Muscle Oxygenation Trends During Dynamic Exercise Measured by Near Infrared Spectroscopy

Yagesh N. Bhambhani

Catalogue Data

Key words: blood volume, noninvasive measurement

Mots-clés: volume sanguine, mesure non-invasive

Abstract/Résumé

During the last decade, NIRS has been used extensively to evaluate the changes in muscle oxygenation and blood volume during a variety of exercise modes. The important findings from this research are as follows: (a) There is a strong correlation between the lactate (ventilatory) threshold during incremental cycle exercise and the exaggerated reduction in muscle oxygenation measured by NIRS. (b) The delay in steady-state oxygen uptake during constant work rate exercise at intensities above the lactate/ventilatory threshold is closely related to changes in muscle oxygenation measured by NIRS. (c) The degree of muscle deoxygenation at the same absolute oxygen uptake is significantly lower in older persons compared younger persons; however, these changes are negated when muscle oxygenation is expressed relative to maximal oxygen uptake values. (d) There is no significant difference between the rate of biceps brachii and vastus lateralis deoxygenation during arm cranking and leg cycling exercise, respectively, in males and females. (e) Muscle deoxygenation trends recorded during short duration, high-intensity exercise such as the Wingate test indicate that there is a substantial degree of aerobic metabolism during such exercise. Recent studies that have used NIRS at multiple sites, such as brain and muscle tissue, provide useful information pertaining to the regional changes in oxygen availability in these tissues during dynamic exercise.

Au cours de la dernière décennie, on observe une grande utilisation de la spectroscopie en proche infrarouge (NIRS) afin d’évaluer les variations de l’oxygénation du muscle et du

Yagesh Bhambhani is with the Faculty of Rehabilitation Medicine, Rm 373 Corbett Hall, University of Alberta, Edmonton, AB, Canada T6G 2G4.
volume sanguin pendant différents exercices physiques. Les principaux résultats de la présente étude sont: (a) Il y a une forte corrélation entre le seuil lactique (ventilatoire) au cours d’un effort progressif sur ergocycle et la réduction excessive de l’oxygénation du muscle observée par la NIRS. (b) Il y a une corrélation importante entre le délai pour atteindre le plateau de consommation d’oxygène au cours d’un effort d’intensité constante au-dessus du seuil lactique/ventilatoire et la variation de l’oxygénation du muscle observée par la NIRS. (c) Le niveau de désoxygénation musculaire pour une consommation d’oxygène donnée est significativement plus bas chez les individus plus âgés comparativement aux plus jeunes, mais ces variations sont annulées quand l’oxygénation musculaire est exprimée en fonction de la consommation maximale d’oxygène. (d) Il n’y a pas de différence significative entre le taux de désoxygénation du biceps brachial et du vaste externe au cours d’un exercice de pédalage des bras ou des jambes tant chez les hommes que chez les femmes. (e) La courbe de désoxygénation musculaire observée au cours d’un vigoureux effort de courte durée comme dans le test de Wingate montre qu’il y a un niveau substantiel de sollicitation du métabolisme aérobie dans un tel effort. Des études récentes utilisant la NIRS à de multiples régions corporelles (cerveau, muscle) nous fournissent de précieuses observations concernant les variations régionales de la disponibilité de l’oxygène dans ces tissus au cours d’un exercice dynamique.

Introduction

During the last decade there has been considerable interest in the application of continuous-wave near infrared spectroscopy (NIRS) to evaluate trends in muscle oxygenation and blood volume during exercise in healthy individuals. The increased popularity of using NIRS lies in the fact that scientists can noninvasively evaluate the relative changes in the balance between oxygen delivery and utilization at the level of the small blood vessels—the arterioles, capillaries, and venules—thereby providing a more complete picture of the response to exercise, especially when it is combined with simultaneous measurements of cardiorespiratory responses (Ferrari et al., 1997). NIRS has been used by exercise physiologists to study the peripheral response to exercise during a variety of dynamic exercise modes such as cycling, arm cranking, treadmill walking and running, rowing, and in-line skating. The purpose of this brief review is to summarize the important findings of these studies and provide direction for future research in this area.

Validity of NIRS in Evaluating Muscle Oxygenation

The validity of NIRS in evaluating muscle oxygen saturation in vitro has been demonstrated. Belardinelli et al. (1995a) reported a highly significant correlation (0.997) between oxyhemoglobin (HbO₂) saturation and NIRS absorbency signal at a hemoglobin concentration of 21 gm/liter. Similarly, Mancini et al. (1994) reported a correlation of 0.92 between venous oxygen saturation and the HbO₂/MbO₂ absorbency recorded by NIRS during rhythmic contractions of the forearm under normoxic conditions. These responses were not influenced by skin blood flow. However, these findings have not been supported by other studies that have altered the inspired oxygen content to test the validity of NIRS during dynamic exercise.
Costes et al. (1996) reported that muscle oxygenation responses during 30 min of submaximal cycle exercise in healthy males were related to femoral venous oxygen tension under hypoxic conditions (10.5% inspired oxygen; $r^2 = 0.31$) but not during normoxia. They speculated that this discrepancy could be due to the fact that during normoxia only the venular blood is deoxygenated by metabolic demand, whereas under hypoxia both the reduced arterial oxygen content and the muscle oxygen demand decreases the overall muscular oxygen content during exercise. Costes et al. (1996) also suggested that differences in skinfold thickness at the NIRS measurement site among the subjects could have influenced the absorbency readings and affected the muscle oxygenation results.

