The “second wind” in McArdle’s disease patients during a second bout of constant work rate submaximal exercise

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ABSTRACT

Patients with McArdle’s disease (McA) typically show the “second wind” phenomenon, a sudden decrease in heart rate (HR) and an improved exercise tolerance occurring after a few minutes of exercise. In the present study we investigated whether in McA a first bout of exercise determines a second wind during a second bout, separated by the first by a few minutes of recovery.

Eight McA (44±4 years) and a control group of 6 mitochondrial myopathy patients (MM) (51±6 years) performed two repetitions (CWR1 and CWR2) of 6-min constant work rate exercise (~50% of peak work rate) separated by 6-min (SHORT) or 18-min (LONG) recovery. Pulmonary O2 uptake (VO2), HR, cardiac output (Q), rates of perceived exertion (RPE), vastus lateralis oxygenation (Δ[deoxy(Hb+Mb)], by near-infrared spectroscopy) were determined.

In McA VO2 (0.86±0.2 vs. 0.95±0.1 L·min⁻¹), HR (113±10 vs. 150±13 b·min⁻¹), Q (11.6±0.6 vs. 15.0±0.8 L·min⁻¹), RPE (11±2 vs. 14±3) were lower, whereas Δ[deoxy(Hb+Mb)] was higher (14.7±2.3 % vs. -0.1±4.6 %) in CWR2-SHORT vs. CWR1; the “overshoot” of Δ[deoxy(Hb+Mb)] and the “slow component” of VO2 kinetics disappeared in CWR2-SHORT. No differences (vs. CWR1) were observed in McA during CWR2-LONG, or in MM during both CWR2-SHORT and -LONG.

A second wind phenomenon was observed in McA during the second of two consecutive 6-min constant-work rate submaximal exercises. The second wind was associated with changes of physiological variables suggesting an enhanced skeletal muscle oxidative metabolism. The second wind was not described after a longer (18-min) recovery period.

KEY WORDS: myophosphorylase deficiency; exercise tolerance; VO2 kinetics slow component; near-infrared spectroscopy.

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INTRODUCTION

Patients with McArdle’s disease (McA) are affected by an autosomal recessive muscle glycogenosis (type V) caused by mutations in the gene that encodes muscle glycogen phosphorylase. Absence of activity of this enzyme blocks the breakdown of intramuscular glycogen and significantly impairs both substrate level phosphorylation from glycolysis and oxidative phosphorylation (44-46). The impairment of oxidative metabolism results in a reduced capacity to increase muscle O₂ extraction, or arterio-venous O₂ concentration difference ([C(a-v)O₂]), during exercise (14), and leads to a significantly lower than normal maximal (or peak) O₂ uptake (VO₂) (13). McA also present an exaggerated cardiovascular response to submaximal exercise, that is, higher heart rate (HR), cardiac output (Q), and muscle blood flow values, compared to healthy subjects, for the same submaximal VO₂ (19, 28,29,40,44-46,49), together with a markedly slower adjustment of VO₂ during constant work rate submaximal exercise (17).

A typical feature of McA is the “second wind” phenomenon (20,31). As first described by Pearson et al. (31), the second wind is characterized by the sudden decrease in HR and improvement of exercise tolerance after about 8 min of aerobic, dynamic, exercise (walking or cycling). According to Vissing and Haller (49), the second wind is pathognomonic for the disease and it is attributable to an enhanced sympatho-adrenal response and to an improved delivery of extramuscular energy substrates, free fatty acids and glucose, to working muscles, which partially compensates for the impaired glycogen breakdown (20). Other studies have demonstrated that the second wind can be induced by oral glucose (20).

No study has so far investigated if in McA a previous bout of exercise can induce a second wind phenomenon during a subsequent bout. This would be of interest also from a clinical point of view, considering that many activities of everyday life entail bouts of exercise separated by recovery periods. It could also allow patients to develop strategies (for example, having an exercise bout preceded by a “warm up” activity) which could increase their exercise tolerance. Moreover, in no previous studies the second wind phenomenon has been characterized in terms of variables intrinsically related to an enhanced skeletal muscle
oxidative metabolism and to an increased exercise tolerance, such as a reduced amplitude of
the “slow component” of the VO₂ kinetics (23) and a reduced O₂ cost of exercise.

Also in healthy subjects a vigorous “priming” or warm-up exercise can determine a reduced
amplitude of the slow component and an increased exercise tolerance during a subsequent
high-intensity exercise bout (4). The mechanism(s) responsible for this phenomenon may
comprise increased muscle O₂ availability, greater muscle oxidative enzyme activity and
carbon substrates supply, and altered motor unit recruitment profiles (7,18,21). Thus, at least
in part (see the increased carbon substrates supply) the mechanisms potentially responsible
for the “priming effect” in healthy humans could also be responsible for the second wind
phenomenon in McA. It should be noted, however, that the priming phenomenon does not
determine in healthy humans a lowering of HR (4), whereas lower HR (and Q) are
prominent effects in the second wind. In any case, in the present study the presence of a
second wind phenomenon during a second bout of exercise will be evaluated also in a control
group of patients affected by a mitochondrial myopathy (MM), who have similar exercise
tolerance of McA (8,14), but in whom a second wind phenomenon has never been
demonstrated. Thus, if the effects described during the second exercise bout would appear
only in McA, this would represent strong (although indirect) evidence that they are related to
a second wind phenomenon; on the other hand, if the effects would appear also in MM they
would likely be related to a priming effect.

