



Double gas transfer factors (DLCO-DLNO) at rest in patients with congenital heart diseases correlates with their ventilatory response during maximal exercise

Gregoire de La Villeon, Arthur Gavotto, Nam Ledong, Charlene Bredy, Sophie Guillaumont, Jonathan Man, Fares Gouzi, Maurice Hayot, Thibault Mura, Pascal Amedro, et al.

► To cite this version:

Gregoire de La Villeon, Arthur Gavotto, Nam Ledong, Charlene Bredy, Sophie Guillaumont, et al.. Double gas transfer factors (DLCO-DLNO) at rest in patients with congenital heart diseases correlates with their ventilatory response during maximal exercise. International Journal of Cardiology Congenital Heart Disease, 2022, 8, pp.100346. 10.1016/j.ijcchd.2022.100346 . hal-03586902

HAL Id: hal-03586902

<https://hal.science/hal-03586902>

Submitted on 29 Mar 2022

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Double gas transfer factors (DLCO-DLNO) at rest in patients with congenital heart diseases correlates with their ventilatory response during maximal exercise[☆]

Gregoire De La Villeon^{a,d}, Arthur Gavotto^{a,b,*}, Nam Ledong^e, Charlene Bredy^a, Sophie Guillaumont^{a,d}, Jonathan Man^c, Fares Gouzi^{b,c}, Maurice Hayot^{b,c}, Thibault Mura^f, Pascal Amedro^{g,h,1}, Stefan Matecki^{b,c,1}

^a Paediatric and Congenital Department, M3C Regional Reference Centre, Montpellier University Hospital, Montpellier, France

^b PhyMedExp, CNRS, INSERM, University of Montpellier, Montpellier, France

^c Department of Physiology, Montpellier University Hospital, Montpellier, France

^d Paediatric Cardiology and Rehabilitation Unit, Institut-Saint-Pierre, Palavas-Les-Flots, France

^e Private Consultant, Sorinnes, Belgium

^f Department of Epidemiology and Biostatistics, Nîmes University Hospital, Nîmes, France

^g Paediatric and Congenital Cardiology Department, M3C National Reference Centre, Bordeaux University Hospital, Bordeaux, France

^h IHU Liryc, Electrophysiology and Heart Modelling Institute, Bordeaux University Foundation, Pessac, France

ARTICLE INFO

Keywords:

Congenital heart disease
Exercise capacity
Combined measurement of lung diffusion
Pulmonary circulation
Single ventricle
Pulmonary regurgitation

ABSTRACT

Aim: Exercise capacity is altered in congenital heart diseases (CHD) with potentially impaired pulmonary perfusion adaptation during exercise, such as in single ventricle or in significant pulmonary regurgitation. This study aimed to evaluate the value of double gas transfer factor analysis, at rest in conjunction with postural manoeuvres, to explore the various components of pulmonary gas transfer and its association with exercise capacity.

Methods: A total of 40 subjects (24 CHD, 16 controls) underwent a combined measurement of lung diffusing capacity for carbon monoxide and nitric oxide (DLCO-DLNO) to determine pulmonary membrane diffusion (Dm) and Vcap, in sitting then supine position. CHD patients performed a maximal cardiopulmonary exercise test.

Results: Compared to normal controls, the CHD group's DLNO, DLCO, Vcap, and alveolar volume (AV) at rest, in the sitting position were depressed, whereas the DLCO/AV and DLNO/AV were similar. The magnitude of Dm and Vcap adaptation induced by postural change was similar in both groups, indicating a preserved pulmonary capillary recruitment capacity in CHD. In the CHD group, at rest, for each ml of postural-induced increase in Vcap we observed during exercise a VE/VCO₂ slope decrease of 0.46 (95% CI[0.83; 0.098]), indicating a better ventilatory response to exercise.

Conclusion: CHD patients with impaired pulmonary circulation have a reduced Dm and Vcap mainly due to decreased pulmonary volume but maintain a normal capacity to adapt these parameters through a simple recruitment manoeuvre. Vcap adaptation evaluated at rest predicts the level of ventilatory efficiency during exercise, which represents a main limiting factor in these CHD patients.

Abbreviations: CPET, Cardiopulmonary exercise test; CHD, Congenital Heart disease; DLCO, lung diffusion for carbon monoxide; DLNO, lung diffusion for nitric oxide.

[☆] The authors take responsibility for all aspects of the reliability and freedom from bias of the data presented and their discussed interpretation.

* Corresponding author. Paediatric and Congenital Department, M3C Regional Reference Centre, Montpellier University Hospital, 371 Avenue du Doyen Giraud, 34295, Montpellier, France.

E-mail address: a-gavotto@chu-montpellier.fr (A. Gavotto).

¹ Both authors contributed equally to this study.

1. Introduction

Cardiopulmonary exercise test (CPET), through the highest oxygen uptake observed ($VO_{2\text{peak}}$) during a CPET, provides a global evaluation of oxygen transport function by the ventilatory and cardiovascular systems, as well as muscular oxygen consumption during exercise [1].

In patients with cardiac diseases, CPET has become the reference functional test to evaluate exercise capacity, which correlates with disease severity, prognosis and quality of life [2–4]. Therefore, CPET is now recommended in the follow-up of patients with heart failure, as $VO_{2\text{peak}}$ reflects their ability to engage in physical activity, as well as their quality of life [5]. CPET has also found an interest in patients with congenital heart diseases (CHD) [6,7]. Indeed, from a large cohort of young patients with CHD, we recently observed a correlation between the quality of life and $VO_{2\text{peak}}$, which were significantly decreased in a time dependent manner, compared to healthy control subjects [2,3]. The existence of an impaired exercise capacity in patients with CHD usually involves multifactorial mechanisms: hemodynamic changes, electrical abnormalities, desaturation, and muscular deconditioning [8]. Moreover, exercise capacity is more altered in CHD with an impaired pulmonary perfusion adaptation during exercise, such as in single ventricles or in CHD with significant pulmonary regurgitation [3,8]. Indeed, the inability to increase the blood flow into the pulmonary circulation during exercise may blunt gas exchange adaptation, imbalance ventilation-perfusion ratio and, ultimately increase ventilatory response during exercise, which may be an important contributing factor to exercise intolerance [9,10].

The measure of various components of pulmonary gas transfer, may be of interest in CHD concerned with impaired pulmonary perfusion, reduced blood volume in the ventilated alveoli and presence of restrictive ventilatory defect [11,12].

In these patients, an impaired pulmonary gas exchange adaptation, such as a blunted V_{cap} increase during exercise, may contribute to $VO_{2\text{peak}}$ limitation. Standard CPET cannot unambiguously identify this phenomenon. Therefore, we hypothesized that measuring the various components of a CHD patient's of pulmonary gas transfer adaptation, using a simple functional test at rest, could help identify a pulmonary perfusion defect contributing to the patient's physical activity limitations.

