

Research

Characterization of tissue oxygen saturation and the vascular occlusion test: influence of measurement sites, probe sizes and deflation thresholdsHernando Gómez¹, Jaume Mesquida^{1,2}, Peter Simon¹, Hyung Kook Kim¹, Juan C Puyana³, Can Ince⁴ and Michael R Pinsky¹¹Department of Critical Care Medicine, University of Pittsburgh, 606 Scaife Hall, 3550 Terrace Street, Pittsburgh, PA 15261, USA²Department of Medicine, Universitat Autònoma de Barcelona, Barcelona, 08193 Spain³Department of Surgery, University of Pittsburgh, 200 Lothrop Street, Pittsburgh, PA 15213, USA⁴Department of Intensive Care, Erasmus MC, University Medical Center Rotterdam, dr. Molewaterplein 40-60 3000 Dr Rotterdam, The NetherlandsCorresponding author: Michael R Pinsky, pinskymr@upmc.edu

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Critical Care 2009, **13(Suppl 5):S3** (doi:10.1186/cc8001)**Abstract**

Introduction: Tissue oxygen saturation (StO₂) and the vascular occlusion test (VOT) can identify tissue hypoperfusion in trauma and sepsis. However, the technique is neither standardized nor uses the same monitoring site. We hypothesized that baseline and VOT StO₂ would be different in the forearm (F) and thenar eminence (TH) and that different minimal StO₂ values during the VOT would result in different reoxygenation rates (ReO₂).

Methods: StO₂ and its change during the VOT were simultaneously measured in the F and TH, with 15 mm and 25 mm probes, using the 325 InSpectra monitor in 18 healthy, adult volunteers. Two VOTs were done to a threshold thenar StO₂ of 40% interchanging the 15 mm and 25 mm probes between sites. Two additional VOTs were done to thresholds of 50% and 30%. Baseline StO₂ (BaseO₂), the deoxygenation rate (DeO₂) and ReO₂ (%O₂/minute) were compared between sites, probes and thresholds. Results are presented as the median (interquartile range), *P*-value.

Results: BaseO₂, DeO₂, ReO₂, area under the curve and hyperemia duration values were different when comparing TH vs. F and 15 mm vs. 25 mm probes. ReO₂ was different between different thresholds for the TH and 15 mm probes. TH_{15 mm} vs. F_{15 mm}: BaseO₂, 90.4 (85.2, 93.5) vs. 85.2 (80.7, 90.2), *P*=0.031; DO₂, -12.1 (-16.2, -11.3) vs. -8.5 (-10.3, -7.8), *P*=0.011; ReO₂, 297.2 (213.7, 328.6), *P*<0.0001; 15 mm vs. 25 mm probe: BaseO₂, 97.2 (89.4, 94.7) vs. 87.3 (81.7, 90.9), *P*=0.016; DeO₂, -18.0 (-24.1, -14.8) vs. -9.9 (-15.3, -6.5), *P*<0.0001; and ReO₂, 401.6 (331.7, 543.2) vs. 160.5 (132.3, 366.9), *P*=0.012, respectively. TH_{15 mm} vs. TH_{25 mm}: BaseO₂, *P*=0.020; DeO₂, *P*<0.0001; and ReO₂, *P*<0.0001. Threshold StO₂ values (15 mm probe only): ReO₂, *P*=0.003; DeO₂, *P*=0.60. ReO₂ at 40% and 50% StO₂ thresholds, *P*=0.01.

Conclusion: BaseO₂, DeO₂ and ReO₂ were different when measured in different anatomical sites (F and TH) and with different probe sizes, and ReO₂ was different with differing VOT release StO₂ threshold values. Thus, standardization of the site, probe and VOT challenge need to be stipulated when reporting data.