In the present study we hypothesize that in McA, but not in MM, a preceding bout of constant
work rate submaximal exercise would determine, during a subsequent bout, a second wind
phenomenon. Apart from the hallmark index of increased exercise tolerance, represented by
lower rates of perceived exertion, more “traditional” signs of the second wind (lower HR,
lower Q, increased O₂ extraction) were sought after, together with other “ancillary” signs of
increased exercise tolerance (lower VE, lower R). In addition, we sought to determine if the
second wind was associated with a decrease or with a disappearance of the slow component
of the VO₂ kinetics, with a lower O₂ cost of exercise, with a decrease of transient unbalances
between O$_2$ delivery and O$_2$ utilization within skeletal muscles (as determined by NIRS [11,36,43]); these findings would point to an enhanced performance of skeletal muscle oxidative metabolism as one of the mechanisms of the second wind.

As a secondary aim, by arbitrarily choosing a “short” (6 minutes) and a “long” (18 minutes) recovery period between exercise bouts, we also tried to get insights (also for practical purposes) into the length of the recovery period which would allow the second wind phenomenon to manifest itself. Whereas 6 minutes represent a “standard” recovery between two 6-min exercise bouts (see e.g. 4), 18 minutes were arbitrarily chosen in order to represent a longer recovery, considering that prior exercise combined with an extended recovery period [$>15$ min (47)] might maximize the potential for exercise tolerance to be enhanced (4).

By applying on patients methods which have been developed in the exercise physiology laboratory, the present study will follow a classic translational approach, with the ultimate aim of increasing the exercise tolerance and the quality of life of the patients.

**METHODS**

**Subjects.** Eight McA and six MM were studied. Gender distribution, age, and body mass of McA were as follows: 3 males and 5 females, age (mean ± SD) 44 ± 4 yr and body mass 75.9 ± 8.9 kg. Gender distribution, age, and body mass of MM were as follows: 5 males and 1 female, age $51 \pm 6$ yr and body mass $69.1 \pm 7.4$ kg. Patients were from the Department of Neuromuscular Diseases, Neurological Institute “Carlo Besta” (IRCCS), Milano. The diagnosis of McA and MM was based on clinical, morphological, biochemical, and molecular evaluations. Clinical details of the patients were similar to those reported in our previous article (14). The degree of functional impairment varied from mild (no limitations in activities of everyday life but reduced exercise tolerance) to severe (very limited exercise tolerance, impairment in activities of daily living). Exclusion criteria were the presence of neoplastic and other major neurological/psychiatric, orthopedic, rheumathologic, endocrine, pulmonary, or cardiovascular disorders. The subjects were fully informed of any risk and discomfort associated with the experiments before giving their written consent to participate to the study, which was approved by the ethics committees of the involved institutions.
procedures were in accordance with the recommendations found in the Declaration of Helsinki (2000) of the World Medical Association.

**Measurements.** Experiments were conducted in the morning, a few hours (at least 2 hr) after a light breakfast (about 600 kcal, 35 % fat, 55 % carbohydrate and 10 % protein). Patients were not following any specific diet. All tests were carried out under medical supervision. Subjects were monitored by 12-lead ECG. An electromagnetically braked cycle ergometer (Corival; Lode BV, Groningen, The Netherlands) was used. Pedaling frequency was digitally displayed to the subjects. Subjects were allowed time to gain familiarity with the investigators and the experimental arrangement, were carefully instructed about the procedures, and were familiarized with the protocol using short practice runs.

On the first day the subjects performed an incremental test up to voluntary exhaustion to assess peak $O_2$ uptake ($V_0_2$ peak). After three minutes of unloaded pedaling, exercise was conducted at 25-50 W for 5 minutes, and thereafter the work rate was increased by 5-15 W (according to the subject’s estimated level of physical fitness) every minute. The exhaustion was defined by: (1) inability to maintain the pedaling frequency despite encouragement by the operators; (2) maximal levels of self-perceived exertion, using the validated Borg’s scale; and (3) heart rate (HR) values >85 % of the age-predicted maximum. Values of cardiovascular, ventilatory, gas exchange, and muscle oxygenation variables determined during the last 30 seconds of the exhausting load were considered “peak” values.

During the second and the third days, the patients performed two repetitions of subsequent 6-min constant work rate submaximal exercise (CWR1 and CWR2) (at a work rate corresponding to ~50 % of peak work rate); in the first case CWR1 and CWR2 were separated by a 6-min recovery period (SHORT), whereas and in the second case, (after observing at least two hours of rest) CWR1 and CWR2 were separated by a 18-min recovery period (LONG). Pedaling frequency was kept at ~60 rpm and transitions from rest to the imposed load were attained in ~3 s.
Pulmonary ventilation (VE, in BTPS), O₂ consumption (VO₂), and CO₂ output (VCO₂), both in STPD, were determined breath-by-breath by a metabolic cart (Vmax29c; SensorMedics, Bilthoven, The Netherlands). Expiratory flow was determined by a mass flow sensor (hot wire anemometer). VO₂ and VCO₂ were determined by continuously monitoring PO₂ and PCO₂ at the mouth throughout the respiratory cycle and from established mass balance equations. Gas exchange ratio (R) was calculated as VCO₂/VO₂. HR was determined from the ECG signal. Stroke volume (SV) was estimated beat-by-beat by impedance cardiography (Physio Flow; Manatec, Paris, France). The accuracy of this device has been previously evaluated, in healthy subjects, during incremental exercise on a cycle ergometer, and was found to be “acceptable” (38). Cardiac output (Q) was calculated as HR·SV. Systemic peak arterial-venous O₂ concentration difference ([C(artery)-O₂]/Q) was calculated as VO₂peak/Qpeak. At rest and at various times (1, 3, and 5 min) during recovery, 20 μL of capillary blood was obtained from a preheated earlobe for the determination of blood lactate concentration ([La]b) by an enzymatic method (Biosen 5030; EKF, Eppendorf Italia, Milano, Italy).

