

The use of near infrared spectroscopy (NIRS) as a diagnostic tool to measure microvascular haemodynamics in bone tissue.



Project Background:

Bone is a dynamic and highly vascular tissue, but measuring markers of microvascular haemodynamics within bone is currently difficult. There are logistical and technical limitations with existing tests based around MRI and radioisotope scans, in part due to bone's high density and mineral content. This complicates studying bone diseases where microvascular dysfunction plays a pathogenic role.

Near infrared spectroscopy (NIRS) has the potential to measure bone haemodynamic markers in real time and is safe and inexpensive. It also provides information on oxygen levels within bone, previously only possible with bone biopsy. NIRS utilises similar technology to a pulse oximeter, transmitting and receiving designated optical frequencies using non-invasive probes at a specific anatomical site and measuring tissue depths of up to 4cms (Figure 1). NIRS takes advantage of the differences in attenuation caused by oxyhaemoglobin (O₂Hb) and deoxyhaemoglobin (HHb). This provides haemodynamic markers such as:

- Total oxygenation index (TOI): The ratio of O₂Hb to total haemoglobin (cHb);
- Normalised total haemoglobin index (nTHI): Real time percentage change in cHb concentration from an initial baseline measurement; and,
- Real time absolute concentration changes of HHb, O₂Hb and cHb [1].

Primary Aim and Objectives:

To investigate the potential of NIRS as a diagnostic tool in the measurement of microvascular haemodynamics in bone tissue, including whether NIRS:

- 1) Can exclusively measure bone tissue, based on the known physiological differences between bone and muscle.
- 2) Can provide reproducible measurements across different anatomical sites, participants and operators.

Methods:

Testing was carried out on healthy volunteers, recruited using convenience sampling, with institutional ethical approval. Participants were positioned supine with baseline TOI measurements taken at four different superficial bony anatomical locations (proximal tibia (TP), tibial diaphysis (TD), medial malleolus (MM) and patella) and one muscle location, as shown in Figure 2.

Femoral artery occlusions were undertaken whilst simultaneously taking NIRS measurements at a bony site (TD or TP) and the muscle site before, during and after occluding blood flow at the distal thigh (Figure 3 shows set up).

Arterial occlusion integrity was confirmed by a decrease in TOI and simultaneous matched changes in HHb and O₂Hb during and after occlusion, with a nTHI change of less than +/- 15% during occlusion suggesting blood volume remained constant (Figure 4).



Figure 1: The Hamamatsu NIRO NX-200 (a) demonstrates the NIR emitting sensor probe (left) and the detector probe (right); (b) shows the typical real time display data including TOI, nTHI, O₂Hb, HHb and cHb changes.

