Modelling of mitochondrial oxygen consumption and NIRS detection of cytochrome oxidase redox state

Murad Banaji[1], Alfred Mallet[2], Clare E. Elwell[2], Peter Nicholls[1], Ilias Tachtsidis[2], Martin Smith[3] and Chris E. Cooper[1].

[1]Department of Biological Sciences, University of Essex, Colchester, CO4 3SQ, UK, [2]Department of Medical Physics and Bioengineering, University College London, Gower St., London WC1E 6BT, UK, [3]Department of Neuroanaesthesia and Neurocritical care, The National Hospital for Neurology and Neurosurgery, Queen Square, London WC1N 3BG, UK

Abstract In recent years there has been widespread use of near infrared spectroscopy (NIRS) to monitor the brain. The signals of interest include changes in the levels of oxygenated and deoxygenated haemoglobin and tissue oxygen saturation. In addition to oxy- and deoxy-haemoglobin, the Cu_A centre in cytochrome-c-oxidase (CCO) is a significant NIR absorber, giving rise to another signal termed the ΔoxCCO signal. This signal has great potential as a marker of cellular oxygen metabolism, but is also the hardest to interpret. Here we use a recently constructed model to predict NIRS signal changes, and compare the model output to data from an *in vivo* hypoxia study in healthy adults. Our findings indicate strongly that the ΔoxCCO signal contains useful information despite the noise, and has responses consistent with the known physiology.

1 Introduction: NIRS and brain monitoring

Near infrared spectroscopy has been used extensively to monitor brain oxygenation, haemodynamics and metabolism [1,2]. The chromophores of interest include oxyhaemoglobin (HbO2) and deoxy-haemoglobin (HHb), with changes ΔHbO2 and ΔHHb being measured using differential spectroscopy systems [3,4,5]. Also measured are absolute tissue oxygen saturation (TOS), the mean level of haemoglobin oxygen saturation across all vascular compartments in the tissue of interest, and changes in the level of oxidation of the Cu_A centre in cytochrome-c-oxidase, a signal termed the ΔoxCCO signal.

The usefulness of the ΔoxCCO signal has been the subject of much debate for two reasons: Firstly it is hard to interpret as a number of different causes may give rise to the same signal changes; Secondly the high signal-to-noise ratio raises the question of whether it contains sufficient information to be of clinical interest. However, as we will show, combining measurement with modelling allows useful information to be extracted from the signal, giving it the potential to make a significant contribution in a clinical setting.

2 The model

A model capable of predicting behaviour of all the NIRS signals was constructed . The mathematical description is presented in [6], and the model itself is available for download at [7]. The model was designed to respond to four input stimuli, chosen both for their physiological importance, and because there is considerable data on the response of NIRS signals to these inputs:
1) Changes in mean arterial blood pressure (ABP) [8];
2) Changes in arterial O_2 levels (SaO2) [9];
3) Changes in arterial CO_2 levels (PaCO2) [10] and
4) Functional activation [5].
The most important model outputs are the NIRS signals TOS and ΔoxCCO, along with cerebral blood flow (CBF) and oxygen consumption rate (CMRO2). A schematic of the structure of the model is presented in Fig. 1.

The model consists essentially of two components: A submodel of the cerebral circulation, which is known to respond in complicated ways to a variety of stimuli, and a submodel of mitochondrial metabolism related to the models presented in [11] and [12]. The two components of the model are linked via oxygen transport and consumption. In addition to the full model designed to represent an *in vivo* situation, a simplified model representing an *in vitro* situation was also constructed, in order to allow model parameter setting from *in vitro* data. Model parameters were chosen to be consistent with thermodynamic principles.

Parameter setting and model validation are described extensively in [6]. Examples of model behaviour are presented in Fig. 2 and Fig. 3 below. In Fig. 2 steady state behaviour of cerebral blood flow (CBF) in response to changes in arterial blood pressure and PaCO2 is shown to be consistent with published *in vivo* experimental data. In Fig. 3, experimentally measured cytochrome *c* redox state and modelled Cu_A redox state (which for theoretical reasons can be assumed to be similar) are compared. The data suggest that the redox states are sensitive to arterial O_2 levels at physiological values.