The two main subcomponent of lung diffusion capacity, e.g. alveolar-capillary membrane conductance (D_m) and pulmonary capillaries volume (V_{cap}) [13], are assessed by the combined measurement of lung diffusion for carbon monoxide (DLCO) and nitric oxide (DLNO) [14]. To our knowledge, their evaluation in CHD with impaired pulmonary perfusion has not yet been reported.

The primary aim of this study was to evaluate, in comparison with healthy controls, how CHD associated with impaired pulmonary blood flow affect the various components of pulmonary gas transfer, by using double gas transfer factors (DLCO-DLNO) technic analysis at rest in conjunction with postural manoeuvres.

The secondary aim was to determine, in CHD with impaired pulmonary perfusion, how the abnormalities of pulmonary gas exchange and their response to postural changes evaluated at rest relate to CPET-derived parameters of exercise function.

2. Methods

2.1. Study design and patients

This prospective cross-sectional study was carried out from December 2014 to December 2016 in a tertiary care paediatric and adult congenital cardiology reference centre (Montpellier University Hospital, France).

Adolescent and adult CHD patients with a single ventricle or a significant pulmonary regurgitation, as defined by the international classification [15], were screened. They were recruited during their annual

follow-up, which consisted in a physical examination, an electrocardiogram (ECG), an echocardiography, and a CPET [16]. Patients with any lung disease associated to their CHD and those with an absolute contraindication for CPET procedure were not eligible. The following clinical data were collected: gender, age, weight, height, body mass index (BMI), medical treatments. The following echocardiography data were collected: systemic ventricle ejection fraction, systemic ventricle global 2D strain, right ventricle systolic pressure, subaortic velocity time integral (VTI) and tricuspid annulus plane systolic excursion (TAPSE).

In the CHD group, patients and their legal guardians for minors were offered to participate in the study, which consisted in a combined measurement of double gas transfer factor measurement (DLCO-DLNO), performed at rest, before CPET. All functional test procedures were harmonized before the study started, as in our previous similar studies [3,12].

In the control group, a call for voluntary application was performed to recruit healthy subjects, who were offered to undergo a similar DLCO-DLNO measurement procedure at rest. These subjects were classified in the control group after a completely normal check-up, including physical examination, electrocardiogram, echocardiography, and spirometry. Subjects with any chronic disease, medical condition (cardiac, neurologic, respiratory, muscular, or renal), or medical treatment, were not eligible.

2.2. Double gas transfer factor (DLCO-DLNO) measurement

For all subject, spirometry and plethysmography were performed with measurement of forced expiratory volume in 1 s (FEV_1), forced vital capacity (FVC), FEV_1/FVC ratio ($FEV_1/FVC\%$), functional residual capacity (FRC) and total lung capacity (TLC) according to ATS/ERS recommendations [17]. Results were expressed as percent predict values and Z-scores from the spirometry reference equations.

Then, using Hyp'Air system (Medisoftware-MGCD, Sorinnes, Belgium) each subject underwent a single-breath method to measure the diffusing capacity of the lung for carbon monoxide (DLCO) and the diffusing capacity of the lung for nitric oxide (DLNO), accordingly to the recent 2017 ERS standards [18,19].

Variability of DLCO-DLNO measurement at rest was evaluated by performing two tests within 4–10 min of each other for 21 subjects (e.g. 16 controls and 5 CHD patients), and found an acceptable variability with difference value between the two tests of less than 3 and 17 $\text{mL min}^{-1} \text{mmHg}^{-1}$ respectively [19].

A similar time interval was imposed between DLCO-DLNO measurement in the seated and supine positions to ensure that all CO backpressure had dissipated between the tests.

The analysers and flowmeter were calibrated and checked before each experiment. Blood sample was drawn for haemoglobin (Hb) concentration for carbon monoxide (CO) specific conductance. The subjects inspired, from residual volume to total lung capacity, a bolus of a test gas mixture containing 18% oxygen (O_2), 13% helium (He), 0.28% CO and 50 ppm of nitric oxide (NO) (balanced in nitrogen, N_2). Then, they held the breath for a 6-s period at normal intrapulmonary pressure, and exhaled to residual volume within 4 s. The initial volume (900 mL) of exhaled air was discarded, then 900 mL of alveolar gas volume was sampled into a bag for analysis. Considering the absence of respiratory pathology in the study subjects and the constant level of inspired oxygen at barometric pressure, the alveolar oxygen partial pressure (PaO_2) was assumed to be 100 mmHg to derive the optimal value of specific Hb uptake rate for CO (θ_{CO}), using Hervé Guénard's equation [18]: $\frac{1}{\theta_{CO}} =$

$$(0.0062 PaO_2 + 1.16) \left(\frac{14.6}{\text{measHb}} \right)$$

A finite θ_{NO} value of 4.5 mL NO ml/blood/min/mmHg was used, according to in vitro value of NO uptake by red cell [20]. Alveolar-capillary membrane conductance for CO and NO (D_mCO , D_mNO) and capillary blood volume (V_{cap}) were then estimated from

Table 1

Clinical and pulmonary function characteristics (CHD population and healthy controls).

Variables	CHD patients (N = 24)	Healthy controls (N = 16)
Age	24.4 ± 9.1	28.7 ± 3.3
Gender (Male/Female)	16/8	11/5
Weight (kg)	63.3 ± 12.7	64.1 ± 11.6
BMI	22.2 ± 4.5	21.7 ± 2.0
FVC (Z-score)	-2.11 ± 1.3	0.5 ± 1.1*
FEV1 (Z-score)	-2.38 ± 1.5	0.6 ± 1.4*
FEV1/FVC (Z-score)	-0.67 ± 1.6	0.1 ± 1.3
FRC (% predict)	96.2 ± 22	102.5 ± 21
TLC (% predict)	88.4 ± 11	104 ± 15

Legend: Variables were described as means ± SD (standard deviation); *P < 0.05, indicates a significant difference between the two groups (Mann-Whitney test).

Abbreviations: BMI, body mass index; FVC, forced vital capacity; FEV1, forced expiratory volume in 1 min; FEV1/FVC, Tiffeneau ratio; FRC, functional residual capacity; TLC, total lung capacity; % predict, percentage of predicted lung function values.

DLNO, DLCO, 0NO and 0CO, following the ERS standards [18]. Each manoeuvre for DLCO-DLNO measurement was performed, first with the subject being in sitting position, and then 1 min after positioning the subject in supine posture.

As patients with CHD may be affected by erythrocytosis, the actual Hb level was used to calculate DLCO parameters. In the control group, we used Hb standard values of 13.4 g/dL in female subjects and 14.6 g/dL in male subjects.