Introduction

Non-invasive measurements of tissue oxygen saturation (StO₂) using near-infrared spectroscopy have been studied to assess tissue hypoperfusion in different populations. The absolute StO₂ has been proven instrumental in predicting outcome in trauma patients [1,2]. These same studies, however, have shown limited discriminating capacity for the absolute StO₂ value (sensitivity, 78%; specificity, 39%) and have even shown it to be at least as good as systolic blood pressure (sensitivity, 74%; specificity, 32%) [1]. The addition of a provocative test (ischaemic challenge) to the measurement of the StO₂ would potentially enhance this discriminatory power between different cardiovascular states by creating emerging parameters to follow, such as the deoxygenation and reoxygenation slopes. Furthermore, the use of such provocative tests [3], specifically the vascular occlusion test (VOT), has been shown to improve and expand the predictive ability of StO₂ to scenarios such as trauma, severe sepsis and septic shock [4,5].

The VOT is a provocative test in which StO₂ is measured on a distal site (such as the thenar eminence (TH)) whilst a transient rapid vascular occlusion is performed, using a sphygmomanometer, for either a defined time interval (for

BaseO₂ = baseline tissue oxygen saturation; DeO₂ = deoxygenation rate; F = forearm; ReO₂ = reoxygenation rate; StO₂ = tissue oxygen saturation; TH = thenar eminence; VOT = vascular occlusion test.

example, 3 minutes) or until the StO_2 decreases to a defined minimal threshold. Once this threshold is reached, the vascular occlusion is released and the StO_2 is allowed to recover. Several emergent parameters arise from this technique, including the rate of deoxygenation (DeO_2), thought to reflect the local metabolic rate, the rate of reoxygenation (ReO_2), which reflects the time required to wash out stagnant blood and is thought to be determined by local cardiovascular reserve and microcirculatory flow, and the postobstructive hyperemic response.

The VOT, however, has not been standardized. In fact, multiple studies report results using different anatomical StO_2 measuring sites (TH, forearm (F), deltoid) [4,6-8], different near-infrared spectroscopy probe sizes (15 mm vs. 25 mm) [4,8-12], and different types and levels of deflation thresholds (StO_2 of 40%, 50%) versus time thresholds (3 minutes, 4 minutes, 5 minutes) [4,6,11,13]. Differences between anatomical sites may potentially affect near-infrared spectroscopy-derived parameters given the fact that fat and the muscle thickness distribution along the body is variable. Furthermore, given that increasing the sensing and illuminating probe distance augments the depth that near-infrared light travels, the use of different probes may screen different capillary beds at any given time. Finally, since the primary reason to perform the VOT is to produce a transient ischaemic challenge to note the subsequent ReO_2 , the level of ischaemic challenge might vary for the same ischaemic time or threshold StO_2 . As a consequence, caution must be taken when comparing results from the available literature on the VOT, as the test may be inadequate [14,15].

We hypothesized that absolute StO_2 values and VOT-derived StO_2 values obtained by monitoring different anatomical sites or using dissimilar depth may differ, and that different minimal StO_2 values during the VOT (deflation thresholds) would result in dissimilar ReO_2 rates.

Materials and methods

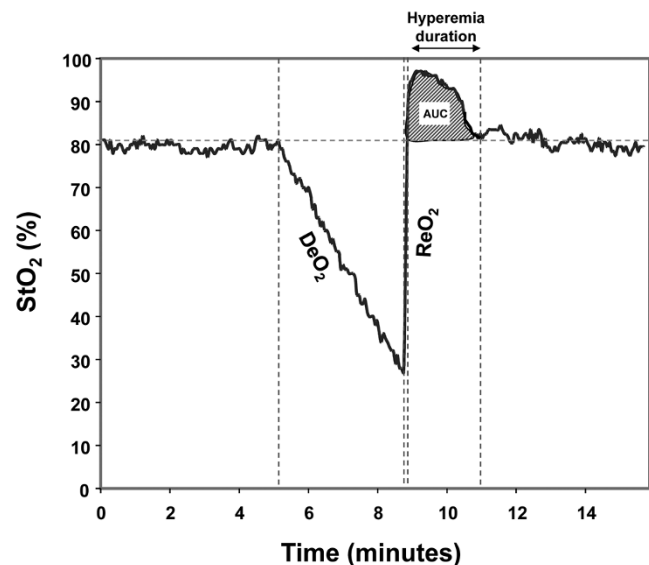
The present study was approved by the Institutional Review Board of the University of Pittsburgh. Informed consent was obtained from every volunteer enrolled in the study prior to performing any procedure.