3 Comparison with data from a hypoxia study

A study of nine healthy volunteers undergoing a hypoxic challenge (SaO2 slowly reduced to about 80 percent three times) was carried out [17]. During this process both TOS (TOI, Hamamatsu NIRO300) and ΔoxCCO (multiwavelength CCD spectrometer) were measured, along with systemic data on SaO2, PaCO2 and ABP. The systemic data were used as input into the model, in order to compare modelled and measured signals. An example of such a comparison for one subject is presented in Fig. 4.

In Fig. 5, modelled and measured ΔoxCCO are compared for a further eight subjects from the same study. No attempt at model fitting has been carried out – in each case the model was run at default parameter values. It is clear that in some cases model behaviour is both qualitatively and quantitatively consistent with the data. Where this fails to be the case, there appears to be some systematic drift in the measured data, with the magnitude of this drift suggesting experimental artefact. It is also interesting that where the measured data appears to lack information (e.g. panel 4 of Fig. 5) the model suggests that this may have been the physiological response, *not* measurement artefact. At this stage, the comparison is promising.

4 Estimating ΔoxCCO using the BRAINCIRC model

Finally, we wished to find out if our recent model provided any improvement over a previous model, BRAINCIRC, described in [18]. Although the BRAINCIRC model cannot directly predict Cu_A redox state, ΔoxCCO can be estimated using indirect calculation from NAD/NADH levels and phosphorylation potential, based on the assumption that the electron transport chain is near thermodynamic equilibrium [19], and that Cu_A redox changes are identical to those of cytochrome *c*.

In Fig. 6 a comparison between predictions of version 2 of the BRAINCIRC model (available at [7]) and experimental data is presented for the first two subjects whose data are shown in Fig. 5. Although the estimates from the BRAINCIRC model were qualitatively reasonable, the comparison suggests that direct modelling of Cu_A using the new model yields more accurate results. It should be mentioned however that no attempt was made to optimise the BRAINCIRC model or adapt it to the current situation.

5 Conclusions

Despite the low signal-to-noise ratio, the ΔoxCCO signal contains physiological information of potential clinical importance. The model we have constructed is able to reproduce the behaviour of this signal both qualitatively and sometimes

quantitatively during hypoxia. It appears to give more accurate results than estimates using previous modelling work Once the model has been more extensively validated against further *in vivo* data, it has the potential to be used to identify artefactual trends in the measured ΔoxCCO traces, and importantly for clinical studies, to make predictions about blood flow and CMRO2.

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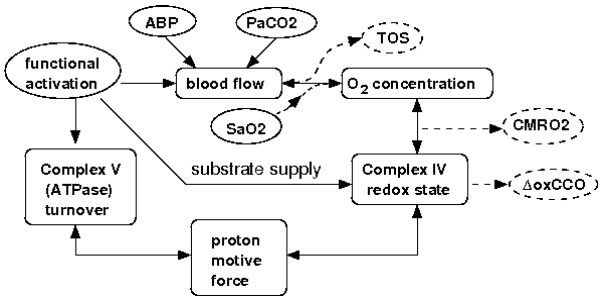

Fig.1. Summary of the main inputs, variables and processes in the model. Model inputs are enclosed in solid ovals, while outputs are enclosed in dashed ovals.

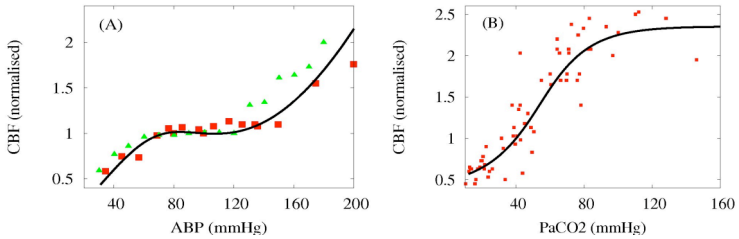

Fig. 2. (A) The modelled response (solid line) of CBF to changes in blood pressure, with data from [13] (squares) and [14] (triangles). (B) The modelled response (solid line) of CBF to changes in PaCO2 with data from [15] (normal resting CBF set at 40 ml/min/100g).

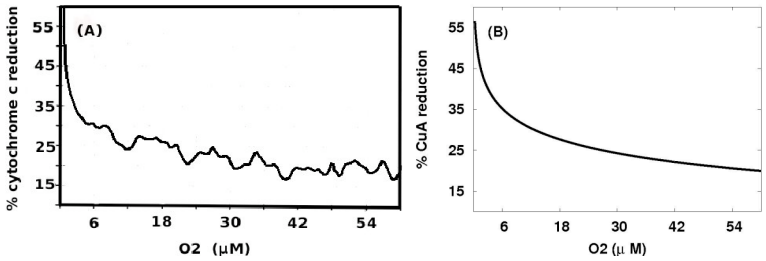

Fig. 3. (A) The variation in measured cytochrome *c* redox state during hypoxia, redrawn from Figure 5A of [16]. (B) Modelled Cu_A redox state during hypoxia for simplified *in vitro* model.

