge there have not been described elsewhere. Potentially, we suggest that interval maximal work could induce sufficient mechanical and/or physiological stimulus to promote a long-term improvement in lung diffusion capacity (e.g., alveolar growth, increased permeability of the alveolar–capillary membrane) or lung growth (4).

Changes in DL_{CO} in relation to exercise at HA

In this study, there were no changes in lung diffusion upon arrival to 4,000 m at rest nor after exercise in a short-term HA exposure of 60 minutes, although some relevant risk factors to the development of pulmonary oedema were also induced in our experimental design such as rapid ascent rate, high-altitude and intervals of strenuous exercise. However, our data supports the idea that short-term exposures to HA seems to be in-sufficient to provoke capillary wall stress failure in the lungs (29).

During HA exposure, in some cases, an exacerbation in the permeability properties of the lung capillary endothelium can create an imbalance between pulmonary vascular leakage and alveolar fluid reabsorption (30, 31), although a large inter-individual response has been described (32, 33). We suggest that the activity of the pulmonary lymphatics regulated the rate of fluid clearance from the interstitial space well under short-term severe hypoxic exposure in healthy subjects, avoiding significant changes in lung diffusing capacity. The appearance of pulmonary oedema or reduced transit time under specific conditions of low PO_2 and high blood flow due to exercise may provoke diffusion unbalance (34, 35), although in some cases an additional functional reserve can be recruited to improve membrane O_2 diffusing capacity during exercise in hypoxia (33, 36).

The literature is unclear regarding the conditions needed to provoke changes in lung diffusing capacity. Senn et al. (37) found a slight decrease in DL_{CO} after a rapid ascent (3 h) to 4,559 m compared to baseline at 490 m. Agostoni et al. (38) also found a slight decrease in DL_{CO}, and an increase in ultrasound lung comets (ULCs) at 4,559 m after 36 h, suggesting that interstitial lung oedema can occur relatively rapid in healthy lowlanders. However, Snyder et al. (39) found that exercise in hypoxia increased DL_{CO} and reduced lung fluid accumulation due to acceleration in alveolar fluid clearance in a 17-h exposure to normobaric hypoxia (FIO_2 = 12.5%). Prolonged exposure to HA could be necessary to elicit changes in lung diffusion capacity, although the evidence is also unclear. In this regard, Clarenbach et al. (32) found 8 cases of HAPO in a group of 18 mountaineers, but DL_{CO} was only decreased after 3 days of exposure to 4,559 m. In turn, de Bisschop et al. (40) showed a post-exercise decrease in lung diffusing capacity for nitric oxide (DL_{NO}), but no changes in DL_{CO} after 7 days at 5,050 m. Nonetheless, Taylor et al. (41) found a significant increase in DL_{CO} after an 8-day hike and 5-day stay at 5,150 m in mountaineers. At the best of our knowledge, this is the first study assessing DL_{CO} changes during short-term altitude exposure with exercise. As it can been assumed after the results at sea level, exercise intensity seemed a relevant factor to induce DL_{CO} modifications. Therefore, the moderate intensity interval exercise proposed at high-altitude could have influenced the lack of DL_{CO} modifications during hypobaric hypoxic exercise. From a security point of view, the participants of this study were healthy subjects, but unaccustomed to strenuous exercise at high altitude neither highly trained athletes. As a result, a limitation in the exercise intensity performed at 4,000 m was not possible to elude.

We suggest that there was no decrease in DL_{CO} due to a pulmonary interstitial fluid fine balance between pulmonary capillary fluid leakage and the rate of fluid removal from the thoracic lymphatic ducts during short-term exposure to HA (12, 42). Also, the induced increase in interstitial lung fluid could be masked by an additional recruitment of the pulmonary vasculature during hypoxic exercise due to limitations in O_2 uptake in the lungs under low barometric pressure conditions (33).

Strengths and limitations

The duration and intensity of the exercise may be decisive to find an increase, no changes, or a decrease in DL_{CO}. Dynamics of lung diffusing equilibrium may change depending on these factors, and inter-individual physiological status.