MacDonald et al. (1999) evaluated the changes in femoral venous oxygen saturation during 6 min of submaximal leg kicking exercise under three inspired oxygen concentrations: 14%, 21%, and 70%. At all three concentrations, significant correlations ($r = 0.61, 0.53, \text{and} 0.57$, respectively) were observed between the femoral venous oxygen tension and the NIRS oxygen saturation during the first 40 sec of exercise. However, when the correlations over the whole duration of exercise were examined, significant correlations were observed only under hypoxia ($r = 0.42$), but not under hyperoxia ($r = 0.05$) or normoxia ($r = 0.02$).

The discrepancy between the direct measurements of venous oxygen tension and muscle oxygen saturation measured by NIRS could be due to the following: First, muscle oxygen saturation measured by NIRS is an estimate of the balance between oxygen delivery and extraction in the small blood vessels directly beneath the sampling area of the probe, whereas estimates of oxygen saturation from the femoral vein includes blood drawn from the less active areas of the quadriceps muscle group. Second, differences in muscle NIRS sampling sites among these studies could have influenced the findings. Costes et al. (1996) did not specify the exact location of the NIRS probe on the vastus lateralis muscle, whereas MacDonald et al. (1999) indicated that the probe was placed at a distance of 10 to 12 cm above the knee. It should be noted that Quaresima et al. (2001) used multichannel NIRS to demonstrate considerable variation in the magnitude of the muscle oxygenation responses at different sites of the vastus lateralis and rectus femoris muscles during isometric exercise. Their findings highlight the importance of ensuring the consistency of probe placement among NIRS studies so that appropriate comparisons can be made.

Third, alterations in muscle geometry and sampling volume that occur as a result of differences in exercise intensity and duration could influence these findings (Chuang et al., 2002). Finally, the changes in muscle oxygenation measured by NIRS include the contribution of MbO$_2$, which cannot be differentiated by the HbO$_2$ contribution because the absorbency of the two chromophores overlaps in the near infrared region. Although the contribution of MbO$_2$ is quite small (Mancini et al., 1994), it could confound the validity of the findings of studies that attempt to correlate femoral venous oxygen tension with that obtained from NIRS absorbency readings. Despite these controversial findings pertaining to the validity of NIRS during dynamic exercise in vivo, many researchers have used this technique to evaluate the trends in muscle oxygenation and blood volume during dynamic exercise.
It has been well documented (Belardinelli et al., 1995b; 1995c; Bhamhiani et al., 1997) that during the transition from rest to maximal incremental cycle exercise and recovery, the muscle oxygenation (Mox; measured by the difference between the 850-nm and 760-nm absorbency signals) trend demonstrates a characteristic four-phase response, as shown in Figure 1. At the onset of exercise there is an initial increase in Mox above resting baseline levels so as to meet the increased demand for ATP via aerobic sources. Although this has been attributed to a redis-
tribution of blood flow from the active to the inactive areas of the muscle (Bellardinelli et al., 1995b; Bhambhani et al., 1997), computer simulations of microvascular unit (capillary) recruitment during contractions of increasing intensity do not support this hypothesis (Fuglevand and Segal, 1997). However, the possibility of this initial increase in Mox resulting from an increase in skin blood flow cannot be ruled out (Chuang et al., 2002; Maehara et al., 1997).

With increasing work rate, Mox tends to decrease linearly or exponentially below the resting baseline value, followed by a leveling off as the person reaches volitional fatigue or attains maximal oxygen uptake (VO₂ max). During the recovery period there is a very rapid increase in Mox during the first 1 to 2 minutes which usually exceeds the resting baseline value. This increased oxygenation is attributed to a sudden reduction in the demand for aerobic production of ATP. This hyperemic response is followed by a gradual decline in Mox toward the resting baseline value, and it could take several minutes before resting baseline conditions are restored. Chance et al. (1992) demonstrated that the half time for Mox recovery from maximal rowing exercise was significantly correlated with repayment of the oxygen debt and the removal of lactate following exercise.

Although the Mox trends during incremental exercise have been well documented, the trends in muscle blood volume (MbV; measured by the sum of the 760-nm and 850-nm absorbency signals) have not been described in detail. Grassi et al. (1999) reported that during stepwise incremental cycle exercise the MbV increased until approximately 60% to 65% of VO₂ max, after which it remained constant or decreased slightly until VO₂ max was attained. The latter is likely due to the fact that during the transition from rest to maximal exercise, the increase in intramuscular pressure during the contraction phase exceeds the rise in intravascular pressure, thereby decreasing the ability of blood to perfuse the exercising muscle (Radegran, 1997). While arterial blood flow during the relaxation phase is unimpeded due to the lower intramuscular pressure, it should be pointed out that these variations in blood flow during the contraction/relaxation phases cannot be identified by alterations in MbV as measured by NIRS. During recovery from exercise, there is a rapid decline toward the resting baseline value in the first 1 to 2 minutes, followed by a more gradual decrease for several minutes before the resting baseline is attained.

Detecting Metabolic Thresholds From NIRS Trends

It is generally accepted that during stepwise incremental exercise there are two distinct metabolic thresholds (Reinhard et al., 1979; Wasserman et al., 1967). The first, referred to as the lactate threshold (LT), is defined as the lowest exercise intensity that results in increased production of lactate relative to lower exercise levels. Although the exact mechanism for the LT is not clear, it has been suggested that the accumulation of blood lactate is due to the inability of the central circulation to supply enough oxygen to the exercising muscle for the aerobic production of ATP. As a result, some of the energy requirements to meet the muscular demands are metabolized via anaerobic sources and result in the production of lactate. This lactate diffuses out of the muscle and can be traced in the arterial blood, where it is buffered by bicarbonate. When the limited amount of arterial blood is
depleted, there is an accumulation of hydrogen ions which results in a significant reduction in blood pH. The exercise intensity at which blood pH decreases significantly is referred to as the threshold of decompensated metabolic acidosis (TDMA), i.e., the second metabolic threshold (Reinhard et al., 1979).