2.3. CPET protocol

Just after double gas transfer factor (DLCO-DLNO) measurement, subjects performed a CPET using a cycle ergometer protocol adapted to CHD to obtain a homogeneous incremental overall duration between 8 and 12 min: a 1-min rest; a 3-min warm-up (10–20 W) in increments of 10, 15, or 20 W each minute; a pedalling rate of 60–80 revolutions per minute; a 3-min active recovery (20 W); and a 2-min rest. Exercise was pursued until the limit of the patient's tolerance was reached. For CPET we used a calibrated gas analyser with 12-lead ECG equipment (ErgoCard Medisoft, Belgium), a cycle ergometer (Ergoline, Jaeger, Erich Jaeger GmbH, Hoechberg, Germany), a pulse oximeter (Masimo, Irvine, USA), and a manual sphygmomanometer. VO_{2peak} and VAT values were normalized in a percentage of the predicted VO_{2peak} using normal values from Wasserman and Cooper [21,22].

On all points of the curve of the increment period, the slope of ventilation according to carbon dioxide expiratory flow (VE/VCO₂ slope), was calculated and used to evaluate ventilatory response to exercise. An increased VE/VCO₂ slope >34 suggested the existence of an impaired ventilatory efficiency [23].

2.4. Formal aspects

The study was conducted in compliance with the Good Clinical Practices protocol and Declaration of Helsinki principles. It was approved by South Mediterranean IV Ethics Committee (2014-A00113-44) and registered on [ClinicalTrials.gov](https://clinicaltrials.gov) (NCT02561975). Informed consent was obtained from all major patients, parents or legal guardians.

2.5. Statistical analyses

In patients and healthy controls, clinical characteristics, pulmonary function, and DLCO-DLNO parameters were described, using mean and standard deviations (SD) for continuous variables, and frequencies and proportions for categorical variables. The impact of switching from sitting to supine position on the values of DLCO-DLNO parameters were first analysed, separately in patients and healthy controls, using the

Table 2

Clinical and paraclinical characteristics in the CHD group.

CHD	CHD group (overall)	Single ventricle	Pulmonary regurgitation
N	24	12	12
NYHA (N)	I	3	9
	II	8	3
	III	1	0
	IV	0	0
Haemoglobin (g/dL)	15.6 ± 2.0	16.0 ± 1.9	15.2 ± 2.0
Sinus rhythm (N,%)	23	11 (92%)	12 (100%)
Right ventricle TAPSE (mm)	16.7 ± 5.1	14.7 ± 3.9	18.9 ± 5.5*
RV systolic pressure (mmHg)	NA	NA	39.4 ± 12.3
Systemic ventricle EF (%)	61.5 ± 10.2	54.2 ± 8.1	67 ± 8.0*
Global longitudinal Strain (%)	-16.2 ± 2.6	-15.6 ± 2	-16.8 ± 2.8
Subaortic VTI (cm)	19.5 ± 5.4	19.8 ± 6.2	19.2 ± 4.6
VO _{2max} (% predict)	55.3 ± 18	48 ± 18	63.3 ± 15*
VAT (% predict)	38.1 ± 9.5	33.8 ± 9.3	42.7 ± 7.5*
VE/VCO ₂ slope	36.4 ± 8.8	39.1 ± 8.9	33.2 ± 8.0
SpO _{2max} - SpO _{2min} (%)	2.2 ± 3.2	1.2 ± 2.1	2.9 ± 3.7

Legend: Data were described as mean ± SD (standard deviation); *P < 0.05, indicates a significant difference between single ventricle and pulmonary regurgitation sub-groups (Mann-Whitney test).

Abbreviations: NA, non applicable; % predict, percentage of predicted VO_{2max} values; SpO₂, oxygen saturation measured with pulse oximetry; TAPSE, tricuspid annular plane systolic excursion; RV, right ventricle; EF, ejection fraction; VAT, ventilatory anaerobic threshold; VTI, velocity time integral.

paired Student *t*-test or the sign rank test according to the distribution of the difference between the paired values (supine value-sitting value). We then compared the values of DLCO-DLNO parameters – separately in sitting and supine position-, and of the difference (supine value-sitting value), according to the presence of CHD using the Student *t*-test when the data distribution was normal, and using the Mann-Whitney *U* test when the variables were skewed.

We then fitted a linear mixed model with subject-specific random intercept to take into account for correlations between repeated measures of each subject in different position. The dependent variables were DLCO-DLNO parameters. Fixed effects were the disease status (CHD versus no CHD) and posture (supine versus sitting position). An interaction term between disease status and posture was also tested in a separate model (E-Table 1).

The linear relationship between CPET parameters (VO_{2peak}, VE/VCO₂) and the difference in DLCO-DLNO parameters due to postural change, was analysed using Bayesian determination of the Pearson's correlation coefficient (*r*).

Statistical analyses were performed at the conventional two-tailed α level of 0.05 using SAS version 9.4 (SAS Institute, Cary, NC).

3. Results

3.1. Study population

During the study period, 48 patients were included, 32 patients with CHD and 16 healthy subjects. However, we excluded 5 patients for technical problems and 3 patients for measurement errors (negative DmCO). Therefore, a total of 40 patients (24 CHD; 16 healthy controls) with complete CPET and double gas transfer measures were analysed in the study.

No significant differences were observed between healthy controls and CHD patients in terms of main demographic characteristics (Table 1). The main clinical and paraclinical characteristics of the CHD group were reported in Table 2. In terms of lung function, the CHD group presented with a decreased forced vital capacity (FVC), a decreased

Table 3

Description of DLCO-DLNO parameters in CHD and healthy controls in sitting and supine positions.

DLNO-DLCO parameters	Sitting Position		Supine position	
	Healthy controls (N = 16)	CHD group (N = 24)	Healthy controls (N = 16)	CHD group (N = 24)
AV (L)	6.67 (1.22)	5.13 (1.18)*	6.00 (1.38)	4.68 (1.12) ** \$§
IV (L)	4.70 (0.96)	3.34 (0.79) *	4.40 (0.98)	3.03 (0.79) ** \$§
DLCO (mL/min/mmHg)	33.38 (9.56)	25.33 (6.14) *	36.42 (9.45) §	27.55 (5.94) ** \$§
DLNO (mL/min/mmHg)	184.78 (50.22)	141.35 (35.71) *	184.07 (45.82)	144.52 (37.83) **
DLCO/AV (L/min/mmHg)	4.96 (0.84)	5.00 (0.93)	6.36 (1.99)	5.97 (0.83)
DLNO/AV (L/min/mmHg)	27.49 (4.25)	27.80 (5.21)	32.11 (10.04)	31.21 (6.07)
DmCO (mL/min/mmHg)	248.11 (89.15)	341.63 (331.51)	201.89 (65.36) §	209.30 (201.12) \$§
DmNO (mL/min/mmHg)	488.77 (175.63)	673.02 (653.07)	397.72 (128.76) §	412.31 (396.20) \$§
Vcap (mL)	68.21 (18.95)	48.52 (13.53) *	78.71 (20.18) §	61.30 (12.94)** \$§

Legend: All parameters were analysed with original scale (measured values). Data were described as mean and standard deviation (SD).

