Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Journal of Electromyography and Kinesiology 59 (2021) 102567

Contents lists available at ScienceDirect

Journal of Electromyography and Kinesiology

journal homepage: www.elsevier.com/locate/jelekin

EMG breakpoints for detecting anaerobic threshold and respiratory compensation point in recovered COVID-19 patients

Murillo Frazão [a,b,c,d,*], Paulo Eugênio Silva [e], Lucas de Assis Pereira Cacau [f], Tullio Rocha Petrucci [d], Mariela Cometki Assis [f,g], Amilton da Cruz Santos [a,b], Maria do Socorro Brasileiro-Santos [a,b]

[a] Laboratory of Physical Training Studies Applied to Health, Physical Education Department, Universidade Federal da Paraíba (UFPB), João Pessoa, PB, Brazil
[b] Associate Graduate Programme in Physical Education UPE/UFPB, PB, Brazil
[c] Lauro Wanderley University Hospital, UFPB, PB, Brazil
[d] CLINAR – Exercise Physiology, João Pessoa, PB, Brazil
[e] Clinical Physiology and Technological Innovation FICTI-CNPq, Brasília, DF, Brazil
[f] Intervent – Infectology and Phyiotherapy, Aracaju, SE, Brazil
[g] Unimed, Aracaju, SE, Brazil

ARTICLE INFO

Keywords:
Electromyography
Cardiopulmonary exercise test
COVID-19
Anaerobic threshold

ABSTRACT

Introduction: A huge number of COVID-19 patients should be referred to rehabilitation programmes. Individualizing the exercise intensity by metabolic response provide good physiological results. The aim of this study was to investigate the validity of EMG as a non-invasive determinant of the anaerobic threshold and respiratory compensation point, for more precise exercise intensity prescription.

Methods: An observational cross-sectional study with 66 recovered COVID-19 patients was carried out. The patients underwent a cardiopulmonary exercise test with simultaneous assessment of muscle electromyography in vastus lateralis. EMG breakpoints were analyzed during the ramp-up protocol. The first and second EMG breakpoints were used for anaerobic threshold and respiratory compensation point determination.

Results: EMG and gas exchange analysis presented strong correlation in anaerobic threshold ($r = 0.97$, $p < 0.0001$) and respiratory compensation point detection ($r = 0.99$, $p < 0.0001$) detection. Bland-Altman analysis demonstrated a bias = −4.7 W (SD = 6.2 W, limits of agreement = −16.9 to 7.6) for anaerobic threshold detection in EMG compared to gas exchange analysis. In respiratory compensation point detection, Bland-Altman analysis demonstrated a bias = -2.1 W (SD = 4.5 W, limits of agreement = −10.9 to 6.6) in EMG compared to gas exchange analysis. EMG demonstrated a small effect size compared to gas exchange analysis in oxygen uptake and power output at anaerobic threshold and respiratory compensation point detection.

Conclusions: EMG analysis detects anaerobic threshold and respiratory compensation point without clinical significant difference than gas exchange analysis (gold standard method) in recovered COVID-19 patients.

1. Introduction

Patients who have successfully recovered from the acute COVID-19 pneumonia will require health support to define and quantify the consequences of the disease. The follow-up is currently the new challenge as it was in the beginning for intensive care units. Indeed, it is not clear if COVID-19 will leave permanent lung and/or physical damage, and if so, to what extent. Persisting limitations in respiratory function and gas exchange will likely be more pronounced in the subgroup of severe patients (Mo et al., 2020). In addition, as in non-COVID-19 related acute respiratory distress syndrome, we can anticipate a high incidence of intensive care unit acquired weakness that is associated with poor short- as well as long-term outcomes (Polastri et al., 2020).

Considering the expected high burden of respiratory, physical and psychological impairment following the acute phase of COVID-19, a huge number of patients should be early referred to a rehabilitation program (Polastri et al., 2020). Multinational task force recommends early rehabilitation for patients affected by severe COVID-19. The

* Corresponding author at: Av. Sen. Ruy Carneiro, 412 - Miramar, João Pessoa, PB, Brazil.
E-mail address: murillo.frazao@gmail.com (M. Frazão).

https://doi.org/10.1016/j.jelekin.2021.102567
Received 28 December 2020; Received in revised form 6 May 2021; Accepted 3 June 2021
Available online 8 June 2021

pulmonary rehabilitation model may suit as a framework, particularly in a subset of patients with long term respiratory consequences (Spruit et al., 2020).

Individualizing the exercise intensity by exercise testing evaluation provides good physiological results. The best choice of exercise intensity might also play a fundamental role in the rehabilitation program adherence (Vandoni et al., 2016). Moreover, the use of relative terms such as maximum oxygen uptake percentage or heart rate has been substantially criticized (Meyer et al., 1999). Due to most physiological responses to exercise being intensity dependent, reliance on these parameters alone without considering the anaerobic threshold and respiratory compensation point is not sufficient. The threshold-based exercise intensity method (zone between anaerobic threshold and respiratory compensation point) provides great improvements in cardiorespiratory fitness (Wolpern et al., 2015). Due to physical limitations in recovered severe COVID-19 patients, a threshold-based intensity program could be a reliable and efficient method for exercise prescription in this population (Huang et al., 2016).

Cardiopulmonary exercise test (CPET) is the gold standard method for anaerobic threshold and respiratory compensation point detection. Gas exchange analysis has been used for decades as a precise tool for metabolic behavior determination. However, CPET is a high cost method, not available in a large number of rehabilitation centers. Surface electromyography (EMG) has been used for anaerobic threshold and respiratory compensation point detection in the last decades in healthy and chronic disease populations (Hug et al., 2003) (Lucía et al., 1997), (Tikkanen et al., 2012). EMG is a low cost, portable and non-invasive method which can be incorporated in rehabilitation programs.

In view of the above, the aim of this study was to investigate the validity of EMG as a non-invasive determinant of the metabolic response to incremental exercise. We studied the relationship between EMG activity and the gold standard method of anaerobic threshold and respiratory compensation point detection.