Oxygenation changes in the vastus lateralis muscle were evaluated by NIRS (5,10). A portable NIRS single-distance continuous-wave photometer (HEO-100; Omron, Kyoto, Japan), which adopts an algorithm based on diffusion theory (42), was utilized. The instrument provides separate measurements of changes in deoxygenated Hb and Mb concentrations, as well as of oxygenated Hb and Mb concentrations, expressed in arbitrary units. Details on the method can be found in previous studies from our group (15,27,36). Concentration changes of oxygenated Hb + Mb (Δ[oxy(Hb+Mb)]) and deoxygenated Hb + Mb (Δ[deoxy(Hb+Mb)]), with respect to an initial value arbitrarily set equal to zero, were calculated and expressed in arbitrary units. The sum of the two variables (Δ[oxy(Hb+Mb) + deoxy(Hb+Mb)]) is related to changes in the total Hb volume in the muscle region of interest (6,12,25). Δ[deoxy(Hb+Mb)] was taken as a “deoxygenation index”, because this variable is relatively insensitive to changes in blood volume (6,25). Δ[deoxy(Hb+Mb)] was considered an estimate of skeletal muscle fractional O₂ extraction, that is of the ratio between O₂ consumption and O₂ delivery (11,15). Δ[deoxy(Hb+Mb)] data were expressed as a percentage of the values determined.
after the exercise (at least 10 min) by obtaining a maximal deoxygenation of the muscle, by pressure cuff inflation (at about 300 mmHg) at the root of the thigh (subject in the sitting position on the cycloergometer), for a few minutes until the $\Delta[\text{deoxy(Hb+Mb)}]$ increase reached a plateau.

**Kinetics analysis.** $\dot{V}O_2$ kinetics were evaluated during transitions from rest to CWR. Breath-by-breath $\dot{V}O_2$ values obtained during the repetitions of the exercises were time aligned and then superimposed for each subject. Average $\dot{V}O_2$ values every 10 s were calculated. Data obtained during the first 20 s of the transition ("cardiodynamic" phase) (37) were excluded from analysis. Thus, $\dot{V}O_2$ kinetics analysis focused on the "phase 2" (or "fundamental" component) of the response, which more closely reflects gas exchange kinetics occurring in skeletal muscles (16,34,50).

To mathematically evaluate the $\dot{V}O_2$ kinetics, data were first fitted by a monoexponential function of the type:

$$y(t) = y_{BAS} + A_f [1 - \exp^{-\left(\frac{x - TD_f}{\tau_f}\right)}]$$  \hspace{1cm} (Eq. 1)

where $y_{BAS}$ indicates the $\dot{V}O_2$ value at baseline; $A_f$ the amplitude of the $\dot{V}O_2$ response calculated between the baseline value and the steady-state value for the fundamental component; $TD_f$ is the time delay, and $\tau_f$ the time constant of the function for the fundamental component.

To check the presence of a slow component of the kinetics (21), data were also fit by a double exponential function of the type:

$$y(t) = y_{BAS} + A_f [1 - \exp^{-\left(\frac{x - TD_f}{\tau_f}\right)}] + A_s [1 - \exp^{-\left(\frac{x - TD_s}{\tau_s}\right)}]$$  \hspace{1cm} (Eq. 2)

where $A_s$, $TD_s$, and $\tau_s$ indicate, respectively, the amplitude, the time delay, and the time constant of the slow component of the kinetics.
Sometimes, after the first exponential rise, $\dot{V}O_2$ increases linearly without reaching a steady-state value. In this case, Eq. 2 does not provide a good fit of data. Thus, a third equation was also utilized, with an exponential function for the fundamental component and a linear function for the slow component (exponential + linear fitting) (38):

$$y(t) = y_{BAS} + Af[1 - \exp^{-(t - TD_f)/T_f}] + S(x-TDs)$$

(Eq. 3)

where $S$ (slope) is the angular coefficient of the linear regression of $\dot{V}O_2$ vs. time.

The equation that best fit the experimental data was determined by the F-test. That is to say, when Eq. 2 or Eq. 3 provided a better fit of the data, a slow component (46) of $\dot{V}O_2$ kinetics was present, superimposed on the fundamental component. The actual amplitude of the slow component ($A$'s) was estimated as the difference between the average $\dot{V}O_2$ value obtained during the last 20–30 s of CWR and the asymptotic value of the fundamental component (15,38). The percentage contribution of the slow component to the total amplitude of the response ($A$/Atot) was also calculated (36).

