Effect of sepsis on skeletal muscle oxygen consumption and tissue oxygenation: interpreting capillary oxygen transport data using a mathematical model

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Submitted 15 September 2003; accepted in final form 11 August 2004

Goldman, Daniel, Ryon M. Bateman, and Christopher G. Ellis. Effect of sepsis on skeletal muscle oxygen consumption and tissue oxygenation: interpreting capillary oxygen transport data using a mathematical model. Am J Physiol Heart Circ Physiol 287: H2535–H2544, 2004. First published August 19, 2004; doi:10.1152/ajpheart.00889.2003.—Inherent in the inflammatory response to sepsis is abnormal microvascular perfusion. Maldistribution of capillary red blood cell (RBC) flow in rat skeletal muscle has been characterized by increased 1) stopped-flow capillaries, 2) capillary oxygen extraction, and 3) ratio of fast-flow to normal-flow capillaries. On the basis of experimental data for functional capillary density (FCD), RBC velocity, and hemoglobin O2 saturation during sepsis, a mathematical model was used to calculate tissue O2 consumption (VO2), tissue PO2 (P) profiles, and O2 delivery by fast-flow capillaries, which could not be measured experimentally. The model describes coupled capillary and tissue O2 transport using realistic blood and tissue biophysics and three-dimensional arrays of heterogeneously spaced capillaries and was solved numerically using a previously validated scheme. While total blood flow was maintained, capillary flow distribution was varied from 60/30/10% (normal/fast/stopped) in control to 33/33/33% (normal/fast/stopped) in average sepsis (AS) and 25/25/50% (normal/fast/stopped) in extreme sepsis (ES). Simulations found approximately two- and fourfold increases in tissue VO2 in AS and ES, respectively. Average (minimum) P, decreased from 43 (40) mmHg in control to 34 (27) and 26 (15) mmHg in AS and ES, respectively, and clustering fast-flow capillaries (increased flow heterogeneity) reduced minimum PO2 to 14.5 mmHg. Thus, although fast capillaries prevented tissue dysoxia, they did not prevent increased hypoxia as the degree of microvascular injury increased. The model predicts that decreased FCD, increased fast flow, and increased VO2 in sepsis expose skeletal muscle to significant regions of hypoxia, which could affect local cellular and organ function.

sepsis, the systemic inflammatory response to bacterial infection, is the leading cause of death in North American intensive care units. Recent advances in the treatment of septic patients, including early goal-directed therapy to adjust cardiac preload, afterload, and contractility to balance oxygen delivery with oxygen demand (39), and the administration of activated protein C (8), have significantly reduced mortality. Both morbidity and mortality rates due to multiple organ failure remain high, however, and the underlying pathophysiology of sepsis remains unresolved (6, 11, 31, 23). Septic patients may also experience difficulty during withdrawal of mechanical ventilation because their respiratory muscles have weakened or been injured (26, 1, 42). Whether remote organ dysfunction and muscle weakness result from acute cellular injury, abnormal mitochondrial function (17), or abnormal oxygen transport (15) is still undetermined.

Recent clinical evidence of decreased microvessel density in the sublingual microcirculation of fluid-resuscitated septic patients (14, 27, 44) is consistent with findings of decreased functional capillary density and abnormal blood flow in the gut, liver, and skeletal muscle microcirculation in animal models of sepsis. This clinical finding raises the possibility that abnormal microvascular O2 transport develops in multiple organs despite fluid resuscitation, leading to heterogeneous microvascular dysfunction and local tissue hypoxia in severe cases of sepsis. In skeletal muscle, where the impact of sepsis has been widely studied, sepsis has been shown to produce a severe derangement of the microcirculation, which is manifested by increased flow heterogeneity (5, 15, 23, 30, 34, 35, 9) within individual capillary beds such that some capillaries stop flowing, others retain their normal flow behavior, and others experience increased red blood cell (RBC) supply rates (15). While studies have shown that tissue oxygen tension (PO2) decreases in skeletal muscle in animal models of sepsis (2, 4, 47), it is not clear how abnormalities in microvascular blood flow and its regulation affect the spatial heterogeneity of tissue oxygenation. In particular, because PO2 microelectrodes provide data on a relatively large scale (~100 μm), the importance of capillary-scale (~5 μm) heterogeneities and the possible presence of highly localized regions of hypoxia or even anoxia could not be determined in these studies.

Whereas anoxia denotes a PO2 of zero, dysoxia indicates not a specific PO2 value but rather a state in which the local tissue O2 concentration, given the overall cellular environment, is insufficient to support normal mitochondrial oxidative metabolism (12). When O2 supply is the only relevant factor, as in the present study, dysoxia is usually considered to require PO2 values of less than about 1 mmHg. Therefore, because it is more relevant physiologically and can, in our case, be related to a specific PO2 value, we will generally use the term dysoxia rather than anoxia.

To determine the mechanisms responsible for abnormal tissue oxygenation, one must have simultaneous information on the determinants of microvascular O2 transport, including...
microvascular geometry, RBC supply rate and distribution, RBC hemoglobin O₂ saturation (SO₂), and tissue O₂ consumption (V\(\dot{O}_2\)). Because the complete data needed to resolve these issues are difficult to obtain experimentally, we have developed a detailed, experiment-based computational model to study the effect of sepsis-induced changes in microvascular blood flow on tissue oxygen distributions. This model allows us to estimate the local tissue V\(\dot{O}_2\) rate (M\(_0\)) and calculate steady-state tissue oxygen distributions for several normal and septic cases.