Abbreviations: AV, alveolar volume; IV, inspiratory volume; DLNO, total diffusing capacity of the lung for nitric oxide; DLCO, total diffusing capacity of the lung for carbon monoxide; DmCO, diffusing capacity of the membrane for carbon monoxide; VCap, total volume of blood in the lung capillaries exposed to alveolar air. * Parameter statistically different ($p < 0.05$) between healthy controls and CHD group in sitting position ** Parameter statistically different ($p < 0.05$) between healthy controls and CHD group in supine position § Parameter statistically different ($p < 0.05$) between sitting and supine position in healthy controls (paired statistical test) \$§ Parameter statistically different ($p < 0.05$) between sitting and supine position in CHD group (paired statistical test). When the change in value of the parameters during the switch of position was compared between the CHD and healthy groups, no significant difference was found.

forced expiratory volume in 1 s (FEV1), and a normal Tiffeneau ratio, suggesting the existence of a restrictive pattern. However, the trend for lower functional residual capacity and total lung capacity in the CHD group did not reach statistical significance. As shown in Table 2, CHD patients had standard echocardiographic evaluation without evidence of significant ventricular dysfunction.

When performing exercise test, CHD patients presented with an impaired VO₂peak of $53\% \pm 20.9\%$ of predicted values, overall. Exercise capacity was significantly lower in patients with a single ventricle than in those with pulmonary regurgitation (predicted VO₂peak of $48\% \pm 18\%$ vs. $63\% \pm 15\%$, $P = 0.04$, respectively). Impaired ventilatory efficiency with a VE/VCO₂ slope >34 was predominantly observed in patients with a single ventricle (mean VE/VCO₂ slope of 39.1 ± 8.9) (Table 2).

3.2. DLCO-DLNO analyses in CHD versus healthy controls at rest in sitting position

Individual and mean values of lung transfer factors (DLCO-DLNO) and their components in CHD and healthy controls are presented and compared in Table 3. DLCO and DLNO in the sitting position were significantly lower in the CHD group in comparison with healthy controls. Moreover, in the CHD group, the alveolar volume (AV), the inspiratory volume (IV) and the pulmonary capillary volume (Vcap) were lower than in the control group. No significant difference between CHD and healthy controls was found for DmCO, DmNO, DLCO/AV and DLNO/AV.

Table 4

Impact of the presence of CHD and switching to supine position on DLCO-DLNO parameters.

	Independent effects: estimated values (95% CI)				
	Presence of CHD	pvalue	Switching to supine position	pvalue	
AV (L)	-1.4 (-2.1 to -0.8)	<.01	-0.5 (-1.0 to -0.1)	0.02	
IV (L)	-1.4 (-1.8 to -0.9)	<.01	-0.3 (-0.6 to 0.0)	0.06	
DLCO (mL/min/mmHg)	-8.5 (-13.2 to -3.7)	<.01	2.5 (1.3–3.8)	<.01	
DLNO (mL/min/mmHg)	-41.5 (-68.2 to -14.8)	<.01	1.6 (-3.5 to 6.7)	0.52	
DLCO/AV (L/min/mmHg)	-0.2 (-0.8 to 0.5)	0.57	1.1 (0.7–1.6)	<.01	
DLNO/AV (L/min/mmHg)	-0.3 (-3.9 to 3.3)	0.87	3.9 (1.6–6.2)	<.01	
DmCO (mL/min/mmHg)	50.5 (-78.2 to 179.2)	0.43	-97.9 (-159.5 to -36.3)	<.01	
Vcap (mL)	-18.6 (-27.9 to -9.2)	<.01	11.9 (7.3–16.5)	<.01	
DmNO (mL/min/mmHg)	99.4 (-154.1 to 353.0)	0.43	-192.8 (-314.1 to -71.6)	<.01	
DLNO/DLCO ratio	0.1 (-0.3 to 0.5)	0.66	-0.4 (-0.6 to -0.3)	<.01	

Note: The table represents the estimated mean and 95% confidence interval of the effect of 2 independent factors: the presence of CHD and the change from sitting to supine. A pvalue <0.05 indicates an effect significantly different from 0. The Fig. 2 was established from these results.

Abbreviations: AV, alveolar volume; IV, inspiratory volume; DLNO, total diffusing capacity of the lung for nitric oxide; DLCO, total diffusing capacity of the lung for carbon monoxide; DmCO, diffusing capacity of the membrane for carbon monoxide; VCap, total volume of blood in the lung capillaries exposed to alveolar air.

3.3. Effects of postural change on DLCO-DLNO parameters at rest in CHD and healthy controls

In both healthy controls and CHD patients, postural change from sitting to supine position led to similar increases in pulmonary capillary volume (Vcap), and similar decreases in AV and alveolar capillary membrane conductance (Dm), with a similar magnitude of difference. In both groups, the body position did not have any effect on IV.

The existence of a CHD, as well as the supine posture, presented independent and different effects on the pulmonary gas transfer factors, with a similar postural adaptation of DLCO, DLNO and Vcap recruitment between CHD and healthy population (as shown by the non-significance of the interaction terms between CHD and posture in e-Table 1).

3.4. Correlation between DLCO, DLNO and Vcap postural-induced changes at rest with VO₂ max and VE/VCO₂ slope in the CHD group (Fig. 2)

In the CHD group, postural-induced changes in DLCO, DLNO and Vcap correlated with VE/VCO₂ slope during exercise ($r = -0.62$, $r = -0.63$, and $r = -0.54$, all $P < 0.05$, respectively). The VO₂peak only correlated with postural-induced change in DLNO ($r = 0.48$, $P < 0.05$) (Fig. 1). Based on the regression models, for each DLCO unit increase between sitting and supine position we observed a VE/VCO₂ slope decrease of 1.45 (95% CI [0.47; 2.4]) which reflects an improvement in

