Video Article

Exploring Cognitive Functions in Babies, Children & Adults with Near Infrared Spectroscopy

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Abstract

An explosion of functional Near Infrared Spectroscopy (fNIRS) studies investigating cortical activation in relation to higher cognitive processes, such as language[1,2,3,4,5,6,7,8,9,10], memory[11], and attention[12] is underway worldwide involving adults, children and infants [3,4,13,14,15,16,17,18,19] with typical and atypical cognition[20,21,22]. The contemporary challenge of using fNIRS for cognitive neuroscience is to achieve systematic analyses of data such that they are universally interpretable[23,24,25,26], and thus may advance important scientific questions about the functional organization and neural systems underlying human higher cognition.

Existing neuroimaging technologies have either less robust temporal or spatial resolution. Event Related Potentials and Magneto Encephalography (ERP and MEG) have excellent temporal resolution, whereas Positron Emission Tomography and functional Magnetic Resonance Imaging (PET and fMRI) have better spatial resolution. Using non-ionizing wavelengths of light in the near-infrared range (700-1000 nm), where oxy-hemoglobin is preferentially absorbed by 680 nm and deoxy-hemoglobin is preferentially absorbed by 830 nm (e.g., indeed, the very wavelengths hardwired into the fNIRS Hitachi ETG-400 system illustrated here), fNIRS is well suited for studies of higher cognition because it has both good temporal resolution (~5s) without the use of radiation and good spatial resolution (~4 cm depth), and does not require participants to be in an enclosed structure[27,28]. Participants cortical activity can be assessed while comfortably seated in an ordinary chair (adults, children) or even seated in mom s lap (infants). Notably, NIRS is uniquely portable (the size of a desktop computer), virtually silent, and can tolerate a participants subtle movement. This is particularly outstanding for the neural study of human language, which necessarily has as one of its key components the movement of the mouth in speech production or the hands in sign language.

The way in which the hemodynamic response is localized is by an array of laser emitters and detectors. Emitters emit a known intensity of non-ionizing light while detectors detect the amount reflected back from the cortical surface. The closer together the optodes, the greater the spatial resolution, whereas the further apart the optodes, the greater depth of penetration. For the fNIRS Hitachi ETG-4000 system optimal penetration / resolution the optode array is set to 2cm.

Our goal is to demonstrate our method of acquiring and analyzing fNIRS data to help standardize the field and enable different fNIRS labs worldwide to have a common background.

Video Link

The video component of this article can be found at https://www.jove.com/video/1268/

Protocol

Part 1: Prior to participant arriving to the lab

1. Ensure that the room is free of extraneous articles that may be distracting to the participant.
2. Set-up and load experimental protocol on the fNIRS Hitachi ETG-4000 system.
3. Set-up your experimental paradigm. Experimental paradigms can be programmed with different presentation software, including Eprime, Presentation, Psyscope or a Matlab based psychology toolbox. Here we use Matlab based psychology toolbox.
4. Timing is key for data analysis, thus the experimental paradigm must be perfectly timed with data collection. The fNIRS Hitachi ETG-4000 has triggering capabilities, allowing for the experimental paradigm to trigger the data collection or vice versa. Test triggering of presentation program from fNIRS Hitachi ETG-4000. Triggering can be done using parallel, serials, or USB ports. Here we show triggering via the parallel port.
5. Prior to starting the fNIRS study it is important to conduct participant background screening. In the Petitto lab, we conduct background screening by having the participants or their parents fill out study-appropriate standardized questionnaires[29].

Participant Arrives

6. It is important to conduct the session and to treat the participants in a professional manner. The participant or the participants' parents/legal guardians must sign a consent form before the experiment begins. It is vital to thank the participant for their time in these important and exciting experiments.
7. The participant is seated comfortably close to the fNIRS testing room. An infant participant may be seated on a parent's lap.

Part 2: Placing Optodes & Using the 10-20 system

Another component of the analysis method that enables consistent data interpretation is the standardization of fNIRS recording protocol. This entails optode placement, participant positioning, and triggering of stimulus presentation software. Both the accurate neuro-anatomical placement of probes and the confirmation of regions of interest (ROIs) are achieved by using the 10–20 system[3,4,30]. Further, stereotactic localization of the probe array was confirmed on the participant's skull by overlaying 3D tracking information from a Polhemus Fast trak system onto an anatomical MRI co-registration scan of the participant conducted with vitamin E capsules placed at each probe location[3,4].Optimal participant positioning involved placing participants comfortably in a reclining chair, with fiber optics hanging loosely without contact with the body or chair.