Recently, in vivo experimental results were published on capillary density, hemodynamics, and RBC oxygenation in the rat extensor digitorum longus (EDL) muscle under control and septic conditions using a 24-h normotensive, fluid-resuscitated cecal ligation and perforation (CLP) model (15). Results from the Ellis et al. study (15) showed that the character of capillary blood flow changed significantly in sepsis despite immediate fluid resuscitation and maintenance of mean arterial pressure over the experimental period. Indeed, proinflammatory mediators independent of hypotension (32, 9), changes in hemorheology (5) and loss of microvascular control mechanisms have been described as factors in abnormal microvascular perfusion. In the Ellis et al. study (15), increased flow heterogeneity was characterized by an increase in the proportion of capillaries with stopped flow, an increase in capillaries with RBC velocities (\(v_{\text{RBC}}\)) >325 \(\mu\)m/s (“fast” capillaries), and an increase in oxygen extraction from “normal” capillaries (\(v_{\text{RBC}} < 325 \, \mu m/s\)) that was directly proportional to the increase in stopped-flow capillaries. Hemodynamic and oxygenation data were not obtained for the fast-flow capillaries due to technical limitations.

The above results suggest that tissue V\(\dot{O}_2\) may increase under the sepsis conditions studied; however, because of insufficient data for the fast-flow capillaries it is not possible to determine exactly how V\(\dot{O}_2\) changes during sepsis. In addition, because no tissue O₂ measurements were made, the experimental data do not indicate to what extent tissue oxygenation was altered or whether tissue dysoxia occurred during sepsis, although previous studies (2, 4, 47) suggest some degree of hypoxia is expected. As will be shown below, the difficulty in extracting information on V\(\dot{O}_2\) and tissue oxygenation from the experimental data is directly related to the complexity of microvascular geometry and blood flow. Because of this complexity, highly simplified oxygen transport models are not appropriate for interpreting measurements of microvascular blood flow and RBC oxygenation during sepsis.

Theoretical studies over the last 15 years (7, 16, 20, 24, 41; see also Ref. 36) have shown that accurately representing microvascular oxygen transport requires including a realistic degree of physiological complexity, in particular, heterogeneity of microvessel spacing and blood flow, and diffusive interactions between microvessels. These features are particularly important for modeling situations such as sepsis in which the local oxygen supply may be relatively low. Therefore, to properly interpret data on capillary blood flow and oxygen transport during sepsis, it is necessary to use a theoretical model that includes realistic spatial heterogeneity and intercapillary transport.

In what follows, we first describe our theoretical model of heterogeneous, three-dimensional oxygen transport in EDL capillary networks under control and sepsis conditions. We then use the heterogeneous model to estimate V\(\dot{O}_2\), calculate tissue PO₂ distributions, and determine the effect of fast-flow vessels on tissue oxygenation for various control and sepsis cases, based on experimental oxygen transport data in normally flowing capillaries. Next, we explore the sensitivity of our results on tissue oxygenation to our V\(\dot{O}_2\) estimates and other model properties. Finally, we discuss the implications of our results for understanding skeletal muscle O₂ delivery and utilization during sepsis. The main goal of this work is to understand how changes in microvascular geometry and blood flow may be affecting tissue oxygenation and capillary oxygen extraction in sepsis.

**MATHEMATICAL MODEL**

**EDL capillary morphology and hemodynamics.** We have previously described a capillary network model for the hamster cheek pouch retractor muscle that is based on the muscle fiber structure of striated muscle and measured morphometric parameters (19). This model includes capillaries parallel to the muscle fibers and anastomotic cross-connections between the parallel capillaries. In the present case, we adapt this muscle fiber-based construction to the rat EDL muscle. As an initial simplification we do not consider anastomoses, because sampled capillaries did not have anastomoses (15). Experimental data show that overall functional capillary density (FCD) is not strongly affected by sepsis (5, 15), which indicates that in the 24-h CLP model there is little change in interstitial volume due to edema. However, because there is some uncertainty about the appropriate baseline value for FCD per unit area (FCD\(_{\lambda}\)), two values at the ends of the measured range of 1,000–1,500 capillaries/mm\(^2\) (45) are used for each case studied. On the basis of experimental data (15), the arteriovenous length and the radius of the capillaries are fixed at 400 \(\mu\)m and 2.8 \(\mu\)m, respectively.

Results for the 24-h EDL CLP model (15) show that the average percent distribution of normal/fast/stopped vessels is \(\sim 60/30/10\) for the control case and 33/33/33 for the sepsis (CLP) case. The most extreme septic injury seen in the 24-h CLP model resulted in a normal/fast/stopped vessel distribution of \(\sim 25/25/50\). Here, we simulate the experimental results using tissue/capillary configurations consisting of 27 capillaries: 16 normal-, 8 fast-, and 3 stopped-flow capillaries to represent the (average) “control” case; 9 normal-, 9 fast-, and 9 stopped-flow capillaries for the “average sepsis” (AS) case; and 7 normal-, 7 fast-, and 13 stopped-flow capillaries for the “extreme sepsis” (ES) case. Both AS and ES microvascular injuries are considered for the same underlying degree of sepsis because it is felt that average (measured) properties alone may not adequately describe the physiological effects of sepsis in the microcirculation.

Figure 1 shows how the 24 flowing capillaries for the control case are distributed in two dimensions, where the fast and normal capillaries are selected randomly and the 3 stopped capillaries are not shown. For the sepsis cases, fast-, normal-, and stopped-flow capillaries are selected from the same set used for the control case. The cross-sectional area of our tissue domain is either 17,496 or 26,215 \(\mu\)m\(^2\) depending on the value used for FCD\(_{\lambda}\). The tube hematocrit is fixed at 0.25, consistent with the average experimental value (15), and the discharge hematocrit is calculated using an established empirical relation.
EFFECT OF SEPSIS ON SKELETAL MUSCLE OXYGEN TRANSPORT

Fig. 1. Distribution of flowing capillaries for the control case. A: tissue cross section; B: three-dimensional tissue domain.