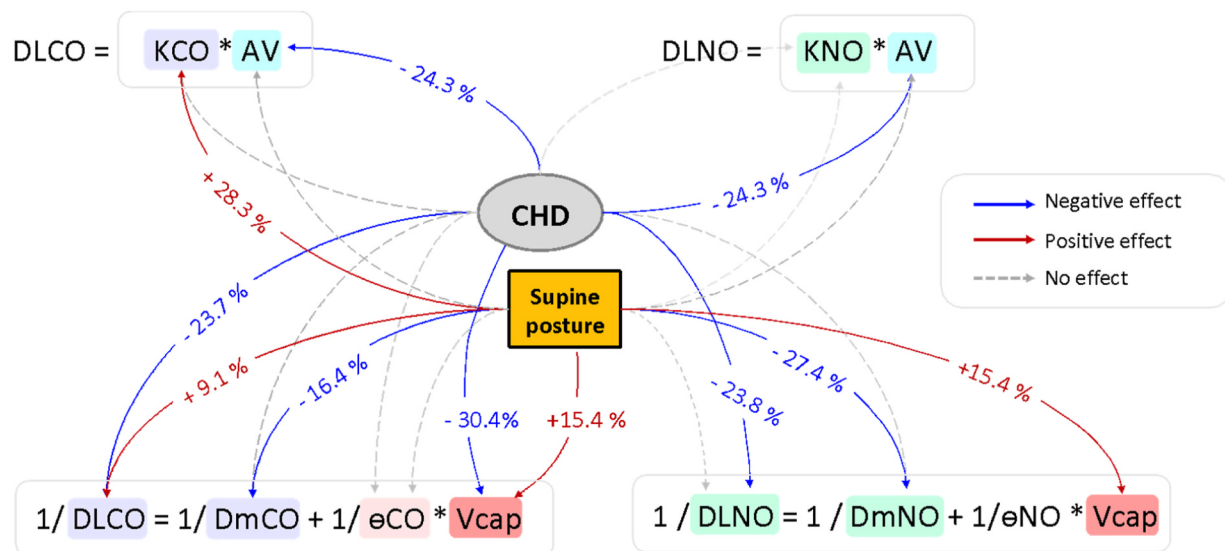


Fig. 1. Independent effects of CHD and supine position on the sub-components of gas transfer factors for NO and CO. Legend: Graph summarising the independent effects of CHD and supine posture (from the centre) on the transfer factors (DLNO, DLCO) and their sub-components (DmCO, θCO , DmNO and Vcap), based on the Roughton-Forster's equation and alveolar volume (AV), represented in separated blocks. Red arrows indicate the significant positive effect (= increase), while blue arrows indicate the significant negative effect (= decrease) on the target variable. The numbers indicate the mean effect size expressed in % of basal value (i.e. x for CHD, y for supine) and could be interpreted as: the target variable would decrease by x % in patients with CHD or would increase by y % when switching from sitting to supine posture. When no significant effect could be established, the connection link was drawn in grey colour and dotted line. Abbreviation: AV, alveolar volume; DLCO, total diffusing capacity of the lung for carbon monoxide; DLNO, total diffusing capacity of the lung for nitric oxide; KCO, ratio of total diffusing capacity of the lung for carbon monoxide over alveolar volume; KNO, ratio of total diffusing capacity of the lung for nitric oxide over alveolar volume; DmCO, diffusing capacity of the membrane for carbon monoxide; DmNO, Diffusing capacity of the membrane for nitric oxide; Vcap, total volume of blood in the lung capillaries exposed to alveolar air.

ventilatory efficiency (gas exchange) during exercise. Similarly, for each Vcap increase of 1 mL we observed a VE/VCO₂ slope decrease of 0.46 (95% CI [0.83; 0.098]).

The VO₂peak correlated with the postural-induced change in DLNO, with an increase in VO₂peak of 0.18 mL/kg/min (95% CI[0.02; 0.37]) for each unit of DLNO increase.

4. Discussion

The prospective controlled trial reported, for the first time, the use of double gas transfer factors (DLCO-DLNO) in patients with CHD at rest to explore the effect of their impaired pulmonary blood flow on the various components of pulmonary gas transfer, and its consequence on maximal exercise adaptation.

Defect in total lung diffusion capacity evaluated by DLCO, and more specifically diffusion capacity of the alveolar capillary membrane, is associated with disease severity and prognosis of chronic congestive heart failure [24,25]. Indeed, DLCO is related to the combining effects of resistance both to gas flow across the alveolar capillary membrane and Hb binding by CO. Since both gas-transfer resistances are connected in series, the total resistance for CO transfer follows the Roughton-Forster equation [26] ((e.g. $1/DLCO = 1/DmCO + 1/\theta CO Vcap$). DmCO reflects the diffusion limitation across the effective alveolocapillary membrane, the alveolar wall and the plasma layer surrounding the erythrocyte. The $\theta CO Vcap$ is defined as the product of alveolar capillary blood volume exposed to alveolar air (Vcap) and specific gas conductance for CO in the blood (θCO). The θCO represents the CO diffusive process across the membrane of the erythrocyte and its competitive replacement reaction with oxyhemoglobin (HbO₂). To determine DmCO and Vcap, which are the two main components of pulmonary gas transfer, Guénard et al. [14] and Borland and Higen-Bottamet [27] developed one-step CO-NO technique requiring the measurement of DLCO and DLNO during a single hold breath, a very easy and clinically convenient technique, which has been recently standardized [19]. This specific increase in membrane resistance to CO (DmCO), previously observed in chronic

congestive heart failure, has been explained by fluid accumulation, cellular increase, fibrosis, and remodelling of connective tissue [25,28].

In patients with CHD concerned with impaired pulmonary circulation, e.g. those with a single ventricle or a severe pulmonary regurgitation, we observed a significant decrease of FVC and FEV₁. This results was previously reported from our group and others in paediatric and adult patients with CHD [12,29,30] and suggests the presence of smaller lung volumes due to the existence of an impaired thoracic compliance after cardio-thoracic surgical procedures. Similarly, in these patients, we also observed at rest, in the sitting position, a smaller capillary volume.

Interestingly, the CHD patients' DLCO and DLNO also at rest, tended to be lower than in healthy subjects. However, in our study, DLCO/AV and DLNO/AV at rest were similar between both groups. Indeed, when normalized for alveolar volume, DLCO and DLNO in healthy subjects and in patients with CHD were similar. This observation suggests that the diffusion of these gases per functional lung unit was normal in the CHD patients, and that the low DLCO and DLNO was due to their relatively small lungs and consequent reduced number of functional lung units, rather than to diffusion abnormalities within the individual functional lung units.

In the supine position, the specific alteration of the preload factor previously reported in a similar CHD population [20], may blunt the increase of blood flow into the pulmonary circulation. However, we observed, that the postural change from sitting to supine induced in both group similar increase in Vcap, DLCO and DLNO. Therefore, our study highlighted the existence of a preserved pulmonary capillary recruitment capacity in CHD patients, as induced by a simple lying position manoeuvre, with a magnitude of difference in postural-induced changes of pulmonary capillary volumes similar to that of healthy controls.

