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

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Submitted 20 September 2013; accepted in final form 17 March 2014

Porcelli S, Marzorati M, Belletti M, Bellistri G, Morandi L, Grassi B. The “second wind” in McArdle’s disease patients during a second bout of constant work rate submaximal exercise. J Appl Physiol 116: 1230–1237, 2014. First published March 20, 2014; doi:10.1152/japplphysiol.01063.2013.—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 yr) and a control group of six mitochondrial myopathy patients (51 ± 6 yr) 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 VO2 uptake (VO2), HR, cardiac output, rates of perceived exertion, vastus lateralis oxyhemoglobin [changes in deoxygenated Hb and myoglobin Mb concentrations, Δ[deoxy(Hb+Mb)], by near-infrared spectroscopy] were determined. In McA, VO2 (0.86 ± 0.2 vs. 0.95 ± 0.1 l/min), HR (113 ± 10 vs. 150 ± 13 beats/min), cardiac output (11.6 ± 0.6 vs. 15.0 ± 0.8 l/min), and rates of perceived exertion (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 mitochondrial myopathy patients during both CWR2-SHOR 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.

myophosphorylase deficiency; exercise tolerance; VO2 kinetics slow component; near-infrared spectroscopy

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 arteriovenous O2 concentration difference (lavo2), 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 with healthy subjects, for the same submaximal VO2 (20, 28, 29, 40, 44–46, 49), together with a markedly slower adjustment of VO2 during constant work rate (CWR) submaximal exercise (17).

A typical feature of McA is the “second-wind” phenomenon (21, 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 sympathoadrenal 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 (21). Other studies have demonstrated that the second wind can be induced by oral glucose (21).

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 has the second-wind phenomenon 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, 19). 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 CWR 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 (RPE), more “traditional” signs of the second wind (lower HR, lower \( Q \), increased \( O_2 \) extraction) were sought after, together with other “ancillary” signs of increased exercise tolerance [lower pulmonary ventilation (\( V_t \)), lower gas exchange ratio (R)]. In addition, we sought to determine whether the second wind was associated with a decrease or with a disappearance of the slow component of the \( V_{O_2} \) kinetics, with a lower \( O_2 \) cost of exercise, and with a decrease in transient unbalances between \( O_2 \) delivery and \( O_2 \) utilization within skeletal muscles [as determined by near-infrared spectroscopy (NIRS) (12, 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 min) and a “long” (18 min) recovery period between exercise bouts, we also tried to get insights (also for practical purposes) into the length of the recovery period that would allow the second-wind phenomenon to manifest itself. Whereas 6 min represent a “standard” recovery between two 6-min exercise bouts (see e.g., Ref. 4), 18 min were arbitrarily chosen 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 to patients methods that 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. Sex distribution, age, and body mass of McA were as follows: 3 men and 5 women, age (mean ± SD) 44 ± 4 yr, and body mass 75.9 ± 8.9 kg. Sex distribution, age, and body mass of MM were as follows: 5 men and 1 woman, age 51 ± 6 yr, and body mass 69.1 ± 7.4 kg. Patients were from the Department of Neuromuscular Diseases, Neurological Institute “Carlo Besta” (Istituto Di Ricovero e Cura a Carattere Scientifico), 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 laboratory’s 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, rheumatological, 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. All 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 h) after a light breakfast (~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 \( V_{O_2peak} \). After 3 min of unloaded pedaling, exercise was conducted at 25-50 W for 5 min, 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) HR values >85% of the age-predicted maximum. Values of cardiovascular, ventilatory, gas exchange, and muscle oxygenation variables determined during the last 30 s of the exhausting load were considered “peak” values.

