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Fast cerebral functional signal in the 100-ms range detected in the visual cortex by frequency-domain near-infrared spectrophotometry

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Abstract

Brain activity is associated with physiological changes, which alter the optical properties of the tissue in the near-infrared part of the spectrum. Two major types of optical signals following functional brain activation can be distinguished: a slow signal due to hemodynamic changes and a fast signal, which is directly related to neuronal activity. The fast signal is small and therefore difficult to detect. We used a specially noise-optimized frequency-domain near-infrared spectrometer with a π -sensor, which was expected to be particularly sensitive to deeper tissue layers, to investigate the human visual cortex during visual stimulation generated by a checkerboard. We were able to detect significant fast signals in single light bundles, but not in π -signals. The fast signals were mostly collocated with strong slow hemodynamic signals, but showed a higher degree of localization than the latter. The latencies of 40 ± 16 ms of the fast signals were similar between locations. Our results also indicate that the brain responds differently to a single and double (forth and back) reversal of the checkerboard, with a stronger reaction upon the double reversal.

Descriptors: Near-infrared spectrophotometry, Frequency-domain, Functional fast signal, Brain, Adult

Brain activity is associated with physiological changes, which alter the optical properties of the tissue. These changes can be detected by near-infrared spectrophotometry (NIRS). Two major types of optical signals following functional brain activation can be distinguished: first, the *slow signal*, which occurs within seconds after the onset of the stimulation and is mainly generated by light absorption related in particular to changes in oxyhemoglobin and deoxyhemoglobin concentration. This slow hemodynamic signal has been demonstrated by many authors (Colier et al., 1999, 2001; Heekeren et al., 1997; Hoshi & Tamura, 1993; Kato, Kamei, Takashima, & Ozaki, 1993; Meek et al., 1995; Ruben et al., 1997; Takahashi et al., 2000; Villringer, Planck, Hock, Schleinkofer, & Dirnagl, 1993).

Second, the *fast signal*, which appears within milliseconds after the onset of the stimulus and was suggested to be associated with changes in light scattering of the neurons (Cohen, 1973; Hochman, 1997; Salzberg, Obaid, & Gainer, 1985; Tasaki, 1999). This signal is much smaller than the slow signal and therefore much more difficult to detect. It was described only by a few authors (Gratton, Corballis, Cho, Fabiani, & Hood, 1995; Gratton & Fabiani, 2001; Gratton, Fabiani, & Corballis, 1997;

Gratton, Fabiani, Corballis, & Gratton, 1997; Gratton, Fabiani, Corballis, Hood, et al., 1997; Gratton, Sarno, Maclin, Corballis, & Fabiani, 2000; Rinne et al., 1999; Steinbrink et al., 2000).

To improve the sensitivity for the fast signal, we used a probe with a special geometry that had been developed and tested in our group earlier. With the new type of probe, the π -sensor, we tried to detect the fast signal during visual stimulation. The π -sensor was based on taking the product of the signals of two crossed source-detector pairs, which were obtained simultaneously over the region under study. The product of the variations in modulation amplitude (AC), mean light intensity (DC), and phase (ϕ) were considered. By using this design we decreased the influence of optical fluctuations near the surface and increased the sensitivity to deeper and more localized volumes of the tissue (Filiaci, 2001; Filiaci et al., 1998).

Our frequency-domain NIRS instrument was noise optimized, and special filtering algorithms were applied to detect the fast signal in the human visual cortex during visual stimulation generated by a checkerboard.

Materials and Methods

Instrument

We used a modified frequency-domain spectrophotometer (Oximeter, ISS, Champaign, IL). The instrument operated at

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two different wavelengths, 670 nm and 830 nm. The light generated by four laser diodes (two per wavelength with an output power of approximately 5 mW per laser diode) was intensity modulated at a frequency of 110 MHz. The light from the instrument to the tissue and back to the instrument was guided through optical glass fibers. The core diameter of the source fibers was 0.4 mm and 5 mm for the detector fibers, for which we used liquid light guides. The collected light was conducted to two photomultiplier tubes (PMT). The second dynode of the photomultiplier tube was modulated at 110.005 MHz to demodulate the high frequency. The signal at the difference frequency of 5 kHz was low-pass filtered (cut-off 10 kHz). The output signals from the PMTs were sent to a computer for data processing and the AC, DC, and ϕ of the wave were determined.