The LT and TDMA can be detected noninvasively via alterations in respiratory gas exchange measurements during incremental exercise (Reinhard et al., 1979; Wasserman et al., 1967). Research has indicated that at the LT, the buffering of lactate by bicarbonate results in the production of carbonic acid, which at physiological pH dissociates into water and carbon dioxide (CO₂). As a result there is an increase in arterial carbon dioxide tension (PaCO₂), which tends to stimulate the peripheral chemoreceptors, thereby causing a nonlinear increase in the ventilation rate (Vₑ) during exercise.

The increased CO₂ production, commonly referred to as the excess CO₂, also results in a nonlinear increase in respiratory exchange ratio (RER) at this intensity. Hence, when these two variables are plotted against the work rate or oxygen consumption during incremental exercise, the point at which the Vₑ, VCO₂, and RER increase in a nonlinear manner can be used to identify the LT. Once the buffering capacity of the blood is exceeded and TDMA occurs, the significant reduction in blood pH provides a very strong stimulus to the respiratory center, resulting in an exponential increase in the Vₑ during incremental exercise. Consequently there is a large increase in expired carbon dioxide at this threshold, and a concomitant reduction in PaCO₂. When LT and TDMA are identified noninvasively using the alterations in these respiratory gas exchange measurements, the terms commonly used are ventilatory threshold (VT) and respiratory compensation threshold (RCT).

Several studies have demonstrated that the trends in Mox during incremental exercise can be used to identify the two metabolic thresholds. These studies are summarized in Table 1. Grassi et al. (1999) demonstrated that during stepwise incremental exercise, there was an exaggerated decrease in Mox, i.e., greater deoxygenation, at a particular intensity that coincided with the LT. The correlation reported for oxygen uptake at the Mox and LT was 0.93. Other studies (Belardinelli et al., 1995b; 1995c; Bhambhani et al., 1997) have demonstrated an exaggerated decrease in the Mox which coincided with the VT identified by different gas exchange methods. The correlations for oxygen uptake ranged from 0.75 to 0.95, while those for power output ranged from 0.90 to 0.93. The physiological basis for the exaggerated decrease in Mox observed at the LT and VT is that the accumulation of a significant amount of lactate in the blood facilitates the release of oxygen from HbO₂ via the Bohr effect, so that it can be utilized in the mitochondria for aerobic energy production (Stringer et al., 1994; Wasserman et al., 1991).

Since NIRS is based on the differential absorption properties of hemoglobin (and myoglobin in the muscle), the greater release of oxygen which occurs at the LT and VT increases the deoxygenation of hemoglobin, thereby resulting in a sharp decrease in tissue absorbency at this threshold which is evident as an exaggerated decline in Mox. An important point to note is that when the VT is detected by changes in Mox during incremental exercise, it usually occurs just before the changes in respiratory gas exchange measurements are evident. This is because NIRS reflects the metabolic changes that occur directly at the muscle site, whereas
<table>
<thead>
<tr>
<th>Authors, Subjects</th>
<th>LT or VT Criteria</th>
<th>RCP Criteria</th>
<th>NIRS Criteria</th>
<th>Correlation</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bellardinelli et al., 1995b</td>
<td>V-slope method</td>
<td>Accelerated fall in tissue oxygen saturation</td>
<td>Abs. VO$_2$ = 0.94 PO = 0.95</td>
<td></td>
<td>VT identified by NIRS occurred earlier than VT identified from gas exchange criteria</td>
</tr>
<tr>
<td>8 M, 3 F</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Runman</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bellardinelli et al., 1995c</td>
<td>V-slope method</td>
<td>Accelerated fall in tissue oxygen saturation</td>
<td>PO = 0.94</td>
<td></td>
<td>Acceleration in muscle deoxygenation occurred later in healthy subjects than in CHF patients</td>
</tr>
<tr>
<td>12 M, 7 CHF</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Runman</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bhambhani et al., 1997</td>
<td>V-slope method</td>
<td>Absorbency crossed resting baseline value observed immediately prior to exercise</td>
<td>Rel. VO$_2$ = 0.90 PO = 0.88 Rel. VO$_2$ = 0.89 PO = 0.86</td>
<td></td>
<td>VT identified by NIRS occurred earlier than VT identified from gas exchange criteria in 65% of cases; No significant gender differ. in muscle oxygen at VT</td>
</tr>
<tr>
<td>21 M, 19 F</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Runman</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Grassi et al., 1999</td>
<td>Lowest blood lactate that was 0.5 mM lower than the next one</td>
<td>Accelerated decrease in muscle deoxygenation</td>
<td>PO = 0.97</td>
<td></td>
<td>Muscle deoxygenation occurred after the localized blood volume measured by NIRS reached a max</td>
</tr>
<tr>
<td>5 M</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Omron</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Miura et al., 1998</td>
<td>V-slope method $V_{i}/VCO_{2}$ started to increase, $PetCO_{2}$ started to decrease</td>
<td>First decrease in oxyHb Second decrease in oxyHb</td>
<td>Rel. VO$_2$ = 0.753*</td>
<td></td>
<td>Significant correlation with peak VO$_2$ r = 0.645 Significant correlation with peak VO$_2$ r = 0.899</td>
</tr>
<tr>
<td>6 M (A), 14 M &amp; 7 F (S), 14 M &amp;2 F (CHF)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Shimadzu</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Note: A = active, healthy subjects; S = sedentary, healthy subjects; CHF = chronic heart failure patients; VO$_2$ = oxygen uptake; PO = power output. *Indicates overall correlation for all subjects.
the alterations in respiratory gas exchange measurements, which theoretically are based on the metabolic changes occurring in the muscle, are delayed due to the biochemical mechanisms described earlier (Belardinelli et al., 1995b; Bhambhani et al., 1997).

