Muscle O$_2$ consumption by NIRS: a theoretical model

Binzoni, T., W. Colier, E. Hiltbrand, L. Hoofd, and P. Cerretelli. Muscle O$_2$ consumption by NIRS: a theoretical model. J. Appl. Physiol. 87(2): 683–688, 1999.—In the past, the measurement of O$_2$ consumption (O$_\dot{2}$) by the muscle could be carried out noninvasively by near-infrared spectroscopy from oxyhemoglobin and/or deoxyhemoglobin measurements only at rest or during steady isometric contractions. In the present study, a mathematical model is developed allowing calculation, together with steady-state levels of O$_2$, of the kinetics of O$_2$ readjustment in the muscle from the onset of ischemic but aerobic constant-load isotonic exercises. The model, which is based on the known sequence of exoergonic metabolic pathways involved in muscle energetics, allows simultaneous fitting of batched data obtained during exercises performed at different workloads. A Monte Carlo simulation has been carried out to test the quality of the model and to define the most appropriate experimental approach to obtain the best results. The use of a series of experimental protocols obtained at different levels of mechanical power, rather than repetitions of the same load, appears to be the most suitable procedure.

The study of energy metabolism, both at rest and during exercise, represents a valuable method of determining the functional status of human skeletal muscle (11, 16). However, in humans, this approach implies the difficult task of monitoring noninvasively, in situ, the rate of the basic energy-yielding metabolic processes, i.e., the Lohmann reaction, aerobic glycolysis, and anaerobic glycolysis (11). The most powerful tool fulfilling the above-outlined requirements is nuclear magnetic resonance spectroscopy (NMRS) (16). $^{31}$P-NMRS is well suited to follow, intracellularly, the metabolic reactions involved in the Lohmann reaction, particularly hydrolysis of phosphocreatine (PCr), provided the time resolution is sufficient to monitor the changes underlying muscle activity (2, 4, 6, 15, 18). As is well known, anaerobic glycolysis may also be assessed by $^{31}$P-NMRS, even though indirectly, from pH measurements (19, 21), or by $^{1}$H-NMRS by using an edited technique specific for lactate (La) (14). Indeed, $^{31}$P-NMRS has been widely used to study muscle metabolism in normal (16) and pathological conditions (17). By contrast, no NMRS technique is available to directly monitor tissue O$_2$ consumption (O$_2$). Recent theoretical (3) and experimental (2, 4) studies were aimed at identifying the relationship existing among the various energy-yielding mechanisms to establish from relatively simple $^{31}$P-NMRS measurements the rate of aerobic and anaerobic glycolysis. Despite recent progress, the above methods are still not satisfactory because of their high cost and organizational problems.

Near-infrared spectroscopy (NIRS) appears to be the emerging technique for monitoring aerobic metabolism in muscle (12). Indeed, NIRS allows measurement, noninvasively, at the tissue level, and during short periods of ischemia, of tissue oxyhemoglobin ($\Delta$[HbO$_2$]) and deoxyhemoglobin concentration changes ($\Delta$[Hb]). The latter changes, in the absence of inflow and outflow to and from the tissue, reflect the functional changes induced by oxidative metabolism (7, 12). $\Delta$[Hb] is the mirror image of the disappearance of O$_2$ stored in the tissue before ischemia is induced (increase in $\Delta$[Hb] = decrease in $\Delta$[HbO$_2$]). Previous studies have made it possible to measure resting O$_2$ in the arm (9, 13) and in the calf (5) muscles by using NIRS. The O$_2$ values found correspond to those obtained by the Fick method under normal perfusion conditions. The same technique was applied for O$_2$ measurements during isometric contractions (8). The latter approach was based on the hypothesis that the same algorithm used for rest was still applicable.

By contrast, the relationship among $\Delta$[HbO$_2$], $\Delta$[Hb], and O$_2$ starting from the onset of a series of isotonic muscle contractions has not been assessed, and so far no algorithm has been developed for the calculation of O$_2$ either during the rest-to-work transient or at steady state. As is well known, steady-state O$_2$ as well as the rate of change of O$_2$ during a rest-to-work transient, classically defined by the time constant of an exponential curve, are important functional parameters known to be influenced by the fiber-type content and training level of the muscle, by pathological factors, and so on.