**Statistical analysis.** Results were expressed as mean values ± standard deviation (x ± SD). The statistical significance of differences between two means was checked by Student’s t-test (two tailed, unpaired analysis). The effects of the “warm-up” exercise bout (CWR2 vs. CWR1) and of the group (McA vs. MM) on the investigated variables were checked by two-way analysis of variance (ANOVA). This analysis, however, did not yield a statistically significant difference for $\dot{V}O_2$ and $\dot{V}E$. This is likely attributable to the relatively number of patients in the two groups (McA and MM are rare diseases [see also the recent commentary by Ploutz-Snyder *J Appl Physiol* in press]). Thus, analysis of differences between CWR1 and CWR2 in MM and McA was also performed by one-way ANOVA. A Tukey’s post hoc test was utilized when significant differences emerged upon ANOVA. Data fitting by linear regression or exponential functions was performed by the least squared residuals method. Comparisons between fittings with different exponential models were performed by F test.
The level of significance was set at P<0.05. Statistical analyses were performed by a software package (Prism 5.0; GraphPad, San Diego, CA).

**RESULTS**

**Incremental exercise.** Peak values are shown in Table 1. Values were very similar to those obtained in McA and MM in a previous study by our group (14) and by others (19,21). $\dot{V}O_2$ peak was ~50% of that usually obtained in healthy age-matched subjects (30), indicating a severely reduced maximal aerobic power. HR values, significantly higher in McA than in MM, corresponded to ~96% of the age–predicted maximum. $Q$ peak values were only slightly lower than those usually obtained in healthy controls (39). As expected for both patients groups, $[C(a-v)O_2]$ and peak skeletal muscle fractional $O_2$ extraction values were very low. As expected, in McA $R$ peak values were relatively low, and $[La]b$ peak values were not higher than those determined at rest (1.2 ± 0.1 mM). For the other variables no differences were observed between McA and MM.

**Constant work rate exercises.** Figure 1 shows typical examples of HR time-courses of a MM (upper panels) and of a McA (lower panels) during SHORT (left panels) and LONG (right panels). In McA, during SHORT (but not during LONG) HR values at the end of CWR2 were markedly lower (by about 50 beats·min⁻¹) than during CWR1. This second wind phenomenon is indicated by the arrow. No differences between CWR1 and CWR2 were observed in MM, either during SHORT and LONG.

Mean (± SD) values determined in the last ~30 s of CWR1 and CWR2 (SHORT and LONG recovery) are presented in Table 2. In McA during SHORT $VO_2$, $\dot{V}CO_2$, $VE$, $R$, HR, $Q$ and RPE values were significantly lower in CWR2 vs. CWR1. On the other hand, $[La]b$, $[C(a-v)O_2]$ and $\Delta[deoxy(Hb+Mb)]$ were significantly higher in CWR2 vs. CWR1. No significant differences were observed between CWR1 and CWR2 in LONG. As for MM, no significant differences were observed between CWR1 and CWR2, both in SHORT and in LONG.
\( \dot{V}O_2 \) and \( \Delta[\text{deoxy(Hb+Mb)}] \) kinetics. Typical individual examples of \( \dot{V}O_2 \) kinetics of a MM (upper panels) and of a McA (lower panels) during SHORT (left panels) and LONG (right panels) are shown in Figure 2. As for McA, \( \dot{V}O_2 \) values did not reach a steady-state and a slow component was evident in CWR1. During CWR2 the slow component disappeared in SHORT, but not in LONG. This second wind phenomenon is indicated by the arrow. A slow component was not observed in MM, both during CWR1 and CWR2 (SHORT and LONG).

The parameters of \( \dot{V}O_2 \) kinetics are shown in Table 3. In both groups TD, \( \tau_f \) and \( A_f \) values were not significantly different in CWR1 vs. CWR2 (both in SHORT and in LONG). Gain values (G) were calculated as \( \Delta \dot{V}O_2 \) (\( \dot{V}O_2 \) at the end of CWR minus resting \( \dot{V}O_2 \)) divided by work rate. A slow component, corresponding to \( \sim 15 \% \) of the total amplitude of the response, was present in all McA in CWR1. In six McA the slow component was best described by a linear function (Eq. 3). In CWR2-SHORT, but not in CWR2-LONG, the slow component disappeared. No slow component was evident in any MM. In McA \( A'_s, A'_s/A_{tot} \) and G values were significantly lower in CWR2 vs. CWR1 in SHORT, but not in LONG. In MM no differences were observed for G values in CWR1 vs. CWR2 (both in SHORT and in LONG). In both groups of patients G values were substantially higher that those usually observed in normal subjects (\( \sim 10 \text{ ml} \cdot \text{min}^{-1} \cdot \text{watt}^{-1} \)), independently from the presence of the slow component.