A previous study reported that DmCO was also higher in supine compared to erected position [31]. Indeed, the increased pulmonary blood volume and pulmonary capillary recruitment in supine position increase effective alveolar-capillary surface area and alveolar-capillary conductance. However, in our study, we observed a DmCO decrease with postural change in both groups. This may be related to the short

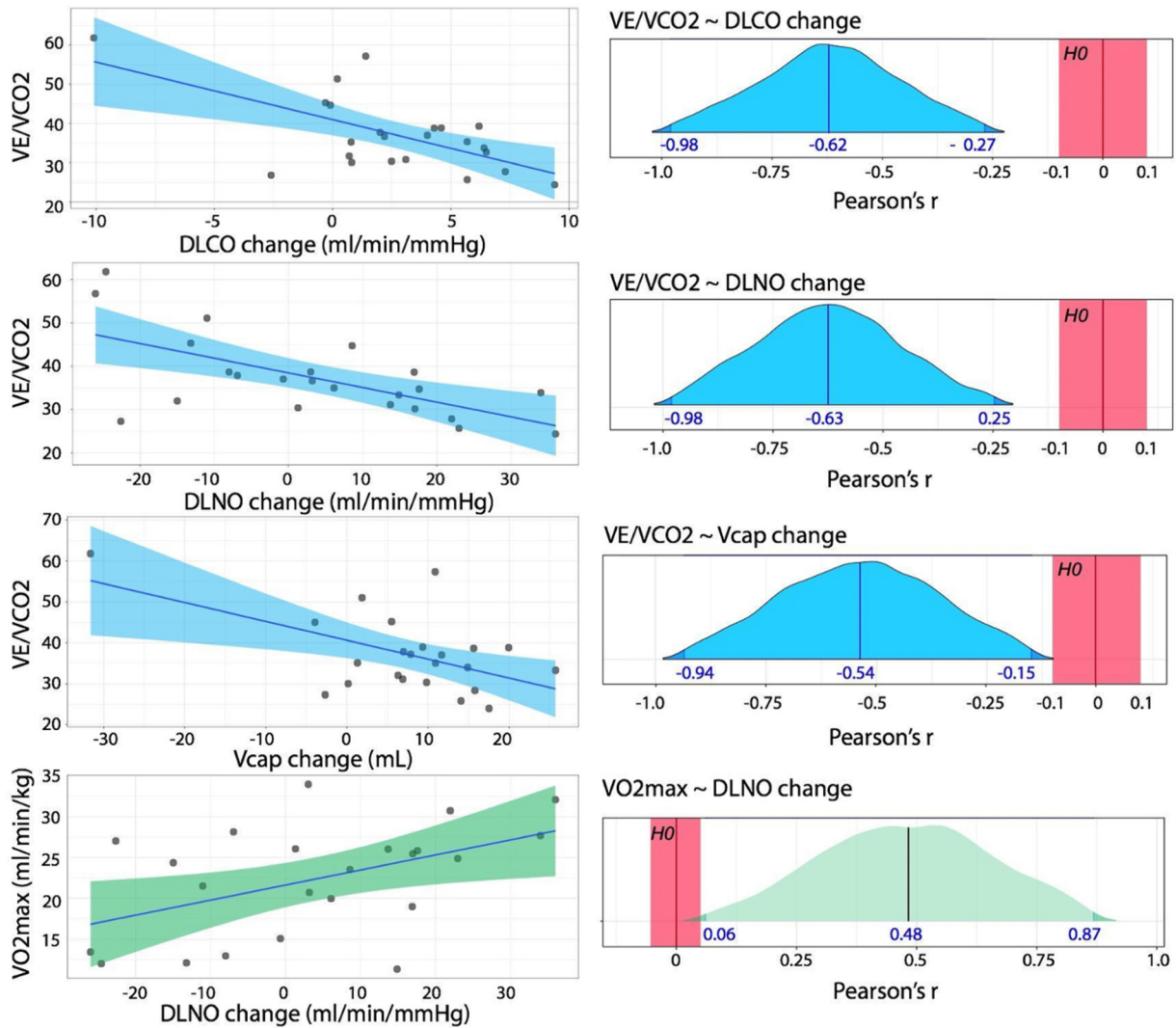


Fig. 2. Relationship between $VO_2\text{peak}$, VE/VCO_2 slope and position related change in gas transfer factors. Legend: the graph represents the result of a Bayesian linear correlation analysis, including the marginal effect of 4 linear regression models (left side) to estimate the values of VE/VCO_2 slope and $VO_2\text{peak}$ (Y-axis) as a linear function of the change in DLNO, DLCO and Vcap (X-axis). On the left column, the lines indicate the mean predicted values, while the curvilinear bands represent the confidence interval of this prediction. The right column shows the estimated posterior distribution of the Pearson's r coefficient measuring the linear correlation between the studied variables. The lighter area in each Kernel Density Estimate (KDE) plots represents the highest density interval (95% of Highest Density Interval, HDI) while the 3 labelled vertical lines indicate the 5th, 50th and 95th centiles of the distribution. The red vertical lines and red boxes determine the thresholds for null hypothesis (H_0 : $r = 0$, with tolerance interval of ± 0.1).

delay after postural change (1 min) in our study, before repeating DLCO-DLNO measures, as opposed to 2–3 h in the previous study [31], where the pulmonary circulation could adapt to increased venous return. We thus speculated that the sudden change of position within 1 min was sufficient to induce a transient congestion of interstitial tissue and a membrane diffusion limitation, which may represent similarity with the transient pulmonary capillary volume increase during exercise. This suggests that $DmCO$ decrease induced by the lying position was compensated by an Vcap increase, due to pulmonary capillary bed recruitment, and resulted in a moderate DLCO increase, as illustrated in Fig. 1.

Although CHD patients with impaired pulmonary circulation have a lower pulmonary capillary volume at rest, they tend to maintain a normal capacity to increase Vcap during a simple recruitment manoeuvre at rest. These results are in line with the existence of a preserved ventricular contractile function in our patients, as previously reported [32].

Despite this preserved vascular adaptability in the CHD group, postural-induced changes of pulmonary capillary volumes negatively

correlated with ventilatory efficiency at exercise.

Indeed, in this study we observed, for the first time in CHD patients with impaired pulmonary circulation, a negative correlation between ventilatory efficiency (VE/VCO_2) and DLCO-DLNO postural-induced change at rest. Interestingly, among all DLCO-DLNO parameters, Vcap change appeared to be the main component of this correlation. Therefore, CHD patients whose Vcap increased from sitting to supine positions tended to have lower VE/VCO_2 slopes (i.e., better ventilation efficiency) and consequently higher exercise tolerance than those who did not increase their Vcap in response to this postural change.

In CHD patients, during exercise, their deficit of pulmonary perfusion may increase the ventilatory response to exercise, participating in the limiting factors for aerobic fitness.

However, maximum oxygen uptake and DLNO change at rest were weakly correlated, and no correlation was found between $VO_2\text{peak}$ and DLCO change or Vcap change. These negative results may be explained by the fact that $VO_2\text{max}$ is a less efficient parameter to account for alteration of ventilation-perfusion ratio than VE/VCO_2 slope.