The purpose of the present study was to develop a method for the measurement of intramuscular O$_2$ kinetics and O$_2$ at steady state, utilizing $\Delta$[Hb] measurements during ischemic constant-load isotonic exercise. Because myoglobin has the same NIRS spectrum as hemoglobin, as will be pointed out in discussion, the present method is not influenced by muscle myoglobin. Because of the nonstationarity of the energetic processes, a model is required that is different from the one based on a linear regression used for measurements in resting muscle (5, 9, 13). In practice, it is proposed to 1) construct a theoretical model describing $\Delta$[Hb] as a function of time in the muscle region of interest during the rest-to-work transient of constant-load isotonic exercise; 2) show theoretically how to derive O$_2$ and its time constant from $\Delta$[Hb] kinetics; and 3) analyze the differences between the time courses of O$_2$ and $\Delta$[Hb].

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THE PHYSIOLOGICAL BACKGROUND

As was pointed out above, the purpose of the present study is to describe muscle O\textsubscript{2} kinetics from \(\Delta[Hb]\) measurements. Figure 1A is a typical set of experimental data obtained in a healthy sedentary subject. The measurements were carried out by a NIRS instrument (Oxymon, University of Nijmegen) (20) by using three wavelengths (775, 848, and 905 nm). The detectors were placed on the right forearm, and \(\Delta[Hb]\) and \(\Delta[HbO2]\) were recorded in the hand flexors. Each curve describes \(\Delta[Hb]\) kinetics just after the inflation of a cuff that coincides with the onset of series of constant-load contractions at the rate of 0.5 Hz against increasing loads (0.10, 0.41, 0.62, 1.24, 1.65, and 2.06 W). As may be seen from the graph in Fig. 1A, at higher loads the curves become steeper. The straight line (bottom of panel) represents \(\Delta[Hb]\) at rest. The choice of the muscle is arbitrary as well as that of the NIRS instrument.

For constructing the model, two different functional conditions must be considered: 1) rest and 2) a series of rest-to-work transients. With regard to condition 1, it has been demonstrated (5, 9, 13) that during the first 5 min of ischemia, the muscle depends only on aerobic sources for its metabolic requirements. This is because the tissue contains enough O\textsubscript{2} stores, mainly bound to hemoglobin, to sustain oxidation without requiring energy from anaerobic sources. For example, in the resting plantar flexors, it was demonstrated that during 5-min ischemia, PCr concentration ([PCr]) is unchanged and pH keeps essentially constant at the control level (5). After 5-min ischemia, \(\Delta[Hb]\) tends to level off, and anaerobic metabolism becomes the main energy source (not shown in Fig. 1A).

During rest-to-work transients (condition 2) the picture is more complicated. In fact, depending on the workload, the tissue O\textsubscript{2} stores are depleted more rapidly than at rest. This implies that, in applying the same method as at rest, the analysis must be restricted to shorter periods of time, i.e., the first 20–40 s after the onset of ischemia. The basic requirement for the applicability of the proposed model is that, during this short time interval, the slope of \(\Delta[Hb]\) vs. time (proportional to O\textsubscript{2}) must not decrease. This is tantamount to accepting the classic physiological observation that, during a rest-to-work transient, O\textsubscript{2} does not decrease. Once this condition is fulfilled, the experiment can be reasonably considered equivalent to one with normal perfusion.

Because the measurement of \(\Delta[Hb]\) in Fig. 1A is influenced by the proportion of both muscle and adipose tissue, the latter must be eliminated. On the assumption that adipose tissue metabolism keeps constant during exercise, resting \(\Delta[Hb]\) can be subtracted from the corresponding exercise values (Fig. 1B). This subtraction also eliminates basal muscle metabolism. Hence, net muscle \(\Delta[Hb]\) can be assessed for each tested load (Fig. 1B). The model will be based on the latter curves.