While there has been considerable evidence validating the use of NIRS to identify the LT and VT, there has been limited use of this technique to identify the TDMA or RCT. Miura et al. (1998) demonstrated that during stepwise incremental exercise, there was a second inflection in the Mox trend that coincided with the RCT. The correlation for oxygen uptake between these two techniques was 0.90. Miura et al. suggested that the reduction in Mbv observed at the RCT reflected the point at which the intramuscular pressure during the contraction phase increased by a greater proportion than the intravascular pressure during incremental exercise.

**Muscle Oxygenation Trends During Submaximal Exercise**

Several studies (see Table 2) have used NIRS to examine changes in Mox during constant work-rate cycle exercise. Belardinelli et al. (1995a) compared the Mox changes at exercise intensities below and above the VT during 6 min of constant work-rate exercise. Their results indicated a consistent pattern between whole body oxygen uptake kinetics and the Mox response measured by NIRS. Specifically, they reported that the delay in steady-state oxygen uptake response (defined as the difference in oxygen uptake between the 4th and 6th minute of constant work-rate exercise) observed at intensities above the LT was due to localized alterations in Mox measured by NIRS. These findings were subsequently confirmed by Chuang et al. (2002) and Miura et al. (1998).

Bhambhani et al. (1999) hypothesized that there would be a significant correlation between Mox measured by NIRS and the mixed arteriovenous oxygen difference during steady-state exercise because most of the oxygen utilized was extracted by the muscles directly involved in the exercise. These researchers measured cardiac output by carbon dioxide rebreathing [from which the mixed (a – v)O2 diff was calculated], as well as Mox by NIRS during 4 min of constant work-rate exercise at intensities below and above VT in healthy men and women. The findings did not substantiate their hypothesis, most likely because the changes in Mox reflect the balance between oxygen delivery and removal at the muscle site, whereas the calculated (a – v)O2 diff was the difference in oxygen content between arterial and mixed venous blood. Miura et al. (2000) examined the changes in Mox and myoelectric activity during constant work-rate exercise at different power outputs. Their findings indicated a close relationship between the integrated EMG activity and alterations in Mox at each work rate, thereby validating the use of NIRS in measuring muscle activity during exercise.

Ogata et al. (2002) used NIRS to evaluate changes in active and inactive leg Mox by combining leg exercise with arm exercise. During 6 min of submaximal arm cranking at 30% peak VO2, Mox and Mbv in the inactive vastus lateralis were unaffected, whereas at 50% peak VO2 both variables decreased significantly toward the initial and latter stages of the test, respectively. When leg cycling at 40% of VO2 max was combined with arm cranking at 30% peak VO2, no significant changes were observed in either variable. However, leg exercise combined with
Table 2  Studies That Have Examined the Relationship Between Changes in Whole Body Oxygen Uptake and Vastus Lateralis Muscle Oxygenation Changes Measured by NIRS During Constant Work-Rate Cycle Exercise

<table>
<thead>
<tr>
<th>Authors, Subjects</th>
<th>Instrument</th>
<th>Duration</th>
<th>Findings</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bellardinelli et al. (1995a)</td>
<td>peak VO₂ (below VT)</td>
<td>6 min each @ 20% and 40%</td>
<td>At intensities below VT, the OxyHb/OxyMb initially decreased, then remained constant or increased with time; At intensities above VT, the OxyHb/OxyMB decreased continuously during the 6 min in proportion to the work intensity and failed to reach a steady state; Strong relationship between slope of whole body VO₂ and OxyHb/OxyMb saturation.</td>
</tr>
<tr>
<td>8 M, 2 F Runman</td>
<td>peak VO₂ (above VT)</td>
<td>6 min each @ 60% and 75%</td>
<td></td>
</tr>
<tr>
<td>Bhambhani et al. (1999)</td>
<td>Muscle oxygenation, expressed as % of maximal range during exercise and recovery</td>
<td>4 min @ 40% below VT</td>
<td>Muscle oxygenation, expressed as % of maximal range during exercise and recovery (%Mox), decreased systematically with exercise intensity; No significant correlation between % Mox and mixed arterio-venous oxygen difference at the different exercise intensities.</td>
</tr>
<tr>
<td>19 M, 19 F Runman</td>
<td>% Mox and VO₂ max</td>
<td>4 min @ 80% below VT</td>
<td></td>
</tr>
<tr>
<td>Chuang et al. (2002)</td>
<td>6 min @ 60% LAT</td>
<td>6 min @ 60% LAT</td>
<td>Kinetics of tissue deoxygenation were significantly more rapid than the oxygen uptake and heart rate kinetics at all exercise intensities; Steady state in tissue deoxygenation occurs within 1 min at intensities below LAT but is delayed at intensities above LAT.</td>
</tr>
<tr>
<td>8 M, 4 F Runman</td>
<td>6 min between 35% of LAT and VO₂ max</td>
<td>6 min @ 80% LAT</td>
<td></td>
</tr>
<tr>
<td>Miura et al. (2000), 7 M OMRON</td>
<td>6 min each @ 50 W, 100 W, 150 W, 250 W</td>
<td>6 min each @ 50 W</td>
<td>Significant negative correlations between % of OxyHb/Mb concentration and: (a) integrated EMG activity; (b) % of OxyHb/Mb and blood lactate; and (c) % of OxyHb/Mb and whole body oxygen uptake.</td>
</tr>
<tr>
<td>Miura et al. (1998), 7 M Shimadzu</td>
<td>VT and peak VO₂</td>
<td>6 min @ 60% VT</td>
<td>VO₂ attained a steady state, no change in both OxyHb and DeoxyHb; VO₂ did not attain a steady state; OxyHb decreased and DeoxyHb increased.</td>
</tr>
</tbody>
</table>
arm cranking at 50% peak VO₂ induced a significant increase in vastus lateralis blood volume between the 4th and 6th minutes of exercise, but no change in oxygenation. These findings suggest that the decreases in Mox and Mbv observed in the inactive vastus lateralis muscle during moderate-intensity arm cranking exercise are attenuated when moderate-intensity leg exercise is performed. Unfortunately, Ogata et al. (2002) did not examine the changes in Mox and Mbv during the submaximal arm cranking protocol.