During the 2nd and 3rd days, the patients performed two repetitions of subsequent 6-min CWR 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 in the second case (after observing at least 2 h 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. 

\[ V_{E}(\text{in BTPS}), V_{O_2}, \text{and } V_{CO_2} \text{ output (V_{CO_2})}, \text{both in STPD}, \text{were determined breath by breath by a metabolic cart (Vmax29c; Sensor-Medics, Bilihoven, The Netherlands). Expiratory flow was determined by a flow sensor (hot wire anemometer). } V_{O_2} \text{ and } V_{CO_2} \text{ were determined by continuously monitoring } P_{O_2} \text{ and } P_{CO_2} \text{ at the mouth throughout the respiratory cycle and from established mass balance equations.} \]

\[ R \text{ was calculated as } V_{CO_2}/V_{O_2}. \text{ HR was determined from the ECG signal. Stroke volume (SV) was estimated beat by beat by impedance cardiography (Physio Flow; Manatec, Paris, France). The accuracy of this device has been previously evaluated, in healthy subjects, during incremental exercise on a cycle ergometer, and was found to be “acceptable” (38).} \]

\[ Q \text{ was calculated as } HR\cdot SV. \text{ Systemic peak (\[a-vCO_2]\]) was calculated as } V_{O_2peak}/peak \ Q. \text{ At rest and at various times (1, 3, and 5 min) during recovery, 20 } \mu \text{L of capillary blood were 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 NIR 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 deoxyggenated \( Hb \) and myoglobin (Mb) concentrations \([\Delta[\text{deoxy(Hb+Mb)}]]\), as well as of oxygenated \( Hb \) and Mb concentrations \([\Delta[\text{oxy(Hb+Mb)}]]\), expressed in arbitrary units. Details on the method can be found in previous studies from our group (15, 27, 36). \([\Delta[\text{oxy(Hb+Mb)}]]\), with respect to an initial value arbitrarily set equal to zero, were calculated and expressed in arbitrary units.
sum of the two variables \( \Delta [\text{oxy}(\text{Hb} + \text{Mb}) + \text{deoxy}(\text{Hb} + \text{Mb})] \) is related to changes in the total Hb volume in the muscle region of interest (6, 11, 25). \( \Delta [\text{oxy}(\text{Hb} + \text{Mb})] \) was taken as a “deoxygenation index,” because this variable is relatively insensitive to changes in blood volume (6, 25). \( \Delta [\text{deoxy}(\text{Hb} + \text{Mb})] \) was considered an estimate of skeletal muscle fractional \( \text{O}_2 \) extraction, that is, of the ratio between \( \text{V}O_2 \) and \( \text{O}_2 \) delivery (12, 15). \( \Delta [\text{deoxy}(\text{Hb} + \text{Mb})] \) 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 \( \sim 300 \) mmHg) at the root of the thigh (subject in the sitting position on the cycloergometer), for a few minutes until the \( \Delta [\text{deoxy}(\text{Hb} + \text{Mb})] \) increase reached a plateau.

**Kinetics analysis.** \( \text{V}O_2 \) kinetics were evaluated during transitions from rest to CWR. Breath-by-breath \( \text{V}O_2 \) values obtained during the repetitions of the exercises were time aligned and then superimposed for each subject. Average \( \text{V}O_2 \) values every 10 s were calculated. Data obtained during the first 20 s of the transition (“cardiodynamic” phase) (37) were excluded from analysis. Thus \( \text{V}O_2 \) 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, 35, 50).

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

\[
y(t) = y_{\text{BAS}} + A_f \left[ 1 - \exp\left(-\frac{t}{T_d}\right)\right] \tag{1}
\]

where \( y_{\text{BAS}} \) indicates the \( \text{V}O_2 \) value at baseline; \( A_f \) is the amplitude of the \( \text{V}O_2 \) response calculated between the baseline value and the steady-state value for the fundamental component; \( T_d \) is the time delay, and \( t \) the time constant of the function for the fundamental component.

To check the presence of a slow component of the kinetics (23), data were also fit by a double-exponential function of the type:

\[
y(t) = y_{\text{BAS}} + A_f \left[ 1 - \exp\left(-\frac{t}{T_d}\right)\right] + A_s \left[ 1 - \exp\left(-\frac{t}{T_s}\right)\right] \tag{2}
\]

where \( A_s \), \( T_d \), and \( T_s \) indicate, respectively, the amplitude, the time delay, and the time constant of the slow component of the kinetics.