Another concern is the use of indirect measurements of interstitial lung fluid. Although DL_{CO} has been consistently associated with an increase in extravascular lung water (32, 39), the study of DL_{NO} is more sensitive to detect very mild interstitial fluid accumulation (43). A combination of DL_{CO} and DL_{NO} would be more descriptive of changes in lung diffusion since DL_{NO} is minimally affected by haemoglobin and pulmonary blood volume (V_C).-One relevant strength from this study is that all the DL_{CO} measurements were taken into the first minute after exercise. Most of the studies have assessed DL_{CO} 30 to 120 min after exercise suggesting that the potential decrease in DL_{CO} is due to blood volume redistribution to the peripheral organs after exercise, a hypothesis that may be dismissed in our study.

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Conflicts of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Figure footnotes

Figure 1. Scheme of the study's experimental design.

Figure 2. Changes in DL_{CO} from sea level at rest (SL-R), to after 30-s maximal exercise (SL-ANA), to after 15-min moderate continuous exercise (SL-AER). *Significant differences between conditions ($P < 0.05$).

Figure 3. (a) Correlation between DL_{CO} at sea level at rest (SL-R) and the average watts (W) performed after 30-s maximal exercise (SL-ANA), and (b) correlation between the changes in DL_{CO} (Δ DL_{CO}) from SL-R to SL-ANA and the W performed at SL-ANA.

Figure 4. Changes in DL_{CO} from sea level at rest (SL-R), to simulate altitude at rest (HA-R), to after 30-min moderate interval exercise (HA-AER).

Table 1. Pulmonary parameters response to the different conditions studied at sea level (SL): Basal (SL-R), after 30-seconds maximal intensity exercise (SL-ANA), and after moderate intensity continuous exercise (SL-AER).

	SL-R	SL-ANA	SL-AER
DL_{CO} ($mL \cdot min^{-1} \cdot mmHg^{-1}$)	32.5 ± 6.4	40.3 ± 11.6 [a]	34.7 ± 9.3
DL_{CO} (%-predicted)	126 ± 11	154 ± 13	134 ± 13
K_{CO} ($mL \cdot min^{-1} \cdot mmHg^{-1} \cdot L^{-1}$)	6.02 ± 0.48	6.70 ± 0.64 [a]	6.26 ± 0.71
K_{CO} (%-predicted)	124 ± 10	138 ± 10	129 ± 11
VA (L)	5.39 ± 0.94	5.97 ± 1.33	5.58 ± 1.29
VA (%-predicted)	101 ± 8	111 ± 9	104 ± 11
TLC (L)	5.54 ± 0.94	6.13 ± 1.33	5.73 ± 1.29
TLC (%-predicted)	101 ± 8	111 ± 9	104 ± 11
VC_{IN} (L)	4.01 ± 0.92	3.89 ± 0.89	3.84 ± 0.84
RV (L)	1.54 ± 0.50	2.23 ± 0.66 [a]	1.91 ± 0.79

[a] Significantly higher than SL-R ($p<0.05$)

Table 2. Pulmonary parameters response to the different environmental and exercise conditions studied at 4,000 m high-altitude (HA): Sea level at rest (SL-R), simulated high-altitude at rest (HA-R) and simulated high-altitude immediately at the end of exercise (HA-AER).

	SL-R	HA-R	HA-AER
$DL_{CO}adj$ ($mL \cdot min^{-1} \cdot mmHg^{-1}$)	32.1 ± 6.7	31.4 ± 8.2	32.9 ± 9.1
$DL_{CO}adj$ (%-predicted)	121 ± 13	118 ± 14	125 ± 17
$K_{CO}adj$ ($mL \cdot min^{-1} \cdot mmHg^{-1} \cdot L^{-1}$)	5.93 ± 0.48	5.73 ± 0.85	6.09 ± 0.70
$K_{CO}adj$ (%-predicted)	122 ± 10	118 ± 16	128 ± 21
VA (L)	5.39 ± 0.95	5.51 ± 1.28	5.42 ± 1.43
VA (%-predicted)	99 ± 9	101 ± 15	98 ± 12
TLC (L)	5.54 ± 0.95	5.66 ± 1.28	5.57 ± 1.43
TLC (%-predicted)	99 ± 9	101 ± 15	99 ± 12
VC_{IN} (L)	4.06 ± 0.88	3.83 ± 0.88	3.99 ± 0.94
RV (L)	1.48 ± 0.51	1.83 ± 0.51	1.59 ± 0.66