1. The following head measurements are taken with a tape measure and written down on the participants data sheet:
   - Nasion to Inion around
   - Nasion to Inions over top
   - Ear to Ear over top
2. Surgical tape can be used to mark specific target locations. In this experiment we will mark Fp, T3/T4, F8/F7
3. Optode arrays are placed on the participants' head with specific optodes anchored at 10-20 points as directed by the purposes of the experiment.

Part 4: Testing the Optode Array

1. Introduction to the Hitachi ETG-4000 GUI Interface and probe testing.
2. Testing signal: Once optodes are placed on the participants' scalp, the signal quality is tested. If an optode does not have a clear signal, researchers gently remove hair from the connection of the optode and the scalp. On occasion the optodes may need to be wiped with an alcohol swab.

Part 5: Running the experiment.

1. At least two experimenters must be always present in the room; one observing the fNIRS Hitachi ETG-4000 real-time read out and the other observing the participant. Having a video camera focused on participant is highly recommended for post-hoc observations. An advantage of the fNIRS Hitachi ETG-4000 is that the video and fNIRS signal are synched and co-registered. A log containing all relevant information and files generated is kept.
2. There are well-established methods of building experimental hemodynamic paradigms, namely Block design and Event-related designs. For a more complete description please see the recent review paper[31].

Part 6: Analysis

Once all of the data have been collected, the participant is thanked for their time and willingness to participate and leaves the lab. As analysis is not done on the fNIRS Hitachi ETG-4000, as, instead, the data are exported to an analysis computer.

1. Conversion from μV to hemoglobin concentrations. As raw attenuation values are collected in attenuation of laser strength (as measured in μV), these values must be converted to oxygenated and deoxygenated hemoglobin values. This is done using the modified Beer-Lambert equation.
2. The application of the modified Beer-Lambert is conducted in two steps. Under the assumption that scattering is constant over the path length, first the attenuation for each wavelength ($\Delta A_\lambda(t)$) is calculated by comparing the optical density of light intensity during the task (I_{task}) to the calculated baseline of the signal ($I_{baseline}$). The ΔA values for each wavelength and sampled time point (*t*) to solve the modified Beer-Lambert equation.

Equation 1	Equation 2
$\Delta A_{\lambda(t)} = \log_{10}\left(\frac{I_{task}}{I_{baseline}}\right)$	$\left(\frac{\Delta A_{\lambda_1}(t)}{\Delta A_{\lambda_2}(t)}\right) = \begin{bmatrix} \varepsilon_{deoxy}^{\lambda_1} & \varepsilon_{oxy}^{\lambda_1} \\ \varepsilon_{deoxy}^{\lambda_2} & \varepsilon_{oxy}^{\lambda_2} \end{bmatrix} \begin{pmatrix} C_{deoxy(t)} \\ C_{oxy(t)} \end{pmatrix}$

$\lambda_1\#_{deoxy}$, $\lambda_1\#_{oxy}$, $\lambda_2\#_{deoxy}$ and $\lambda_2\#_{oxy}$ are the constants for the extinction coefficients that measure the fraction of light lost to absorption per unit concentration distance in the tissue. The resultant C_{deoxy} and C_{oxy} values are the concentrations of deoxygenated and oxygenated hemoglobin for each *t*.

Part 7: Representative Results

Typical hemodynamic response results in several distinct characteristics. In the oxy-hemoglobin response, there is first a characteristic dip. This dip occurs as a region of neurons activates and depletes available oxygen. As blood flow increases, carrying oxygenated hemoglobin, the oxy-hemoglobin response rises rapidly above the initial baseline levels to a steady state level. When the region is no longer being activated, the oxy-hemoglobin response decays over the next 12-15 seconds and slowly drops back to baseline levels. There is occasionally an undershoot that occurs prior to the hemodynamic response returning to initial baseline levels.

Bad results are usually in the form of optodes not properly seated on the scalp or excessive movement. These types of noise – called, 'Flatling' – are evident in the signal as the microvolt values saturate and a number of different channels, both oxy- and deoxy- response move in a coordinated fashion.

DEMONSTRATION: Shaking the fiber optics.

Statistical Analyses: The extracted oxy and deoxy-hemoglobin values for each channel, for each participant and for each task can then be submitted to conventional statistical analysis, including t-tests, ANOVAs, correlations etc.

Discussion

In this study, we demonstrated the use of a novel, non-invasive fNIRS brain imaging technology to investigate human brain function in relation to human cognition and perception. fNIRS brain imaging may represent the future of non-invasive brain imaging, particularly with infant and child populations, that may one day be widely available in research labs, physicians' offices, and in the school systems allowing clinicians to apply basic scientific findings about the brain to their clinical practice.

Disclosures

The authors have nothing to disclose.

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