Importance of mitochondrial P\textsubscript{O2} in maximal O\textsubscript{2} transport and utilization: A theoretical analysis

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1. Introduction

At rest or during exercise, production of ATP requires both physical O\textsubscript{2} transport from the environment to the mitochondria and subsequent chemical utilization of O\textsubscript{2} by oxidative phosphorylation. Oxygen transport has been well described (Dejours and Kayser, 1966; Gnaiger et al., 1998; Weibel et al., 1981) based on the O\textsubscript{2} transport pathway, consisting of the lungs/chest wall, the heart, vascular tree and blood, and the tissues. These structures conduct O\textsubscript{2} as an in-series system in which the main sequential transport steps are ventilation, alveolar-capillary diffusion, circulatory transport, and tissue capillary to mitochondrial diffusion. At each step, the mass of O\textsubscript{2} must be conserved, and this allows a set of simple equations to be defined (Wagner, 1993, 1996b) that quantifies how the transport process at each step integrates with those of the other steps to determine how much O\textsubscript{2} is delivered to the mitochondria per minute (Wagner, 1996a). In this construct, it is shown that each of the four steps contributes to limitation to V\textsubscript{O2 max} and that the quantitative effects of changes at each step are similar.

Systems physiological investigations (Wagner, 1993, 1996b) targeting the understanding of the limits to maximal V\textsubscript{O2 max}, have previously been performed on the basis of an important simplifying approximation. This has been that the downstream mitochondrial P\textsubscript{O2} (Pm\textsubscript{O2}) is so small in comparison to tissue capillary P\textsubscript{O2} that it can be ignored and therefore set to zero, thus making the analyses of O\textsubscript{2} transport much more tractable. However, because O\textsubscript{2} is one of the molecules that drive oxidative phosphorylation according to the law of mass action, this approximation cannot be physiologically correct, or otherwise V\textsubscript{O2 max} would itself be zero.

Given that Pm\textsubscript{O2} must exceed zero, the P\textsubscript{O2} difference between red cells and mitochondria must be less than when Pm\textsubscript{O2} is assumed to be zero, and thus the diffusive movement of O\textsubscript{2} between them must also be reduced. Therefore, if Pm\textsubscript{O2} is now considered as greater than zero, there is an additional resistance, from the process of mitochondrial respiration, to O\textsubscript{2} movement through the entire pathway of O\textsubscript{2} transport and utilization. We therefore hypothesize that this additional resistance must reduce maximal V\textsubscript{O2 max} below that which would be expected if this resistance were ignored. Clearly, the degree to which V\textsubscript{O2 max} would be reduced will depend on how the high mitochondrial P\textsubscript{O2} rises above zero. This in turn will depend broadly on the capacity for O\textsubscript{2} transport (how many O\textsubscript{2} molecules can be delivered to the mitochondria per minute) compared to the capacity for metabolism (how many O\textsubscript{2} molecules can be consumed by the mitochondria per minute).

The importance of including consideration of oxidative phosphorylation goes beyond asking how much does mitochondrial respiration contribute to the overall impedance to V\textsubscript{O2 max}. Because the value of Pm\textsubscript{O2} is dependent on the mitochondrial respiration curve/O\textsubscript{2} transport interaction, hypoxia-induced biological...
experimental data (Gnaiger et al., 1998; Scandurra and Gnaiger, 2010) are well fitted by such a curve. As a hyperbola, it can be represented by Eq. (2):

\[
\dot{V}_O_2 = \frac{\dot{V}_M_{AX} \cdot P_mO_2}{P_mO_2 + P_{50}}
\]

where \(\dot{V}_O_2\) is mitochondrial \(V_o_2\) (the ordinate in Fig. 1); \(\dot{V}_M_{AX}\) is the asymptote of the curve, and represents the maximal rate of use of \(O_2\) when \(O_2\) is in excess; \(P_mO_2\) is mitochondrial \(P_{O_2}\) (the abscissa in Fig. 1) and \(P_{50}\) is the \(P_{O_2}\) at 50% of \(V_M_{AX}\). Thus, the mitochondrial respiration curve is defined by two parameters: \(V_M_{AX}\) and \(P_{50}\).