Figure 1

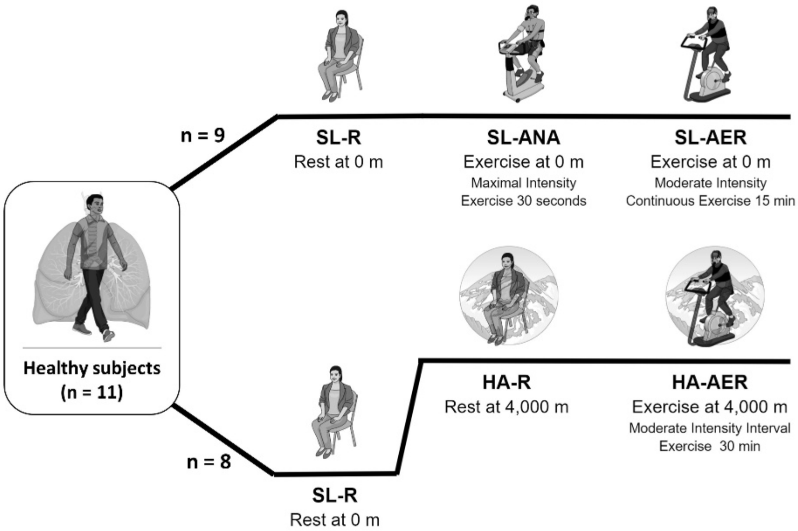
n = 9
SL-R
Rest at 0 m
SL-ANA
Exercise at 0 m
Maximal Intensity
Exercise 30 seconds
SL-AER
Exercise at 0 m
Moderate Intensity
Continuous Exercise 15 min
Healthy subjects
(n = 11)
HA-R
Rest at 4,000 m
HA-AER
Exercise at 4,000 m
Moderate Intensity Interval
Exercise 30 min
n = 8
SL-R
Rest at 0 m