Sensor

The sensor consisted of two crossed source-detector pairs with each source and detector fiber located at a corner of a rectangle of approximately 2.5 cm by 2.5 cm (Figure 1). The source-detector distance was set to 2.8 to 3.7 cm by adjusting the size of the rectangle depending on the optical density of the tissue of the subject. For each source position the light of two laser diodes was combined to achieve a better signal-to-noise ratio. All laser diodes were turned on continuously without multiplexing. We only measured the crossed source-detector channels. Light filters were inserted in front of the PMT detectors. At the detector collecting light at 670 nm a light filter removed the light at 830 nm. Analogously at the other detector a filter removed the light at 670 nm. Each light filter had a blocking optical density of >6 and cross talk between the two channels was excluded. This sensor was named π -sensor because the product of the two crossed signals was taken.

Auxiliaries

Heart rate (HR) and arterial hemoglobin oxygen saturation (SaO₂) were recorded with a pulse oximeter (N200, Nellcor, Inc.) and the respiration rate with a strain gauge (New Life Technologies).

Participants

Four healthy adult volunteers (two women and two men, age range between 34 and 36 years) were included in this study. Written informed consent was obtained from all participants prior to the measurements.

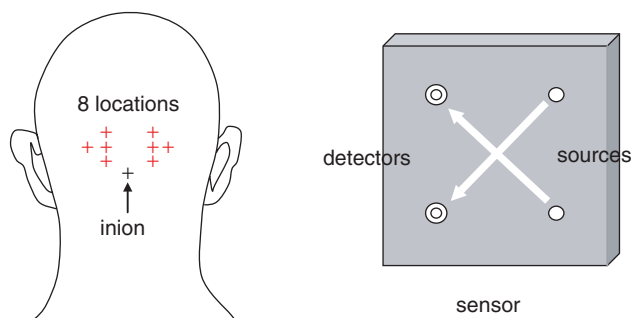


Figure 1. We used the inion method to locate the visual cortex. We placed the sensor at each of the eight locations and measured for 20 min. After that, the sensor was moved 0.5 cm to the next location. The arrows indicate the two crossed light bundles that were analyzed.

Protocol

The sensor was placed sequentially at eight different locations (four over each hemisphere) on the back of the head over the visual cortex using the inion as an orientation mark (Gratton et al., 1995; Kato et al., 1993). The distance between the locations over one hemisphere was 0.5 cm (Figure 1).

The stimulus was a checkerboard pattern (Chiappa, 1997). The participant was lying face down in a quiet and dim room and was asked to close her/his eyes during the resting periods and to focus on a light emitting diode (LED) in the center of the checkerboard during the stimulation periods. The checkerboard consisted of yellow LEDs. The checkerboard pattern was formed by illuminating half of the checkers (every second checker). The checkerboard pattern was reversed by illuminating all the dark checkers and vice versa. One measurement cycle included three periods: (1) 10 s while the checkerboard pattern was illuminated, (2) 20 s while the checkerboard pattern was reversing, and (3) 30 s rest while the checkerboard was turned off. The entire time while the checkerboard was illuminated (30 s) was regarded as the functional stimulation for the slow signal. For each location this cycle was repeated 20 times. Preceding the first and following the last stimulation cycle a baseline of 1 min was recorded. After completing 20 cycles at one location, the sensor was moved to the next position and the measurement was repeated (Figure 1).

The reversing frequency of the checkerboard was set to 4 Hz or 5 Hz depending on the participant's heart rate to avoid pulse harmonics. The data acquisition and the reversing of the checkerboard were fully electronically synchronized at a rate of 16 samples per reversing period.

Control measurements were performed on a solid phantom block of approximately the same optical properties as the human head. The visual stimulation system was also active during these measurements to test the system for direct electrical or optical cross talk.

This protocol was approved by the Institutional Review Board of the University of Illinois at Urbana-Champaign (IRB #94125).

Data Analysis

Fast neuronal signal. The raw AC, DC and ϕ values for each wavelength of the frequency-domain instrument were analyzed. The data were recorded at a sample rate of 64 Hz or 80 Hz depending on the stimulation frequency.

To reduce physiological noise, the arterial pulsation was removed using an adaptive filter, which is described in detail elsewhere (Gratton & Corballis, 1995). In short, it extracted a mean shape of the pulse by screening each trace separately for pulses. The period of each pulse was adjusted before averaging. This mean shape corresponded to the best estimate of each pulse and was used to remove each pulse from the data. This required adjusting the period of the mean shape to the one of each pulse and scaling the shape by a linear regression.

The natural logarithm of the AC and DC values was calculated.