2. Methods

2.1. Study design

This is an observational cross-sectional study. Patients underwent a cardiopulmonary exercise test with simultaneous assessment of muscle electromyography in a single-day evaluation. This study was approved by the local research ethics committee and was registered in the Brazilian Clinical Trial Registration Platform (Number: RBR-6xqcr4).

2.2. Study patients

A group of COVID-19 patients who were referred for functional evaluation by CPET at the Exercise Physiology Laboratory of the Federal University of Paraíba from July 4th to August 14th were considered eligible for this study.

COVID-19 diagnosis was established by clinical symptoms (fever, fatigue, muscle soreness, cough, dyspnea, etc.) associated with a positive laboratory test (nasal swab or serology) and/or chest tomography (ground-glass opacity). Patients were classified as mild (major clinical symptoms without dyspnea or respiratory failure) or severe (major clinical symptoms with dyspnea or respiratory failure), as postulated by Tian et al. (2020). Patients who met the following inclusion criteria were enrolled: recovered (<30 days) from mild to severe COVID-19. Exclusion criteria were based on comorbidity confounding factors. Thus, patients with critical COVID-19 (i.e. who had required intubation and mechanical ventilation) and those with previous cardiac, pulmonary, neurological, hematological or muscular diseases were excluded.

2.3. Cardiopulmonary exercise test

The technical procedures for CPET followed the American Thoracic Society/American College of Chest Physicians guidelines for cycle ergometer testing (ATS/ACCP, 2003). The CPET was performed on a CG-04 cycle ergometer (INBRAMED, Porto Alegre, Brazil). Each subject performed a ramp-up protocol, starting with a warm-up by unloaded pedaling for 2 min, followed by an individually-selected workload increment to achieve maximum effort within 8 to 12 min. Subjects were instructed to keep a cadence of 60 rotations per minute and were strongly encouraged by verbal stimuli to achieve maximum effort. The VO2000 (MedGraphics, St. Paul, Minnesota, USA) was used for gas exchange analysis, and it was calibrated according to the manufacturer's instructions. Data were filtered (mean of 7 points) to avoid noise and analyzed by 10 s-averages. Resting spirometry was conducted before the CPET, in which forced expiratory volume in one second (FEV_1) was measured (ASMA-1, Vitalograph, United Kingdom) to calculate maximum voluntary ventilation ($MVV = FEV_1 \times 35$).

Next, the following variables were considered for analyses: power output, peak oxygen uptake (VO_2), percentage of predicted VO_2 (Hansen et al., 1984), respiratory exchange ratio at maximal effort (RER), oxygen pulse at maximal effort (O_2Pulse), peak ventilation (VE) and breathing reserve used during maximal effort (BR = VE/MVV). Ventilatory equivalents for oxygen (VE/VO_2) and carbonic gas exchange (VE/VCO_2) were used for anaerobic threshold and respiratory compensation point analysis (Gas Exchange analysis). The anaerobic threshold was determined by the first inflection in the VE/VO_2 curve (Wasserman et al., 1994). The respiratory compensation point was determined by the second inflection in the VE/VO_2 curve (Fig. 1).

2.4. Electromyography

Neuromuscular activity during CPET was analyzed by EMG (Fig. 1) using a signal acquisition module with a 12-bit resolution A/D converter (EMG800C, EMG System, São José dos Campos, Brazil). The sampling frequency was adjusted to 1000 Hz, frequency band to 20–500 Hz and gain to 1000 times. Bipolar Ag/AgCl self-adhesive surface electrodes were used and placed 20 mm apart (center to center) on the right vastus lateralis (2/3 of the way from the anterior superior iliac spine to the lateral side of the patella), according to Surface Electromyography for the Non-Invasive Assessment of Muscle recommendations (Hermens et al., 1999). A reference electrode was placed on the ulna. The subject's skin was shaved, abraded and cleaned with alcohol prior to electrode placement.

Root mean square (RMS) values were used for analysis. EMG breakpoints were analyzed during the ramp-up protocol. A visual method based on previous reports by Lucía et al. was employed (Lucía et al., 1999). The increased EMG amplitude reflects the recruitment of additional motor units (Hug, 2009). Based on this, the first EMG breakpoint was assumed to be type IIa fiber activation, and the second EMG breakpoint was assumed to be type IIb fiber activation (Henneman's principle) (Henneman and Somjen, 1965). The first and second EMG breakpoints were used for anaerobic threshold and respiratory compensation point determination (EMG analysis) (Fig. 1).

The raw signal and exercise test time were exported and merged into a non-commercial software called Shengo® (INBRAFIC, Brasília, Brazil). The root mean square voltage was then computed at every five seconds throughout whole test. The signal analysis area was automatically determined by the Shengo® algorithm using a trigger signal acquired from a device adjusted in the cycle ergometer. The window analysis was 500 ms positioned from the signal peak median. Next, the signal was smoothed by seven-point means.

We used the Shengo® algorithm which models RMS response to exercise using multisegment linear regression to establish objective criteria to determine breakpoints in the EMG power output response. With this method, a single linear regression was initially fitted to all data points. A brute force method was then used to fit two lines to the data points. The program calculated regression lines for all possible divisions of the data into two contiguous groups, and the pair of lines yielding the