(38). RBC velocities were initially fixed at 130 μm/s for normal-flow capillaries ($v_{\text{norm}}$), based on experimental findings, and set at either 500 or 1,000 μm/s for fast-flow capillaries ($v_{\text{fast}}$), because specific experimental data were not available but it was known that $v_{\text{fast}} > 325$ μm/s (15) and that RBC velocities on the order of 1,000 μm/s occur in the EDL (23) with averages in the range of 250–400 μm/s (30, 46). Global adjustments in capillary velocity (from −0.04% to +38%) were then made for the average and extreme sepsis cases to ensure the same total blood flow (or RBC supply rate) as for the corresponding control case. Because data on $v_{\text{fast}}$ were not available, it was necessary to make an assumption about total blood flow during sepsis, and this was the simplest choice. Although there is evidence that total flow decreases in the EDL during sepsis (30), we show below that our tissue oxygenation results are not qualitatively changed if a flow decrease is assumed.

Capillary oxygen transport. The oxygen transport model (18, 21) includes both hemoglobin (Hb)-bound and dissolved oxygen in the blood and an equilibrium-binding description of the effect of tissue myoglobin (Mb), with the intravascular and extravascular (tissue) spaces treated as separate volumes. In the blood vessels, which are assumed to have an approximately circular cross section, oxygen transport is described by a time-dependent equation for the fractional blood SO$_2$, $S(\xi,t)$:

$$\frac{\partial S}{\partial t} = \left( \frac{H_b C_{Ib} + \alpha_b \frac{dP_b}{dS}}{D_b} \right)^{-1} \left[ -v_b \left( H_b C_{Ib} + \alpha_b \frac{dP_b}{dS} \right) \frac{\partial S}{\partial \xi} - \frac{1}{\pi R} \phi j \cdot d\theta \right]$$

(1)

where $v_b$ is the mean blood velocity, $R$ is the vessel radius, and $\xi$ is the local axial coordinate. The two terms in parentheses represent the volume- and flow-weighted oxygen-carrying capacities of blood, where $H_b$ and $H_D$ are the tube and discharge hematocrits, $\alpha_b$ and $\alpha_d$ are the volume- and flow-weighted blood oxygen solubilities, and $C_{Ib}$ is the $O_2$-binding capacity of the Hb solution inside RBCs. $P_b(S) = P_{50}(S/(1 - S))^{1/n}$ is the blood PO$_2$ obtained by inverting the Hill equation for the Hb-O$_2$ equilibrium binding curve. The oxygen flux ($j$) at any point on the vessel/tissue interface is

$$j = k(P_b - P_w)$$

(2)

where $P_w$ is the tissue PO$_2$ at the vessel surface and $k$ is a mass transfer coefficient used to model intravascular transport resistance (19). The boundary condition on $S$ is the SO$_2$ in all inlet segments. In the present study, the blood velocities and hematocrits needed in Eq. 1 are assigned based on experimental data as described above.

Tissue oxygen transport. O$_2$ transport in the tissue is described by the following time-dependent equation for PO$_2$, $P(x,y,z,t)$:

$$\frac{\partial P}{\partial t} = \left( 1 + \frac{c_{Mb} \frac{dS_{Mb}}{dP}}{\alpha \frac{dP}{dP}} \right)^{-1} \left[ -\frac{1}{\alpha} \left( M(P) + D \nabla^2 P \right) + \frac{1}{\alpha} D_{Mb} c_{Mb} \nabla \cdot \left( \frac{dS_{Mb}}{dP} \nabla P \right) \right]$$

(3)

where $\alpha$ and $D$ are the tissue oxygen solubility and diffusivity and $c_{Mb}$, $D_{Mb}$, and $S_{Mb}$ are the Mb concentration, diffusivity, and oxygen saturation. Michaelis-Menten consumption kinetics are used to describe the PO$_2$-dependent $V_{O_2}$, $M(P) = M_0 P/(P + P_c)$, and Mb saturation is given by $S_{Mb}(P) = P/P_{50,Mb}$. At the vessel surface, a flux boundary condition on $P$ is applied using Eq. 2, and periodic boundary conditions are specified on the outer tissue surfaces in the $x$ and $y$ directions. On the outer tissue surfaces in the $z$ direction, which is taken to be the blood flow direction, no-flux boundary conditions are specified.

Numerical solution of model equations. To solve the above oxygen transport equations, a slightly modified version (18) of a previously validated (19) finite-difference-based, fully time-dependent numerical scheme is used. A three-dimensional Cartesian finite-difference discretization is used in the tissue, and the capillary network is discretized into effectively one-dimensional cylindrical segments. At the intersections of the Cartesian grid lines with the cylinder surfaces, the fluxes given
by Eq. 2 are calculated. These are used in Eq. 1 and in applying
the flux boundary condition on Eq. 3 at the vessel/tissue
interface. For the steady-state calculations used in the present
study, solutions in the tissue and capillary spaces are advanced
from an arbitrary initial condition until constant values are
reached.