Assessments of pulmonary function lung, diffusion capacity and its

components, and the effect of postural changes on these parameters provides new insights into the effects of CHD upon exercise physiology and pulmonary gas exchange during exercise.

We found in this study that DLNO-DLCO measurement at rest may predict VE/VCO₂ slope during maximal exercise, which is an independent variable of VO₂peak, and is sensible to mismatch from inadequate pulmonary perfusion of normally ventilated alveoli. Thus, DLNO-DLCO measured at rest may be a useful tool, in addition to VO₂peak, to more specifically evaluate pulmonary perfusion defect and its consequence on ventilatory response to exercise, in the follow-up of patients with single ventricle or pulmonary regurgitation.

4.1. Study limitations

Our results rely on a single-centre study with a small cohort of patients and heterogeneous CHD conditions. Further studies with a more homogeneous CHD population in terms of haemodynamics will be necessary in the near future to better assess the parameters involved in Vcap change during exercise and their implication in aerobic fitness limitation.

In this study we used a finite θ NO value of 4.5 as recommended by ERS guidelines [18]. However, as previously reported [33], we found negative DmCO values in three patients which were excluded from study analyses. The reason for this negative value remains unclear and may indicate that θ NO could be finite in vitro but infinite in vivo [34].

The main limitation of this study is the lack of DLCO-DLNO measurement during exercise. However, it is difficult for patient with severe cardiac conditions to hold the breath for a 6-s period and exhale to residual volume within 4 s, during exercise. Indeed, it seems that patients with complex CHD may find it difficult to perform a rapid inspiration from residual volume to total lung capacity. In greater detail, we found that maximal inspired volume (VI) from residual volume was statistically lower than forced vital capacity during plethysmography in the CHD group (CVF 3.46 ± 0.86 versus 3.34 ± 0.78 , $P = 0.049$). However, maximal inspiration to 100% of inspiratory vital capacity is not necessary for DLCO/DLNO measurements. Therefore, we think that the hypothetical decrease of the ability to perform the ventilatory manoeuvre did not significantly affect the study results.

A change from seated to supine position cannot be assimilated to the effect of exercise on the pulmonary circulation. Indeed, the Vcap increase during exercise mainly depends on several parameters: a) pulmonary blood flow increase, b) cardiac output, c) pulmonary arterial pressure, d) intrathoracic pressure swings linked to ventilation, e) spatial distribution of blood flow in the lung and intrinsic capillary recruitment.

Our objective was not to reproduce at rest the physiological stress of exercise but to propose a simple tool, which may reflect the inability of pulmonary vascularization to expand.

However, as we couldn't directly evaluate Vcap response to exercise in CHD and directly compare with Vcap response to exercise in control subjects, our hypothesis still remains speculative.

In this study, healthy subjects did not perform a CPET. Indeed, we previously showed that CHD patients presented a lower VO₂peak associated to an increase in ventilatory response to exercise, in comparison with healthy controls [10,12]. Moreover, in healthy subject, the variability of VO₂peak and ventilatory response to exercise is moderate and pulmonary circulation adaptation to exercise is not a limiting factor for aerobic fitness [35].

5. Conclusion

CPET contributes to identifying exercise capacity limiting factors and therefore helps the physician to adapt the therapeutic project. Currently, VO₂peak and VE/VCO₂ slope are two main predictors of prognosis in chronic heart failure, including in patients with CHD.

In this study, CHD patients with impaired pulmonary circulation

have a reduced pulmonary membrane diffusion and pulmonary capillary volume, mainly as a result of a decreased pulmonary volume, in comparison with healthy subjects. However, they maintain a normal capacity to adapt those parameters through a simple recruitment manoeuvre. Moreover, pulmonary capillary volume adaptation evaluated at rest predicts the level of ventilatory efficiency during exercise, which represents a main limiting factor in CHD patients. This study emphasized the importance of pulmonary function assessment in the follow-up of the CHD population, with a particular focus on lung diffusion capacity and its components.

Declarations of competing interest

The authors have no competing interests to declare.

Acknowledgement

Acknowledgement of grant support: this study was supported by the paediatric and congenital cardiology clinical research program in Montpellier University Hospital.

References

- [1] Albouaini K, Egred M, Alahmar A, et al. Cardiopulmonary exercise testing and its application. *Postgrad Med* 2007;83:675–82. <https://doi.org/10.1136/hrt.2007.121558>.
- [2] Amedro P, Picot MC, Moniotte S, et al. Correlation between cardio-pulmonary exercise test variables and health-related quality of life among children with congenital heart diseases. *Int J Cardiol* 2016;203:1052–60. <https://doi.org/10.1016/j.ijcard.2015.11.028>.
- [3] Amedro P, Gavotto A, Guillaumont S, et al. Cardiopulmonary fitness in children with congenital heart diseases versus healthy children. *Heart* 2018;104:1026–36. <https://doi.org/10.1136/heartjnl-2017-312339>.
- [4] Myers J, Arena R, Dewey F, et al. A cardiopulmonary exercise testing score for predicting outcomes in patients with heart failure. *Am Heart J* 2008;156:1177–83. <https://doi.org/10.1016/j.ahj.2008.07.010>.
- [5] Guazzi M, Adams V, Conraads V, et al. Clinical recommendations for cardiopulmonary exercise testing data assessment in specific patient populations. *Circulation* 2012;126:2261–74. <https://doi.org/10.1161/CIR.0b013e31826fb946>.
- [6] Khan AM, Paridon SM, Kim YY. Cardiopulmonary exercise testing in adults with congenital heart disease. *Exp Rev Cardiovasc Ther* 2014;12:863–72. <https://doi.org/10.1586/14779072.2014.919223>.
- [7] Marcadet D-M, Pavy B, Bosser G, et al. French Society of Cardiology guidelines on exercise tests (part 2): indications for exercise tests in cardiac diseases. *Archives of Cardiovascular Diseases* 2019;112:56–66. <https://doi.org/10.1016/j.acvd.2018.07.001>.
- [8] Kempny A, Dimopoulos K, Uebing A, et al. Reference values for exercise limitations among adults with congenital heart disease. Relation to activities of daily life—single centre experience and review of published data. *Eur Heart J* 2012;33:1386–96. <https://doi.org/10.1093/eurheartj/ehr461>.
- [9] Weatherald J, Sattler C, Garcia G, et al. Ventilatory response to exercise in cardiopulmonary disease: the role of chemosensitivity and dead space. *Eur Respir J* 2018;51. <https://doi.org/10.1183/13993003.00860-2017>.
- [10] Gavotto A, Huguette H, Picot M-C, et al. The Ve/Vco₂ slope: a useful tool to evaluate the physiological status of children with congenital heart disease. *J Appl Physiol* 1985;129:1102–10. <https://doi.org/10.1152/japplphysiol.00520.2020>.
- [11] Degano B, Mittaine M, Guénard H, et al. Nitric oxide and carbon monoxide lung transfer in patients with advanced liver cirrhosis. *J Appl Physiol* 1985;107:139–43. <https://doi.org/10.1152/japplphysiol.91621.2008>.
- [12] Abassi H, Gavotto A, Picot MC, et al. Impaired pulmonary function and its association with clinical outcomes, exercise capacity and quality of life in children with congenital heart disease. *Int J Cardiol* 2019;285:86–92. <https://doi.org/10.1016/j.ijcard.2019.02.069>.
- [13] Roughton FJW, Forster RE. Relative importance of diffusion and chemical reaction rates in determining rate of exchange of gases in the human lung, with special reference to true diffusing capacity of pulmonary membrane and volume of blood in the lung capillaries. *J Appl Physiol* 1957;11:290–302. <https://doi.org/10.1152/jappl.1957.11.2.290>.
- [14] Guenard H, Varenne N, Vaida P. Determination of lung capillary blood volume and membrane diffusing capacity in man by the measurements of NO and CO transfer. *Respir Physiol* 1987;70:113–20. [https://doi.org/10.1016/S0034-5687\(87\)80036-1](https://doi.org/10.1016/S0034-5687(87)80036-1).