Near-infrared spectroscopy

StO_2 and its change during the VOT were simultaneously measured in the F and TH, with 15 mm and 25 mm probes, using two 325 InSpectra[®] tissue spectrometers (Hutchinson Technology Inc., Minneapolis, MN, USA) on the same arm in 18 healthy, adult volunteers. Placement of the TH probe was done according to the manufacturer's recommendations as previously described [4,9]. The F probe was placed over the Flexor digitorum profundus in the F.

Research software (InSpectra Version vBeta1.1; Hutchinson Technology Inc.) on two independent laptop computers was used for data collection.

Figure 1



Tissue oxygen saturation (StO_2) during a vascular occlusion test. Calculated parameters: deoxygenation rate (DeO_2), reoxygenation rate (ReO_2), hyperemia area under the curve (AUC), and hyperemia duration.

Vascular occlusion test

The VOT was performed as previously described [9]. Briefly, the baseline arterial pressure was measured using a manual sphygmomanometer. An adult-size tourniquet cuff was then placed on the arm above both probes. After StO_2 signal stabilization at rest (<2% variation in 30 seconds), the tourniquet was inflated with an automatic pump (Portable Tourniquet System, ref. 9-2100-001; Delfi Medical Innovations Inc., Vancouver, BC, Canada) to >30 mmHg above systolic pressure and was kept inflated until the StO_2 decreased to 40% (deflation threshold). Inflation took approximately 3 to 4 seconds. The tourniquet was then rapidly deflated (<0.5 seconds) and the StO_2 response was followed until the StO_2 returned to baseline values. All repeated VOTs were performed after a minimal period of 5 minutes to allow the StO_2 to recover.

DeO_2 and ReO_2 slopes, the area under the curve of the hyperemic response as well as the duration of hyperemia (Figure 1) were calculated using the published methodology and a software package designed for this purpose and this specific tissue spectrometer (Version 3.03 InSpectra Analysis Program; Hutchinson Technology Inc.), as previously validated by us [9].

Protocol

Subjects were placed in a quiet environment while resting in a semirecumbent position with the studied arm resting over a cushion in the anatomical position. Probes were placed using a form-fitting plastic adhesive over the TH and volar surface

of the F. The 15 mm probe was initially placed on the F and the 25 mm probe placed on the TH. A VOT was then performed. After a 5-minute resting period, the probe locations were switched (that is, the 15 mm probe to the TH, and the 25 mm probe to the F) and a second VOT was performed. Subsequently, two additional VOTs were carried out to StO₂ deflation thresholds of 50% and 30% with the same probe disposition.

Healthy volunteers

Volunteers were selected according to the following inclusion and exclusion criteria: inclusion criterion, subjects older than 17 years (that is, ≥ 18 years); and exclusion criteria, acute or chronic cardiovascular or pulmonary disease, or subject taking any vasoactive medications.

Statistical analysis

Mean absolute StO₂ values, the DeO₂ and the ReO₂ (%O₂/minute) as measured by the 15 mm and 25 mm probes between the TH and F monitoring sites were compared using the Wilcoxon signed-ranks test. Comparison of the DeO₂ and ReO₂ when the VOT was performed at different StO₂ deflation thresholds (that is, StO₂ of 30%, 40% and 50%) was performed using the Friedman test and analysis of variance for repeated measures (after running normality tests). Results are presented as the median (interquartile range), *P*-value unless stated otherwise. Significance reports a difference between groups corresponding to *P* < 0.05 unless stated otherwise.

Results

Eighteen healthy volunteers were enrolled in the present study after obtaining informed consent. Demographic characteristics of the studied population are summarized in Table 1. The mean baseline StO₂, DeO₂, ReO₂ and their coefficients of variability for these measures for the various comparisons are summarized in Table 2.