The data was detrended by a digital high-pass filter with a variable cutoff frequency. The filter was very sharp (10 dB/0.1 Hz). As a cutoff frequency, we selected 2.2 times the mean heart rate, that is, to further reduce the effect of the hemodynamic pulsation, but still to be low enough not to affect the fast neuronal signal at 2.5 times the mean heart rate. This filter also removed low-frequency effects such as breathing and slow vasomotion.

To address the question of whether each single reversal of the checkerboard was to be considered as a stimulation or whether a double reversal (forth and back) should be considered as one stimulation, we generated for each option one signal, which was synchronized with the stimulation named reversing signal.

A cross-correlation function (CCF) between the optical signal (OS) and the reversing signals (RS) was calculated:

$$CCF(\tau) = \sum (OS(t) * RS(t + \tau)) / \sum RS(t) \quad (1)$$

The advantage of this equation was that it removed the amplitude of the reversing signal and preserved the amplitude of the optical signal. The CCF was very sensitive and selective for signal components in the optical signal, which were coherent with the reversing signal. OS represents the AC, DC, and ϕ of each light bundle separately and the π -signal (product of the two crossed light bundles).

The reversing signal was also cross-correlated to optical signals measured on a solid phantom block to test for instrumental noise. A noise analysis was carried out to determine the level of noise depending on the number of samples. For this purpose we compared the noise in the data after the filtering to the noise level in the CCF. In our case the CCF followed the statistical laws, that is, the factor by which the level of random noise was reduced, depended on $1/N^{1/2}$, where N was the number of data points. In contrast, when the optical signal contained components that were coherent with the reversing signal, this genuine signal kept its amplitude independent of the number of data points. The measurements on the phantom block were used to determine the factor of the noise reduction depending on N . It was verified that this factor also held for physiological measurements. Thus it was possible to identify signals that were statistically significantly above this level of noise and select significant AC, DC, and ϕ of light bundles.

To test for the relation between reversing signal and the detected signal, we compared the signal amplitudes during stimulation and rest.

The *latency* of a fast signal was the time period between a stimulus (reversal) and the closest maximum and minimum of the CCF.

Slow hemodynamic signal. The data was reduced to 2 Hz and 2.5 Hz, respectively, by averaging 32 samples, and the natural

logarithm was taken. A folding average was taken to convert the entire stimulation sequence into one representative stimulation and rest period. The change in oxyhemoglobin (O_2Hb in $\mu\text{mol/l}$) and deoxyhemoglobin (HHb in $\mu\text{mol/l}$) concentration was calculated from the attenuation changes using the differential path length factor method (Delpy et al., 1988). The time traces of these signals were visually inspected and corresponded to the known patterns (Colier et al., 1999, 2001; Heekeren et al., 1997; Hoshi & Tamura, 1993; Kato et al., 1993; Meek et al., 1995; Ruben et al., 1997; Takahashi et al., 2000; Villringer et al., 1993).

Results

We found five significant fast signals in two out of four subjects. We found significant signals only in AC or DC but not in ϕ and none of the π -signals were significant. Most of the signals were found for the type of analysis when a double reversal was considered as one stimulus. Examples of fast signals for each type of analysis are shown in Figure 2 and Figure 3. All significant signals were found in the female subjects, none in the male subjects. The amplitude of the CCF was larger during stimulation than rest in all significant fast signals, which indicates that the signals were related to the stimulation. The amplitude significance, latency, and type of all fast signals are presented in Table 1. The amplitudes varied for the different signals but were, in general, very low. The latencies for the maximum of the signal showed a large variation, whereas the variation between the latencies for the minimum of the CCF was low. This indicates that the fast signal was a drop in the AC or DC. The collocations of the fast and slow signal are shown in Figure 4 and Figure 5. Four out of five fast signals were collocated with a strong slow signal.

In measurements on a solid phantom block of approximately the same optical properties as the human head, no significant signals were found, which indicates that the signal was not due to instrumental noise or interference.

The slow signal was detected in all subjects and varied within individual subjects depending on the location (Figures 4 and 5).

