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TOOLS OF THE TRADE

What is Optical Imaging?

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This article introduces a promising new methodology called optical imaging. Optical imaging is used for measuring changes in cortical blood flow due to functional activation. The article outlines the pros and cons of using optical imaging for studying the brain correlates of perceptual, cognitive, and language development in infants and young children.

INTRODUCTION

Imaging methods have caused a revolution in cognitive science for research on adult brain function (Cabeza & Kingstone, 2006; Posner, 2003; Raichel & Mintun, 2006). It is clear that many of these neuroscience methods can be applied to younger populations to investigate the relationship between cognition and brain development (Casey, Davidson, & Rosen, 2002; Csibra, Kushnerenko, & Grossman, 2008, Kuhl, 2004; Nelson, de Haan, & Thomas, 2006; Neville, 2005). The goal of this paper is to introduce a new methodology called optical imaging and to describe the feasibility of using optical imaging on infants and young children.

The motivation for using neuroimaging techniques on young populations is that they allow us to address critical issues of continuity and change over development. When a young infant, an older child, and an adult all exhibit a behavioral discrimination, how can we tell whether the underlying

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mechanisms producing the discrimination are the same (a case of developmental continuity) or different (a case of qualitative developmental change)? Imaging studies of infants, combined with and guided by the findings of imaging studies with adults and by behavioral studies, should let us approach this question. If we see convergent findings in infants and adults at both the behavioral level and the neural level, we can infer continuity across development. Conversely, non-convergence between infants and adults may signal points of significant developmental change, as well as provide a window for examining such change.

A preview of the take-home message is that the future for the optical imaging technique looks bright, because currently developmental research is skewed. There is a wealth of behavioral data on the nature of infants' representation abilities, but there are relatively few studies of developmental cognitive neuroscience (Aslin, 2007). I believe that optical imaging has the potential to do for infancy research what functional magnetic resonance imaging (fMRI) has done for cognitive neuroscience. There is a synergistic effect in addressing cognitive abilities in infants, both at the behavioral and neural levels. My goal in using optical imaging technology is to develop a corroborative measure to lend insight to the behavioral findings of infants and young children.

HOW DOES OPTICAL IMAGING WORK?

The optical imaging method that we use is called near-infrared spectroscopy (NIRS), and it provides a measure of changes in blood flow due to functional activation. NIRS is the same technology as pulse oximetry, commonly used in hospitals to measure a person's pulse. NIRS is harmless and approved by the Food and Drug Administration for continuous use. Unlike pulse oximetry, we use NIRS to measure changes in cerebral blood oxygenation (i.e., the blood oxygen level dependence, or BOLD response).

In a typical experiment, an array of fiber optic probes is placed on the infant's head. Half of the probes emit light, the other half of the probes serve as detectors that absorb reflected light. The emission probes emit light at two specific frequencies: One frequency is optimally absorbed by oxygenated blood while the other is optimally absorbed by deoxygenated blood. The dependent measure is the amount of light at each wavelength that is reflected from the emission probe to detection probes. Increases in oxygenated and deoxygenated hemoglobin in functionally activated cortex will produce an increase in the amount of light absorbed and therefore a decrease in the amount of light reflected back to the detection probe. By gathering data on the changes in blood flow over time, we can make inferences about the

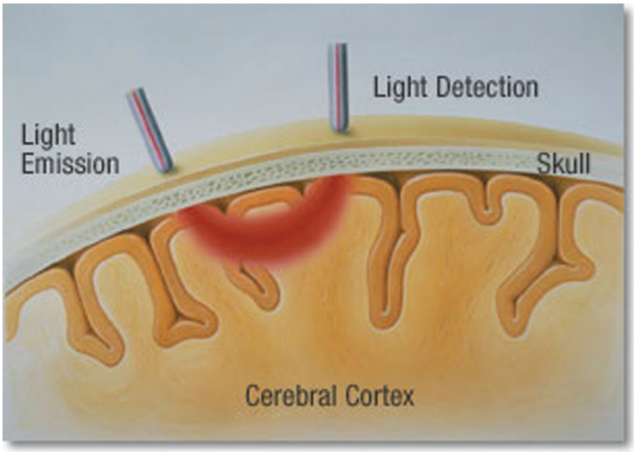

FIGURE 1 This figure demonstrates the principle of NIRS technology. Light enters the skull from the emission probe placed on the surface of the head. Most of the light emitted from the source probe and reflected to the detector probe has passed through the path depicted by the red banana-shaped curve. Inferences about the cortical activity occurring in the region of the banana-shaped curve are made by measuring the changes in light absorption when the area is at rest versus when it is active. In a typical experiment, an array of probes creating multiple source-detector pairings is placed over cortical regions of interest, allowing multiple areas/channels of cortical activity to be measured simultaneously.