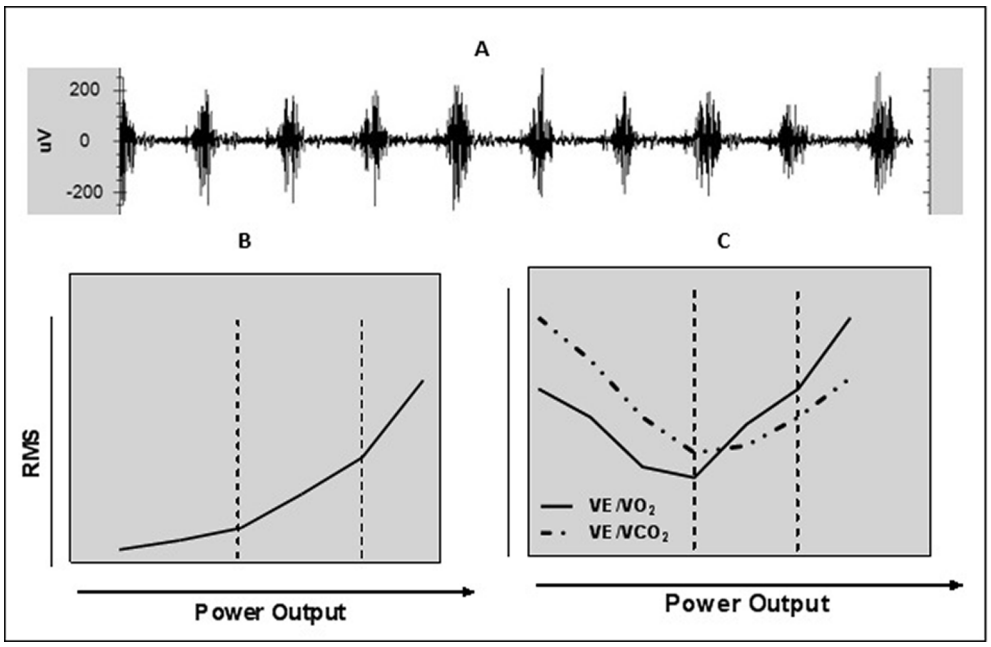

Fig. 1. Individual example of EMG signals recorded during the pedalling exercise (A), illustration of first and second EMG breakpoints (B) and first and second inflection in the VE/VO_2 curve (C).

least pooled residual sum of squares was chosen as representing the best fit. Thereafter, the program attempts to fit a third line to the data in order to detect another breakpoint in the EMG data. The third middle segment was obtained by methodically adding points on the left side of the two-line regression intersection point. The new regression line was then calculated and extended in the direction that yielded the lower sum of squares. Finally, an analysis of variance determined whether a significant ($p < 0.05$) reduction in the total sum of squares is achieved by adding a third line segment. The first and second EMG breakpoints were then reported as the first and second intersection points, respectively, of the computerized model. This method was previously validated in 20 subjects (Silva et al., 2018).

2.5. Statistical analysis

Data normality was verified using the Shapiro-Wilk test. The effect size was calculated by the T-test (difference between two dependent means) and post hoc analysis. The input parameters were total sample size, means and standard deviations, and error probability $\alpha = 0.05$. The effect size points were small = 0.2, medium = 0.5 and large = 0.8 (Cohen, 1992). The effect size was used to determine clinically significant differences (medium or large effect size was assumed as clinically significant). The relationship between values was tested using the Pearson's or Spearman's correlation tests according to Gaussian distribution. Agreement between variables was analyzed by the Bland-Altman test. A statistical significance value of $p \leq 0.05$ was set for all analyses. GraphPad Prism 7.0 and GPower 3.0.10 software programs were used. According to data normality distribution, the data are presented as means ± standard deviations or as medians and interquartile ranges and percentages.

3. Results

A total of 66 patients were enrolled, however 18 were excluded (comorbidities: asthma = 9, heart failure = 3, critical COVID-19 = 3, COPD = 2 and fibromyalgia = 1). Anthropometric characteristics, main COVID-19 symptoms and main drug therapy are presented in Table 1. Exercise characteristics are presented in Table 2.

The EMG analysis demonstrated a small effect size compared to the gas exchange analysis in oxygen uptake at the anaerobic threshold (effect size = 0.18, power = 0.33). The stratified data showed that this pattern was observed in mild (effect size = 0.14, power = 0.20) and severe (effect size = 0.36, power = 0.41) patients (Fig. 2). The EMG analysis also demonstrated a small effect size compared to the gas exchange analysis in oxygen uptake at the respiratory compensation point (effect size = 0.05, power = 0.12). The stratified data showed that this pattern was observed in mild (effect size = 0.05, power = 0.08) and severe (effect size = 0.10, power = 0.11) patients (Fig. 2).

The EMG analysis demonstrated a small effect size compared to the gas Exchange analysis in power output at the anaerobic threshold (effect size = 0.18, power = 0.34). The stratified data showed that this pattern

Table 1
Anthropometric characteristics, main COVID-19 symptoms and main drug therapy.

Variable	All (n = 48)	Mild (n = 31)	Severe (n = 17)
Age, years	43.9 ± 10.7	44.5 ± 10.9	42.3 ± 10.5
Weight, kg	79.3 ± 17.4	82.8 ± 16.8	73.1 ± 17.0
Height, cm	167.3 ± 10.1	160.0 ± 9.4	164.1 ± 10.6
Sex, M/F%	52/48	61/39	35/65
Symptoms, n (%)			
Fatigue	38 (79)	16 (52)	12 (70)
Muscle Soreness	25 (52)	12 (39)	13 (76)
Cough	25 (52)	15 (48)	10 (59)
Fever	21 (44)	10 (32)	11 (65)
Headache	8 (17)	5 (16)	3 (17)
Diarrhea	10 (21)	6 (19)	4 (23)
Nausea	6 (12)	4 (14)	2 (12)
Anosmia	23 (48)	14 (46)	9 (53)
Sore Throat	10 (21)	6 (19)	4 (23)
Dyspnea	17 (35)	0 (0)	17 (100)
Hospitalization	4 (8)	0 (0)	4 (23)
Drug therapy,n(%)			
Hydroxychloroquine	16 (33)	8 (26)	8 (47)
Antibiotics	38 (79)	23 (74)	15 (88)
Ivermectin	26 (54)	14 (45)	12 (70)
Zinc	18 (37)	8 (26)	10 (59)
Corticosteroids	16 (33)	16 (52)	6 (35)
Anticoagulants	6 (12)	4 (13)	2 (12)

M: male and F: female. Mean ± standard deviation.