Model parameters. Many of the parameters used in our
oxygen transport calculations, shown in Table 1, are the same
as used previously to study the hamster cheek pouch retractor
muscle (19, 20). However, it was necessary to alter several
blood and tissue parameters to make our model appropriate for
the rat EDL muscle. Typical values for the parameters in the
Hill equation for rat blood are $P_{50} = 37$ mmHg and $n = 2.7$
(15), in contrast to $P_{50} = 29$ mmHg and $n = 2.2$ for hamster
blood. Oxygen diffusion coefficients and solubilities should be
the same in the rat and hamster; however, the Mb concentration
in the EDL is lower than in the retractor. The measured value of $C_{Mb} \approx 1.3$ mg/g (3) was converted to obtain a binding
capacity of $2 \times 10^{-3}$ ml O$_2$/ml tissue at $37^\circ$C. In addition to
the above species-specific changes in O$_2$ transport parameters,
we used a slightly different Mb diffusivity than in our previous
studies: $D_{Mb} = 3 \times 10^{-7}$ cm$^2$/s, based on several recent exper-
imental results (28, 33, 49).

RESULTS

Estimates of oxygen consumption rate. Oxygen consumption
in the EDL has not been measured directly under control and
sepsis conditions. The average oxygen extraction ratio for the
normal-flow capillaries (ER$_{cn}$) was found to increase signif-
icantly in sepsis (15), but the extraction ratio for fast-flow
capillaries (ER$_{cf}$) was not measured. However, by using our
theoretical model to match the experimental data for ER$_{cn}$, we
were able to estimate M$_0$ per unit volume of parenchymal
tissue for the control and sepsis conditions. For each case, this
is done by starting with two initial guesses for the parameter
M$_0$ in Eq. 3 ($P_e$ is fixed for all cases) and then using linear
interpolation based on the calculated O$_2$ extraction ratios for
the normal flow capillaries (ER$_{cn,calc}$). In all cases, it was
verified that the final estimates of M$_0$ resulted in the correct
values for ER$_{cn,calc}$. For all O$_2$ transport simulations, the arteriolar-
end SO$_2$ in all capillaries was set to $S_a = 0.67$, the averaged
measured value for both control and sepsis conditions (15).

Table 1. Biophysical parameters for O$_2$ transport

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\alpha$</td>
<td>$3.89 \times 10^{-5}$ ml O$_2$/ml$^{-1}$-mmHg$^{-1}$</td>
</tr>
<tr>
<td>$D$</td>
<td>$2.41 \times 10^{-5}$ cm$^2$/s</td>
</tr>
<tr>
<td>$P_e$</td>
<td>0.5 mmHg</td>
</tr>
<tr>
<td>$k$</td>
<td>$7.51 \times 10^{-7}$ ml O$_2$/cm$^{-2}$-s$^{-1}$-mmHg$^{-1}$</td>
</tr>
<tr>
<td>$\bar{\alpha}$</td>
<td>$2.92 \times 10^{-5}$ ml O$_2$/ml$^{-1}$-mmHg$^{-1}$</td>
</tr>
<tr>
<td>$\bar{\alpha}$</td>
<td>$2.96 \times 10^{-5}$ ml O$_2$/ml$^{-1}$-mmHg$^{-1}$</td>
</tr>
<tr>
<td>$C_{Hb}$</td>
<td>0.52 ml O$_2$/ml</td>
</tr>
<tr>
<td>$P_{50,mb}$</td>
<td>5.3 mmHg</td>
</tr>
</tbody>
</table>

$\alpha$, O$_2$ solubility; $D$, diffusivity; $P_e$, critical P$_O_2$ for O$_2$ consumption; $k$, mass
transfer coefficient; $\bar{\alpha}$ and $\bar{\alpha}$, volume- and flow-weighted blood O$_2$ solubility,
respectively; $C_{Hb}$, O$_2$-binding capacity of Hb; $P_{50,mb}$, myoglobin (Mb) half-
maximal pressure.

ER$_{cn}$ is given by

$$ER_{cn} = \frac{\sum Q_{ai}S_{ai} - \sum Q_{vj}S_{vj}}{\sum Q_{ai}S_{ai}}$$

where $Q_{ai}$ and $S_{ai}$ are the RBC volume flow and $S_o$ at the
arteriolar end of each normal-flow capillary sampled and $Q_{vj}$
and $S_{vj}$ are the RBC volume flow and $S_o$ at the venular end of
each normal-flow capillary sampled. For ER$_{cn,calc}$, the flow
rates are exactly the same at both ends of each capillary
sampled, so we have

$$ER_{cn,calc} = \frac{\sum (S_{ai} - S_{ai})}{\sum S_{ai}}$$

In the 24-h CLP model, for control, AS-induced injury, and
ES-induced injury cases, ER$_{cn}$ was found to be 0.15, 0.44, and
0.69, respectively (15). Starting with values in the range 1.5 <
M$_0$ < 6 (in units of $10^{-2}$ ml O$_2$-ml$^{-1}$-s$^{-1}$), we found that for
FCD$_{A}$ = 1,000 capillaries/mm$^2$ and $v_{fast} = 500$ $\mu$m/s, the
correct ER$_{cn,calc}$ was obtained by taking M$_0$ = 1.50, 3.86, and
5.61 for the control, AS, and ES cases, respectively. Similar
results were also obtained for the other three cases considered,
as shown in Table 2. Thus the intrinsic M$_0$ was found to increase
two- to threefold for the AS cases and approximately fourfold
for the ES cases.

Note that the preceding results and those in the following
subsection are for “uniformly distributed” septic injuries to the
microvasculature. This means that, although capillary place-
ment in two dimensions and the selection of normal-, fast-,
and stopped-flow capillaries were random, allowing a signifi-
cant degree of spatial heterogeneity, there was no bias imposed
toward any particular pattern or location. This uniformly dis-
bursed injury will later be contrasted to a heterogeneous or
“clustered” injury, in which capillaries in one area are chosen
to have fast flow, whereas those in the rest of the domain have
either normal or stopped flow (according to a random selec-
tion).