Sometimes, after the first exponential rise, \( \text{V}O_2 \) 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) (41):

\[
y(t) = y_{\text{BAS}} + A_f \left[ 1 - \exp\left(-\frac{t}{T_d}\right)\right] + S \left[ t - T_d \right] \tag{3}
\]

where \( S \) (slope) is the angular coefficient of the linear regression of \( \text{V}O_2 \) 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 (50) of \( \text{V}O_2 \) kinetics was present, superimposed on the fundamental component. The actual amplitude of the slow component (\( A_s \))s was estimated as the difference between the average \( \text{V}O_2 \) value obtained during the last 20–30 s of CWR and the asymptotic value of the fundamental component (15, 41). The percentage contribution of the slow component to the total amplitude of the response (\( A_s/A_{\text{Total}} \)) was also calculated (36).

**Statistical analysis.** Results were expressed as mean values ± 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 (MmA vs. MM) on the investigated variables were checked by two-way ANOVA. This analysis, however, did not yield a statistically significant difference for \( \text{V}O_2 \) and \( \text{VE} \). This is likely attributable to the relative number of patients in the two groups [MmA and MM are rare diseases; see also the recent commentary by Ploutz-Snyder (32)]. Thus analysis of differences between CWR1 and CWR2 in MM and MmA was also performed by one-way ANOVA. 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 MmA and MM in a previous study by our group (14) and by others (19, 20). \( \text{V}O_2\text{peak} \) was ~50% of that usually obtained in healthy age-matched subjects (30), indicating a severely reduced maximal aerobic power. HR values, significantly higher in MmA than in MM, corresponded to ~96% of the age-predicted maximum. Peak Q values were only slightly lower than those usually obtained in healthy controls (39). As expected for both patients groups, [a-\( \text{v}\)\( \text{Co}_2 \)] and peak skeletal muscle fractional \( \text{O}_2 \) extraction values were very low. As expected, in MmA 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 MmA and MM.

**CWR exercises.** Figure 1 shows typical examples of HR time courses of a MM (top) and of a MmA (bottom) during SHORT (left) and LONG (right). In MmA, during SHORT (but not during LONG) HR values at the end of CWR2 were markedly lower (by about 50 beats/min) than during CWR1. This second-wind phenomenon is indicated by the arrow. No differ-

| Table 1. Peak values of the main cardiovascular, ventilatory, and metabolic variables in MmA and MM, as well as rates of perceived exertion and blood lactate levels |
|-----------------|-----------------|-----------------|-----------------|-----------------|
|                | MmA             | MM              |                |
| Work rate, W   | 78.6 ± 18.5     | 71.7 ± 11.2     |                |
| RPE            | 16.6 ± 0.8      | 15.6 ± 0.7      |                |
| \( \text{V}O_2 \), l/min | 1.33 ± 0.25   | 1.08 ± 0.22     |                |
| \( \text{V}O_2 \), ml·kg\(^{-1}\)·min\(^{-1}\) | 18.5 ± 2.9     | 15.5 ± 1.1      |                |
| \( \text{VE} \), l/min | 1.22 ± 0.21   | 1.29 ± 0.2      |                |
| R              | 0.93 ± 0.1*     | 1.21 ± 0.1      |                |
| [La]b,m M      | 47.0 ± 6.8      | 52.4 ± 10.1     |                |
| RPE            | 1.61 ± 0.2      | 1.60 ± 0.2      |                |
| [La]b, Torr    | 29.4 ± 2.8      | 31.8 ± 2.3      |                |
| \( \text{VE} \), l/min | 113.4 ± 2.2    | 118.7 ± 2.5     |                |
| [La]b,COD, Torr| 29.8 ± 1.0      | 30.7 ± 1.7      |                |
| HR, beats/min  | 161.6 ± 3.4*    | 149.9 ± 8.1     |                |
| \( \text{VE} \), l/min | 103.2 ± 9.7   | 109.4 ± 7.5     |                |
| [La]b, l/min   | 17.5 ± 1.7      | 16.3 ± 1.2      |                |
| \[\text{a-\( \text{v}\)\( \text{Co}_2 \)}\], ml O\(_2\)/100 ml | 7.6 ± 0.9      | 6.4 ± 0.7       |                |
| \( \Delta [\text{deoxy}(\text{Hb} + \text{Mb})], \%\text{ischemia}\) | 20.3 ± 8.4     | 20.1 ± 4.6      |                |