Also shown in Fig. 1 is a straight (dashed) line of negative slope. It represents the Fick law of diffusion and depicts diffusive \(O_2\) transport between the tissue capillary and the mitochondria as a function of mitochondrial \(P_{O_2}\), for a given tissue \(O_2\) diffusional conductance (DM) and a given tissue mean capillary \(P_{O_2}\) (\(P_{O_2}^{+}\)), both at maximal exercise. We previously utilized this representation as a tool for interpreting intracellular oxygenation data obtained using magnetic resonance spectroscopy (Richardson et al., 1999).

The equation is as follows:

\[
\dot{V}_O_2 = DM \cdot (P_{O_2}^{+} - P_{O_2})
\]

As the figure indicates, as \(P_mO_2\) is increased, \(\dot{V}_O_2\) in Eq. (3) must fall because the \(P_{O_2}\) difference between mean capillary and mitochondrial \(P_{O_2}\) is reduced. Thus, Fig. 1 shows how \(\dot{V}_O_2\) increases with mitochondrial \(P_{O_2}\) according to oxidative phosphorylation, but decreases with mitochondrial \(P_{O_2}\) according to the laws of diffusion.

The key concept in Fig. 1 is that in a steady state of \(O_2\) consumption, \(\dot{V}_O_2\), given by both Eqs. (2) and (3) must be the same at the same mitochondrial \(P_{O_2}\) (i.e., the law of mass conservation applies). This can occur only at the single point of intersection between the two relationships, as indicated by the solid circle placed there. If, as previously approximated (Wagner, 1996b), mitochondrial \(P_{O_2}\) were truly zero, \(\dot{V}_O_2\) would be higher, as indicated by the open circle at the left end of the dashed straight line in Fig. 1. For a given \(O_2\) transport system defined by the conductances for \(O_2\) allowed by ventilation, alveolar-capillary diffusion, circulation, and capillary to mitochondrial diffusion, the values of mitochondrial \(V_M_{AX}\) and \(P_{50}\) (Eq. (2)) will thereby influence maximal rate of \(O_2\) utilization, \(\dot{V}_M_{AX}\). In the remainder of this paper, it will be important to distinguish between \(V_M_{AX}\) (the asymptote to the mitochondrial respiration curve) and \(V_{O_2}^{max}\) (actual maximal rate of \(O_2\) utilization, solid circle in Fig. 1) to avoid confusion. In general, \(V_M_{AX}\) can exceed \(\dot{V}_O_2^{max}\), but \(\dot{V}_O_2^{max}\) cannot exceed \(V_M_{AX}\).

2. Material and methods

2.1. Principles

Oxidative phosphorylation ensues via the following Eq. (1) that embodies the law of mass action:

\[
3ADP + 3Pi + NADH + H^+ + 1/2O_2 \rightarrow 3ATP + NAD + H_2O + \dot{V}_O_2
\]

In this equation, \(P_{mO_2}\) corresponds to \(O_2\). Clearly, this mass action equation can only move from left to right and produce ATP if \(P_{mO_2}\) is greater than zero.

To illustrate this effect, a graphical depiction of mitochondrial respiration is presented in Fig. 1. Here, the solid line is the relationship between velocity of the reaction (i.e., mitochondrial \(V_{O_2}\)), and \(P_{mO_2}\), similar to what has been found experimentally (Gnaiger et al., 1998; Scandurra and Gnaiger, 2010; Wilson et al., 1977). It shows how \(\dot{V}_{O_2}\) is a positive but non-linear function of mitochondrial \(P_{O_2}\), and indicates that at low \(P_{mO_2}\), \(\dot{V}_{O_2}\) is very sensitive to (and thus limited by) \(P_{O_2}\), while at higher \(P_{mO_2}\), \(\dot{V}_{O_2}\) becomes independent of \(P_{O_2}\), and is limited by factors other than \(O_2\).