The absolute StO₂, DeO₂ and ReO₂ values for a deflation threshold of 40% were different between the TH and F for both the 15 mm and 25 mm probes (Figures 2 and 3), and were also different at the TH site for the 15 mm and 25 mm probes (Figure 4). Not surprisingly, the TH DeO₂ was not altered by varying the deflation StO₂ thresholds from 30% to 50% when measured with the 15 mm probe. This result was expected given that the DeO₂ is dependent on the metabolic conditions of the tissue, independent of the intensity of the ischemic insult and given the specific calculation methodology [1]. If the metabolic rate of the sampled tissue was not varied, therefore, the DeO₂ should not be significantly modified. The ReO₂ was higher using the 30% threshold than either the 40% or 50% thresholds, which themselves were similar (Figure 5).

The coefficients of variability for the absolute StO₂, DeO₂ and ReO₂ values were also different when comparing both sites

Table 1

Demographic characteristics of the study population	
Characteristic	Value
Age (years)	32.9 ± 8.2
Gender	
Female	6 (33%)
Male	12 (67%)
Race	
Caucasian	10 (56%)
Hispanic	4 (22%)
Asian	3 (17%)
Indian	1 (5%)
Weight (kg)	75.5 ± 15.5
Height (cm)	175.8 ± 8.2
Temperature (°C)	36.1 ± 0.6
Systolic blood pressure (mmHg)	118 ± 12
Diastolic blood pressure (mmHg)	73 ± 7
Mean arterial pressure (mmHg)	88 ± 8
Heart rate (beats/minute)	69 ± 9

Data are presented as mean ± standard deviation or *n* (%).

and probes, being less with the TH than F measures and with the 15 mm than 25 mm probe size. We also observed a lower coefficient of variability with 40% and 50% thresholds as compared with the 30% threshold (Table 3).

The mean ± standard deviation hyperemic areas under the curve (Figure 1) as measured by TH_{15 mm}, TH_{25 mm}, F_{15 mm} and F_{25 mm} were 11.2 ± 8.8%O₂/minute, 7.7 ± 5.1%O₂/minute, 6.4 ± 5.7%O₂/minute and 13.2 ± 9.6%O₂/minute, respectively, and were all significantly different when compared among each other (Figure 6). Their coefficients of variability were 78.7%, 66.2%, 90.3% and 72.5%, respectively. The hyperemia durations (mean ± standard deviation) for the same measurements stated above were 2.3 ± 0.8 minutes, 1.8 ± 0.6 minutes, 1.6 ± 0.5 minutes and 2.8 ± 1.2 minutes, respectively. These were also statistically different from each other, as shown in Figure 6. Their coefficients of variability were 32.9%, 33.5%, 34.1% and 33.5%, respectively.

Discussion

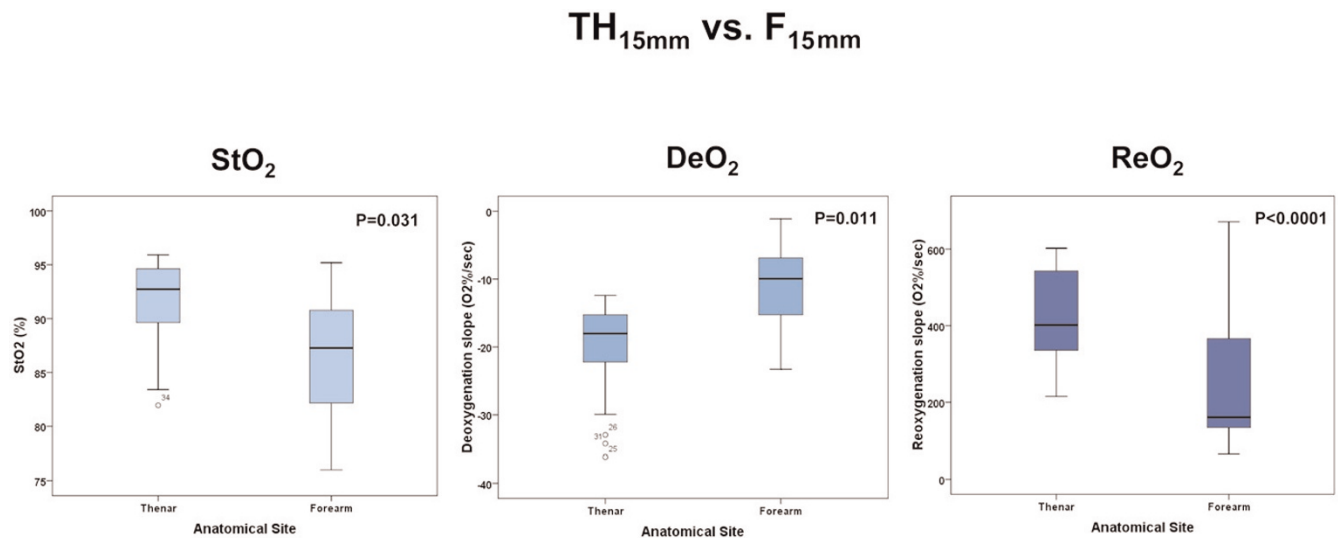
Baseline StO₂ values and their emergent parameters of the DeO₂ and the ReO₂, assessed by the VOT, are dependent on the site of measurement, the probe size and, to a certain extent, the ischaemic challenge. Varying any one of these components will significantly vary StO₂-related measures independent of the baseline cardiovascular status. When making inferences about cardiovascular function using these measures, therefore, care must be taken in comparing or