Table 2
Exercise characteristics.

Variable	All (n = 48)	Mild (n = 31)	Severe (n = 17)
$VO_{2\ peak}$, L/min	1.66 [1.27–2.17]	1.88 [1.53–2.62]	1.26 [1.08–1.64]
$VO_{2\ peak}$, %predicted	81.9 ± 16.8	88.6 ± 15.9	69.8 ± 10.9
RER	1.17 [1.09–1.22]	1.16 [1.05–1.20]	1.18 [1.12–1.26]
O_2Pulse, mL/beat	11.0 [9.0–14.7]	12.6 [10.6–16.0]	8.0 [7.0–12.0]
VE, L/min	55.9 [44.6–71.9]	62.5 [47.5–77.7]	47.2 [42.7–55.9]
BR, %	42.0 [31.0–51.0]	43.0 [28.0–62.5]	40.0 [36.0–46.5]
Power Output, watts	123 [85–148]	129 [105–168]	81 [72–131]
RMS $_{peak}$	174 [141–225]	199 [159–230]	154 [103–174]
RMS EMG_1	105 [86–130]	117 [91–138]	87 ± 31
RMS EMG_2	156 [126–195]	173 [145–204]	129 ± 53
VO_2 AT, L/min	1.08 [0.82–1.33]	1.12 [0.98–1.40]	0.94 ± 0.23
VO_2 RCP, L/min	1.49 [1.13–2.00]	1.80 ± 0.58	1.22 ± 0.36
VO_2 EMG_1, L/min	0.99 [0.70–1.25]	1.17 [0.89–1.40]	0.85 ± 0.22
VO_2 EMG_2, L/min	1.46 [1.13–2.02]	1.78 ± 0.57	1.19 ± 0.36
$\%VO_{2\ peak}$ AT	64.5 ± 10.6	62.2 ± 11.3	68.8 ± 7.8
$\%VO_{2\ peak}$ RCP	84.2 [74.0–94.0]	91.0 [86.0–94.0]	88.8 ± 6.6
$\%VO_{2\ peak}$ EMG_1	60.4 ± 9.4	59.1 ± 9.6	62.6 ± 8.8
$\%VO_{2\ peak}$ EMG_2	85.0 [71.0–89.0]	89.0 [86.0–92.0]	86.1 ± 6.0
Power Output AT, watts	71 [51–93]	83 ± 27	52 [47–71]
Power Output RCP, watts	105 [71–127]	120 ± 38	68 [60–110]
Power Output EMG_1, watts	66 [50–88]	79 ± 25	49 [44–67]
Power Output EMG_2, watts	103 [71–132]	122 ± 39	71 [61–111]

VO_2: oxygen uptake, RER: respiratory exchange ratio, O_2pulse: oxygen pulse, VE: minute ventilation, BR: breathing reserve. RMS: root mean square, EMG_1: first EMG breakpoint, EMG_2: second EMG breakpoint, AT: anaerobic threshold, RCP: respiratory compensation point. Mean ± standard deviation. Median [interquartile range].

was observed in mild (effect size = 0.17, power = 0.24) and severe (effect size = 0.28, power = 0.30) patients (Fig. 2). The EMG analysis also demonstrated a small effect size compared to the gas exchange analysis in power output at the respiratory compensation point (effect size = 0.06, power = 0.09). The stratified data showed that this pattern was observed in mild (effect size = 0.08, power = 0.09) and severe (effect size = 0.10, power = 0.11) patients (Fig. 2).

The EMG and gas exchange analysis presented strong correlation in power output anaerobic threshold detection ($r = 0.97$, $p < 0.0001$). The stratified data also demonstrated strong correlation in both mild ($r = 0.97$, $p < 0.0001$) and severe ($r = 0.92$, $p < 0.0001$) patients (Fig. 3). There was a strong correlation in the power output respiratory compensation point detection between the EMG and gas exchange analysis ($r = 0.99$, $p < 0.0001$). The stratified data also demonstrated strong correlation in mild ($r = 0.99$, $p < 0.0001$) and severe ($r = 0.98$, $p < 0.0001$) patients (Fig. 3).

The Bland-Altman analysis demonstrated a bias = −4.7 W (SD = 6.2 W, limits of agreement = −16.9 to 7.6) for anaerobic threshold detection in the EMG analysis compared to the gas exchange analysis. The stratified data showed a bias = −4.2 W (SD = 6.5 W, limits of agreement = −16.9 to 8.5) for mild and a bias = −5.5 W (SD = 5.9 W, limits of agreement = −17.0 to 6.1) for severe patients (Fig. 4). The Bland-Altman analysis demonstrated a bias = −2.1 W (SD = 4.5 W, limits of agreement = −10.9 to 6.6) in the EMG analysis compared to the gas exchange analysis in the respiratory compensation point detection. The stratified data showed a bias = −1.9 W (SD = 4.8 W, limits of agreement = −11.4 to 7.6) for mild and a bias = −2.4 W (SD = 3.8 W, limits of agreement = −9.8 to 5.0) for severe patients (Fig. 4).

4. Discussion

The main finding of this study was that: 1) EMG analysis detects the anaerobic threshold and respiratory compensation point without clinical significant difference than gas exchange analysis; 2) EMG analysis presented good correlation and agreement in anaerobic threshold and respiratory compensation point detection compared to gas exchange analysis.

Houtz and Fischer (Houtz and Fischer, 1959) were the first to record surface electromyograms during pedaling in 1959, and since then numerous investigators have reported EMG analyses of pedaling for different purposes (Hug et al., 2004) (Duc et al., 2008) (Sarre et al., 2003). Some studies used visual methods to identify breakpoints in myoelectric signal response during incremental exercise ramp protocol. Hug et al. (2006) postulated that EMG activity of the vastus lateralis muscle presents a non-linear increase during incremental ramp-up cycling. The results of the Hug study suggest that determining EMG breakpoints can be used as a reliable method for studying neuromuscular fatigue during cycling exercise in this specific muscle, and better than in other leg muscles. Bearden and Moffatt (Bearden and Moffatt, 2001) demonstrated that the two breakpoints of the RMS/power output ratio coincided with the first and second ventilatory equivalent inflection points, as well as in the present study. Glass (Glass et al., 1998) analyzed rectus femoris and vastus lateralis electromyography during CPET, finding that VO_2 at EMG breakpoints was not significantly different from the gas exchange method (similar to our data).