cortical activity occurring in the underlying brain regions (see Figure 1).[1] Infants tend to have fine hair, and their skulls are thinner than adults. Therefore, the proportion of signal loss due to scattering is less than adult participants. In this review, I focus on a specific type of NIRS called the continuous wave system, because it is currently the most successful method for use with infants and children (see Minagawa-Kawai, Mori, Hebden, & Dupoux, 2008, for a more extensive review, particularly the physics principles behind NIRS).

One of the best ways to place a new technique is to compare it to the existing techniques. The vast majority of neuroscience research on infants has been done with electroencephalograph (EEG; Gliga & Dehaene-Lambertz, 2007; Kuhl, 2004; Nelson & Monk, 2001; Neville, 2005). EEG measures electrical activity along the scalp produced by the firing of neurons within the brain. In contrast, NIRS measures a hemodynamic response related to neural activity

[1]It should be noted that this is a characterization to demonstrate the principles of NIRS. There are a variety of systems that have a single source detected by many probes (e.g., Wilcox, Bortfeld, Woods, Wruck, & Boas, 2005), and the placement and separation between sources and detectors vary across experiments and labs.

in the brain. NIRS is more closely aligned with fMRI, because both techniques measure the same phenomena of increased blood flow due to functional brain activity. NIRS detects the blood flow changes by measuring the light absorption for the oxygenated and deoxygenated blood in a specific brain area. In contrast, fMRI detects the blood flow changes by measuring the magnetic signal for the deoxygenated blood in the specific brain area. Because infants lack the ability to remain still while awake, fMRI is not feasible for behavioral measures like looking time. EEG has excellent temporal resolution and can measure changes on the order of fractions of milliseconds, but the spatial resolution of EEG is limited. NIRS systems can have the temporal resolution of a few Hz, although it is effectively 0.3–0.5 Hz because it measures changes in blood flow due to neural activity (Minagawa-Kawai et al., 2008). The spatial resolution of NIRS is approximately 2 cm, so it is better than EEG on localization (see Gratton & Fabiani, 2001, for a more detailed comparison of EEG and NIRS on adults). With respect to fMRI, NIRS has much better temporal resolution but worse spatial resolution (on the order of millimeters for fMRI instead of centimeters for NIRS; for within-subject comparisons on adults, see Strangman, Culver, Thompson, & Boas, 2002).

WHAT ARE THE PROS AND CONS OF USING OPTICAL IMAGING?

Investing in a new technology takes time. Happily, optical imaging has progressed to a point where the technology and data analysis are at a favorable balance in terms of cost versus benefit. In this section, I review the pros and cons of three major issues: quality of data, ease of use, and operation costs.

Quality of the Data

One of the biggest advantages that NIRS has over other neuroimaging techniques is freedom of movement. Unlike EEG and fMRI, NIRS has relatively little data compromise due to motion artifact or resting position (prone, sitting, or standing). Because infants can move around during the course of an experiment without compromising the data, they are more likely to finish a protocol thereby lending itself to better data. For infants, we created a hat that holds the fiber optic probes.[2] The hat has a chin strap, and after the hat is slipped on the infant's head, an elastic bandage is wrapped over the hat. This hat and bandage combination allows the probes to stay in place while the infant moves with some freedom (see Figure 2).

[2]These hats can be custom designed/purchased from Alex Sargent by e-mail: alex.sargent@vanderbilt.edu

FIGURE 2 Photos of infants who were in our experiments. The photos on the top show the cap with chin strap and elastic bandage combination that provides a snug fit between the probes and the infant's head. The pictures on the bottom represent the placement of the probes over the auditory and visual cortices (on the left and right respectively). The red circles indicate the approximate placement of the source probes and the blue circles represent the detector probes. The black squares indicate where the NIRS system would detect blood flow between each source-detector pairing. This particular probe arrangement consists of two sets of 3×3 optode arrays comprising 24 channels that each detect changes in oxy- and deoxyhemoglobin.