Values are means ± SD. MmA, patients with McArdle’s disease; MM, patients affected by a mitochondrial myopathy. RPE, rate of perceived exertion; \( \text{V}O_2 \), oxygen uptake; \( \text{VE} \), \( \text{CO}_2 \) output; R, gas exchange ratio; \( \text{VE} \), pulmonary ventilation; \( \text{VT} \), tidal volume; \( \text{IR} \), breathing frequency; \( \text{PETCO}_2 \), end-tidal \( \text{CO}_2 \) partial pressure; \( \text{PaCO}_2 \), arterio-venous \( \text{CO}_2 \) partial pressure; [La]b, blood lactate concentration; HR, heart rate; SV, stroke volume; Q, cardiac output; [a-\( \text{v}\)\( \text{CO}_2 \)], systemic arterial-venous \( \text{O}_2 \) concentration difference; \( \Delta [\text{deoxy}(\text{Hb} + \text{Mb})] \), changes in deoxygenated Hb and myoglobin Mb concentrations, muscle oxygenation index obtained by near-infrared spectroscopy. *\( P < 0.05 \), significantly different from the corresponding value obtained in MM. See text for further details.
ences between CWR1 and CWR2 were observed in MM, during either SHORT or 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, VO₂, VCO₂, VE, R, HR, Q, and RPE values were significantly lower in CWR2 vs. CWR1. On the other hand, [La]ₐ, [a-vCO₂], and Δ(deoxy(Hb+Mb)] were significantly higher in CWR2 vs. CWR1. No significant differences were observed between CWR1 and CWR2 in MM, during both CWR1 and CWR2 (SHORT and LONG recovery). See text for further details. b/min, beats per minute.

Values are means ± SD. Data obtained with 6 (SHORT) or 18 min (LONG) of recovery are presented. CWR1 and CWR2, first and second constant work rate exercise, respectively. *P < 0.05, significantly different from the corresponding value obtained in CWR1. See text for further details.

The second-wind phenomenon is indicated by the arrow. A slow component was not observed in MM, during both CWR1 and CWR2 (SHORT and LONG).

The parameters of VO₂ kinetics are shown in Table 3. In both groups, TD, τf, and Af values were not significantly different in CWR1 vs. CWR2 (both in SHORT and in LONG). Gain (G) values were calculated as ΔVO₂ (VO₂ at the end of CWR minus resting VO₂) divided by work rate. A slow component, corresponding to ~15% of the Aₛ tot 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/ Aₛ 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

Table 2. Values of the main cardiovascular, ventilatory, and metabolic variables in CWR1 and CWR2 in McA and MM