The hyperbolic curve through the origin displayed in Fig. 1 represents mitochondrial respiration. It is of note that despite mitochondrial respiration kinetics is not really a Michaelis–Menten type (Johnson and Goody, 2011; Michaelis and Menten, 1913), changes may be affected by this interaction. Thus, the significance of the present study is in the degree to which \(V_{O_2}^{max}\) is reduced by the resistance imparted by oxidative phosphorylation and the consequent effect on mitochondrial \(P_{O_2}\), which in turn may affect processes such as generation of reactive oxygen species and hypoxia-induced gene expression.

The purpose of the present paper is therefore to expand the prior theoretical analysis of the integrated \(O_2\) transport pathway (Wagner, 1993, 1996a) by analyzing the consequences for \(O_2\) transport of allowing mitochondrial \(P_{O_2}\) to be greater than zero. This requires integration of the previously described \(O_2\) transport equations with an equation for mitochondrial respiration, followed by the application of mass conservation principles to solve this new equation system. The same data that were used in (Wagner, 1993, 1996a) are used here.

The present study augments our prior approach (Wagner, 1993, 1996b) by adding Eq. (2) to the equation system used previously. Fig. 2 recapitulates the \(O_2\) transport pathway, and the associated four mass conservation equations governing \(O_2\) transport at each step. It adds Eq. (2), describing \(O_2\) utilization as a function of \(P_{mO_2}\). The important point is that in this way, the system has expanded from four equations with four unknowns into a system of five equations and five unknowns.

Briefly, using specified input values for \(O_2\) transport stage parameters (i.e., values of inspired \(O_2\) fraction (\(F_{O_2}\)), ventilation (VI; inspired; \(VA\); expired), lung diffusing capacity (DL), cardiac output (Q; Hb), acid base status, tissue (muscle) diffusing capacity (DM), and mitochondrial respiration curve parameters (\(V_M_{AX}\) and \(P_{50}\)), five mass conservation equations are written for \(O_2\) (see Fig. 2). They describe (a) ventilatory transport; (b) alveolar–capillary diffusion; (c) circulatory transport; (d) muscle
Capillary–mitochondrial diffusion; and (e) mitochondrial respiration.

There are five unknowns in these equations: Alveolar PO$_2$ ($PAO_2$), arterial PO$_2$ ($PaO_2$), venous PO$_2$ ($PvO_2$), mitochondrial PO$_2$ ($PmO_2$), and $V_A$ itself. In Fig. 2, Eqs. (b) and (d) are differential equations describing the process of diffusion across the lung blood: gas barrier and across the tissue capillary wall, respectively. They specifically describe the time rate of change of O$_2$ concentration, $[O_2]$, along the respective capillary as a function of the diffusing capacity, blood flow, red cell capillary transit time ($T_L$ (lungs); $T_M$ (tissues)) and the instantaneous difference between upstream and downstream PO$_2$ values (alveolar and pulmonary capillary in (b); capillary and mitochondrial in (d)). The two equations are each expressions of the Fick law of diffusion.

The additional inputs of mitochondrial $V_{MAX}$ and $P_{50}$, and the additional coding for the fifth equation were added to the prior model, and the same (numerical) method of solution employed before (Wagner, 1996b) was used to find the solutions for any set of input variables, defined as the unique values of the five unknowns listed above that simultaneously satisfy all five equations for the given input data defining O$_2$ transport and utilization.

### 2.3. Input data for simulations

The input data defining the O$_2$ pathway parameters used in this analysis were essentially identical to those used previously (Wagner, 1996b), and come from Operation Everest II (Sutton et al., 1988). They reflect maximal exercise by normal subjects at sea level, at a chamber “altitude” of 4573 m (approximately 15,000 ft.) and at the chamber altitude of the Everest summit, 8848 m (approximately 29,000 ft.). They are reproduced in Table 1. It is clear that data do not exist for the two new key variables: mitochondrial $V_{MAX}$ and $P_{50}$. Therefore, for each of the three data sets we computed solutions to the equation system over a systematic range of five mitochondrial $V_{MAX}$ (1000, 2000, 3000, 4000, and 5000 ml/min) and four mitochondrial $P_{50}$ values (0.1, 0.3, 0.5 and 1.0 mm Hg), resulting in 20 combinations of the two, and thus 20 mitochondrial respiration curves. The values of $V_{MAX}$ were chosen to encompass the range of $V_{O2 max}$ from the very sedentary to the elite athlete. Values of $P_{50}$ on the other hand were based on physiological studies inscribing the mitochondrial respiration curve from samples of normal muscle (Gnaiger et al., 1998; Scandurra and Gnaiger, 2010).