Tissue oxygen distributions for uniformly distributed injury.
The above VO$_2$ estimates for control and sepsis cases allow us to
calculate steady-state tissue oxygen distributions and evaluate
the effect of sepsis on O$_2$ delivery and utilization in the
EDL. Figure 2 shows the spatial distributions of tissue P$_O_2$ for
the control, AS, and ES cases when FCD$_{A}$ = 1,000 capillaries/
mm$^2$ and $v_{fast} = 500$ $\mu$m/s. Sepsis shifts tissue P$_O_2$ to lower
values (average P$_O_2$ of 34.5 and 26.4 mmHg for all AS and ES
cases, respectively, vs. 43.0 mmHg for controls) and increases
its spatial heterogeneity [coefficient of variation (CV) = SD/
mean of tissue P$_O_2$ of 0.12 and 0.23 for all AS and ES cases,
respectively, vs. 0.04 for controls] as well as increasing aver-
age tissue P$_O_2$ gradients. However, our results imply that
dysoxia (P$_O_2$ < 1 mmHg) does not occur, because for all eight of
our AS/ES cases (see Table 2), the minimum tissue P$_O_2$
($P_{min}$) does not fall below 14 mmHg. Figure 3 shows $P_{min}$ as a
function of axial location and probability distributions of tissue
P$_O_2$ for all 12 control and sepsis cases. It can be seen that the
above results are not sensitive to the particular choice of either
Table 2. O$_2$ transport results for control and uniform septic injury cases

<table>
<thead>
<tr>
<th>Conditions</th>
<th>FCD$_A$, capillaries/mm$^2$</th>
<th>$v_{fast}$, $\mu$m/s</th>
<th>ER$_cn$</th>
<th>$M_0 \times 10^4$</th>
<th>ER$_cf$</th>
<th>$P_{min}$</th>
<th>$P_{min,Krogh}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>1,000</td>
<td>500</td>
<td>0.15</td>
<td>1.50</td>
<td>0.09</td>
<td>39.7</td>
<td>41.1</td>
</tr>
<tr>
<td>Control</td>
<td>1,000</td>
<td>1,000</td>
<td>0.15</td>
<td>1.69</td>
<td>0.06</td>
<td>39.8</td>
<td>43.2</td>
</tr>
<tr>
<td>Control</td>
<td>1,500</td>
<td>500</td>
<td>0.15</td>
<td>2.23</td>
<td>0.02</td>
<td>39.8</td>
<td>43.1</td>
</tr>
<tr>
<td>Control</td>
<td>1,500</td>
<td>1,000</td>
<td>0.15</td>
<td>2.58</td>
<td>0.06</td>
<td>39.7</td>
<td>43.2</td>
</tr>
<tr>
<td>AS</td>
<td>1,000</td>
<td>500</td>
<td>0.44</td>
<td>3.86</td>
<td>0.25</td>
<td>27.0</td>
<td>32.6</td>
</tr>
<tr>
<td>AS</td>
<td>1,000</td>
<td>1,000</td>
<td>0.44</td>
<td>4.38</td>
<td>0.17</td>
<td>27.2</td>
<td>36.7</td>
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<tr>
<td>AS</td>
<td>1,500</td>
<td>500</td>
<td>0.44</td>
<td>5.87</td>
<td>0.25</td>
<td>27.1</td>
<td>32.5</td>
</tr>
<tr>
<td>AS</td>
<td>1,500</td>
<td>1,000</td>
<td>0.44</td>
<td>6.64</td>
<td>0.16</td>
<td>27.2</td>
<td>36.7</td>
</tr>
<tr>
<td>ES</td>
<td>1,000</td>
<td>500</td>
<td>0.44</td>
<td>6.94</td>
<td>0.22</td>
<td>15.6</td>
<td>33.2</td>
</tr>
<tr>
<td>ES</td>
<td>1,000</td>
<td>1,000</td>
<td>0.44</td>
<td>6.01</td>
<td>0.22</td>
<td>14.2</td>
<td>26.7</td>
</tr>
<tr>
<td>ES</td>
<td>1,500</td>
<td>500</td>
<td>0.69</td>
<td>8.56</td>
<td>0.34</td>
<td>14.2</td>
<td>26.7</td>
</tr>
<tr>
<td>ES</td>
<td>1,500</td>
<td>1,000</td>
<td>0.69</td>
<td>9.26</td>
<td>0.22</td>
<td>14.8</td>
<td>33.0</td>
</tr>
</tbody>
</table>

Results for control, average sepsis (AS), and extreme sepsis (ES) cases. Shown for 4 combinations of appropriate baseline values of functional capillary density (FCD$_A$) and red blood cell velocity of fast-flow capillaries ($v_{fast}$) and measured values of O$_2$ extraction ratio for normal-flow capillaries ER$_cn$ are the O$_2$ consumption rate ($M_0$), minimum tissue PO$_2$ ($P_{min}$), and calculated O$_2$ extraction ratio for the fast-flow capillaries (ER$_cf$), where the values of $M_0$ have been selected to match measured ER$_cn$. Also shown are the estimates of minimum tissue PO$_2$ obtained using the Krogh tissue cylinder model ($P_{min,Krogh}$). $M_0$ is given in units of ml O$_2$·ml$^{-1}$·s$^{-1}$, and $P_{min}$ and $P_{min,Krogh}$ are in mmHg.

FCD$_A$ or $v_{fast}$, due to the fact that the appropriate $M_0$ was chosen to match the experimental ER$_cn$ for each pair of these parameters. Therefore, further simulations are performed only for FCD$_A$ = 1,000 capillaries/mm$^2$ and $v_{fast}$ = 500 $\mu$m/s.