<table>
<thead>
<tr>
<th></th>
<th>McA</th>
<th></th>
<th>MM</th>
<th></th>
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</thead>
<tbody>
<tr>
<td></td>
<td>CWR1</td>
<td>CWR2 SHORT</td>
<td>CWR2 LONG</td>
<td>CWR1</td>
</tr>
<tr>
<td>Work rate, W</td>
<td>41.0 ± 14.0</td>
<td>41.0 ± 14.0</td>
<td>41.0 ± 14.0</td>
<td>38 ± 14</td>
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<td>RPE</td>
<td>13.9 ± 2.6</td>
<td>10.8 ± 1.7*</td>
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<tr>
<td>VO₂, l/min</td>
<td>0.95 ± 0.11</td>
<td>0.86 ± 0.15*</td>
<td>0.94 ± 0.12</td>
<td>0.83 ± 0.09</td>
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<tr>
<td>VCO₂, l/min</td>
<td>0.92 ± 0.14</td>
<td>0.81 ± 0.14*</td>
<td>0.89 ± 0.10</td>
<td>0.82 ± 0.16</td>
</tr>
<tr>
<td>R</td>
<td>0.93 ± 0.02</td>
<td>0.86 ± 0.02*</td>
<td>0.90 ± 0.11</td>
<td>0.98 ± 0.11</td>
</tr>
<tr>
<td>VE, l/min</td>
<td>36.2 ± 3.0</td>
<td>27.6 ± 2.2*</td>
<td>33.8 ± 2.1</td>
<td>30.4 ± 12.1</td>
</tr>
<tr>
<td>[La]ₐ, mM</td>
<td>0.8 ± 0.2</td>
<td>1.2 ± 0.4*</td>
<td>0.9 ± 0.3</td>
<td>3.33 ± 0.41</td>
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<tr>
<td>HR, beats/min</td>
<td>150 ± 13</td>
<td>113 ± 10*</td>
<td>143 ± 8</td>
<td>115 ± 21.1</td>
</tr>
<tr>
<td>SV, ml</td>
<td>102.6 ± 6.5</td>
<td>104.6 ± 5.1</td>
<td>104.7 ± 3.9</td>
<td>97.7 ± 4.4</td>
</tr>
<tr>
<td>Q, l/min</td>
<td>15.0 ± 0.8</td>
<td>11.6 ± 0.6*</td>
<td>14.8 ± 0.9</td>
<td>11.7 ± 2.0</td>
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<tr>
<td>[a-vCO₂], ml O₂/100 ml</td>
<td>6.7 ± 0.6</td>
<td>7.7 ± 0.5*</td>
<td>6.5 ± 0.6</td>
<td>6.98 ± 0.87</td>
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<tr>
<td>Δ(deoxy(Hb+Mb)]</td>
<td>−0.1 ± 0.4</td>
<td>4.7 ± 2.3*</td>
<td>1.9 ± 1.0</td>
<td>6.2 ± 3.0</td>
</tr>
</tbody>
</table>

Fig. 1. Typical individual examples of heart rate (HR) kinetics during the first (CWR1) and the second constant work rate (CWR2) exercise in patients affected by a mitochondrial myopathy (MM; top) and patients with McArdle’s disease (McA; bottom). Data obtained with 6 (SHORT) or 18 min (LONG) of recovery are presented on the left and right, respectively. The vertical dotted 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.
were substantially higher that those usually observed in normal subjects (∼10 ml·min⁻¹·W⁻¹), independent from the presence of the slow component.

Δ[deoxy(Hb+Mb)] kinetics are shown in Fig. 3. In MM, in all conditions, there was an initial and transient increase (“overshoot”) of Δ[deoxy(Hb+Mb)] (occurring after ∼45 s of exercise), which was followed by a steady state. Δ[deoxy(Hb+Mb)] values at the peak of the overshoot were significantly higher than at steady state for both CWR1 (24.9 ± 5.1 vs. 6.2 ± 3.0%) and CWR2 (21.9 ± 4.5 vs. 6.0 ± 5.4% and 21.5 ± 5.4 vs. 6.9 ± 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 ± 6.0 vs. −0.1 ± 4.6%) and during CWR2-LONG (24.9 ± 6.7 vs. 1.9 ± 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 CWR exercises, carried out at ∼50% of peak work rate and separated by 6 min 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 min (LONG).

Can the differences mentioned above be considered an expression of a “second-wind phenomenon” (1, 2, 21, 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., Ref. 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, are shown in Table 3.

Table 3. VO₂ kinetics parameters for CWR1 and CWR2 in McA and MM

<table>
<thead>
<tr>
<th></th>
<th>CWR1</th>
<th>CWR2 SHORT</th>
<th>CWR2 LONG</th>
<th>MM</th>
</tr>
</thead>
<tbody>
<tr>
<td>γf, s</td>
<td>24.1 ± 4.1</td>
<td>29.5 ± 4.5</td>
<td>28.7 ± 3.4</td>
<td>48.2 ± 11.1</td>
</tr>
<tr>
<td>TDf, s</td>
<td>2.9 ± 3.1</td>
<td>1.6 ± 2.7</td>
<td>1.2 ± 1.7</td>
<td>1.1 ± 4.9</td>
</tr>
<tr>
<td>yBAS, l/min</td>
<td>0.30 ± 0.03</td>
<td>0.33 ± 0.04</td>
<td>0.32 ± 0.03</td>
<td>0.33 ± 0.03</td>
</tr>
<tr>
<td>A', l/min</td>
<td>0.56 ± 0.07</td>
<td>0.59 ± 0.06</td>
<td>0.58 ± 0.07</td>
<td>0.49 ± 0.09</td>
</tr>
<tr>
<td>A'/A, %</td>
<td>0.11 ± 0.02</td>
<td>0.0 ± 0.0*</td>
<td>0.09 ± 0.04</td>
<td>NA</td>
</tr>
<tr>
<td>Gain, ml·min⁻¹·W⁻¹</td>
<td>16.0 ± 3.5</td>
<td>0.0* ± 0.0</td>
<td>13.7 ± 5.2</td>
<td>13.3 ± 1.7</td>
</tr>
</tbody>
</table>