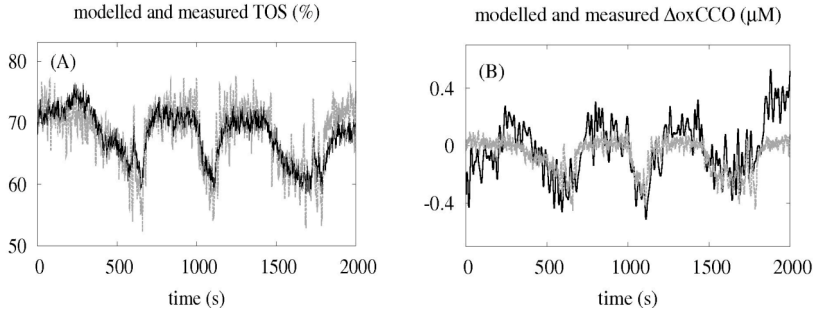

Fig. 4. The modelled (light line) and measured (dark line) responses of (A) TOS and (B) ΔoxCCO for a subject undergoing a series of three hypoxic swings. Further details are in [17].

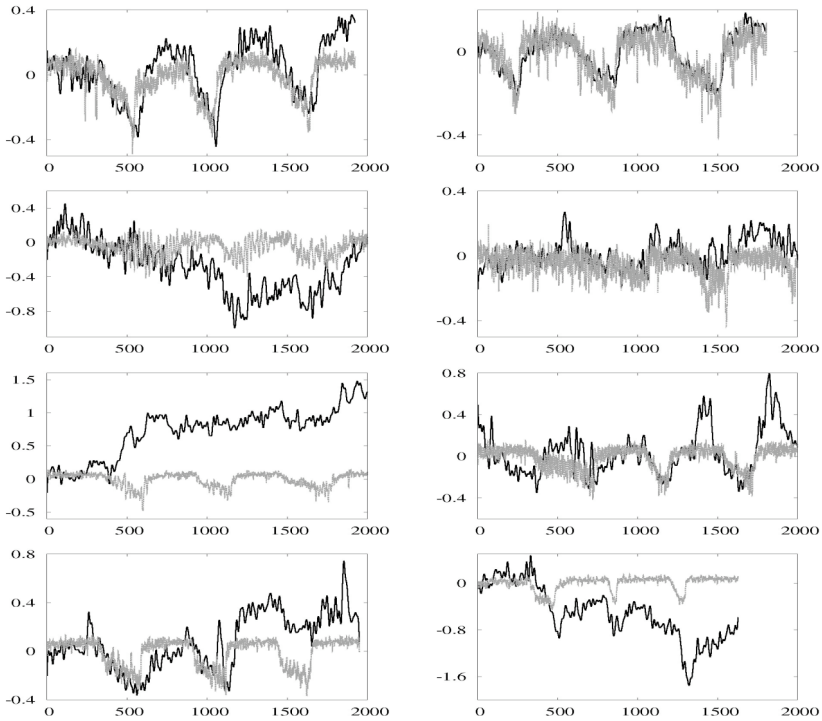

Fig. 5. The modelled (light line) and measured (dark line) responses of ΔoxCCO for eight further subjects undergoing a series of hypoxic swings.

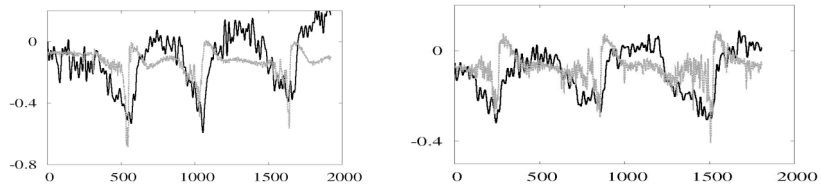

Fig. 6. The modelled (light line) and measured (dark line) responses of ΔoxCCO for the first two subjects (top two panels) presented in Fig. 5. This time the model used is the BRAINCIRC model, and ΔoxCCO was estimated using indirect calculations (see text).