Tissue oxygen distributions for heterogeneous or clustered injury. Results in the previous two subsections were obtained assuming that septic injuries to the microvasculature (i.e., capillary stoppages) are distributed approximately uniformly in space. We now investigate the effect on oxygen transport of having the fast-flow capillaries clustered in one-fourth of the domain to simulate the greatly increased heterogeneity in capillary blood flow during sepsis that has been observed experimentally. Using the same values of ER$_cn$ as above, we can then estimate $M_0$ and perform simulations of steady-state oxygen transport. The results for extreme sepsis with clustered injury (ES + Cl) are shown in Table 3, along with those for the corresponding uniform injury case. It can be seen that there are only slight changes in ES + Cl relative to ES: $M_0$ decreased from 5.61 to 5.26, $P_{min}$ decreased from 15.2 to 14.5 mmHg, and the CV of tissue PO$_2$ increased from 0.23 to 0.24.

Sensitivity of tissue PO$_2$ to V˙O$_2$, blood flow rate, and mass transfer coefficient. We now investigate whether the preceding results on tissue oxygenation during sepsis are qualitatively altered by 1) moderate increases in our estimates for $M_0$, 2) moderate decreases in the total RBC supply rate to the muscle (contrary to our previous assumption of fixed supply rate), or 3) moderate increases in the effective intravascular transport resistance. To do this, we perform simulations for the ES/clustered injury case for $J$ $M_0$ increased by 25% (ES + Cl + M), 2) total blood flow decreased by 50% (ES + Cl + F), and 3) $k$ decreased by 50% (ES + Cl + k), where in the latter two cases $M_0$ is adjusted to match measured ER$_cn$ for the ES case. These results, shown in Fig. 4, indicate that the alterations 2 and 3 (above) produce changes in $P_{min}$ of approximately ±33% (19.1 mmHg for ES + Cl + F and 10.0 mmHg for ES + Cl + k vs. 14.5 mmHg for ES + Cl) and changes in CV(P) of approximately ±13% (0.21 for ES + Cl + F and 0.27 for ES + Cl + k vs. 0.24 for ES + Cl). For case ES + Cl + M, where $M_0$ is arbitrarily increased by 25%, $P_{min}$ falls to 6.8 mmHg and CV(P) increases to 0.36, which means that the effect on tissue PO$_2$ is more severe but still very similar to that found in the other sepsis cases.

**DISCUSSION**

Our experiment-based simulations of oxygen transport in the EDL during sepsis show that there are large increases in V˙O$_2$...
(2- to 4-fold) and in the spatial heterogeneity of tissue oxygenation (CV of tissue PO2 increasing 3- to 6-fold), with average and minimum tissue PO2 decreasing by up to 39% and 63% in the AS and ES cases, respectively. Our results imply both hypoxia and greatly increased tissue PO2 gradients in sepsis, which may have physiological consequences because cells can respond to PO2 gradient changes (13, 50) as well as decreased PO2 values. The key quantity that we estimated using our computational model was VO2 (i.e., the parameter MO) for the various control and sepsis cases. Our full, heterogeneous O2 transport model yielded MO values in the range of 1–3 × 10^-4 ml O2·ml^-1·s^-1 for the controls, as expected for normal resting muscle (16), and MO for sepsis in the range of 3–6 × 10^-4 ml O2·ml^-1·s^-1.

![Fig. 3. Results of O2 transport calculations for control and uniform septic injury cases. Solid lines are FCDA = 1,000 capillaries/mm² and v×t = 500 μm/s; dotted lines are FCDA = 1,000 capillaries/mm² and v×t = 1,000 μm/s; dashed-dotted lines are FCDA = 1,500 capillaries/mm² and v×t = 500 μm/s; and dashed lines are FCDA = 1,500 capillaries/mm² and v×t = 1,000 μm/s. A: minimum tissue PO2 as a function of distance from the arteriolar end of the tissue domain. B: tissue PO2 probability distribution functions.](image)

**Table 3. O2 transport results for ES cases**

<table>
<thead>
<tr>
<th>case</th>
<th>ERcn</th>
<th>MO × 10^4</th>
<th>ERcf</th>
<th>Pmin</th>
<th>CV(P)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ES</td>
<td>0.69</td>
<td>5.61</td>
<td>0.34</td>
<td>15.2</td>
<td>0.23</td>
</tr>
<tr>
<td>ES + Cl</td>
<td>0.69</td>
<td>5.26</td>
<td>0.32</td>
<td>14.5</td>
<td>0.24</td>
</tr>
<tr>
<td>ES + Cl + M</td>
<td>0.86*</td>
<td>6.58</td>
<td>0.39</td>
<td>6.8</td>
<td>0.36</td>
</tr>
<tr>
<td>ES + Cl + F</td>
<td>0.69</td>
<td>3.15</td>
<td>0.42</td>
<td>19.1</td>
<td>0.21</td>
</tr>
<tr>
<td>ES + Cl + k</td>
<td>0.69</td>
<td>4.78</td>
<td>0.27</td>
<td>11.0</td>
<td>0.27</td>
</tr>
</tbody>
</table>

Results for ES with uniform injury (ES), ES with clustered injury (ES + Cl), ES with clustered injury and increased M0 (ES + Cl + M), ES with clustered injury and decreased blood flow (ES + Cl + F), and ES with clustered injury and decreased k (ES + Cl + k). Shown are ERcn, ERcf, MO, Pmin, and coefficient of variation of tissue PO2 [CV(P) = SD/mean]. *Case ES + Cl + M gives ERcn = 0.86, in contrast to the experimental value of 0.69.
10^{-4} \text{ml O}_2 \text{ml}^{-1} \text{s}^{-1}, consistent with some measurements in skeletal muscle (22).