Values are means ± SD. Data obtained with SHORT or LONG recovery are shown. Values are baseline (yBAS), time delay (TDf), time constant (γf), and amplitude (A') of the fundamental component; actual amplitude (A') of the slow component; and total amplitude of the response (A/A), Gain, ΔVO₂ (VO₂ at the end of CWR minus resting VO₂) divided by work rate. *P < 0.05, significantly different from the corresponding value obtained in CWR1. NA, not applicable. See text for further details.
kinetics, the substantially lower \( \dot{V}E \), HR, \( Q \), RPE, etc., and the slightly higher fractional \( \text{O}_2 \) 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 lower during CWR2-SHORT vs. CWR1, whereas the priming effect does not usually affect HR values in healthy subjects (see e.g., Ref. 4). Moreover, in the “control” population represented by MM, which has a similar exercise tolerance compared with McA (see also Refs. 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 that 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 (21). The phenomenon has been previously demonstrated in McA patients during prolonged exercise (21, 49) or after sucrose administration (1, 2, 21) and is considered pathognomonic for the disease (49). The second wind has been described in literature as a lower HR (21), lower RPE (48), increased \( [\text{a-vCO}_2] \) (21), and increased \( [\text{La}]_b \) (21) during submaximal exercise, or increased peak work rate and \( \text{V}O_2\text{peak} \) (21).

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 \( \text{O}_2 \) extraction (as determined by NIRS), confirming the data obtained by different methods by Haller and Vissing (21). 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, 21, 45, 46). Skeletal muscle fractional \( \text{O}_2 \) 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 aging 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. (12), the overshoot is a sign of a relatively inadequate muscle \( \text{O}_2 \) delivery vs. muscle \( \text{V}O_2 \) and could lead to a reduced microvascular \( \text{O}_2 \) pressures and to a lower blood-to-myocyte “driving force” for peripheral \( \text{O}_2 \) diffusion. The overshoot phenomenon, which suggests an impaired intramuscular matching between \( \text{O}_2 \) delivery and \( \text{O}_2 \) 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. (34) and seem to be related to nitric oxide bioavailability. Also, this component of the second-wind phenomenon was no longer present after 18 min 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” glycolgenolysis. GET is usually utilized to discriminate between moderate exercise (below GET, with no slow component of $V_O2$ 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 $V_O2$ keeps increasing as a function of time during the constant work-rate exercise (as in McA during CWR1, see Fig. 2), until $V_O2_{peak}$ 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 CWR exercises, separated by 6 min of recovery. The second exercise was indeed characterized by significantly lower (compared with the first exercise bout) RPE, HR, $V_{E}$, and R, and by slightly higher skeletal muscle fractional $O_2$ 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 the slow component of pulmonary $V_O2$ kinetics (and, therefore, with a lower $V_O2$ and a lower $O_2$ cost of exercise), and the disappearance of signs of transient mismatch between $O_2$ delivery and $O_2$ utilization in skeletal muscle. We did not observe the second-wind phenomenon when the recovery period between the two exercise bouts was longer (18 min).

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 that are often already empowered by McA patients 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 that have been developed in the exercise physiology laboratory, with the ultimate aim of increasing their exercise tolerance and quality of life.

ACKNOWLEDGMENTS

The authors thank Marco Pellegrini and Giusi Ferrari for technical assistance.


32. **Ploutz-Snyder RJ.** Justifying small-n research in scientifically important settings: challenging the notion that only “big-n” studies are worthwhile. *J Appl Physiol;* doi:10.1152/japplphysiol.01335.2013.