The fiber optic cables are gathered in a tail that can trail down the infant's back or over the caretaker's shoulder. In addition, we have used the infant probes to measure from prefrontal cortex in 3-year-old children. We have no motion artifact while the children are participating in behavioral manipulations that require talking, writing, and eating. We have found that the elastic bandage holds the probes in place with a snug fit. When the experiment is complete and we remove the hat, the gentle compression of the elastic band causes an impression of where the probes were on the infant's head that disappears after a couple minutes. We typically use this impression to verify that the probes were in the correct location. For testing adults, we have a separate set of fiber optic cables. The adult probes are specially designed with a spring-loaded, 90° bend in the end of the cable so that the probes can part through hair and rest on the scalp.

Other labs have reported issues with motion artifact (see Aslin & Mehler, 2005). In addition to the well-designed probe-holding methods described above, there are automatic algorithms to reject signals that fall outside the range of hemodynamic response and statistical techniques to help deal with these issues.

An advantage that NIRS has over many behavioral techniques is that imaging tasks allow us to test young infants in a state of full attention with low task demands. These same tasks have been used with great success in fMRI with adults using free viewing, providing opportunities of comparisons across tasks. In contrast, infant behavioral studies, like looking time, could be described as measuring the variability in how quickly infants lose interest in a display. Another advantage that NIRS has over looking-time studies is that the data sets from imaging are rich. For example, our NIRS machine samples from 24 different locations at two different wavelengths, 10 times per second. These detailed data are held in contrast to typical looking-time studies that are limited to dichotomous data gleaned from looking times at three novel and three familiar test displays.

One of the current disadvantages to the quality of the data is that NIRS is limited to measuring only surface cortical activity. In theory, there are ways to use temporal and frequency modulation techniques to get deeper into the cortex, but these machines are not commercially available yet (for more details, see Aslin & Mehler, 2005; Hebden, 2003; Minagawa-Kawai et al., 2008). In addition, the current spatial resolution of optical imaging on infants is typically limited to 2 cm or 3 cm. While centimeters are a large unit of measure, there are ways to use the experimental design, like subtraction analyses to get around this limitation (for example, see Gervain, Macagno, Cogoi, Pena, & Mehler, 2008).

Ease of Use

The beauty of the optical imaging technique is that the technology is quite easy to use. For example, in my lab, undergraduate research assistants have been trained to proficiency in testing infants using ongoing optical imaging procedures within one quarter. Unlike fMRI, the NIRS machine is silent, so presentation of auditory stimuli is easy. There is a serial port connection between the display computer and the imaging machine, so the data presentation information is time locked to the raw data. In addition, the machine is somewhat portable and does not require special housing. The dimensions of our machine, a Hitachi ETG 4000, are approximately 1.5 m high, 1 m deep and 0.5 m wide. The machine has wheels so it can be moved between rooms quite easily.

The difficulties in terms of ease of use come when it is time to analyze the data. Given that the development of the technique is in its infancy, analysis techniques vary. At this point in time, there is no agreed-upon method for data analysis. In particular, there is a great deal of variation in terms of how group analyses are done with regard to co-registration. In my lab, we export the raw data from the Hitachi machine, filter in Matlab, and process the data using Afni. These techniques are available on our lab Web site (see "Additional Information" at the end of the article). We have an older method that uses Matlab and Brain Voyager (Hespos, Ferry, Cannistraci, & Gore, 2009) and have found converging evidence between the two methods (Hespos, Park, et al., 2010; Hespos, Reber, Cannestraci, & Gore, 2010). These findings demonstrate consistency across machines and analysis methods that establish optical imaging as a reliable method for measuring the hemodynamic response. Young populations have smaller capillaries, faster heart rates, faster respiration, greater synaptic density, and immature myelination (Casey et al., 2002). Given all these differences, initial reports suggest that the BOLD responses look similar to adults' responses in terms of time course and peak amplitude. For this reason, it is an exciting time to be doing optical imaging, because the territory is relatively uncharted.