A typical example from one of these papers is reproduced with permission in Fig. 3, where the hyperbolic character of the curve and its $P_{50}$ can both be seen by the fitted curve. In this and other similar published cases (Gnaiger et al., 1998; Scandurra and Gnaiger, 2010), $P_{50}$ is close to 0.3 mm Hg. This accounts for our choice of $P_{50}$ values – from a third of this typical value to about threefold greater. However it should be stressed that the modeling can be based on

![Graphical depiction of the hyperbolic equation for oxidative phosphorylation fitted to the data of Scandurra and Gnaiger (2010), p16.](image-url)
any combination of $P_{50}$ and $\dot{V}_{\text{MAX}}$ and need not be limited to the choice of specific parameters appearing here.

### 2.4. Analysis

Across the matrix of $V_{\text{MAX}}$ and $P_{50}$ values, we posed two questions: First we asked how much would $P_{\text{mO}_2}$ have to rise above zero to satisfy mass conservation and drive mitochondrial respiration for the given set of physiological O2 transport variables, $V_{\text{MAX}}$ and $P_{50}$ and as a result, how much would that cause $\dot{V}_O_2$ to be reduced (compared to assuming $P_{\text{mO}_2} = 0$) as per Fig. 1 (comparing the open and closed circles). This question allows a quantitative description of the theoretical consequences for $\dot{V}_O_2$ max of any combination of mitochondrial $V_{\text{MAX}}$ and $P_{50}$. While this is a very useful question to answer, in reality $\dot{V}_O_2$ max is a directly measured variable. Therefore, asking how much would it be reduced by any pair of $V_{\text{MAX}}$ and $P_{50}$ values is hypothetical. On the other hand, muscle diffusing capacity, DM, is a variable calculated on the assumption that mitochondrial $P_{\text{O}_2}$ can be neglected and set to zero – the very approximation that the present study is addressing.

Thus, another way to interrogate the model system can be proposed, leading to a second question: It recognizes that the muscle O2 diffusion step was previously modeled, and muscle diffusing capacity estimated, on the basis of $P_{\text{mO}_2} = 0$. However, if $P_{\text{mO}_2}$ is greater than zero, the capillary to mitochondrial O2 diffusion gradient would be reduced, and this would necessitate, by the Fick law of diffusion, a higher value of DM to accomplish a given, measured $\dot{V}_O_2$ max (compared to the value calculated assuming $P_{\text{mO}_2} = 0$).

Therefore, for each of the combinations of $V_{\text{MAX}}$ and $P_{50}$, we asked how much would muscle diffusing capacity have to increase to maintain $\dot{V}_O_2$, constant at the measured value as a result of $P_{\text{mO}_2}$ being greater than zero.

### 3. Results

#### 3.1. Effects of mitochondrial respiration on $P_{\text{mO}_2}$ and maximal $\dot{V}_O_2$

Fig. 4 shows how the different combinations of mitochondrial $V_{\text{MAX}}$ and $P_{50}$ affect $\dot{V}_O_2$ max. The upper panel covers the mitochondrial $P_{\text{O}_2}$ ($P_{\text{mO}_2}$) range from 0 to 20 mm Hg; the lower panel shows the same data, but expands the abscissa to better reflect the lower $P_{\text{mO}_2}$ range between 0 and 5 mm Hg. In both panels, each solid curved line emanating from the origin represents one of the 20 mitochondrial respiration curves (as in Fig. 3) for a particular $V_{\text{MAX}}$ and $P_{50}$ combination. Solid circles reflect sea level conditions; solid squares represent moderate altitude and solid triangles are for the equivalent of the Everest summit. It turns out that at each altitude, an approximately straight line can be drawn through the resulting $\dot{V}_O_2$ max/$P_{\text{mO}_2}$ solution points for each mitochondrial respiration curve. These are the dashed lines in the figure.