Our results are in overall agreement with those of previous studies. Lucía (Lucía et al., 1997) evaluated vastus lateralis electromyography during CPET in cardiac transplant patients. They showed that EMG analysis presented strong correlation ($r = 0.89$, $p < 0.05$) with gas exchange analysis in detecting the anaerobic threshold. There was no evidence of a significant difference between methods when expressed as oxygen uptake (again, similar to our data). The data in Lucía's study also demonstrated that the first EMG breaking point occurred around 60% of maximum oxygen uptake, similar to our results. Unfortunately, they did not measure the second EMG breaking point. Zamunér et al. (2013a,b) evaluated vastus lateralis electromyography during CPET in sedentary middle-aged men. As in the present study, their data did not show significant differences in power output at the anaerobic threshold in the EMG and gas exchange methods.

Quantifying the breakpoints can be achieved by dynamically analyzing these variables, as their disproportion increases are relative to the cardiorespiratory adjustments necessary to supply the growing metabolic demand from increased motor unit recruitment. The EMG breakpoints may occur as a result of a change in the motor unit recruitment pattern from predominantly slow-twitch motor units to fast-twitch motor units, which could contribute to the accumulation of circulating lactate during exercise (Lucía et al., 1999) (Viitasalo et al., 1985). Lactic acid is produced during fast-twitch motor unit activation (glycolytic pathway utilization). The H^+ produced in cells as lactate must be immediately buffered upon its formation. Since HCO_3^- is a volatile buffer, the resulting H_2CO_3 does not remain in the cell, but leaves upon its formation as CO_2, thereby removing H^+ from the intracellular environment (Wasserman and Whipp, 1975). The ventilation normally increases at a rate required to remove CO_2 added to the capillary blood by metabolism while minimizing the increase in arterial H^+ concentration.

Vastus lateralis activity analysis is representative in prescribing thresholds for whole body activities. Jürimäe et al. (2007) demonstrated that the EMG breakpoint happens at similar workload in different lower extremity muscles (vastus lateralis, vastus medialis, biceps femoris and gastrocnemiuslateralis). An EMG breakpoint analysis in lower extremity

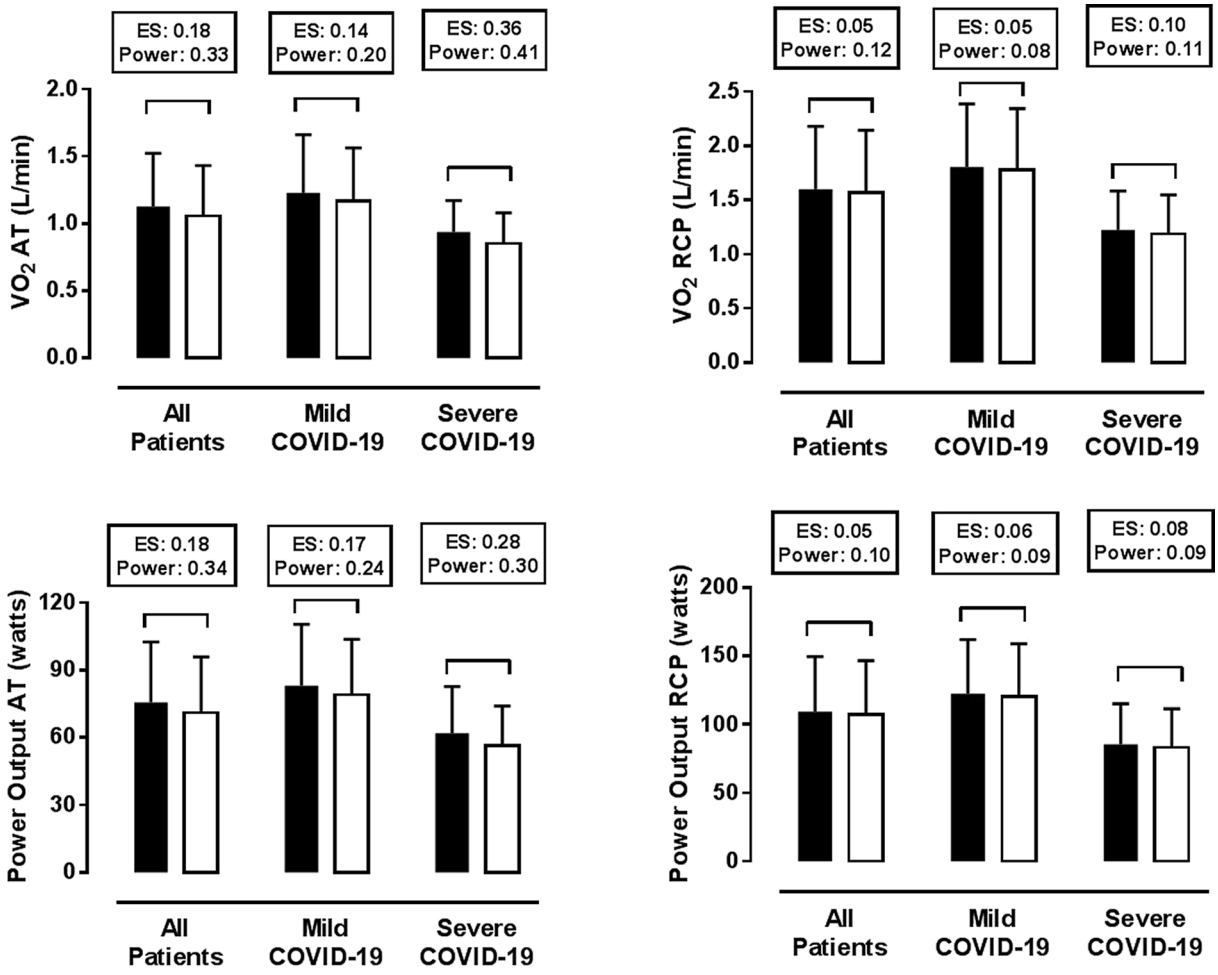

Fig. 2. Effect size of EMG (white bars) compared to gas exchange analysis (black bars). AT: anaerobic threshold. RCP: respiratory compensation point. VO_2: oxygen uptake. Means ± standard deviation.