Operation Costs

The advantage of NIRS over imaging methods like fMRI is that the cost of operation is considerably cheaper. Once the hurdle of the initial instrument cost is surmounted, there are no day-to-day operation costs to maintain the machine. We train students to run the experiments, so there is no need for a technician to operate the machine. We pay subjects $30 for participating in our experiments (this should be held in comparison to the $20 that we pay them for participating in behavioral experiments). There are some parts of the instrument that wear out with extended use. For example, the fiber optic cables will break over time if they are not handled with care. The signal decline will be manifest as an increase of signal to noise. This decline can be verified with a light meter and should be checked at regular intervals.

FOR WHAT AGES AND POPULATIONS WILL THIS TOOL BE BEST SUITED?

The most likely niche for NIRS as a research tool is for testing infants, children, and certain clinical populations. Infants have better signal-to-noise ratio compared with adults due to less hair and thinner skulls. In addition, infants' heads are smaller, so 3-cm depth in an infant head reaches more cortical area than 3 cm in an adult head. My lab has been successful in testing infants ranging from newborns to 10 months of age. We have tested a variety of stimuli and measured from visual, motor, and auditory/language areas (Hespos, Park, et al., 2010; Hespos, Reber, et al., 2010). We have found that infants have adult-like BOLD response patterns as early as the first weeks of life. In addition, we have found that NIRS can capture developmental changes in language areas when infants are presented with their native language, a foreign language, and a scrambled version of both languages. As infants near their 1st birthday, they tend to grab and pull at the hat and fiber optic cables. Since these actions can cause permanent damage to the fiber optic cables, we focus testing on infants less than 1 year old. By the time children are 3 years old, they have the communication skills that allow them to wear the hat without grabbing the probes or cables. We have been successful at engaging preschoolers in a battery of behavioral tasks that measure emotion regulation, while the NIRS instrument is measuring blood flow to areas in the frontal cortex. Finally, NIRS has the potential to become a useful clinical tool. NIRS can be used on adult populations who are not comfortable lying down in the confined space inside the bore of a magnet as long as the cortical areas of interest lie near the cortical surface. For example, Folley and Park (2005) have used NIRS to measure activity in the frontal cortex for populations who suffer from schizophrenia.

CURRENT CHALLENGES INVOLVED IN USING OPTICAL IMAGING: MACHINERY AND INTERDISCIPLINARY NATURE

One of the exciting aspects of using NIRS on infants is that the technology improves dramatically and often. There are two broad categories of machines that have been used in NIRS research on infants. Some machines have 1 to 4 emission/detection probe pairings (called channels) and other machines have 24 channels or more. The advantage of fewer channels is that the machines generate less data; therefore, data set size and analyses are more manageable. The disadvantage is that the placement of the probes is critical, and displacement of less than 1 cm can result is a substantial loss of the NIRS signal (Kleinschmidt et al., 1996). One solution to this problem is to increase the number of probes so that a larger area is sampled. With more probes, it is possible to use marker tasks in the procedure to identify specific regions of interest with functional activation (Watanabe, Homae, Nakano, & Taga, 2008). Two inherent problems incurred by having more channels are: 1) the generation of large data sets requires an interdisciplinary team of computer scientists, engineers, and physicists to deal with the data in an appropriate manner, and 2) getting multiple probes to rest well on the curved surface of an infant's head is a behavioral and engineering challenge.

PREVIOUS STUDIES ON INFANTS USING NIRS

The research on infants using a small number of probes has demonstrated that NIRS is a promising and feasible method for noninvasive tracking of functional activation in diverse cortical regions (for a review, see Meek, 2002). Studies have shown that neonates have measurable responses to flickering checkerboard (Meek et al., 1998), pictures of faces (Csibra, 2004), and various smells (Bartocci et al., 2000). Studies on both young and older infants have revealed the potential of using this method for a broad range of ages. Baird et al. (2002) performed a longitudinal study on infants from 5 to 12 months of age comparing the hemodynamic response in the frontal cortex before and after infants succeeded on the Piagetian object permanence/manual search tasks. They found enhanced activity in prefrontal areas on visits when infants succeeded in the task, providing converging evidence for the behavioral tasks that each infant performed simultaneously. Wilcox et al. (2005) found increased activation in temporal and visual areas in 6-month-old infants when watching visual displays involving objects compared with baseline. Bortfeld, Wruck, and Boas (2007) extended these findings to infants who were 6 to 9 months of age and showed that during visual presentation alone versus language and visual presentation, there was constant activation in visual areas, but the activation in temporal areas increased during the conditions with language. Wilcox et al. (2009) showed that activation in temporal areas was modulated by multi-feature and shape changes but not by color changes in 6-month-old infants. In addition, Franceschini et al. (2007) measured regionally specific increases in blood volume and oxygen consumption during resting states in healthy infants over the course of the 1st year of life, demonstrating that as early as 6 weeks of age, infants show predictable patterns of cortical blood flow. These findings were done with machines that had a single source probe and multiple detectors.