The values of $\dot{V}_O_2$ max at each altitude at the point where $P_{\text{mO}_2}$ equals zero (open symbols at zero $P_{\text{mO}_2}$) are the same as those described in (Wagner, 1996b) where $P_{\text{mO}_2}$ was taken to be zero. The figure shows how relaxing that approximation affects $\dot{V}_O_2$ max for each combination of mitochondrial $V_{\text{MAX}}$ and $P_{50}$.

At sea level (solid circles), results show that allowing for a non-zero $P_{\text{mO}_2}$ has a small but significant impact on $\dot{V}_O_2$ max. For example, $\dot{V}_O_2$ max at $P_{\text{mO}_2} = 0$ mmHg (open circle) would be 3827 ml/min, but if $V_{\text{MAX}}$ were 4000 ml/min and $P_{50}$ 1.0 mm Hg, $\dot{V}_O_2$ max would be significantly less, by 9%, and would be 3477 ml/min. Moreover, this would require a mitochondrial $P_{\text{O}_2}$ of 6.7 mm Hg to drive oxidative phosphorylation, as the figure shows.

Fig. 4. Effects of considering mitochondrial respiration on maximal $\dot{V}_O_2$ and mitochondrial $P_{\text{O}_2}$. For each $V_{\text{MAX}}$ value, the four hyperbolic curves represent $P_{50}$ values of 0.1, 0.3, 0.5 and 1.0 mm Hg, left to right. See text for details.

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Table I

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Normal subjects at...</th>
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</thead>
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<tr>
<td></td>
<td>Sea level</td>
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<tr>
<td>Barometric pressure (PB), Torr</td>
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<tr>
<td>Fractional inspired oxygen (FrO2)</td>
<td>0.2093</td>
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<tr>
<td>Alveolar ventilation ($\dot{V}$, L/min)</td>
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<tr>
<td>Blood flow ($Q$, L/min)</td>
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<tr>
<td>Hemoglobin concentration ([Hb]) (g dL⁻¹)</td>
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<tr>
<td>Body temperature ($T_b$, °C)</td>
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<tr>
<td>O2 dissociation curve $P_{\text{O}_2}$ (Torr)</td>
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</tr>
<tr>
<td>Total lung O2 diffusing capacity (DL) (ml/min Torr⁻¹)</td>
<td>51</td>
</tr>
<tr>
<td>Total muscle O2 diffusing capacity (DM) (ml/min Torr⁻¹)</td>
<td>102</td>
</tr>
<tr>
<td>Maximum O2 uptake ($\dot{V}_{\text{O}_2}$, ml/min)</td>
<td>3.82</td>
</tr>
</tbody>
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In general, for the fixed set of O2 transport parameters used (see Table 1), the lower the VMAX and the higher the P50, the greater is the reduction in VO2, and the higher is the PmO2, required to drive ATP generation. The range of possible values of mitochondrial P02 is considerable, from a fraction of a mm Hg to more than 10 mm Hg, depending on VMAX and P50.

The same outcome is seen at each altitude, but with VO2 lower at any PmO2 as PMO2 is reduced. The reduction in VO2, per unit change in PmO2, is somewhat less at altitude than at sea level, but if examined as a percent of VO2 at PmO2 = 0 at each altitude, the effects of allowing for mitochondrial respiration on maximal VO2 are relatively similar across altitudes.

In summary, the higher the mitochondrial VMAX and the lower the P50, the more O2 can be metabolized for a given upstream (heart, lungs, blood, muscle) transport system. Mitochondrial P02 at VO2 max can be neglected when considering O2 transport only when mitochondrial P50 is low and mitochondrial VMAX is high. When VMAX is low and/or P50 is high, the mitochondrial P02 required to drive oxidative phosphorylation may reach double digit values, and the impact on VO2 max can be considerable.