- [15] Houyel L, Khoshnood B, Anderson RH, et al. Population-based evaluation of a suggested anatomic and clinical classification of congenital heart defects based on the International Paediatric and Congenital Cardiac Code. 2011. p. 9.
- [16] Bonhoeffer P, Deanfield JE, Galie N, et al. The task force on the management of grown-up congenital heart disease of the European Society of Cardiology (ESC). ;43.
- [17] Brusasco V. Coming together: the ATS/ERS consensus on clinical pulmonary function testing. *Eur Respir J* 2005;26:1–2. <https://doi.org/10.1183/09031936.05.00034205>.
- [18] Horváth I, Barnes PJ, Loukides S, et al. A European Respiratory Society technical standard: exhaled biomarkers in lung disease. *Eur Respir J* 2017;49:1600965. <https://doi.org/10.1183/13993003.00965-2016>.
- [19] Zavorsky GS, Hsia CCW, Hughes JMB, et al. Standardisation and application of the single-breath determination of nitric oxide uptake in the lung. *Eur Respir J* 2017;49:1600962. <https://doi.org/10.1183/13993003.00962-2016>.
- [20] Carlsen E, Comroe JH. The rate of uptake of carbon monoxide and of nitric oxide by normal human erythrocytes and experimentally produced spherocytes. *J Gen Physiol* 1958;42:83–107. <https://doi.org/10.1085/jgp.42.1.83>.
- [21] Cooper DM, Weiler-Ravell D, Whipp BJ, et al. Aerobic parameters of exercise as a function of body size during growth in children. *J Appl Physiol* 1984;56:628–34. <https://doi.org/10.1152/jappl.1984.56.3.628>.
- [22] Cooper DM, Berry C, Lamarra N, et al. Kinetics of oxygen uptake and heart rate at onset of exercise in children. *J Appl Physiol* 1985;59:211–7. <https://doi.org/10.1152/jappl.1985.59.1.211>.
- [23] Robbers-Visser D, Jan ten Harkel D, Kapusta L, et al. Usefulness of cardiac magnetic resonance imaging combined with low-dose dobutamine stress to detect an abnormal ventricular stress response in children and young adults after fontan operation at young age. *Am J Cardiol* 2008;101:1657–62. <https://doi.org/10.1016/j.amjcard.2008.01.050>.
- [24] Guazzi M. Alveolar-capillary membrane dysfunction in chronic heart failure: pathophysiology and therapeutic implications. *Clin Sci (Lond)* 2000;98:633–41.
- [25] Agostoni P, Bussotti M, Cattadori G, et al. Gas diffusion and alveolar-capillary unit in chronic heart failure. *Eur Heart J* 2006;27:2538–43. <https://doi.org/10.1093/eurheartj/ehl302>.
- [26] Roughton FJ, Forster RE. Relative importance of diffusion and chemical reaction rates in determining rate of exchange of gases in the human lung, with special reference to true diffusing capacity of pulmonary membrane and volume of blood in the lung capillaries. *J Appl Physiol* 1957;11:290–302. <https://doi.org/10.1152/jappl.1957.11.2.290>.
- [27] Borland CD, Higenbottam TW. A simultaneous single breath measurement of pulmonary diffusing capacity with nitric oxide and carbon monoxide. *Eur Respir J* 1989;2:56–63.
- [28] Agostoni P, Cattadori G, Bianchi M, et al. Exercise-induced pulmonary edema in heart failure. *Circulation* 2003;108:2666–71. <https://doi.org/10.1161/01.CIR.0000097115.61309.59>.
- [29] Hawkins SMM, Taylor AL, Sillau SH, et al. Restrictive lung function in pediatric patients with structural congenital heart disease. *J Thorac Cardiovasc Surg* 2014;148:207–11. <https://doi.org/10.1016/j.jtcvs.2013.07.080>.
- [30] Alonso-Gonzalez R, Borgia F, Diller G-P, et al. Abnormal lung function in adults with congenital heart disease: prevalence, relation to cardiac anatomy, and association with survival. *Circulation* 2013;127:882–90. <https://doi.org/10.1161/CIRCULATIONAHA.112.126755>.
- [31] Chang S-C, Chang H-I, Liu S-Y, et al. Effects of body position and age on membrane diffusing capacity and pulmonary capillary blood volume. *Chest* 1992;102:139–42. <https://doi.org/10.1378/chest.102.1.139>.
- [32] Paridon SM, Mitchell PD, Colan SD, et al. A cross-sectional study of exercise performance during the first 2 decades of life after the fontan operation. *J Am Coll Cardiol* 2008;52:99–107. <https://doi.org/10.1016/j.jacc.2008.02.081>.
- [33] Coffman KE, Chase SC, Taylor BJ, et al. The blood transfer conductance for nitric oxide: infinite vs. finite \dot{V} NO. *Respir Physiol Neurobiol* 2017;241:45–52. <https://doi.org/10.1016/j.resp.2016.12.007>.
- [34] Borland C, Patel S, Zhu Q, et al. Hypothesis: why \dot{V} NO could be finite in vitro but infinite in vivo. *Respir Physiol Neurobiol* 2017;241:58–61. <https://doi.org/10.1016/j.resp.2017.02.013>.
- [35] Coffman KE, Carlson AR, Miller AD, et al. The effect of aging and cardiorespiratory fitness on the lung diffusing capacity response to exercise in healthy humans. *J Appl Physiol* 2017;122:1425–34. <https://doi.org/10.1152/japplphysiol.00694.2016>.