Our estimates of $M_0$ as well as tissue $P_O2$ are in contrast to results that would be obtained using simplified models that do not properly account for the role of fast-flow capillaries. As seen in Tables 2 and 3, the average $ER_{cf}$ are lower than those for $ER_{cn}$. However, due to the relatively high velocities of the fast capillaries, they deliver a substantial amount of $O_2$ to the tissue and are a major factor in $O_2$ transport during sepsis. If we estimate $M_0$ for the cases in Table 2 using a simple mass balance that neglects the fast capillaries, we obtain unreasonably low values, ranging from 9% to 34% of the values obtained using the full heterogeneous model. On the other hand, if we assume the fast capillaries have the same average $O_2$ extraction ratios as the normal ones, we obtain large overestimates of $M_0$ during sepsis (24–135% above the values found with the heterogeneous model), which in some cases would incorrectly predict the occurrence of dysoxia.

Ellis et al. (15) have observed a linear relationship between the number of stopped-flow capillaries and the average drop in

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**Fig. 4.** Results of $O_2$ transport calculations for extreme sepsis cases. 

- **A**: minimum tissue $P_O2$ as a function of distance from the arteriolar end of the tissue domain. 
- **B**: tissue $P_O2$ probability distribution functions. Solid lines are extreme uniform injury; dotted lines are extreme clustered injury (ES+Cl); dashed-dotted lines are ES+Cl with local tissue $O_2$ consumption rate increased by 25%; dashed lines are ES+Cl with blood flow decreased 50%; dashed-x lines are ES+Cl with the mass transfer coefficient decreased 50%. In all cases, $FCDA = 1,000$ capillaries/mm$^2$ and $v_{fast} = 500$ $\mu$m/s.
SO2 across the normal-flow capillaries. They have suggested that this linear relationship could be explained by considering a homogeneous, single-capillary Krogh model (28; see Ref. 37 for further details) in which the diameter of the tissue cylinder increases as the density of capillaries decreases. This explanation is conceptually useful, but it cannot be applied quantitatively until the influence of the fast capillaries and the changes in M0 during sepsis have been calculated with the full heterogeneous model. In addition, the Krogh model does not take into account the spatial heterogeneity of blood flow and O2 delivery. If we calculate minimum tissue PO2 using the Krogh model with the correct values of M0, average tissue cylinder radius, and average blood flow per capillary, we obtain large overestimates of minimum tissue PO2, as shown in the last column of Table 2. Thus we see the impact of flow heterogeneity on O2 transport during sepsis and the ability of fast flow capillaries to deliver O2 to their surrounding tissue and supplement O2 delivery by normal flow capillaries. In addition, our results show that the difference between ERcf and ERcn increases as the number of stopped-flow capillaries increases, i.e., as the degree of septic microvascular injury becomes more severe, suggesting a limit to the ability of fast capillaries to overcome the loss of O2 delivery by normal capillaries.

To answer some possible questions about parameter sensitivity, we showed that the above results were not fundamentally altered by arbitrarily increasing M0 an additional 25% above the already large value estimated from experimental data or by decreasing the capillary-tissue mass transfer coefficient by 50% with an accompanying decrease in M0. In addition, we showed that if total muscle blood flow decreased by 50% during sepsis, which would decrease M0 as shown in Table 3, there would be no qualitative change in our results for tissue PO2. This finding is particularly important given published experimental data indicating that total EDL blood flow may decrease as much as 36% during sepsis (30). The data on which the present study was based did not give information on total blood flow during sepsis, because \( v_{fast} \) was not measured. However, by adjusting M0 to match measured values of ERcn in the 24-h CLP model, we have shown that tissue PO2 distributions are similar whether total blood flow was maintained during sepsis or decreased by 50%. Therefore, our results on tissue oxygenation should also apply to a septic flow reduction of the order measured by Lam et al. (30). Note that the more modest increase in M0 corresponding to a flow reduction of 50% versus no-flow reduction (110% above the control value for ES+Cl+F vs. 251% for ES+Cl) may be closer to the expected change in \( V_{O2} \) during sepsis. Estimates of M0 could also be elevated if there is preferential stoppage of capillaries that deliver more \( O2 \), as opposed to random capillary stoppage, without a compensating redistribution of blood flow. Such a possibility could be explored in future experiments and modified versions of the model.

One consequence of our \( V_{O2} \) estimates was that despite significant decreases in tissue PO2 and greatly increased capillary stopped flow and heterogeneity of O2 delivery, tissue dysoxia does not occur in the EDL during normotensive sepsis. This was true for both the AS cases, when approximately one-third of capillaries was stopped, and ES cases, when approximately one-half of capillaries was stopped. It was also true for the ES/clustered injury case, when three-fourths of the tissue domain was primarily supplied by one-fourth the usual number of flowing capillaries, i.e., when the effective capillary density was less than one-third the normal value. In contrast to these findings in the early stage of sepsis, decreased tissue O2 extraction in skeletal muscle has been observed in advanced stages of sepsis in humans (10), suggesting that at a later stage of sepsis, cellular dysfunction or mitochondrial inhibition (17, 43) is decreasing O2 utilization. Because the present results imply that dysoxia does not occur in skeletal muscle in sepsis but significant hypoxia does occur, the model suggests that hypoxia rather than dysoxia is the pathophysiological mechanism responsible for putative O2 transport-mediated problems during the early phase of sepsis.