3.2. Maintenance of maximal VO2 in the face of non-zero PmO2

The preceding subsection showed how VO2 max would have to decrease as a function of mitochondrial VMAX and P50 with constant values for all O2 transport conduances. In this subsection we investigate how much higher the muscle O2 diffusing capacity would have to be to maintain VO2 max constant over the same range of VMAX and P50 values as PmO2 increases above zero.

The results are shown in Fig. 5, which displays the simulation outcomes across the entire matrix of VMAX and P50 values, using VO2 on the abscissa and isopleths for each PmO2. Results are shown for each altitude as indicated by the different symbols. The top panel shows mitochondrial P02 for every combination of VMAX and P50 examined, and the bottom panel the corresponding values of muscle diffusing capacity (DM), that would have to exist to maintain VO2 max at measured levels (indicated at each altitude by the vertical dashed lines). Comparing panels shows that when PmO2 is high (thus reducing the P02 gradient between capillaries and mitochondria, DM must also be high to maintain diffusive O2 transport.

Also, when mitochondrial VMAX substantially exceeds measured VO2 max (at each altitude), PmO2 remains low, and therefore DM does not need to be substantially increased to maintain O2 flux. However, the closer mitochondrial VMAX is to measured VO2 max, the higher PmO2 must be (see Fig. 4), and therefore, DM is also required to be elevated to maintain O2 transport. When VMAX and actual VO2 max are very close, the required DM may be as much as four times the value needed when PmO2 is (close to) zero, and the associated PmO2 would reach double digit values.

4. Discussion

4.1. Summary of major findings

This study shows that including mitochondrial respiration in analyzing O2 transport and utilization generally poses a very small additional resistance to the system (over that of the transport pathway alone), only slightly reducing VO2 max below that computed ignoring this contribution (Fig. 4). The associated mitochondrial P02 is also usually low (<1 mm Hg). If however mitochondrial VMAX is low in relation to O2 transport capacity, or if mitochondrial P50 is high, VO2 max may be considerably reduced. Mitochondrial P02 would then increase more, and may reach double digit values.

In order to maintain VO2 max when mitochondrial P02 is high, muscle diffusing capacity (DM) would need to be higher than when PmO2 is assumed to be zero. Under most conditions, the necessary increase in DM would be minimal, being significant only when PmO2 is considerably elevated (Fig. 5).

4.2. Unifying principles

The main principle demonstrated in the present study is that the final step in the O2 pathway – mitochondrial respiration– may contribute a non-negligible resistance to O2 movement through the system from the air to its conversion to CO2, resulting in a lower VO2 max compared to a system where metabolism imposed no resistance to overall O2 flow. The higher the mitochondrial P02 required to drive oxidative phosphorylation, the greater would be the relative resistance and thus the more effect there will be on reduction in VO2 max. When mitochondrial P50 is about 0.30 mm Hg as reported by Gnaiger (Gnaiger et al., 1998; Scandurra and Gnaiger, 2010) the effects are generally minor.

The simulations at the three inspired P02 values shown here demonstrate that it is the relative capacities (rather than individual absolute values) of the physiological transport system and the mitochondrial respiratory chain that effectively determine both the mitochondrial PO2 and the associated effect on VO2 max, and that both variables, but especially mitochondrial PO2, may vary over a wide range depending on mitochondrial respiratory function.

change systematically with training, or in chronic diseases such as chronic obstructive pulmonary disease (COPD) or chronic heart failure. The calculations presented herein however point out that the quantitative nature of the mitochondrial respiration curve may be a critical determinant of the values of mitochondrial \( \text{P} \text{O}_2 \) and \( \dot{V}_{\text{O}_2} \) max, over and above any influence of upstream \( \text{O}_2 \) transport. Even if the effects on \( \dot{V}_{\text{O}_2} \) max are numerically small, they would likely be important in the competitive endurance athlete where very small differences may separate success from failure. But possibly even more significant might be the potentially large variation in mitochondrial \( \text{P} \text{O}_2 \) depending on \( \text{P} \text{O}_2 \) and \( \dot{V}_{\text{O}_2} \) max due to known hypoxia-induced biological effects (Semenza, 2011). Thus, hypoxia-induced gene expression or reactive \( \text{O}_2 \) species generation may vary according to mitochondrial \( \text{P} \text{O}_2 \).

