J. Physiol. Sci. Vol. 56, No. 3; Jun. 2006; pp. 267–268
Online Aug. 31, 2006; **doi:10.2170/physiolsci.L656**

Evaluation of the Skin Blood Flow Contribution to the Non-Invasive Measurement of Muscle Oxygenation by Near Infrared Spectroscopy

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Key words: oxygenation, near infrared spectroscopy, muscle.

In a recent article in the *Jpn. J. Physiol.* Buono *et al.* [1] present the results of a study investigating the influence of the changes in the skin blood flow on the near infrared spectroscopy (NIRS) based measurement of the rectus femoris muscle oxygenation during local heating or epinephrine injection. Tissue oxygenation was measured by a continuous-wave tissue oximeter (InSpectra™ Tissue Spectrometer System–Model 325, Hutchinson Technologies, Hutchinson, MN, USA).

We agree with the authors on: (i) evidencing scarce and inaccurate prior studies on this topic [2–4]; (ii) recognizing the importance to investigate the influence of high and low skin blood flow on the measurement of muscle oxygenation by NIRS. On the other hand, we disagree with the authors on the NIRS methodology (in particular, the light source–detector spacing) adopted in their study.

The light source–detector distance affects the contribution of skin; in fact increasing this distance properly allows the improvement of the sensitivity of measurement and the increase of the probability of looking at oxygenation deep under the tissue surface [5]. In addition, as already reported by the authors, it is well known that the depth of light penetration also depends on the thickness of subcutaneous adipose tissue [6, 7]. For these reasons, were the authors wise to have used a source–detector distance of 2.5 cm? This distance is very short, and to convince the readers that the reported results truly refer to the oxygenation changes occurring in the investigated muscle tissue, the authors should have explored the relationship between skin blood flow and the oxygenation measured at different source-detector distances (larger than 2.5 cm). In fact, the InSpectra™ Tissue Spectrometer optical cable distal tips are available at 25 and 35 mm spacing. Therefore, their generalized conclusion "skin blood flow can significantly affect tissue oxygenation, as measured by NIRS," is not supported by sufficient experimental NIRS data. We would like to add that a similar study has been very recently published by Davies *et al.* [8], who investigated the influence of the increase in the skin blood flow on the NIRS based measurement of the flexor digitorum muscle oxygenation during local or whole-body heating. Unfortunately also in that study, performed by using another NIRS system, the source–detector distance (2 cm) was inadequate to support their findings. We would like to comment that the other commercially available tissue oximeters, utilized also for brain oxygenation monitoring, use a fixed source–detector spacing of 4 or 5 cm, and in most cases, efficient light sources (laser diodes instead of the light-emitting diodes as in the InSpectra™ Tissue Spectrometer) to ensure an accurate quantification of the oxygenation changes in muscle tissue.

In summary, the study on the potential contribution of very high levels of skin blood flow to the muscle oxygenation is of great interest for better understanding the potentiality of NIRS in exercise patho-physiology. However, this issue is still open and further studies should be carried out using more recent NIRS methodologies [9, 10], which include suitable light source-detector distances, for investigating deep regions of muscle.

We believe that this letter adds important discussion and clarifies to the readers of *Jpn. J. Physiol.* relevant issues raised in the paper by Buono *et al*.

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Received on Feb 8, 2006; accepted on Feb 26, 2006; released online on Aug 31, 2006; doi:10.2170/physiolsci.L656
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Response

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We would like to thank Drs. Quaresima and Ferrari for their letter to the editor concerning our recent study that appeared in the *Japanese Journal of Physiology*. Both Dr. Quaresima and Dr. Ferrari are renowned experts in the area of NIRS and thus their concerns certainly warrant a detailed and thoughtful response.

Essentially they question whether our use of a NIRS probe with a 25 mm source–detector spacing was a wise idea. Our response is that we feel confident that the probe spacing was adequate and did not invalidate our results. We base this response on the following two points.

First, when using a 25 mm probe it has been reported that 95% of the optical signal is from a depth of 0–23 mm [1]. Since our subjects had a mean skin and subcutaneous tissue thickness of 7.7 mm, this means that at least 15 mm of superficial muscle was included in the optical signal. This statement is supported by the findings of Matsushita *et al.* [2] who reported that a source–detector distance of 20 mm was enough for detection of the NIR light passing through the muscle layer, even when the thickness of the adipose tissue was 15 mm.

Second, if the 25 mm probe spacing was not allowing NIR light to reach the underlying muscle layer, than it seems reasonable to assume that the optical signal should not respond to metabolic perturbations occurring in that tissue during exercise. However, several studies [1, 3] using the Hutchinson Technologies InSpectra Spectrometer and a 25 mm probe have reported large decreases in tissue oxygenation during exercise. In fact our laboratory routinely sees vastus lateralis StO_2 values in the 20–30% range following maximal exercise when using a 25 mm probe spacing (resting values are measured at 70–90%). We feel these findings certainly suggest that when using a 25 mm source–detector spacing a reasonable proportion of the NIRS signal is arising from the superficial muscle.

In summary, we agree with Drs. Quaresima and Ferrari that further studies examining the role of skin blood flow on the NIRS optical signal are warranted. Hopefully the concerns voiced by our two colleagues and our response will assist future investigators in designing studies to advance our collective knowledge in this important area.

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