\( \Delta[\text{deoxy(Hb+Mb)}] \) kinetics are shown in Figure 3. In MM, in all conditions there was an initial and transient increase (“overshoot”) of \( \Delta[\text{deoxy(Hb+Mb)}] \) (occurring after \( \sim 45 \) s of exercise), which was followed by a steady state. \( \Delta[\text{deoxy(Hb+Mb)}] \) values at the peak of the overshoot were significantly higher than at steady state, for both CWR1 (24.9 \( \pm 5.1 \) % vs. 6.2 \( \pm 3.0 \) %) and CWR2 (21.9 \( \pm 4.5 \) % vs. 6.0 \( \pm 5.4 \) % and 21.5 \( \pm 5.4 \) % vs. 6.9 \( \pm 3.5 \) %, respectively in SHORT and LONG). In McA values at the peak of the overshoot were higher than those at steady state during CWR1 (27.5 \( \pm 6.0 \) % vs. -0.1 \( \pm 4.6 \) %) and during CWR2-LONG (24.9 \( \pm 6.7 \) % vs. 1.9 \( \pm 1.0 \) %), whereas in CWR2-SHORT no decrease of the variable was observed after the initial increase (no overshoot was described).
DISCUSSION

We observed in McA, during the second (CWR2) of two 6-min constant work rate exercises, carried out at ~50 % of peak work rate, and separated by 6 minutes of recovery (SHORT), significant changes indicating an improved exercise tolerance and an enhanced oxidative metabolism, such as lower (vs. the first exercise bout [CWR1]) RPE, HR, Q, R, VE, the disappearance of the slow component of VO₂ kinetics and a reduced O₂ cost of exercise, a slightly increased skeletal muscle fractional O₂ extraction and the disappearance of signs of transient unbalance between O₂ delivery and O₂ utilization within skeletal muscles (overshoot). No differences between CWR1 and CWR2 were described when the recovery period was extended to 18 minutes (LONG).

Can the differences mentioned above be considered an expression of a “second wind phenomenon” (1,2,20,46,49), or could they be simply related to a “warm up” or “priming” effect of the first exercise bout, as described also in healthy subjects (see e.g. 4), substantially in terms of a reduced amplitude of the slow component? The answer to this question is not straightforward, but several pieces of evidence appear in favor of a second wind phenomenon. The profound changes described in the present study in McA during CWR2-SHORT, such as the disappearance of the slow component of VO₂ kinetics, the substantially lower VE, HR, Q, RPE, etc., and the slightly higher fractional O₂ extraction, appear qualitatively and quantitatively quite different from those usually observed in healthy subjects following a priming exercise. Just to make an example, in McA HR values were on average 37 beats ` min⁻¹ lower during CWR2-SHORT vs. CWR1, whereas the priming effect does not usually affect HR values in healthy subjects (see e.g. 4). Moreover, in the “control” population represented by MM, which has a similar exercise tolerance compared to McA (see also 14,45) but do not manifest any second wind phenomenon, no differences were observed in CWR2-SHORT vs. CWR1. In any case, independently from the definition which is given to the phenomenon, our data demonstrate that in McA a first bout of exercise affects several cardiovascular, ventilatory and metabolic variables, enhances skeletal muscle oxidative...
metabolism and substantially improves exercise tolerance during a subsequent bout carried out a few minutes after the first. The finding has an obvious clinical interest.

The second wind is usually attributed to an improved delivery of extramuscular energy sources, particularly glucose, to working muscles, following an enhanced sympathoadrenal response (20). The phenomenon has been previously demonstrated in McA patients during prolonged exercise (20,49) or after sucrose administration (1,2,20), and is considered pathognomonic for the disease (49). The second wind has been described in literature as a lower HR (20), lower RPE (48), increased [C(a-v)O2] (20) and increased [La]b (20) during submaximal exercise, or increased peak work rate and peak VO2 (20).

Our results demonstrate that the second wind is also characterized by changes of other physiological variables clearly related to exercise tolerance, such as the slow component of VO2 kinetics and the related O2 cost of exercise (23). As shown in Figure 2 (see CWR1 in McA), the slow component determined a progressive increase in VO2 during constant work rate exercise, suggesting a progressive increase in the O2 cost of exercise, which is directly related with fatigue (23). Our group has recently demonstrated (41) that in obese patients the slope of the slow component is inversely related to the time of exhaustion. In the present study the slow component was eliminated in CWR2-SHORT.

In our study the enhanced exercise tolerance observed in McA during CWR2-SHORT vs. CWR1 was associated with a slightly but significantly increased skeletal muscle fractional O2 extraction (as determined by NIRS), confirming the data obtained by different methods by Haller & Vissing (20). The data demonstrate that the second wind partially corrects the impairment of oxidative metabolism which is one of the pathophysiological hallmarks of the disease (14,17,20,45,46). Skeletal muscle fractional O2 extraction in McA, however, remained quite lower than that usually described in healthy subjects (36), as well as in other populations in which skeletal muscle oxidative metabolism is known to be impaired, such as ageing subjects (13), subjects exposed to bed-rest deconditioning (36) or in patients such as heart transplant recipients (27).
In McA the “overshoot” of the $\Delta[\text{deoxy(Hb+Mb)}]$ kinetics, which was evident during CWR1, disappeared in CWR2-SHORT (but not in CWR2-LONG). According to Ferreira et al. (11), the overshoot is a sign of a relatively inadequate muscle $O_2$ delivery vs. muscle $\text{VO}_2$, and could lead to a reduced microvascular $O_2$ pressures and to a lower blood-to-myocyte “driving force” for peripheral $O_2$ diffusion. The overshoot phenomenon, which suggests an impaired intramuscular matching between $O_2$ delivery and $O_2$ utilization, was observed in the present study also in MM, and in previous studies in subjects undergoing bed rest deconditioning (36) and in patients with chronic heart failure (43). In the present study the overshoot disappeared during CWR2-SHORT in McA, but not in MM; this suggests that an improved intramuscular matching between $O_2$ delivery and $O_2$ utilization is likely associated with the second wind phenomenon. The possible mechanisms underlying the impaired intramuscular matching between the mentioned variables are discussed in detail in Poole et al. (35), and seem to be related to nitric oxide bioavailability. Also this component of the second wind phenomenon was no longer present after 18 minutes of recovery (CWR2-LONG).