Table 2

Baseline tissue oxygen saturation, deoxygenation and reoxygenation rates between sites and between probe sizes						
Probe	Thenar eminence		Coefficient of variability (%)	Forearm	Coefficient of variability (%)	P value ^a
15 mm						
Absolute StO ₂ (%)	90.4 (85.2, 93.5), 76.6 to 95.3 ^b		5.6	85.2 (80.7, 90.2), 65.8 to 93.6 ^b	7.8	0.031
DeO ₂ (%O ₂ /minute)	-12.8 (-16.2, -11.3)		26.1	-8.5 (-10.3, -7.8)	42.7	0.011
ReO ₂ (%O ₂ /minute)	297.2 (213.7, 328.6)		21.5	98.6 (53.7, 157.8)	68.4	<0.0001
25 mm						
Absolute StO ₂ (%)	92.7 (89.4, 94.7), 81.9 to 95.9 ^b		4.6	87.3 (81.7, 90.9), 76.0 to 95.2 ^b	6.7	0.016
DeO ₂ (%O ₂ /minute)	-18.0 (-24.1, -14.8)		36.5	-9.9 (-15.3, -6.5)	53.6	<0.0001
ReO ₂ (%O ₂ /minute)	401.6 (331.7, 543.2)		28.2	160.5 (132.3, 366.9)	66.8	0.012
Comparison of 15 mm vs. 25 mm probe (P value)						
StO ₂	0.020					
DeO ₂	<0.0001					
ReO ₂	<0.0001					

Baseline tissue oxygen saturation (StO₂), deoxygenation rate (DeO₂) and reoxygenation rate (ReO₂) between the thenar eminence versus forearm and between the 15 mm probe versus 25 mm probe. Data presented as the median (interquartile range). ^aThenar eminence versus forearm comparison. ^bAbsolute StO₂ range.