**Muscle Oxygenation Trends During Locomotion**

Although several studies (see Table 3) have examined the Mox and Mbv responses during treadmill walking and running in healthy subjects, their results are difficult to compare primarily because of the different exercise protocols used. Nonetheless, the following conclusions can be drawn from these studies: (a) The decrease in Mox observed during treadmill running is similar to that observed during cycling (Demarie et al., 2001; Hiroyuki et al., 2002). However, the attenuated blood volume responses during treadmill running seem to differ slightly from those observed during cycling (Demarie et al., 2001), which could be due to the variability in sampling volume. (b) The delay in steady-state oxygen consumption at intensities above the ventilatory threshold are correlated with the delta values of muscle deoxygenation (Demarie et al., 2001) and are consistent with the findings during cycle exercise (Belardinelli et al., 1995a; Chuang et al., 2002; Miura et al., 1998). (c) It appears there are considerable interindividual differences in oxyHb and deoxyHb patterns during steady-state walking and incremental running, implying a variation in the factors that determine oxygen supply and demand of the exercising muscle (Quaresima et al., 1996). (d) There are significant differences in the degree of deoxygenation between the vastus lateralis and gastrocnemius muscles during treadmill running (Hiroyuki et al., 2002). (e) There is a mismatch between muscle blood flow and oxygen saturation of the vastus lateralis and medial gastrocnemius muscles during treadmill walking (Quaresima et al., 1996).

**Muscle Oxygenation Trends During Arm Exercise**

It is well documented that peak oxygen uptake during upper body exercise in the form of arm cranking is approximately 70% to 75% of that attained during leg exercise (Sawka, 1986). Research suggests that the difference in peak oxygen uptake between these exercise modes is most likely due to the reduced muscle mass recruited during upper body exercise, as well as possible differences in oxidative capacity between the muscles of the upper and lower body. Bhambhani et al. (1998) confirmed that peak oxygen uptake during arm cranking was approximately 73% and 76% of that attained during leg cycling in healthy men and women, respectively, and that the magnitude of deoxygenation in the biceps brachii was also lower during arm cranking compared to that attained in the vastus lateralis during leg cycling. Their results, however, indicated no significant differences in the rate of deoxygenation between the two exercise modes or between genders.

Jensen-Urstad et al. (1995) evaluated the changes in biceps brachii oxygenation during arm cranking exercise performed at a constant work rate for 15 min.
Table 3  Studies on Muscle Oxygenation and Blood Volume Responses Measured by NIRS During Treadmill Walking and Running in Healthy Subjects

<table>
<thead>
<tr>
<th>Authors, Subjects</th>
<th>Test Protocol</th>
<th>Instrument</th>
<th>Muscle Evaluated</th>
<th>NIRS Findings</th>
</tr>
</thead>
<tbody>
<tr>
<td>Demarie et al. (2001) 11 M soccer players</td>
<td>Running velocity that was midway between onset of blood lactate accumulation (4 mMol/L and VO₂max)</td>
<td>HEO-100, OMRON</td>
<td>Vastus lateralis</td>
<td>Muscle oxygenation showed a consistent decrease between 3rd, 6th, and final minute of exercise; Muscle blood volume decreased initially and recovered to baseline values to remain constant for remainder of the test; Delta values of muscle deoxygenation were correlated with delta values of oxygen uptake (+) and blood lactate (–).</td>
</tr>
<tr>
<td>Quaresima et al. (2002) 5 min walking @ 9.6 kmh</td>
<td>5 min walking @ 3.2 kmh</td>
<td>NIRO-300*</td>
<td>Vastus lateralis</td>
<td>TOI was higher when walking compared to running in both muscles at the end of each minute; Differ. was smaller for medial gastrocnemius compared to vastus lateralis; TOI and venous oxygen saturation during recovery were significantly higher after walking compared to running; Mismatch between muscle blood flow and TOI in both muscles.</td>
</tr>
<tr>
<td>Quaresima et al. (1996) 10 untrained volunteers</td>
<td>Protocol 1: 12 min @ 1.6 kmh; 0 to 6° slope in 4° increments every 2 min</td>
<td>NIRO 500*</td>
<td>Medial gastrocnemius</td>
<td>Two distinct patterns of oxyhemoglobin [HbO₂] and deoxyhemoglobin [Hb] were noted: (a) systematic increase throughout walking test suggesting a continual increase in oxygen demand; (b) initial increase followed by a more gradual increase during elevation phase, suggesting a balance between oxygen supply and demand during this phase. Heterogeneous patterns of [HbO₂] and [Hb] during incremental running to exhaustion; Greatest [Hb] increase occurred at maximal treadmill velocity when [HbO₂] was constant, implying that oxygen supply was the limiting factor.</td>
</tr>
<tr>
<td>Hiroyuki et al. (2002) 8 healthy M</td>
<td>4 min walking @ 4 &amp; 6 kmh followed by increments of 2 kmh every 2 min until 16 kmh</td>
<td>HEO-100, OMRON</td>
<td>Vastus lateralis, Gastrocnemius</td>
<td>Muscle oxygenation decreased in proportion to walking and running velocity; Negative relationship between pulmonary oxygen uptake and muscle oxygenation in vastus lateralis (r = –0.803 to –0.986) and gastrocnemius (r = –0.848 and –0.963); Significant difference in degree of muscle oxygenation between vastus lateralis and gastrocnemius at running speeds of 10 and 12 kmh.</td>
</tr>
</tbody>
</table>