In the present study the work rate of CWR1 and CWR2 cannot be clearly characterized as “moderate” or “heavy” or “severe” (50). As was the case with previous authors (9), in our McA patients we could not determine the gas exchange threshold (GET). It should be remembered that these patients are characterized by the absence of any blood lactate increase during exercise, even at exhaustion, as a consequence of the “blocked” glycogenolysis. GET is usually utilized to discriminate between “moderate” exercise (below GET, with no slow component of $\text{VO}_2$ kinetics) and “heavy” exercise (above GET, with a slow component which eventually reaches a steady state). In normal subjects, exercises in which the slow component does not reach a steady-state and $\text{VO}_2$ keeps increasing as a function of time during the constant work-rate exercise (as in McA during CWR1, see Fig. 2), until $\text{VO}_2$ peak is reached and fatigue ensues, are considered to be in the “severe” exercise domain, above the “critical power” (23). Thus, for McA of the present study the exercise could be defined as “severe” in CWR1 and “moderate” in CWR2-SHORT (24,52).
In conclusion, in the present study carried out on McA patients we demonstrated, for the first time, a “second wind” phenomenon during the second of two consecutive submaximal 6-min constant work rate exercises, separated by 6 minutes of recovery. The second exercise was indeed characterized by significantly lower (compared to the first exercise bout) rate of perceived exertion, heart rate, cardiac output, pulmonary ventilation, gas exchange ratio, and by slightly higher skeletal muscle fractional O₂ extraction. For the first time we also demonstrated that the second wind was associated with signs of enhanced skeletal muscle oxidative metabolism such as the disappearance of slow component of pulmonary VO₂ kinetics (and therefore with a lower VO₂ and a lower O₂ cost of exercise), and the disappearance of signs of transient mismatch between O₂ delivery and O₂ utilization in skeletal muscle. We did not observe the second wind phenomenon when the recovery period between the two exercise bouts was longer (18 minutes).

Considering that many activities of everyday life are characterized by bouts of exercise separated by recovery periods, the present results appear of interest also from a clinical and practical point of view. They also give a scientific background to strategies which are often already empowered by McA patients in order to increase their exercise tolerance: for example, having an exercise bout preceded by a few minutes by a “warm up” activity. By following a classic translational approach, the present study applied on patients methods which have been developed in the exercise physiology laboratory, with the ultimate aim of increasing their exercise tolerance and quality of life.
CONFLICT OF INTEREST
The authors declare they do not have conflict of interests.

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REFERENCES


FIGURE LEGENDS

FIGURE 1. Typical individual examples of heart rate (HR) kinetics during the first (CWR1) and the second (CWR2) constant work rate exercise in MM (upper panels) and McA (lower panels). Data obtained with 6 (SHORT) or 18 minutes (LONG) of recovery are presented in the left and right panels, respectively. The vertical hatched lines indicate the transitions from rest to the imposed work rate, and from the imposed work rate to recovery. The second wind phenomenon is indicated by the arrow. See text for further details.

FIGURE 2. Typical individual examples of pulmonary O2 uptake (\(\dot{V}O_2\)) kinetics during the first (CWR1) and the second (CWR2) constant work rate exercise in MM (upper panels) and McA (lower panels). Data obtained with 6 (SHORT) or 18 minutes (LONG) of recovery are presented in the left and right panels, respectively. The vertical hatched lines indicate the transitions from rest to the imposed work rate, and from the imposed work rate to recovery. The functions fitting the fundamental component (continuous line) and the slow component (hatched lines) are shown. A’s/Atot data are also presented. The second wind phenomenon is indicated by the arrow. See text for further details.

FIGURE 3. Mean (± SD) values every second of skeletal muscle fractional O2 extraction (\(\Delta[\text{deoxy(Hb+Mb)}]\)) during the first (CWR1) and the second (CWR2) constant work rate exercise in MM (upper panels) and McA (lower panels). Data obtained with 6 (SHORT) or 18 minutes (LONG) of recovery are presented in the left and right panels, respectively. \(\Delta[\text{deoxy(Hb+Mb)}]\) data are expressed as a percentage of \(\Delta[\text{deoxy(Hb+Mb)}]\) changes during ischemia. The vertical hatched lines indicate the transitions from rest to the imposed work rate, and from the imposed work rate to recovery. The second wind phenomenon is indicated by the arrow. See text for further details.
TABLES