Figure 2

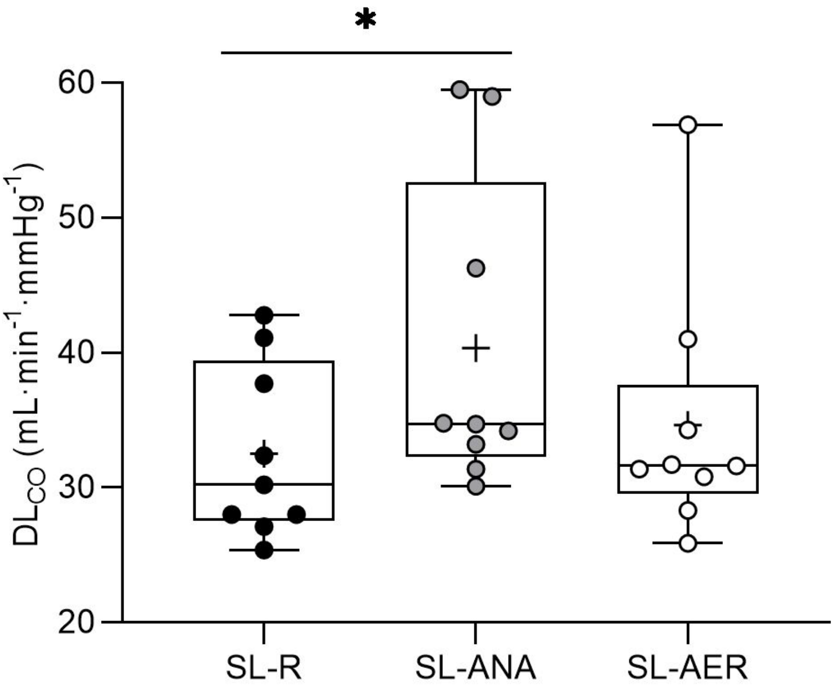

Figure 3

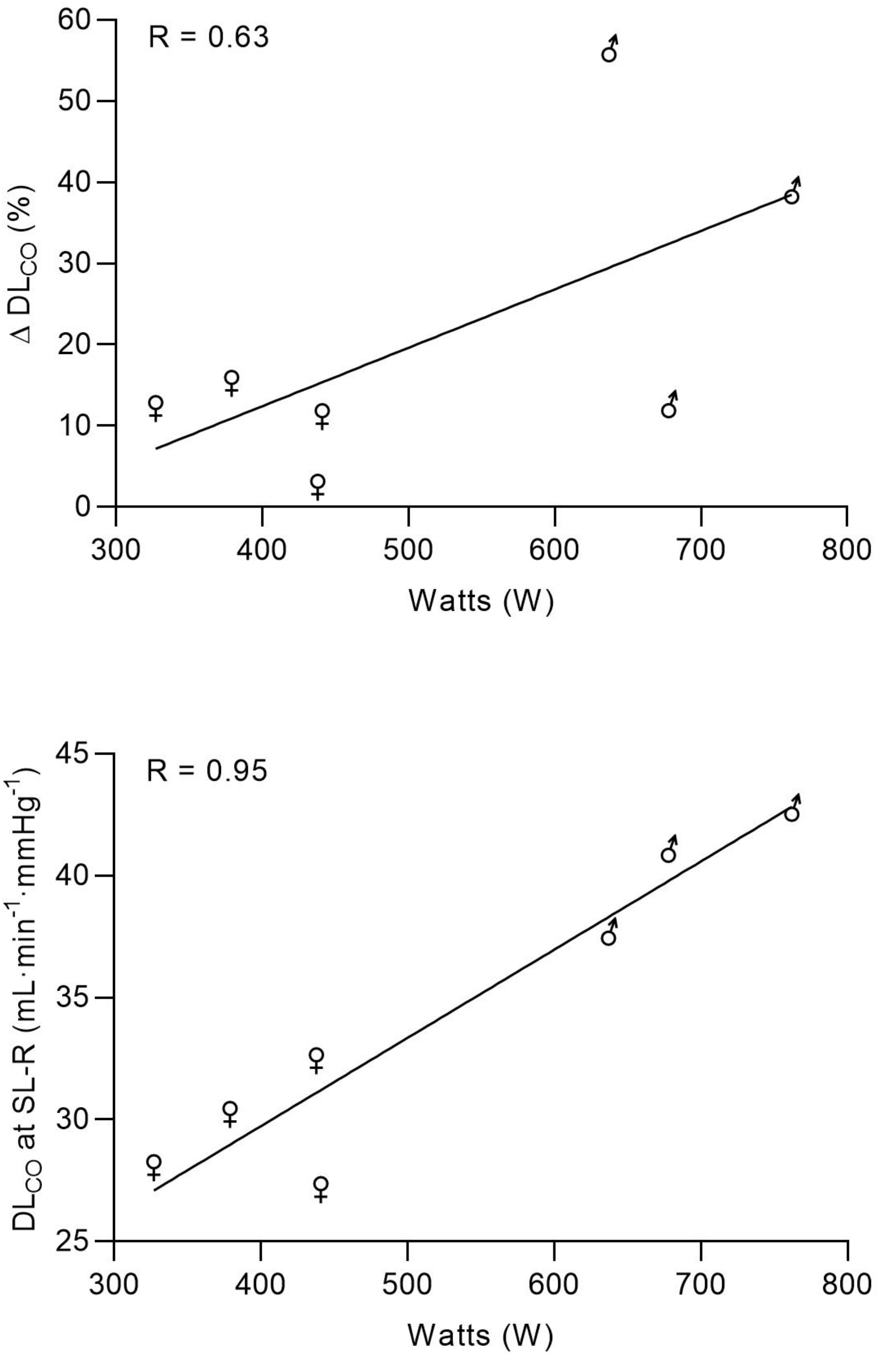
R = 0.63
Δ DL_CO (%)
60
50
40
30
20
10
0
300
400
500
600
700
800
Watts (W)
R = 0.95
DL_CO at SL-R (mL·min-1·mmHg-1)
45
40
35
30
25
300
400
500
600
700
800
Watts (W)

Figure 4