There is a growing set of findings from machines that use 24 channels (i.e., 24 source-detector pairings in an array over different cortical areas). One study with 3-month-old infants measured from frontal and occipital areas while infants viewed various visual displays (Taga, Asakawa, Hirasawa, & Konishi, 2003; Watanabe et al., 2008). Taga et al. found functional differentiation between frontal, early sensory, and higher sensory/association regions as early as 3 months of age. Peña et al. (2003) tested sleeping neonates and measured auditory cortical activation bilaterally while the infants were presented with infant-directed speech, infant-directed speech played backwards, and silence. They found left hemisphere superiority during forward speech but bilateral or no activation during the backward speech and silence. These data demonstrate that even neonates, with their limited language experience, have a specific brain response to normal speech. Gervain et al. (2008) extended these findings to show that sleeping neonates can detect simple structures in speech, provided they have immediate repetitions. Infants could discriminate between an artificial language that had an ABB pattern from one that had an ABC pattern. Nakano, Watanabe, Homae, and Taga (2008) found the neural correlates to habituation using auditory stimuli. They presented 3-month-old infants will a repeating sound and measured from auditory and frontal areas. The frontal areas showed decreased activation over repeated trials, but there was consistent activity in the auditory cortex.

WHAT IS IMPORTANT TO KNOW WHEN THINKING ABOUT USING OPTICAL IMAGING?

As in all areas of science, it is important to have a well-designed experiment. Knowing about the benefits and limitations of the NIRS technique allows one to design an experiment to cope with these issues. For example, it is wise to measure from more than one cortical area and to design experiments so that you can subtract activation patterns in one condition from activation patterns in a matched condition (Gervain et al., 2008; Peña et al., 2003; Taga et al., 2003; Watanabe et al., 2008). This helps distinguish between global signals (e.g., heart rate and respiration) from functional activation. In addition, using within-subject designs is a good idea because the probes are likely to stay in the same place during the course of an experimental session, so differences are more likely to be due to the variable being manipulated. The NIRS technology is in its infancy, and therefore, it is necessary to replicate the phenomena found in other labs, with different machines. I believe the fastest way to establish this new technology is through sharing code and techniques.

ADDITIONAL INFORMATION

One of the best Web sites for information on NIRS analyses with links to other relevant sites is: http://www.bcs.rochester.edu/people/aslin/mcdonnell/NIRS.html. Dick Aslin and Jacques Mehler have been instrumental in bringing together the various people interested in developing NIRS. (Their efforts and this Web site have been supported by funding from the McDonnell foundation).

Another group that has analyses software available is David Boas' group: http://www.nmr.mgh.harvard.edu/PMI/research.htm. The analysis program is called Homer: http://www.nmr.mgh.harvard.edu/DOT/resources/homer/home.htm. David's group also holds an annual training course on learning how to use NIRS: http://www.martinos.org/martinos/training/NIRS-DOTcourse.php.

A great Web site for explanations about the physics and modeling necessary in using NIRS is: http://www.medphys.ucl.ac.uk/research/nirs. The University College London group has an admirable ability to distill the complex physics behind optics in a highly readable way.

Information about the particular studies in my lab can be found at http://www.psych.northwestern.edu/infantcognitionlab/index.html. If you are interested in learning more about our analyses techniques done in collaboration with Paul Reber, please see http://reberlab.psych.northwestern.edu/wiki/?n=Main.NIRSDataAnalysis. For information about the optical imaging machine that we use (Hitachi ETG 4000), please see http://www.hitachi-medical-systems.eu/products-and-services/optical-topography/etg-4000.html.

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