**Table 1.** Peak values of the main cardiovascular, ventilatory and metabolic variables in McA and MM. Rates of perceived exertion and blood lactate levels are also shown.
<table>
<thead>
<tr>
<th>Patient</th>
<th>Work rate</th>
<th>RPE</th>
<th>V\textsuperscript{O}\textsubscript{2}</th>
<th>V\textsuperscript{O}\textsubscript{2}</th>
<th>V\textsuperscript{CO}\textsubscript{2}</th>
<th>R</th>
<th>V\textsuperscript{E}</th>
<th>Vt</th>
<th>fR</th>
<th>PetO\textsubscript{2}</th>
<th>PetCO\textsubscript{2}</th>
<th>[La]b</th>
<th>HR</th>
<th>SV</th>
<th>Q'</th>
<th>[C(a-v)\textsubscript{O}2]</th>
<th>Δ[deoxygenated(Hb +Mb)]</th>
</tr>
</thead>
<tbody>
<tr>
<td>McA</td>
<td>78.6 ± 18.5</td>
<td>16.6 ± 0.8</td>
<td>1.33 ± 0.25</td>
<td>18.5 ± 2.9</td>
<td>1.22 ± 0.21</td>
<td>0.93 ± 0.1#</td>
<td>47.0 ± 6.8</td>
<td>1.61 ± 0.2</td>
<td>29.4 ± 2.8</td>
<td>113.4 ± 2.2</td>
<td>29.8 ± 1.0</td>
<td>1.2 ± 0.1#</td>
<td>161.6 ± 3.4#</td>
<td>103.2 ± 9.7</td>
<td>17.5 ± 1.7</td>
<td>7.6 ± 0.9</td>
<td>20.3 ± 8.4</td>
</tr>
<tr>
<td>MM</td>
<td>71.7 ± 11.2</td>
<td>15.6 ± 0.7</td>
<td>1.08 ± 0.2</td>
<td>15.5 ± 1.1</td>
<td>1.29 ± 0.2</td>
<td>1.21 ± 0.1</td>
<td>52.4 ± 10.1</td>
<td>1.60 ± 0.2</td>
<td>31.8 ± 2.3</td>
<td>118.7 ± 2.5</td>
<td>30.7 ± 1.7</td>
<td>5.8 ± 0.8</td>
<td>149.9 ± 8.1</td>
<td>109.4 ± 7.5</td>
<td>16.3 ± 1.2</td>
<td>6.4 ± 0.7</td>
<td>20.1 ± 4.6</td>
</tr>
</tbody>
</table>
Mean (± SD) Values. $\dot{V}O_2$, oxygen uptake; $\dot{V}CO_2$, CO$_2$ output; R, gas exchange ratio; VE, pulmonary ventilation; Vt, tidal volume; fR, breathing frequency; PetO$_2$, end-tidal O$_2$ partial pressure; PetCO$_2$, end-tidal CO$_2$ partial pressure; [La]b, blood lactate concentration; RPE, rate of perceived exertion; HR, heart rate; SV, stroke volume; $\dot{Q}$, cardiac output; [C$_{(a-v)}$O$_2$], systemic arterial-venous O$_2$ concentration difference; $\Delta[\text{deoxy(Hb+Mb)}]$, muscle oxygenation index obtained by NIRS. #P < 0.05, significantly different from the corresponding value obtained in MM. See text for further details.
Table 2. Values of the main cardiovascular, ventilatory and metabolic variables in CWR1 and CWR2 in McA and MM. Data obtained with 6 (SHORT) or 18 minutes (LONG) of recovery are shown.
<table>
<thead>
<tr>
<th></th>
<th>Work rate</th>
<th>RPE</th>
<th>V'O₂</th>
<th>V'CO₂</th>
<th>R</th>
<th>V'E</th>
<th>[La]b</th>
<th>HR</th>
<th>SV</th>
<th>Q'</th>
<th>[C(a-v)O₂]</th>
<th>Δ[deoxy(Hb+Mb)]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Watt</td>
<td>L min⁻¹</td>
<td>L min⁻¹</td>
<td>L min⁻¹</td>
<td>mM</td>
<td>b min⁻¹</td>
<td>mL</td>
<td>mL min⁻¹</td>
<td>mL O₂ 100 mL⁻¹</td>
<td>% of ischemia</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>McA</strong></td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>CWR1 SHORT</td>
<td>41.0 ± 14.0</td>
<td>13.9 ± 2.6</td>
<td>0.95 ± 0.11</td>
<td>0.92 ± 0.14</td>
<td>0.93 ± 0.02</td>
<td>36.2 ± 3.0</td>
<td>0.8 ± 0.2</td>
<td>150 ± 13</td>
<td>102.6 ± 6.5</td>
<td>15.0 ± 0.8</td>
<td>6.7 ± 0.6</td>
<td>-0.1 ± 4.6</td>
</tr>
<tr>
<td>CWR2 SHORT</td>
<td>41.0 ± 14.0</td>
<td>10.8 ± 1.7*</td>
<td>0.86 ± 0.15*</td>
<td>0.81 ± 0.14*</td>
<td>0.86 ± 0.02*</td>
<td>27.6 ± 2.2*</td>
<td>1.2 ± 0.4*</td>
<td>113 ± 10*</td>
<td>104.6 ± 5.1</td>
<td>11.6 ± 0.6*</td>
<td>7.7 ± 0.5*</td>
<td>14.7 ± 2.3*</td>
</tr>
<tr>
<td>CWR2 LONG</td>
<td>41.0 ± 14.0</td>
<td>12.5 ± 1.5</td>
<td>0.94 ± 0.12</td>
<td>0.89 ± 0.10</td>
<td>0.90 ± 0.11</td>
<td>33.8 ± 2.1</td>
<td>0.9 ± 0.3</td>
<td>143 ± 8</td>
<td>104.7 ± 3.9</td>
<td>14.8 ± 0.9</td>
<td>6.5 ± 0.6</td>
<td>1.9 ± 1.0</td>
</tr>
<tr>
<td><strong>MM</strong></td>
<td></td>
<td></td>
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<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>CWR1 SHORT</td>
<td>38 ± 14</td>
<td>11.7 ± 1.5</td>
<td>0.83 ± 0.09</td>
<td>0.82 ± 0.16</td>
<td>0.98 ± 0.11</td>
<td>30.4 ± 12.1</td>
<td>3.33 ± 0.41</td>
<td>115 ± 21.1</td>
<td>97.7 ± 4.4</td>
<td>11.7 ± 2.0</td>
<td>6.98 ± 0.87</td>
<td>6.2 ± 3.0</td>
</tr>
<tr>
<td>CWR2 SHORT</td>
<td>38 ± 14</td>
<td>12.2 ± 2.0</td>
<td>0.86 ± 0.15</td>
<td>0.85 ± 0.13</td>
<td>0.99 ± 0.11</td>
<td>33.1 ± 11.1</td>
<td>4.09 ± 0.69</td>
<td>122 ± 19.4</td>
<td>98.1 ± 5.2</td>
<td>12.2 ± 2.2</td>
<td>7.00 ± 0.70</td>
<td>6.0 ± 5.5</td>
</tr>
<tr>
<td>CWR2 LONG</td>
<td>38 ± 14</td>
<td>13.5 ± 3.0</td>
<td>0.84 ± 0.13</td>
<td>0.83 ± 0.10</td>
<td>0.98 ± 0.12</td>
<td>32.2 ± 13.4</td>
<td>3.65 ± 0.73</td>
<td>124 ± 19.3</td>
<td>97.9 ± 6.2</td>
<td>12.5 ± 3.2</td>
<td>7.02 ± 0.62</td>
<td>6.9 ± 3.5</td>
</tr>
</tbody>
</table>
Mean (± SD) values of \( \dot{V}O_2 \), oxygen uptake; \( \dot{V}CO_2 \), CO_2 output; R, gas exchange ratio; \( \dot{V}E \), pulmonary ventilation; Gain, \( \Delta \dot{V}O_2 \) (\( \dot{V}O_2 \) at the end of CLE minus resting \( \dot{V}O_2 \)) divided by work rate; [La]b, blood lactate concentration; RPE, rate of perceived exertion; HR, heart rate; SV, stroke volume; \( Q \), cardiac output; \( [C_{(a-v)}O_2] \), systemic arterial-venous O_2 concentration difference; \( \Delta[\text{deoxy}(Hb+Mb)] \), muscle oxygenation index obtained by NIRS. *P < 0.05, significantly different from the corresponding value obtained in CWR1. See text for further details.
Table 3. V’O₂ kinetics parameters for CWR1 and CWR2 in McA and MM. Data obtained with 6 (SHORT) or 18 minutes (LONG) of recovery are shown.