4.4. Potential for estimating mitochondrial \( \text{P} \text{O}_2 \) based on the current modeling approach

The analysis presented here suggests a possible method for estimating the characteristics of the mitochondrial respiration curve in vivo. Currently, mitochondrial \( \dot{V}_{\text{MAX}} \) and \( \text{P} \text{O}_2 \) are measured in vitro in respirometers where mitochondria are exposed to different levels of \( \text{O}_2 \) and \( \text{V}_\text{O}_2 \), measured as in Fig. 3, (Gnaiger et al., 1998; Scandurra and Gnaiger, 2010; Wilson et al., 1977). To obtain this information in humans would therefore necessitate a muscle biopsy, and even if that were done, the result would be subject to the usual sampling constraints as for any other measure of muscle structure or function determined from a single biopsy.

The fitting of a hyperbolic function to paired measured values of \( \dot{V}_{\text{O}_2 \text{max}} \) and mitochondrial \( \text{P} \text{O}_2 \) has the potential for estimating \( \text{P} \text{O}_2 \) and \( \dot{V}_{\text{MAX}} \) in vivo, and this is illustrated in Fig. 7. The intervention to garner several points on the curve would come from acutely varying \( \text{FIO}_2 \) and measuring \( \text{V}_{\text{O}_2} \) and mitochondrial \( \text{P} \text{O}_2 \) during maximal exercise at each \( \text{FIO}_2 \), as indicated by the theoretical example of the two solid circles in the upper panel of Fig. 7. These two points reflect a mitochondrial respiration curve with \( \text{P} \text{O}_2 \) of 0.30 mm Hg and \( \dot{V}_{\text{MAX}} \) of 4000 ml/min. If such data were to span both the steep and flat parts of the respiration curve, as shown in the figure, identifying the \( \dot{V}_{\text{MAX}} \) and \( \text{P} \text{O}_2 \) of a hyperbola that resulted in least squares best fit to the data points would be possible, as shown in the lower panel of Fig. 7. Here, over a range of trial values of both \( \dot{V}_{\text{MAX}} \) and \( \text{P} \text{O}_2 \), the root mean square (RMS) residual \( \dot{V}_{\text{O}_2} \) between the data and the hyperbola corresponding to each trial combination of \( \text{P} \text{O}_2 \) and \( \dot{V}_{\text{MAX}} \) providing the lowest RMS for that \( \text{P} \text{O}_2 \) is shown. In this error-free theoretical case, one could quite accurately estimate \( \dot{V}_{\text{MAX}} \) (4000 ml/min) and \( \text{P} \text{O}_2 \) (0.3 mm Hg) from the values at the nadir of the relationship in the figure. However, if measured data happened to lie on only the flat or only on the steep parts of the curve, ability to estimate \( \dot{V}_{\text{MAX}} \) and/or \( \text{P} \text{O}_2 \) would be considerably reduced.

While whole body or large muscle mass \( \dot{V}_{\text{O}_2} \) can be measured relatively easily, the experimental challenge would be to measure mitochondrial \( \text{P} \text{O}_2 \) (during exercise) (Mik, 2013). The closest approach to date in intact subjects has used MRS-based determination of myoglobin \( \text{O}_2 \) saturation (Jue et al., 1994; Richardson et al., 1995), where the signal comes from a relatively large muscle region. This approach gives intracellular \( \text{P} \text{O}_2 \) estimates of 3–4 mm Hg during exercise (Richardson et al., 1995), but this is the \( \text{P} \text{O}_2 \) associated with myoglobin, inferred from the finding of about 50% myoglobin saturation during peak exercise combined with accepted values of myoglobin \( \text{P} \text{O}_2 \) of about 3 mm Hg (Rossi-Fanelli and Antonini, 1958). This \( \text{P} \text{O}_2 \) is an order of magnitude greater than that projected at the mitochondria based on the preceding discussion. In the end, a method would have to be developed for direct measurement of mitochondrial \( \text{P} \text{O}_2 \). Whether a candidate signaling atom