Figure 2

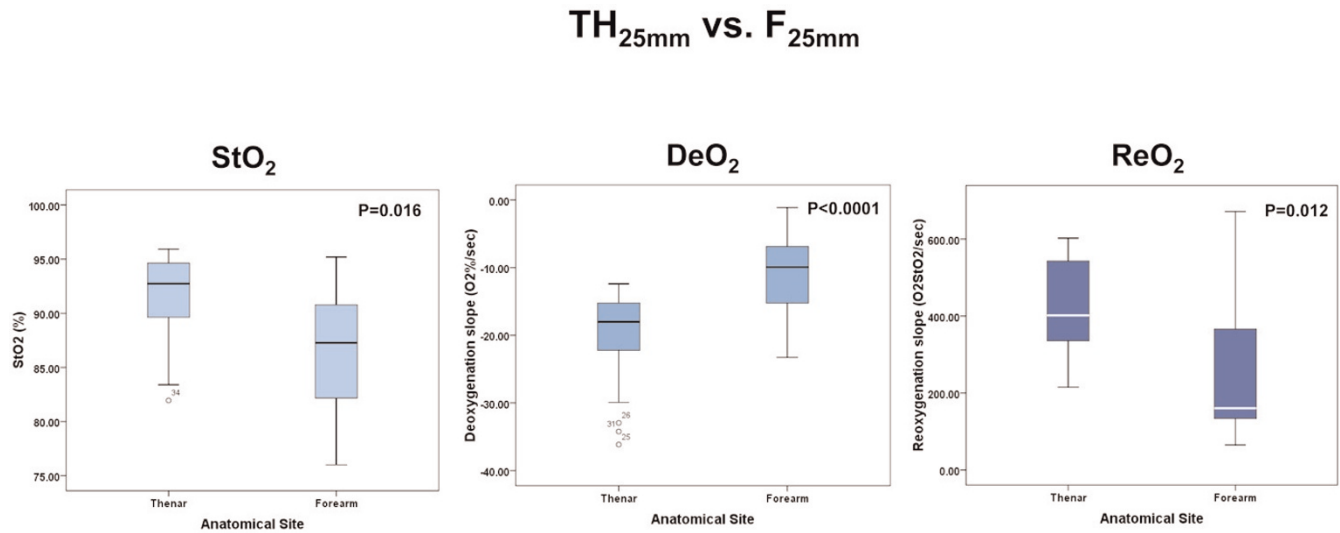


Box plots comparing tissue oxygen saturation (StO₂) parameters measured with the 15 mm probe at different anatomical sites. DeO₂, deoxygenation rate; ReO₂, reoxygenation rate.

combining results from different studies that use different measurement techniques and protocols. Presently, the VOT ischaemic challenge has not been standardized. Several investigators report the DeO₂ and the ReO₂ from a fixed ischaemic time (for example, 3 minutes) rather than to a defined threshold StO₂ value. Since the basal metabolic rate can vary amongst subjects, varying the DeO₂ proportionally

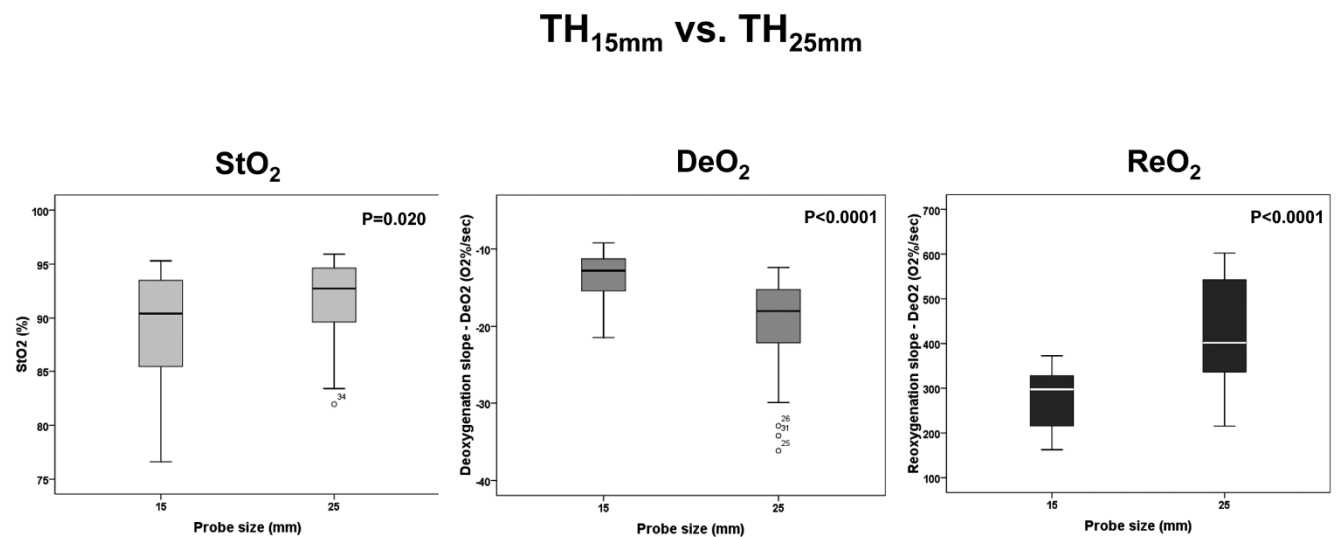
[3] using a fixed ischaemic time will result in differing threshold StO₂ values within and among subjects, making changes in the ReO₂ difficult to interpret among subjects if their metabolic rates are also different because they will reach different StO₂ nadirs for a constant ischaemic time. Our data therefore support using a defined ischaemic StO₂ threshold in future studies to minimize ReO₂ variability.