<table>
<thead>
<tr>
<th></th>
<th>τf (s)</th>
<th>TDf (s)</th>
<th>yBAS (L min⁻¹)</th>
<th>Af (L min⁻¹)</th>
<th>A’s (L min⁻¹)</th>
<th>A’s/Atot</th>
<th>Gain (mL min⁻¹ watt⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>McA</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CWR1</td>
<td>24.1 ± 4.1</td>
<td>-2.9 ± 3.1</td>
<td>0.30 ± 0.03</td>
<td>0.56 ± 0.07</td>
<td>0.11 ± 0.02</td>
<td>16.0 ± 3.5</td>
<td>16.0 ± 0.5</td>
</tr>
<tr>
<td>CWR2 SHORT</td>
<td>29.5 ± 4.5</td>
<td>1.6 ± 2.7</td>
<td>0.33 ± 0.04</td>
<td>0.59 ± 0.06</td>
<td>0.0 ± 0.0*</td>
<td>0.0 ± 0.0*</td>
<td>12.9 ± 0.4*</td>
</tr>
<tr>
<td>CWR2 LONG</td>
<td>28.7 ± 3.4</td>
<td>-1.2 ± 1.7</td>
<td>0.32 ± 0.03</td>
<td>0.58 ± 0.07</td>
<td>0.09 ± 0.04</td>
<td>15.7 ± 5.2</td>
<td>14.2 ± 1.5</td>
</tr>
<tr>
<td>MM</td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>CWR1</td>
<td>48.2 ± 11.1</td>
<td>1.1 ± 4.9</td>
<td>0.33 ± 0.03</td>
<td>0.49 ± 0.09</td>
<td>NA</td>
<td>NA</td>
<td>13.3 ± 1.7</td>
</tr>
<tr>
<td>CWR2 SHORT</td>
<td>42.4 ± 7.1</td>
<td>-4.5 ± 3.5</td>
<td>0.33 ± 0.04</td>
<td>0.50 ± 0.08</td>
<td>NA</td>
<td>NA</td>
<td>14.9 ± 1.8*</td>
</tr>
<tr>
<td>CWR2 LONG</td>
<td>44.7 ± 8.3</td>
<td>-2.9 ± 4.5</td>
<td>0.32 ± 0.03</td>
<td>0.51 ± 0.09</td>
<td>NA</td>
<td>NA</td>
<td>14.2 ± 2.0</td>
</tr>
</tbody>
</table>

Mean (± SD) values of baseline (yBAS); time delay (TDf), time constant (τf), amplitude (Af) of the fundamental component; actual amplitude (A’s) of the slow component; and total amplitude of the response (Atot). *P < 0.05, significantly different from the corresponding value obtained in CWR1. NA = not applicable. See text for further details.