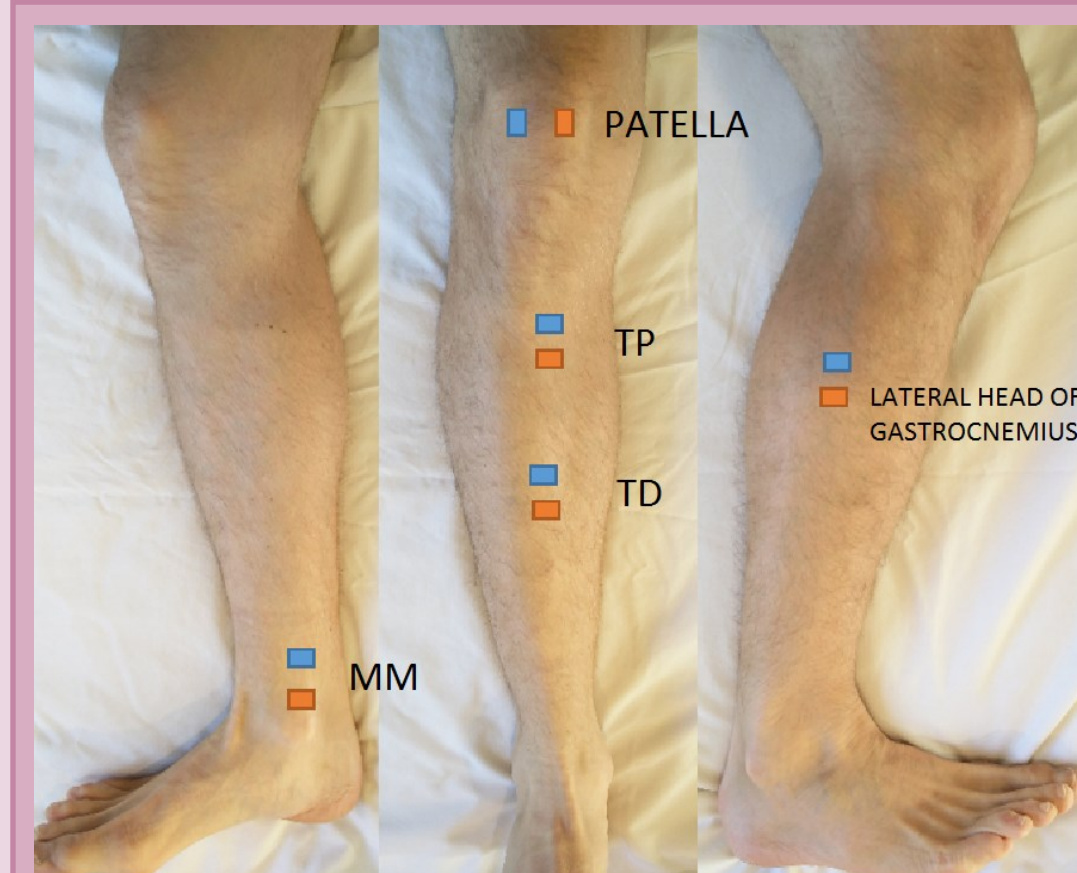


Figure 2: Schematic probe positioning at four bony sites and one muscle site.



Figure 3: Arterial Occlusion set up: A BP cuff is around the distal femur. NIRS probes are secured with bandages.

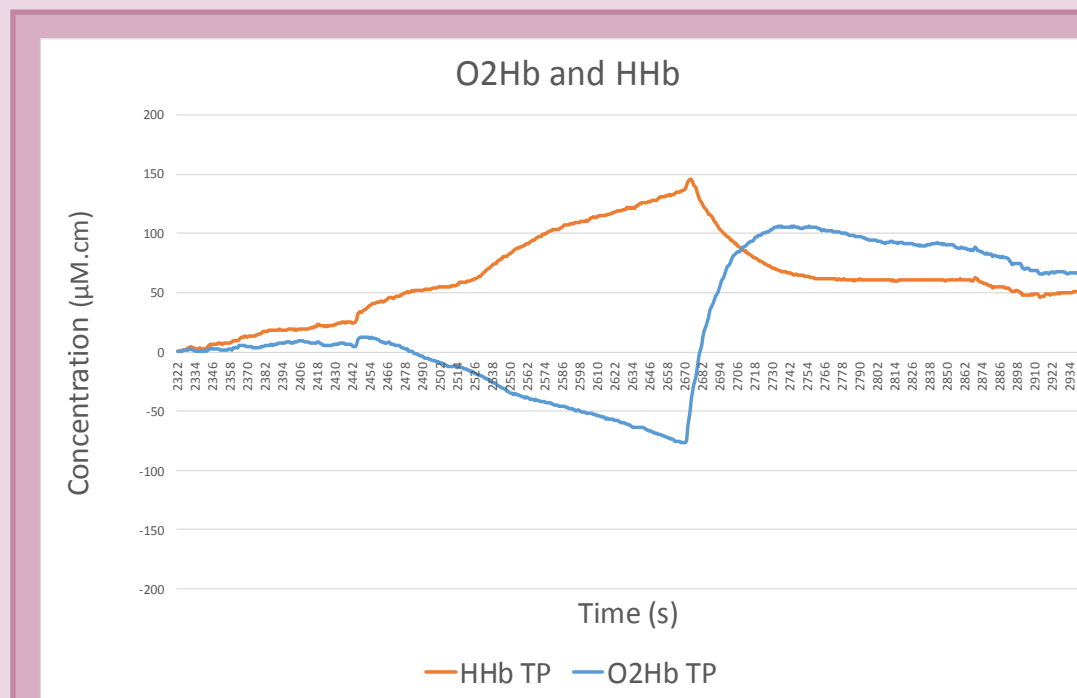


Figure 4: O₂Hb and HHb experience equal and opposite changes during occlusion and at occlusion release.