Figure 3



Box plots comparing tissue oxygen saturation (StO₂) parameters measured with the 25 mm probe at different anatomical sites. DeO₂, deoxygenation rate; ReO₂, reoxygenation rate.

Figure 4

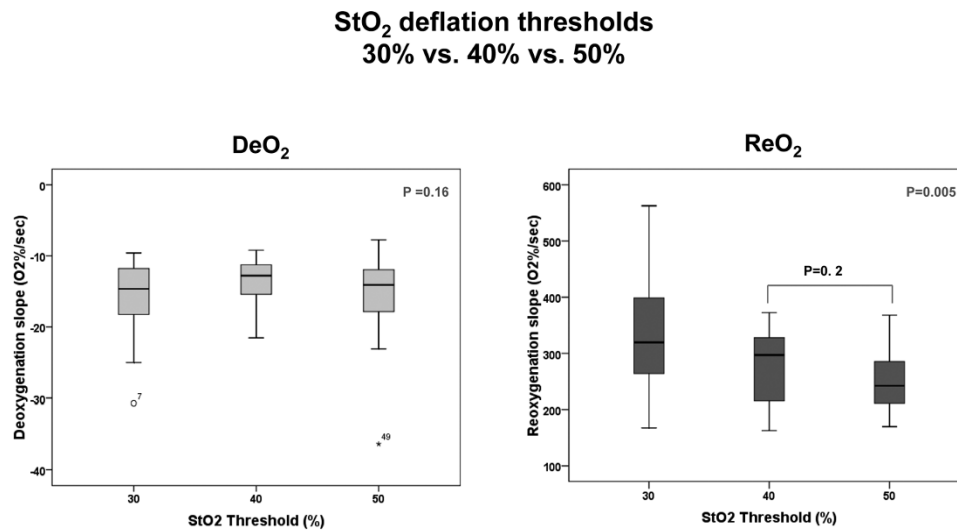


Box plots comparing tissue oxygen saturation (StO₂) parameters measured on the thenar eminence with different probes. DeO₂, deoxygenation rate; ReO₂, reoxygenation rate.

We found that the baseline StO₂, DeO₂ and ReO₂ values were consistently higher in the TH than F sites. Furthermore, the magnitude of the difference between the TH and F was greater for DeO₂ and ReO₂ values than for baseline StO₂ values. There was also a trend toward a greater variability for baseline StO₂ at F than at TH sites. This variability was increased for the DeO₂ and ReO₂. Although our study does not allow us to identify why this difference occurs, most

probably it reflects the greater degree of tissue and the associated vascular variability present in the F over the TH. The F has a variable layer of subcutaneous fat, which has a lower metabolic rate and minimal microcirculatory density when compared with muscle. The TH has almost no fat and is highly consistent amongst subjects. Our data therefore support using the TH site rather than the F site when measuring StO₂ and its VOT-related parameters.

Figure 5



Box plots comparing StO₂ parameters measured using different deflation thresholds. DeO₂, deoxygenation rate; ReO₂, reoxygenation rate.