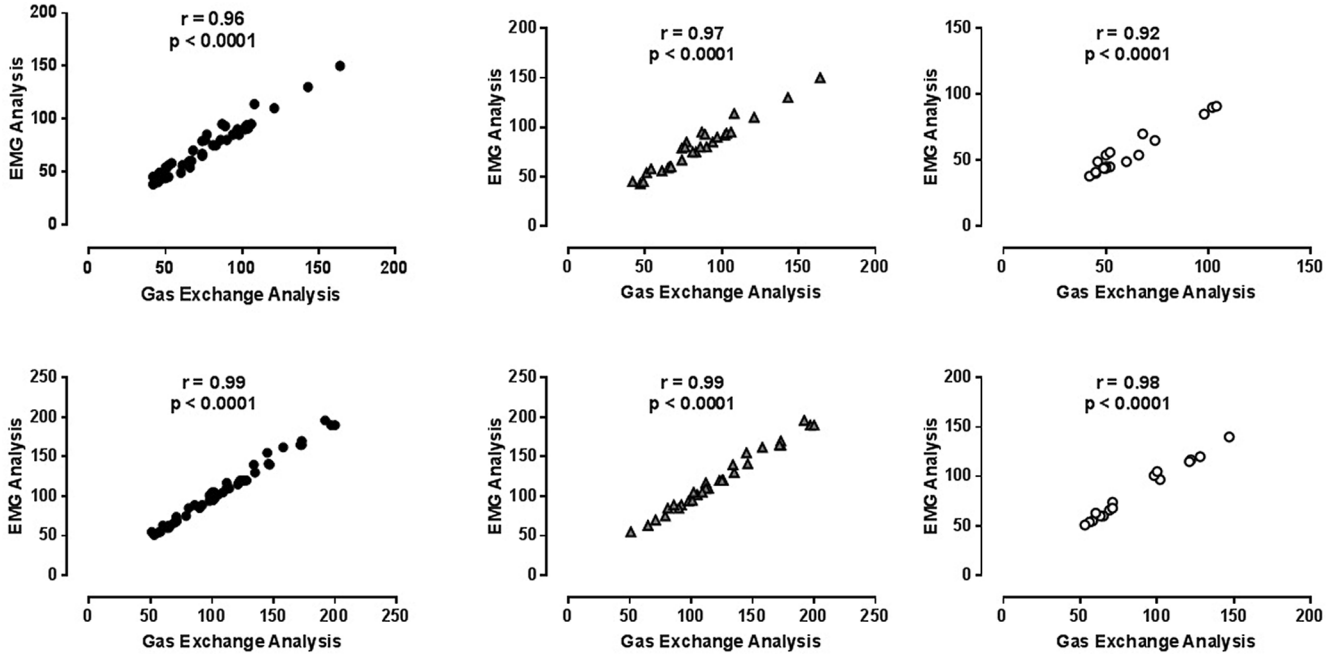

Fig. 3. Correlation in power output anaerobic threshold and respiratory compensation point detection. Black circles = all patients, grey triangles = mild COVID-19 and white circles = severe COVID-19.