Results:

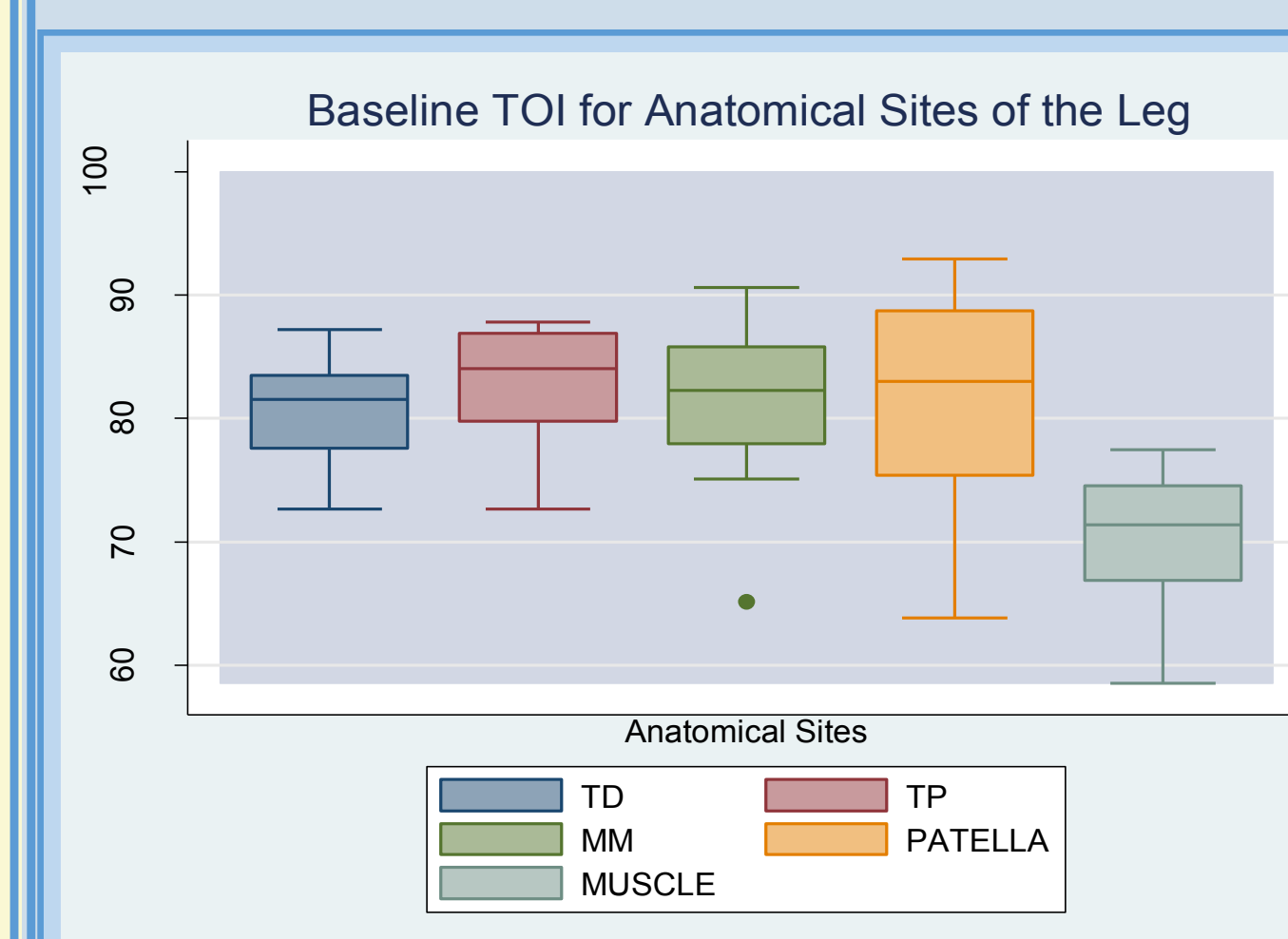


Figure 5: Box plot of mean baseline TOI values (n=15)

32 arterial occlusions were attempted on 15 participants (alternating between TD and TP) of which 27 sets of successfully paired bone and muscle data (12 TP and 15 TD) were recorded. There was a significant statistical difference demonstrated between bone sites and the simultaneous muscle tissue data taken for a number of different parameters including:

- Baseline pre occlusion TOI means;
 - Post occlusion gradients of TOI and HHb desaturation;
 - Occlusion release gradients of TOI and HHb measured during the first 20 seconds post cuff release; and,
 - Hyperaemic response upon occlusion release.
- Variability is higher with occlusion data but is again comparable between bone and muscle readings. Further investigations showed no obvious systematic confounding in results caused by age, gender or leg circumference.

Baseline TOI measurements on 15 participants demonstrated statistically significant differences between baseline TOI at the four bone sites and the muscle site. The variability of results at the preferred TD and TP bone sites was comparable to muscle readings (an established site for NIRS measurements).