*Note: This instrument is a 2-channel spatially resolved oximeter designed to measure tissue oxygenation saturation (TOI), expressed as a %.
during normoxia and hypoxia (12% inspired oxygen). Their results indicated a rapid initial decrease in oxygenation during the first few minutes of exercise with a steady reversal during the latter phases of the test under both conditions. The reversal was slower in all subjects under hypoxic conditions compared to normoxic conditions. The reason for this recovery in Mox after several minutes of submaximal exercise is unclear at present and needs to be further investigated.

**Muscle Oxygenation Trends During Anaerobic Exercise**

The Wingate cycle ergometer test, which is performed for 30 sec at maximal intensity against a resistance proportional to the body mass, is a standardized test for measuring anaerobic power and capacity. Several studies have demonstrated that peak oxygen uptake (peak VO\(_2\)) during the Wingate test is considerably high and can reach 85% of peak VO\(_2\) attained during a stepwise incremental exercise test (Calbet et al., 1997; Seresse et al., 1988). As well, studies which have modified the Wingate test by increasing its duration to 45 sec and 60 sec (Withers et al., 1991) have demonstrated peak VO\(_2\) values approaching those attained during a stepwise incremental test. These findings suggest there should be a considerable degree of deoxygenation in the muscles during these Wingate tests. The limited data currently available support this hypothesis.

Bae et al. (2000) demonstrated a decline in vastus lateralis Mox from the 3rd to the 15th second of a 30-sec Wingate test, with a leveling off during the final 15 sec of the test. They observed significant muscle deoxygenation and reoxygenation during intermittent bouts of exercise and recovery, respectively (10 sec work : 20 sec recovery) over 15 min of exercise at intensities just below ventilatory threshold and at maximal power output attained during interval exercise. There was no significant difference in the rate of deoxygenation (calculated over 10 sec) between the Wingate test and the maximal interval test, with both values being significantly higher than the interval test performed below ventilatory threshold. Bae et al. (2000) also reported a significant correlation (\(r = 0.976\)) between the rate of decline in Mox during the Wingate test calculated over 10 sec and the subjects’ VO\(_2\)max. These findings suggest that: (a) aerobic metabolism makes a significant contribution toward energy production even during high intensity supramaximal exercise; and (b) an enhanced aerobic power could be beneficial during high intensity exercise.

Bhambhani et al. (2001) reported that peak oxygen uptake during 30- and 45-sec Wingate tests in young subjects corresponded to 85% and 93% of peak VO\(_2\) attained during a stepwise incremental cycle ergometer test. However, no significant differences were observed between the three tests for oxygen pulse, i.e., the oxygen utilized per heartbeat. As well, no significant differences were observed between these tests for Mox, suggesting that peripheral oxygen extraction was most likely similar during the three test protocols. Based on these observations, Bhambhani et al. (2001) hypothesized that the most likely reason for the higher peak VO\(_2\) during the incremental test vs. the 30- and 45-sec Wingate tests was the higher oxygen transport resulting from greater cardiac output. In a cross-sectional study on male sprinters, Nioka et al. (1998) reported a significant difference in maximal muscle deoxygenation between a 30-sec Wingate test and a
stepwise incremental test designed to elicit VOₐ₂max. One group of sprinters elicited 40% of the maximum cuff ischemia deoxygenation during the incremental VO₂max test, whereas another group elicited 80% of the cuff ischemic value during a 30-sec Wingate test. However, since both groups of sprinters did not undertake both exercise tests, these results should be interpreted with caution. It is likely that interindividual differences in anaerobic and aerobic fitness could have influenced the findings.

Effects of Aging on Muscle Oxygenation

The effects of aging on cardiorespiratory responses have been the focus of extensive research during the last two decades. However, limited research has applied NIRS to evaluate the peripheral response to dynamic exercise in this population. Costes et al. (1999) compared the vastus lateralis oxygen saturation during incremental cycle exercise to voluntary exhaustion in 10 older subjects (mean age 67 ± 5 yrs) and 13 younger subjects (mean age 27 ± 4 yrs). Although the slope of the oxygen uptake/power output relationship between the two age groups was similar, the rate of decline in muscle oxygen saturation was significantly faster in the older subjects. However, this difference was negated when saturation was expressed relative to the subjects’ VO₂max. Moreover, there was no significant difference in the change (maximum – resting) in muscle oxygen saturation between the two groups during the transition from rest to maximal exercise. As well, the variations in muscle oxygen saturation were not correlated with the VO₂max of the subjects, and the half recovery time of muscle oxygen saturation did not differ significantly between the two groups. Costes et al. (1999) concluded that the lower muscle oxygen saturation at a given absolute oxygen uptake in the older subjects was due to an age related decrease in muscle blood flow, which was demonstrated by NIRS using the arterial occlusion procedure.