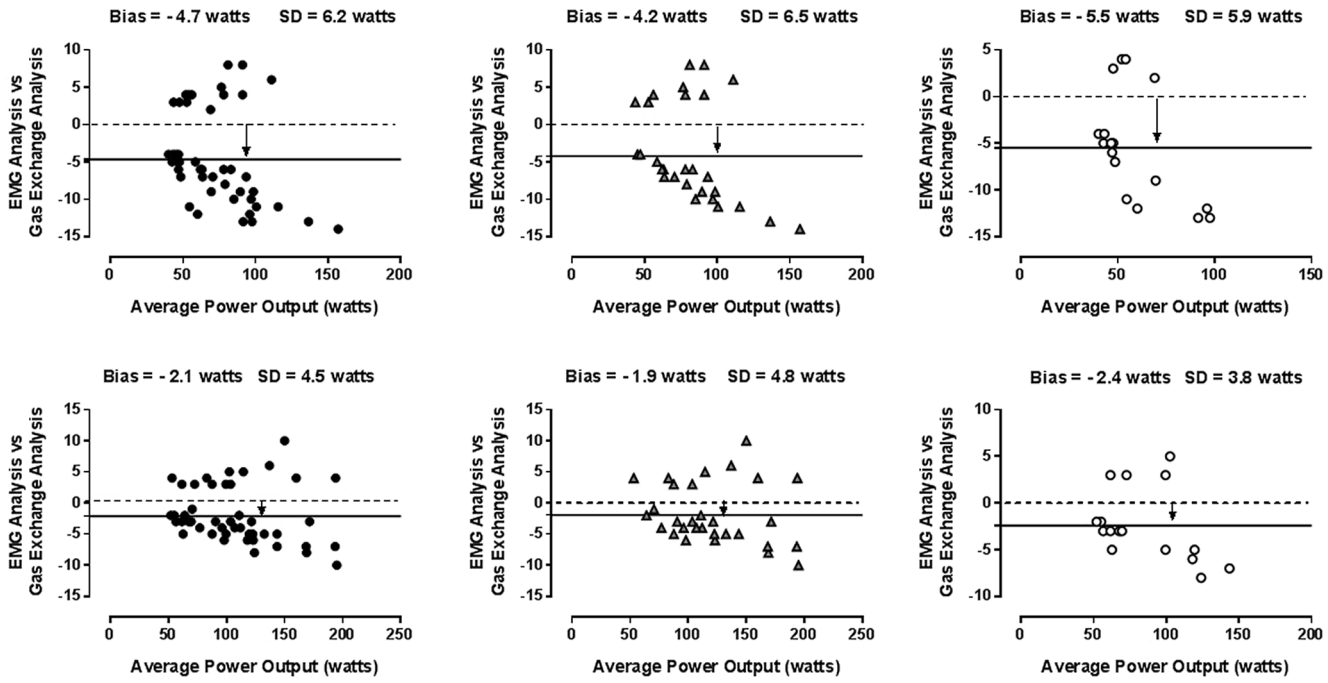

Fig. 4. Bland-Altman analysis for anaerobic threshold and respiratory compensation point detection in EMG compared to gas exchange analysis. Black circles = all patients, grey triangles = mild COVID-19 and white circles = severe COVID-19.

muscles can be done independently of the effort method or ergometer used. Tikkanen et al. (2012) demonstrated that the EMG analysis had similar a breakpoint pattern during treadmill running analysis as in bicycling. The timing of the EMG breakpoints (thresholds) was similar to ventilatory threshold and onset of blood lactate accumulation.

The power output bias in the Bland-Altman analysis of anaerobic threshold and respiratory compensation point detection in the present study was similar to the findings by Lucía (Lucía et al., 1999). This was probably due to "metabolic delay". EMG analyses fiber membrane depolarization, so it can detect muscle fiber activation in real time. Gas exchange measurements analyses the metabolic response to muscle fiber activation. VO_2 responds with linear first-order dynamics for power outputs with a time constant approximately equal to 25 to 35 s and a "delay" of 15 to 20 s (Whipp, 1987). This delay corresponds to the time required for oxygen to travel from the lungs to the muscles. VCO_2 responds with non-linear dynamics for heavy exercise power outputs. There is also a time constant and a time "delay". This delay corresponds to the time required for carbonic gas to travel from the muscles to the lungs.

The present study demonstrated strong correlation and good agreement in the Bland-Altman analysis of the EMG and gas exchange methods. It suggests that the use of EMG breakpoint method is a promising quantification tool for determining anaerobic threshold and respiratory compensation point for exercise prescription in recovered COVID-19 patients. It is important to say that changes in the response patterns of the cardiorespiratory and electromyographic variables were interlinked and interdependent.

In addition to the EMG breakpoint method constituting a promising tool, there are some barriers to implement it in real-life clinical practice. First, it is necessary to process the data to do a reliable analysis. This is time consuming and requires specific knowledge. Electromyography use is not widespread during the clinical training of health professionals, often limiting its use to the research (academic) world. Cultural changes would be necessary to reverse this situation.

Our study has some limitations; for example, *a priori* sample size calculation was not conducted. Gender distribution was different between severe and mild COVID-19 patients, however the data presented the same pattern in both genders. Furthermore, we did not have a non-COVID-19 control group (which would have enabled a clearer picture of the compromised respiratory function resulting from COVID-19 infection and its eventual relationship with neuromuscular response). Some factors may have influenced the EMG patterns during pedaling: 1) power output is directly related to EMG amplitude, so to avoid bias we used an isopower cycle ergometer; 2) the patients were instructed to maintain 60 rotations per minute in pedaling cadence, however some patients had difficulty to keep the precise cadence during all of the test; 3) body position also influences the EMG, and low saddle height increases the muscle activity level of the quadriceps. The patients selected saddle height according to pedaling comfort.

5. Conclusions

The EMG analysis detected the anaerobic threshold and respiratory compensation point without a clinically significant difference compared to the gas exchange analysis (gold standard method) in recovered COVID-19 patients. The EMG analysis has good correlation and agreement in detecting the anaerobic threshold and respiratory compensation point compared to the gold standard method. The present data shows the validity of EMG as a non-invasive determinant method for metabolic response to incremental exercise. This work could represent a starting point for future follow-up studies which could describe the medium and long-term effects of COVID-19 infection on respiratory and neuromuscular systems' functioning.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

ATS/ACCP Statement on Cardiopulmonary Exercise Testing, 2003. Am. J. Respir. Crit. Care Med. 167, 211–277.
Bearden, S.E., Moffatt, R.J., 2001. Leg electromyography and the VO2-power relationship during bicycle ergometry. Med. Sci. Sport. Exerc. 33, 1241–1245.
Cohen, J., 1992. A power prime. Psychol. Bull. 112, 155–159.

S Duc, W Bertucci, J N Pernin, F.G., 2008. Muscular activity during uphill cycling : Effect of slope , posture , hand grip position and constrained bicycle lateral sways. J Electromyogr Kinesiol 18, 116–127.

François Hug, S.D., 2009. Electromyographic analysis of pedaling : A review. J. Electromyogr. Kinesiol. 19, 182–198. https://doi.org/10.1016/j.jelekin.2007.10.010.

Glass SS , Knowlton RG, Sanjabi PB, S.J., 1998. Identifying the integrated electromyographic threshold using different muscles during incremental cycling exercise. J Sport. Med Phys Fit. 38, 47–52.

J E Hansen, D Y Sue, K.W., 1984. Predicted values for clinical exercise testing. Am Rev Respir Dis. 129, S49-55.

Henneman, E., Somjen, G.C.D., 1965. Functional significance of cell size in spinal motoneurons. J. Neurophysiol. 28, 560–580.

Hermens, H.J., Freriks, B., Merletti, R., Stegeman, D., Blok, J., Rau, G., Disselhorst-Klug, C., Hägg, G., 1999. European Recommendations for Surface ElectroMyoGraphy. Roessingh Res. Dev. 8–11 https://doi.org/10.1016/S1050-6411(00)00027-4.