Figure 6: Summary of baseline TOI results (standard deviation is in parenthesis; n=15)

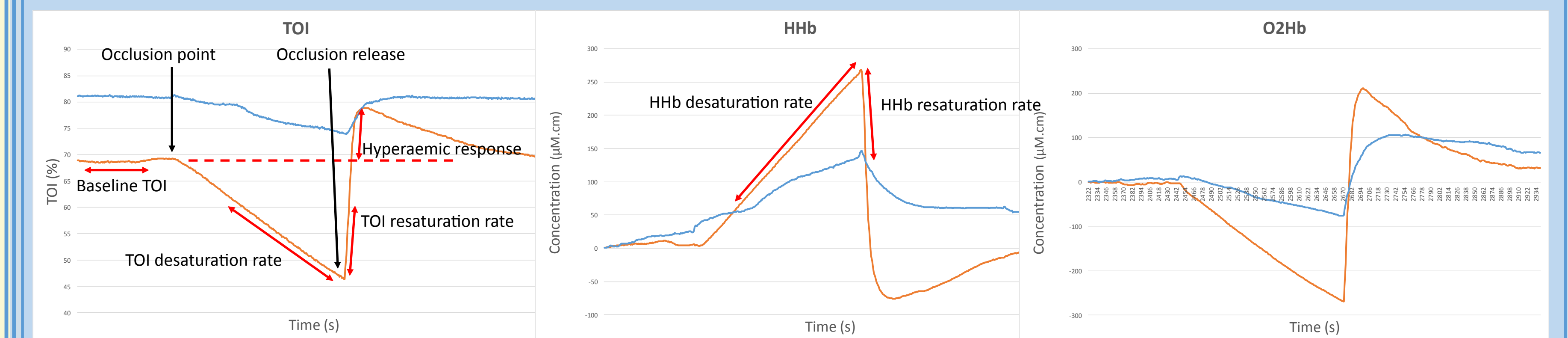
	TD	TP	MM	PATELLA	MUSCLE
Mean TOI (%)	80.55 (4.60)	83.00 (4.13)	81.18 (6.14)	82.20 (8.37)	70.05 (4.82)
Coefficient of Variation (%)	5.71	4.98	7.56	10.18	6.88
Wilcoxon signed-rank test vs muscle	p<0.001	p<0.001	p<0.01	p<0.01	n/a

DATA FROM TP ARTERIAL OCCLUSIONS	TP Bone	SD	Muscle	SD	p-value
TOI Baseline (%)	81.65	4.14	70.96	5.00	0.0029
TOI desat (%/min)	-1.62	0.54	-3.64	1.77	0.0121
HHb desat rate (µM.cm/min)	23.28	7.25	52.79	23.87	0.0047
TOI resat (%/min)	16.24	11.86	75.19	31.40	0.0022
HHb resat rate (µM.cm/min)	-146.26	91.59	-812.00	370.12	0.0022
Hyperaemic Response (% TOI)	0.69	1.44	5.80	3.20	0.0022

DATA FROM TD ARTERIAL OCCLUSIONS	TD Bone	SD	Muscle	SD	p-value
TOI Baseline (%)	80.08	5.05	70.83	4.58	0.0008
TOI desat (%/min)	-1.86	0.55	-3.94	1.96	0.0045
HHb desat rate (µM.cm/min)	28.02	6.27	54.32	24.70	0.0045
TOI resat (%/min)	16.85	7.51	67.89	27.03	0.0008
HHb resat rate (µM.cm/min)	-153.61	56.53	-711.95	301.96	0.0007
Hyperaemic Response (% TOI)	1.17	1.34	5.93	2.53	0.0007

Figure 8: Descriptive results of TD, TP and muscle physiological data. Wilcoxon signed-rank tests demonstrate statistically significant differences between bone and muscle.

Figure 7: Graphs show an individual example of observed differences in bone (blue) and muscle (orange) physiology.



Conclusions:

Despite the statistical significance of the measured differences between bone and muscle, the physiological importance of these differences remains essentially unknown due to a paucity of research in this field. However, the results of this feasibility work are encouraging and justify further research into the use of NIRS as a diagnostic tool for bone pathologies including microvascular pathogenesis, such as osteoporosis, non-union, osteoarthritis, Pagets, and/or blood borne cancers. If successful, further studies investigating the use of NIRS in more diverse populations would be justified. This could lead to inexpensive, quick and tolerable methods of measuring microvascular supply in bone.

References:

Binzoni, T. and L. Spinelli (2015). "Near-infrared photons: a non-invasive probe for studying bone blood flow regulation in humans." *Journal of physiological anthropology* 34(1): 1-6.

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