Recent evidence using the thermodilution technique has indicated attenuation in leg blood flow response during cycling at moderate work intensities in older women (Proctor et al., 2003a), but not in older men (Proctor et al., 2003b). These findings suggest that NIRS can be a useful technique in studying the peripheral changes that occur with aging. Future studies should evaluate the effects of different exercise training programs that will enhance or minimize the attenuation in oxygen saturation that may occur in the peripheral musculature as a result of the aging process.

Muscle and Cerebral Oxygenation Trends During Exercise

Research has demonstrated that NIRS is useful in evaluating the changes in cerebral oxygenation and cerebral blood volume during a variety of tasks involving motor stimulation, cognition, and visual perception. These findings consistently demonstrate a systematic increase in Cox and CBV during these tasks, and tend to support observations from other techniques such as functional magnetic resonance imaging and positron emission tomography that have been used to evaluate cerebral metabolism (Obrig and Villringer, 1997). A few studies (see Table 4) have used NIRS at multiple sites to examine the regional changes in tissue (cerebral and
<table>
<thead>
<tr>
<th>Authors, Subjects</th>
<th>Tissues Evaluated</th>
<th>Intensity</th>
<th>NIRS Findings</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nielsen et al. (1999)</td>
<td>Cerebral, Vastus lateralis</td>
<td>6 min maximal rowing ergometer test</td>
<td>Arterial oxygen saturation decreased from 98% to 91.9% during maximal rowing; Cerebral oxygen saturation decreased 17% during rowing due to an increase in DeoxyHb and an increase in OxyHb; Total Hb was unchanged; Muscle oxygenation decreased significantly during maximal rowing; Inhalation of 30% inspired oxygen reversed the cerebral desaturation during maximal exercise without any change in muscle oxygenation.</td>
</tr>
<tr>
<td>11 trained M rowers</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>INVOS 3100</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>NIRO 500</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nielsen et al. (2001)</td>
<td>Intercostals, Vastus lateralis, Cerebral</td>
<td>3 min @ 150 W with no external resistance to breathing; 4 levels of resistive breathing at same work rate for 3 min; 100 W increments until exhaustion under normal breathing.</td>
<td>Change in deoxyhemoglobin was not affected by low to moderate levels of resistive breathing; Significant deoxygenation of vastus lateralis and intercostals only at higher levels of resistive breathing; Significant increases in OxyHb, DeoxyHb, and total Hb as a result of resistive breathing; Carbon dioxide level in the blood influences blood flow to active muscle although its magnitude is smaller than that for the brain.</td>
</tr>
<tr>
<td>8 M</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>NIRO 300</em></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>NIRO 500</em></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
muscle) oxygenation and blood volume during dynamic exercise, in order to gain
a better understanding of the acute changes in oxygen availability that occur under
these conditions.

Nielsen et al. (1999) examined the regional changes in muscle and cerebral
oxygenation simultaneously during a 6-min maximal rowing effort in elite rowers.
The results indicated that vastus lateralis muscle oxygenation decreased signifi-
cantly during maximal rowing which was consistent with other forms of dynamic
exercise. Cerebral oxygen saturation estimations obtained simultaneously from
the left frontal lobe indicated a decline from 82% at rest to 65% during maximal
exercise. Nielsen et al. (1999) attributed this to a significant decrease in arterial
saturation (%SaO₂) during maximal exercise because, when these experiments
were repeated under hyperoxia (FIO₂ = 0.30), %SaO₂ was maintained during maxi-
mal exercise and the decline in cerebral oxygen saturation observed under normoxic
conditions did not occur.

In another study, Nielsen et al. (2001) used spatially resolved NIRS to study
regional changes in the deoxyHb and oxyHb concentration of the vastus lateralis,
intercostals, and frontal lobe simultaneously during submaximal and maximal cy-
cling with resistive breathing. The results indicated that breathing against resis-
tance induced a significant increase in ventilation rate, while %SaO₂ and PETCO₂
were significantly reduced. These trends were reversed when the breathing resis-
tance was removed. Whole-body VO₂ at all levels of resistive breathing did not
differ significantly from the control (unresisted breathing) condition. Low to mod-
erate resistive breathing did not alter the vastus lateralis and intercostal deoxyHb
concentration compared to the control condition. However, intense resistive breath-
ing induced a significant increase in vastus lateralis and intercostal deoxyHb con-
centration. Cerebral deoxyHb and oxyHb concentrations increased significantly
as a result of moderate and intense resistive breathing, and consequently total Hb
(an index of blood volume) was also significantly elevated. On the basis of these
findings and the PETCO₂ measurements undertaken during exercise, Nielsen et al.
(2001) suggested that CO₂ production during exercise helps regulate regional blood
flow to both the active muscles and the brain, with a more modest effect on muscle
tissue.

**Endurance Training and Muscle Oxygenation**

Currently there is limited evidence that has examined the effects of exercise train-
ing on muscle oxygenation responses during exercise. Neary et al. (2002) evalu-
ated the effects of 3 weeks of intense endurance training on peak cardiorespiratory
responses and vastus lateralis oxygenation changes in moderately trained cyclists/
triathletes. The evidence indicated significant improvements in peak VO₂ with
concomitant increases in muscle deoxygenation at peak exercise. Neary et al. hy-
pothesized that localized muscular factors such as increased mitochondrial den-
sity, capillarization, and oxidative enzyme characteristics most likely contributed
to these peripheral adaptations following training.