Table 3

Deoxygenation rate and reoxygenation rate when using different deflation thresholds

Vascular occlusion test parameter	Tissue oxygen saturation deflation threshold			P-value
	30%	40%	50%	
DeO ₂ (%O ₂ /minute)	-14.6 (-18.5, -11.6)	-12.8 (-16.2, -11.3)	-14.1 (-18.0, -11.9)	0.6
ReO ₂ (%O ₂ /minute)	319.7 (258.2, 401.5)	297.2 (213.7, 328.6) ^a	242.5 (211.1, 296.2) ^a	0.01
Coefficient of variability for DeO ₂ (%)	34.4	26.1	39.3	
Coefficient of variability for ReO ₂ (%)	30.1	21.5	21.0	

Data are presented as the median (interquartile range). DeO₂, deoxygenation rate; ReO₂, reoxygenation rate. ^aComparison of ReO₂ obtained using deflation thresholds of 40% and 50% was not significant (P = 0.2).

We also found consistently lower baseline StO₂ values with the 15 mm probe than with the 25 mm probe, and this difference increased with VOT-related parameters. Although baseline StO₂ values displayed a similar variability (5.6% vs. 4.6%), VOT-derived measurements performed with the 15 mm probe showed less variability than those performed with the 25 mm probe. Presumably, by sampling a smaller amount of tissue volume, the 15 mm probe is less subject to measurement error. Our data support the routine use of the 15 mm probe over the 25 mm probe in future studies.

The hyperemic response, as quantified by the area under the curve and the duration, varied according to the anatomical site of measurement and the probe size used. Furthermore, great variability was found among healthy volunteers with regards to both parameters, as previously pointed out by our group [9]. These results suggest that the hyperemic response is not a reproducible parameter, and has been

proven to be highly dependent on individual variability. Thus, it may not be a reliable parameter for clinical use.

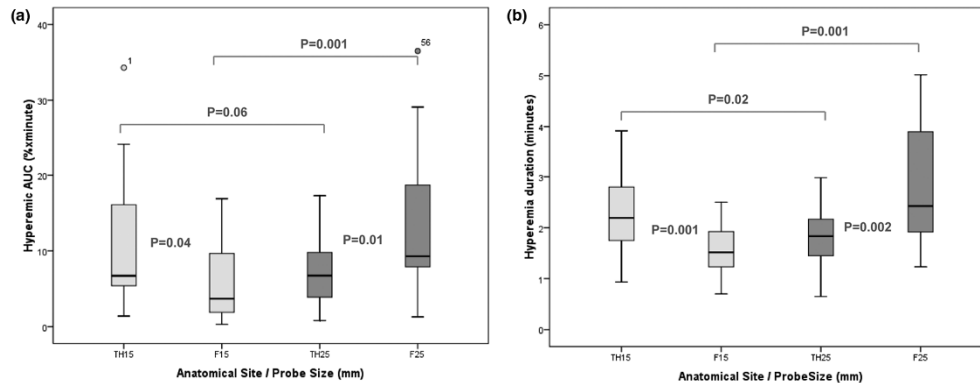
Limitations

The present study has two primary limitations.

First, we studied only two anatomical sites, whereas StO₂ can be measured anywhere on the body. Extrapolation of these findings to other anatomical sites, such as the abdominal wall, thigh and masseter muscles, should be done with caution. Since the VOT can only be carried out realistically on a peripheral site for which transient ischaemia will have no lasting deleterious effects, however, these studies reflect the present clinical practice of near-infrared spectroscopy VOT analysis.

Second, we did not study critically ill patients but healthy volunteers. Although this was the goal of the study, its application to critically ill patients might be limited. Previous

Figure 6



Box plot comparison of the hyperemic response between anatomical sites and probe sizes. AUC, hyperemic area under the curve.

studies by ourselves and others document that cardiovascular insufficiency can markedly increase intersubject variability [4,9]. Our data demonstrated a consistently lower coefficient of variation whenever the combination of TH site and 15 mm probe was used, which suggests that this site may also yield more consistent data in clinical trials. Similarly, using a defined ischaemic threshold for the VOT, method-specific variability of the ReO_2 should also be minimized.

Competing interests

HG, JM and HKK are co-investigators in a Hutchinson-sponsored clinical trial. CI has received honorarium for speaking at a Hutchinson Symposium, and is a co-investigator on a Hutchinson-sponsored clinical trial. MRP and JCP have received an honorarium for speaking at a Hutchinson Symposium, and are co-investigators on a Hutchinson-sponsored prospective clinical trial. PS declares that he has no competing interests.

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