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Fig. 6. Graphical depiction of the concept that even when the capacity for \( \text{O}_2 \) delivery exceeds \( \text{O}_2 \) utilization (upper panel) a change in \( \text{O}_2 \) delivery will change actual \( \dot{V}_{\text{O}_2} \). Conversely, when the capacity for \( \text{O}_2 \) utilization exceeds \( \dot{V}_{\text{O}_2} \) delivery (lower panel) a change in \( \text{O}_2 \) utilization (increase in \( \text{P} \text{O}_2 \) in this example) will change actual \( \dot{V}_{\text{O}_2} \). Open circles: maximal \( \text{O}_2 \) delivery to mitochondria if \( \text{P} \text{O}_2 \) was zero. Closed circles: actual \( \dot{V}_{\text{O}_2} \). Solid and dashed lines: as in Fig. 1.

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An additional important principle is shown in Fig. 6: even when \( \text{O}_2 \) transport capacity (i.e., potential for \( \text{O}_2 \) delivery) is considerably greater than mitochondrial respiratory capacity (i.e., potential for \( \text{O}_2 \) utilization), as illustrated in concept in the top panel, a change in the former will change overall \( \dot{V}_{\text{O}_2} \). The converse is also true – that when mitochondrial respiratory capacity exceeds \( \dot{V}_{\text{O}_2} \) transport capacity, (lower panel), a change in the former will have an effect on \( \dot{V}_{\text{O}_2} \). It is thus not correct to think that when one component is greater than the other, only the lesser of the two determines overall \( \dot{V}_{\text{O}_2} \) max. This conclusion is much the same as described for individual components of the physiological transport pathway of the lungs and chest wall, the heart, blood circulation, and the muscles, where we previously showed (Wagner, 1996a,b) that all components affect \( \dot{V}_{\text{O}_2} \) max, not just the step with the least transport capacity.

4.3. Effects of mitochondrial respiration kinetics on both \( \dot{V}_{\text{O}_2 \text{max}} \) and \( \text{P} \text{O}_2 \) may be small or large

For the examples shown – fit normal subjects – the effects of considering mitochondrial respiration are generally less on \( \dot{V}_{\text{O}_2} \) than on the associated \( \text{P} \text{O}_2 \) (Fig. 4). Examining the sea level results for the example of \( \dot{V}_{\text{MAX}} = 4000 \text{ ml/min} \) and \( \text{P} \text{O}_2 \) increasing from 0.1 to 1.0 mm Hg, \( \dot{V}_{\text{O}_2} \) max would fall by 9% while \( \text{P} \text{O}_2 \) would increase by an order of magnitude, from less than 1 mm Hg to more than 6 mm Hg. Just how much variation there is in mitochondrial \( \text{P} \text{O}_2 \) in the normal population is unknown, let alone whether this may
or molecule can be found for an MRS-based approach is currently unknown.

4.5. Limitations of the analysis

As in previous work (Wagner, 1993, 1996b), the entire analysis is applicable only to steady state conditions (meaning, that O2 partial pressures are constant in time as is VO2 itself). Therefore, the analysis cannot be used to study transient changes in metabolic rate. Another limitation is not taking into account ventilation–perfusion mismatch in the lung and/or metabolism–perfusion mismatch in the muscle as contributors to impaired oxygen transport. However, using methods to quantify both of these phenomena, this limitation could be removed. A final limitation is that non-muscle blood flow during maximum exercise is neglected.

5. Conclusions

Considering the hindrance to overall O2 flux caused by mitochondrial respiration using an established model of O2 transport to the mitochondria revealed that in normal subjects exercising maximally, the step of oxidative phosphorylation, with its requirement for a mitochondrial PO2 > 0, likely plays only a small role in total O2 flux resistance. However, we identified conditions in which mitochondrial PO2 can rise to double digit values. This occurs particularly when the mitochondrial respiration curve has either a low VMAX (relative to O2 transport), or a high P50, and under such conditions, mitochondrial function may significantly impair O2 flux and cell function may be affected, for example, in reactive oxygen species generation and/or oxygen-sensitive gene expression.

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References