Costes et al. (2001) evaluated the effects of 4 weeks of intensity endurance
training on steady-state submaximal exercise responses (50% and 70% of pretraining
VO₂max) in healthy men. Although there was a tendency for significant decreases
in heart rate, blood lactate, and muscle oxygenation during the 15 minutes of steady-state exercise at 50% of VO₂max following training, these changes were not statistically significant. However, at 70% of pretraining VO₂max, significant reductions were observed in heart rate and blood lactate concentrations at the same absolute VO₂ following training. These improvements, which were characteristic of endurance training, were accompanied by a significant reduction in muscle deoxygenation and a corresponding increase in localized blood volume.

Since NIRS reflects the balance between oxygen delivery and utilization at the level of the small blood vessels, these findings suggest that the reduction in oxygen utilization was due to an enhanced oxygen supply following training. However, their findings indicated that the change in muscle oxygenation was not related to the increased capillary density observed from muscle biopsy analysis following training. Costes et al. (2001) did not report the changes in muscle oxygenation and blood volume during maximal exercise following training.

From these observations, it appears that NIRS is sensitive to changes in intracellular metabolism that result from endurance training. However, one limitation of continuous-wave NIRS is that the improvements observed are qualitative in nature, and therefore the magnitude of the peripheral contribution cannot be quantified. Nevertheless, the alterations in muscle oxygenation, when reported in conjunction with the blood volume changes, provide useful information pertaining to the nature of the peripheral adaptations that occur with training. Given the lack of training studies in this area, it is recommended that future NIRS training studies be designed to evaluate: (a) the lactate and ventilatory thresholds in conjunction with the maximal exercise responses, and (b) measures of central circulation (cardiac output and stroke volume) simultaneously, so as to examine the relative contributions of the central and peripheral factors that enhance VO₂max following training.

**Influence of Adipose Tissue on Muscle Oxygenation**

Since NIRS measures changes in muscle oxygenation percutaneously, it is likely that the skin and adipose tissue confound the absorbency measurements during in vivo exercise protocols (McCully and Hamaoka, 2000; van Beekvelt et al., 2001). Of these two, the contribution of the skin to the total NIRS signal is quite small: approximately 5% if the source and detector of the NIRS probe are separated by more than 20 mm (Hampson and Piantidosi, 1988). The major factor confounding the muscle oxygenation measurements is the thickness of the adipose tissue, because the absorption and metabolic properties of fat and muscle differ considerably. Theoretically, a thicker fat layer will result in less light penetrating the active muscle tissue, thereby resulting in reduced light absorption and a stronger NIRS signal. However, the lower metabolic activity of the fat layer, coupled with lower Hb levels, will result in a lower absorbency change during the exercise protocol. The interaction of these two factors can lead to inaccuracies when recording muscle oxygenation trends during exercise.

Research (van Beekvelt et al., 2001) that has systematically examined the influence of adipose tissue thickness on the NIRS-determined oxygen consumption of the flexor digitorum superficialis muscle during sustained isometric handgrip
exercise has indicated an inverse relationship between these two variables at 10%, 20%, and 30% of maximal voluntary contraction. No significant correlations were observed between adipose tissue thickness and forearm blood flow measured by the arterial occlusion technique using NIRS. These findings have been supported by other researchers (Homma et al., 1996; Matsushita et al., 1998; Yamamoto et al., 1996; 1998) who have demonstrated reduced tissue absorbency with increasing levels of adipose tissue thickness. Although these researchers have suggested mathematical corrections to the NIRS data obtained, they cannot be universally applied because of differences in the NIRS systems and techniques used. Since the NIRS probes are generally designed to have a penetration depth of approximately 50% to 60% of the inter-optode distance (usually 2 to 2.5 cm), it is recommended that the adipose tissue thickness at the NIRS probe site be well below this value so as to ensure penetration of the photons into the muscle tissue.

**Conclusions**

On the basis of the available evidence, it appears that continuous-wave NIRS can provide useful information pertaining to muscle oxygen saturation and blood volume during dynamic exercise. Sufficient evidence is available to show that NIRS can be used to identify the lactate (ventilatory) and respiratory compensation thresholds during incremental exercise. There is also conclusive evidence showing that NIRS is sensitive to changes in whole-body oxygen uptake kinetics during constant work-rate exercise at intensities above the lactate/ventilatory threshold. As well, it appears that the degree of muscle deoxygenation at the same absolute oxygen uptake is significantly lower in older persons compared to younger ones. However, these changes are negated when muscle oxygenation is expressed relative to maximal oxygen uptake values.

Evidence from NIRS studies indicates there is no significant difference in the rate of biceps brachii and vastus lateralis deoxygenation during arm cranking and leg cycling exercise, respectively, in males and females. The muscle deoxygenation trends recorded during short-duration, high-intensity exercise such as the Wingate test suggest that a substantial degree of aerobic metabolism occurs during such exercise. Although direct evidence is lacking, it appears that NIRS is sensitive to changes in intracellular metabolism which occur as a result of endurance training in healthy subjects. NIRS also appears to be a suitable technique for evaluating changes in cerebral oxygenation and blood volume during dynamic exercise. The use of NIRS at multiple sites, such as brain and muscle tissue, provides useful information pertaining to regional changes in oxygen availability in these tissues. Future studies should evaluate these changes under a variety of experimental conditions and in different populations so that a better understanding of the central and peripheral factors that influence exercise performance can be obtained.

**References**


Received July 4, 2003; accepted in final form February 17, 2004.