THE MATHEMATICAL MODEL

For constructing the model, a bioenergetic approach is adopted. The choice of the latter is based on the principle that the model holds no matter how workload...
The net (total – resting) energy fluxes during muscular contraction are described by the following equation (11)

\[
[ATP] = \alpha [PCr] + \beta [La] + \gamma [O_2]
\]

where \(\alpha\), \(\beta\), and \(\gamma\) represent the number of ATP moles produced per mole of PCr, La, and \(O_2\), respectively, and the overdot represents the time (t) derivative. The three right-hand terms represent the Lohmann reaction, anaerobic glycolysis, and aerobic glycolysis, respectively (11). Moreover, for modeling purposes, the following conditions hold (3).

1) For any given workload, [ATP] is constant throughout the experiment, i.e.

\[
[ATP] = [ATP]_0
\]

2) At the onset of exercise, the only available energy source is the Lohmann reaction, i.e., for \(t = 0\)

\[
\alpha [PCr] = [ATP]_0
\]

3) For all conditions described by the model, [PCr] attains a steady state, i.e., for \(t = \infty\)

\[
[PCr] = 0
\]

The range of validity of the present model is the aerobic domain. Aerobic exercise implies that [La] = 0 throughout the experiment. Therefore, Eq. 1 becomes

\[
[ATP]_0 = \alpha [PCr] + \gamma [O_2]
\]

As is well known from previous studies (3), during a rest-to-work transient in the aerobic domain, [PCr] may be described by the following equation

\[
[PCr] = \frac{1}{\alpha} [ATP]_0 e^{-\tau t}
\]

where \(\tau\) is a time constant, which, for a given subject, is independent of the workload (3). Equation 6 satisfies the conditions inherent in Eqs. 2, 3, and 4. By substituting Eq. 6 into Eq. 5, it is therefore possible to calculate \(O_2\) as

\[
[O_2] = \frac{1}{\gamma} [ATP]_0 (1 - e^{-\gamma t})
\]

This corresponds to the classic exponential trend of the \([O_2]\) readjustment curve in the muscle during a rest-to-work transient.

The time integral in Eq. 7 allows the calculation of the cumulative \(O_2\) consumed from the onset of exercise to time \(t\) as

\[
[O_2] = \frac{1}{\gamma} [ATP]_0 [t - \gamma (1 - e^{-\gamma t})]
\]

As for adopting the same approach as described in the literature (5, 8, 9, 13), Eq. 8 does not account for a possible contribution by changes in physically dis-
small number of experimental points. Therefore, to improve the quality of the fitting, a further constraint was imposed on the model. This consists of applying the well-known relationship between $\dot{O}_2$ and mechanical work ($\dot{w}$), which is expressed by

$$\dot{O}_2 = \gamma^{-1} [\dot{ATP}]_o = K \dot{w}$$

(11)

where $K$ is a constant independent of $\dot{w}$. Substituting Eq. 11 into Eq. 9, the new expression for $\Delta[Hb]$ becomes

$$\Delta[Hb] = \frac{1.13}{4} K \dot{w} [t - \tau (1 - e^{-\theta t})]$$

(12)

$\Delta[Hb]$ in Eq. 12 describes the data appearing in Fig. 1B after subtraction of the resting values. By this approach, all experimental curves obtained at different $\dot{w}$ values in a given subject contracting the same muscle can now be fitted simultaneously. In this case, only one $K$ and one $t$ value shall be obtained for all $\dot{w}$ levels (for a given subject). As indicated before (Eq. 6), $\tau$ for a given subject is independent of $\dot{w}$. This procedure makes the estimate less sensitive to artifacts generated by the experimental noise. Hence, Eq. 12 should definitively improve the quality of the fitting, and it represents our final model.

The robustness of the approach described above was checked in the time range $t = 0$–40 s (sampling rate: 1/s) on two sets of simulated $\Delta[Hb]$ data generated from Eq. 12 within the aerobic domain. The two Monte Carlo simulations consist of the following. 1) One thousand (no. of hypothetical subjects) series of five repetitions of an identical $\dot{w}$ were chosen at random (range 0.02–0.12 W/cm²). This procedure is equivalent to repeating the same exercise five times. 2) One thousand series of five different $\dot{w}$ were randomly generated (range 0.02–0.12 W/cm²). This is tantamount to each subject's carrying out five different workloads. A random noise of $\pm 5 \mu M$ was superimposed on the curves (in both simulations 1 and 2). The use of normalized $\dot{w}$ (i.e., W/cm²) values allows the simulation to be valid for any muscle cross section, giving the results a more general interest.