We note that our results for tissue PO2 during sepsis are in general agreement with published data, although caution must be used when comparing capillary-scale (~5 \( \mu m \)) calculations to measurements made with 100- to 200-\( \mu m \)-diameter microelectrodes and when comparing different animal models. Using a severe rat sepsis model, Astiz et al. (4) found that average PO2 in the rectus femoralis muscle decreased from 44.9 mmHg initially to 38.6 mmHg after 3 h of sepsis and 23.5 mmHg at 6 h. Additionally, in rat models of endotoxemia, both Anning et al. (2) (from 28.5 mmHg in controls to 7.5 mmHg) and Sair et al. (40) (from 52 mmHg in controls to 24 mmHg) found decreases in skeletal muscle PO2 similar to what our experiment-based calculations predicted. For FCD\(_A\) = 1,000 capillaries/mm\(^2\) and \( v_{fast} = 500 \mu m/s \), our calculations gave average PO2 for control, AS, and ES cases of 42.9, 34.3, and 26.3 mmHg, respectively.

Vallet et al. (47), in a dog model of endotoxemia, found a shift in muscle PO2 histograms to lower values that increased with time after lipopolysaccharide injection, similar to what was found in the present study for increasing severity of microvascular injury (Fig. 3B). In skeletal muscle, Vallet et al. (40) also found increased O2 extraction ratio and tissue PO2 lower than venous PO2, indicative of flow heterogeneity and shunting. These effects were more pronounced in gut mucosa, a tissue known to be at greater risk of dysoxia during sepsis. An additional factor in both muscle and the gut was a decrease in global blood flow that, when coupled with the increased microvascular heterogeneity included in our computational model, could put local regions of skeletal muscle at significant risk for dysoxia. An examination of this issue is planned in future studies.

Although we believe the present model provides a faithful description of O2 transport in the EDL under control and sepsis conditions, there are at least two aspects that may merit further investigation. First, the use of three discrete types of capillary blood flow (normal/fast/stopped) and a uniform hematocrit could be generalized to include a continuous distribution of both RBC velocities and hematocrits. It is not clear what the effect of this would be, but allowing a variation in hematocrit should allow more spatial heterogeneity in the local O2 supply and therefore could be expected to produce more heterogeneity in tissue PO2. We plan to study variations in RBC velocities and hematocrits once better experimental data are available for the fast-flow capillaries. A second aspect of the present model that could be examined is the amount of underlying heterogeneity in the two-dimensional spacing of capillaries. Currently, this heterogeneity is fixed at a relatively moderate amount, but some variation (guided by specific experimental data) may indicate that lower tissue PO2 can occur in very localized
EFFECT OF SEPSIS ON SKELETAL MUSCLE OXYGEN TRANSPORT

H2543

regions. It should also be noted that a separate (non-O2 consuming) interstitial compartment, which would tend to concentrate V\textsubscript{O2} away from the capillaries and decrease calculated tissue PO\textsubscript{2}, is not included in the present model. The overall effect of including the interstitium is unknown and may be considered in future studies; however, given that it occupies ~20% of the muscle volume, it is not expected to change minimum PO\textsubscript{2} sufficiently to alter the basic findings of the present study.

In conclusion, the fundamental questions regarding abnormal O\textsubscript{2} transport in skeletal muscle during sepsis are as follows: 1) how is tissue oxygenation affected by microvascular dysfunction? and 2) is the decreased O2 extraction seen in advanced stages simply a result of microvascular blood flow maldistribution or are additional mechanisms working to decrease the tissue’s intrinsic consumption rate? (6, 15, 25, 48). Using a highly detailed computational model to match measured local capillary O\textsubscript{2} transport parameters and vary parameters that are not known exactly, we obtained results that address the first question and indicate how tissue PO\textsubscript{2} distribution change in the initial phases of sepsis as a result of changes in FCD and hemodynamics. In particular, we have shown that both average and minimum tissue PO\textsubscript{2} in the EDL decrease as the number of stopped flow capillaries increases and that tissue PO\textsubscript{2} becomes more spatially heterogeneous. We have also shown that tissue O2 consumption increases in sepsis, even if both capillary density and total blood flow decrease.

Our modeling results, based on capillary O\textsubscript{2} transport data in a normotensive, fluid-resuscitated sepsis model, suggest that despite abnormal microvascular O\textsubscript{2} transport and the onset of tissue hypoxia in early sepsis, tissue dysoxia is not occurring in skeletal muscle, due to the contribution of the fast-flow capillaries to tissue oxygenation. In the context of the 24-h CLP experimental model used as the basis for this study, other organs with different microvascular geometry, blood flow heterogeneity, and O\textsubscript{2} demand (e.g., intestinal mucosa) may already be at risk of dysoxia (47). However, in skeletal muscle, the present work shows significant hypoxia is occurring, and therefore hypoxia rather than dysoxia is implicated as the more likely mechanism of initial O2-related cellular dysfunction. These results suggest the importance of blood flow maldistribution in skeletal muscle during sepsis. To fully address the cause of decreased O2 extraction in late sepsis (question 2 above), simultaneous information on cellular O2 demand, intrinsic mitochondrial function, and microvascular dysfunction will be required. This is the first report on the impact and importance of fast-flow capillaries (putative convective shunts) on tissue oxygenation under septic conditions and underscores the value of mathematical simulation when it is combined with real pathophysiological data.

GRANTS

This study was supported by the Whitaker Foundation and the National Partnership for Advanced Computing Infrastructure (to D. Goldman), by Canadian Institutes of Health Research Grant MOP-49416 (to C. G. Ellis), and by a Spoerel Research Fellowship (to R. M. Bateman). R. M. Bateman was also supported by postdoctoral fellowships from the Heart and Stroke Foundation and the Michael Smith Foundation for Health Research.

REFERENCES

EFFECT OF SEPSIS ON SKELETAL MUSCLE OXYGEN TRANSPORT