S J HOUTZ, F.J.F., 1959. An analysis of muscle action and joint excursion during exercise on a stationary bicycle. J Bone Jt. Surg Am 41-A, 123–31.

Huang, G., Wang, R., Chen, P., Huang, S.C., Donnelly, J.E., Mehlferber, J.P., 2016. Dose – response relationship of cardiorespiratory fitness adaptation to controlled endurance training in sedentary older adults. Eur. J. Prev. Cardiol. 23, 518–529.

F Hug, D Laplaud, B Savin, L.G., 2003. Occurrence of electromyographic and ventilatory thresholds in professional road cyclists. Eur J Appl Physiol 90, 643–646. https://doi.org/10.1007/s00421-003-0949-5.

François Hug, David Bendahan, Yann Le Fur, Patrick J Cozzone, L.G., 2004. Heterogeneity of muscle recruitment pattern during pedaling in professional road cyclists : a magnetic resonance imaging and electromyography study. Eur J Appl Physiol 92, 334–342. https://doi.org/10.1007/s00421-004-1096-3.

F Hug, D Laplaud, A Lucia, L.G., 2006. EMG Threshold Determination in Eight Lower Limb Muscles During Cycling Exercise : A Pilot Study. Int J Sport. Med 27, 456–462. https://doi.org/10.1055/s-2005-865787.

Jürimäe, J., Von Duvillard, S.P., Mäestu, J., Cicchella, A., Purge, P., Ruosi, S., Jürimäe, T., Hamra, J., 2007. Aerobic – anaerobic transition intensity measured via EMG signals in athletes with di V erent physical activity patterns. Eur. J. Appl. Physiol. 101, 341–346.

Lucía A, Vaquero FA, Pérez M, Sánchez O, Sánchez V, Gómez MA, C.J., 1997. Electromyographic Response to Exercise in Cardiac Transplant Patients * A New Method for Anaerobic Threshold. Chest 111, 1571–1576.

Lucía, A., Sánchez, O., Carvajal, A., Chicharro, J.L., 1999. Analysis of the aerobic-anaerobic transition in elite cyclists during incremental exercise with the use of electromyography. Br. J. Sport. Med. 33, 178–185.

Meyer T., Gabriel H. H. W., K.W., 1999. Is determination of exercise intensities as percentages of VO2max or HRmax adequate. Med. Sci. Sports Exerc. 31, 1342–1345.

Xiaoneng Mo, Wenhua Jian, Zhuquan Su, Mu Chen, Hui Peng, Ping Peng, Chunliang Lei, Ruchong Chen, Nanshan Zhong, S.L., 2020. Abnormal pulmonary function in COVID-19. Eur Respir J 55.

Polastri, M., Nava, S., Clini, E., Gosselink, R., Vitacca, M., 2020. COVID-19 and pulmonary rehabilitation : preparing for phase three. Eur. Respir. J. 55, 19–21.

G Sarre, R Lepers, N Maffiuletti, G Millet, A.M., 2003. Influence of cycling cadence on neuromuscular activity of the knee extensors in humans. Eur J Appl Physiol 88, 476–479. https://doi.org/10.1007/s00421-002-0738-6.

Silva PE, Chedid R, Frazão M, Queiroz AV, Loiola-Jr AEA, Martins HR, A.M., 2018. Desenvolvimento e validação de um sistema para a determinação do limiar ventilatório e do ponto de compensação respiratória, por meio da eletromiografia de superfície. Assobrafir Ciência 9 (supl 1), 29–35.

Martjin A. Spruit, Anne E. Holland, Sally J. Singh, Thomy Tonia, Kevin C. Wilson, T.T., 2020. Early View Original article COVID-19 : Interim Guidance on Rehabilitation in the Hospital and Post-Hospital Phase from a European Respiratory Society and American Thoracic Society-coordinated International Task Force. Eur. Respir. J.

Tian, S., Hu, N., Lou, J., Chen, K., Kang, X., Xiang, Z., Chen, H., 2020. Characteristics of COVID-19 infection in Beijing. J. Infect. 80, 401–406.

Tikkanen, O., Hu, M., Vilavuo, T., Tolvanen, P., 2012. Ventilatory threshold during incremental running can be estimated using EMG shorts. Physiol. Meas. 33, 603–614.

Vandoni, M., Codrons, E., Marin, L., Correale, L., Bigliassi, M., Buzzachera, C.F., 2016. Psychophysiological Responses to Group Exercise Training Sessions : Does Exercise Intensity Matter? PLoS One 16, 1–13.

J T Viitasalo, P Luhtanen, P Rahkila, H.R., 1985. Electromyographic activity related t o aerobic and anaerobic threshold in ergometer bicycling. Acta Physiol Scand 124, 287–293.

Wasserman K, W.B., 1975. Excercise physiology in health and disease. Am Rev Respir Dis. 112, 219–249.

K Wasserman, W W Stringer, R Casaburi, A Koike, C.B.C., 1994. Determination of the anaerobic threshold by gas exchange biochemical considerations, methodology and physiological effects. Z Kardiol 83.

Whipp, B.J., 1987. Dynamics of pulmonary gas Exchange. Circulation 76, VI18-28.

Wolpern, A.E., Burgos, D.J., Janot, J.M., Dalleck, L.C., 2015. Is a threshold-based model a superior method to the relative percent concept for establishing individual exercise intensity ? a randomized controlled trial. BMC Sports Sci. Med. Rehabil. 7, 1–9.

Zamunér, A.R., Catai, A.M., Martins, L.E.B., et al., 2013b. Identification and agreement of first turn point by mathematical analysis applied to heart rate, carbon dioxide output and electromyography. Braz. J. Phys. Ther. 17, 614–622.

Antonio R. Zamunér, Aparecida M. Catai, Luiz E. B. Martins, Daniel I. Sakabe, E.D.S., 2013. Identification and agreement of first turn point by mathematical analysis applied to heart rate , carbon dioxide output and electromyography, Braz J Phys Ther 17, 614–622.