In the Monte Carlo simulations, each hypothetical subject was identified by a random pair of $t$ and $K$ values in the range of 20–50 s and 0.4–0.9 mmol·g⁻¹·s⁻¹·cm², respectively [i.e., 1,000 random ($t$, $K$) pairs for simulation 1 and 1,000 for simulation 2]. Figure 3, A and B, show the results of the fitting from the first set of data (simulation 1), which was performed by a least squares minimization procedure. It clearly appears that computed $\tau$ and $K$ values are affected by very large errors. By contrast, the fitting according to simulation 2 shown in Fig. 4, A and B, yields much better results. This proves the adequacy of Eq. 12, coupled with the experimental approach proposed in simulation 2, to estimate parameters $\tau$ and $K$ for a given subject. Evidently, due to the utilization of five different $\dot{w}$ values, the curves generated by simulation 2 contain much more information than those obtained by simulation 1, allowing a better estimate of $\tau$ and $K$. It goes without saying that a fitting over a time interval longer than 40 s, provided the conditions of aerobiosis set up in PHYSIOLOGICAL BACKGROUND are met, could yield better results.

As is well known, most commercial NIRS spectrometers do not allow $\Delta[Hb]$ to be obtained without the

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Fig. 2. A: graphical representation of $\Delta[Hb]$ normalized by parameter $\gamma^{-1} [\dot{ATP}]_o$ as function of time (Eq. 9) for different time constant ($t$) values ($t = 10$ (bottom curve) to 90 (top curve)) by 10-s steps. $\gamma$, No. of moles produced by 1 mol of $\dot{O}_2$ [ATP]; $[\dot{ATP}]_o$, time derivative of ATP concentration. B: graphical representation of $\Delta[Hb]$ normalized by parameter $\gamma^{-1} [\dot{ATP}]_o$ as function of time for same values as in A ($t$ values are decreasing from bottom to top curves).
introduction of a differential pathlength factor (DPF) (10). However, because DPF is a multiplicative factor in the \( \Delta[\text{Hb}] \) calculation related to the mean distance covered by the photons within the tissue before being detected, as may be seen from Eq. 12, \( \tau \) is DPF independent.

The constant \( \tau \) determined during aerobic exercise is an extremely valuable functional tool, as it defines the tissue's oxidative status. So far, this measurement could be carried out in humans either indirectly, e.g., from gas exchange in the lungs, or noninvasively, by \(^{31}\text{P}-\text{NMRS}\) in muscles. The first of the above procedures is not quite satisfactory because of the bias inherent in the method, whereas the second imposes methodological and economic constraints.

As is well known, oxygenated myoglobin (\( \text{MbO}_2 \)), together with \( \text{HbO}_2 \) in the muscle, contributes to \( \text{O}_2 \)
transport. Deoxygenated myoglobin (Mb) and \( \text{Mbo}_2 \) NIRS signals are superimposed on those of \( \text{Hb} \) and \( \text{HbO}_2 \), respectively. However, it is noteworthy that the proposed method of calculation of \( \text{O}_2 \) in muscle is not influenced by possible changes in myoglobin oxygenation. In fact, as explained in a previous work (5), the change in light absorption when a molecule of \( \text{HbO}_2 \) is transformed into \( \text{Hb} \) is equivalent to that found when four molecules of \( \text{Mbo}_2 \) are reduced to \( \text{Mb} \). Thus, in terms of consumption of \( \text{O}_2 \) molecules, the conditions are the same.

In conclusion, it was proven on theoretical grounds that oxidative metabolism can also be assessed in humans by NIRS \( \Delta[\text{Hb}] \) measurements in working muscles. For this purpose, a mathematical model is presented that allows determination, from the analysis of experimental \( \Delta[\text{Hb}] \) vs. time curves obtained at different ischemic but aerobic \( w \) levels, of 1) steady-state \( \text{O}_2 \) and 2) the kinetics of readjustment of \( \text{O}_2 \) in the rest-to-work transient.

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REFERENCES