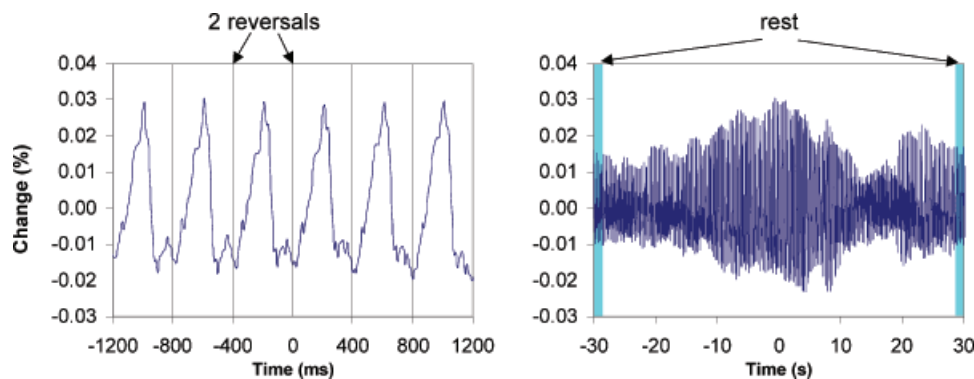


Figure 2. The same fast signal in the light intensity is displayed on two different time axes. The enlarged view has vertical lines that indicate where a stimulation occurs with a periodicity of two reversals of the checkerboard. The traces are completely synchronous with the stimulation. In the graph with a larger time period, the signal during rest (shaded area), which is smaller than during stimulation (time = 0 s), is displayed.

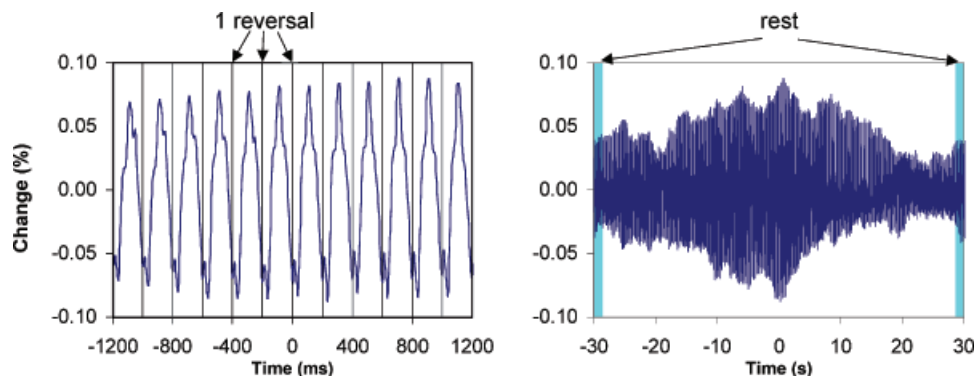


Figure 3. This fast signal was coherent with each reversal of the checkerboard pattern. It is displayed analogously to Figure 2.

Discussion

The product of the two channels (π -signal) is sensitive for common components in the two signals. When the two channels are arranged in a crossed geometry (Figure 1), the common volume of tissue interrogated by both channels is at the crosspoint of the two bundles and deep in the tissue. Changes in the optical properties of this common volume are the origin of the common components of the signals. A single bundle is very sensitive to fluctuations directly below the source or detector, whereas the π -signal is more sensitive to deeper layers of tissue and less influenced by superficial tissue than each light bundle separately (Filiaci, 2001; Filiaci et al., 1998). Consequently, we had expected to find more significant fast signals in the π -signals than in the signals of the separate bundles. There may be several reasons why we did not find any significant π -signal:

Due to the higher spatial resolution of the π -sensor, it is also more likely to miss the activated spot, particularly in the highly localized visual cortex (Frostig, Lieke, Ts'o, & Grinvald, 1990).

We were using two different wavelengths (670 nm and 830 nm), because we wanted to separate the two channels without multiplexing the light sources. It was our aim to measure exactly the same point in time with both bundles. However, the tissue is optically denser at 670 nm than at 830 nm, which leads to a different penetration depth of the two light bundles. From previous measurements, we estimate a mean penetration depth of 0.73 cm (670 nm) and 0.81 cm (830 nm). Thus the difference is less than 1 mm, but still may reduce the common tissue volume interrogated by both channels. Furthermore the detected light intensity was considerably lower at 670 nm than at 830 nm,

which leads to a higher noise level for the 670-nm channel. The difference in the noise level between the two channels may further reduce the efficacy of the π -sensor principle.

The π -signal is particularly sensitive to cross talk, which is an additional π -specific source of noise. Even though there was no optical cross talk, changes in the intensity values due to electrical sources of noise of the instrument may have affected both channels commonly. Cross talk in the analog-to-digital converter card could have the same effect. Even if the cross talk was small, it would be amplified by taking the product of the two channels for the π -signal. This would increase the noise level we found on the phantom block, and thus a fast signal would not that easily achieve a sufficient signal-to-noise ratio for statistical significance compared to the separate channels.

The fast signal that we have detected is a small change in AC or DC in a single light bundle. The magnitude of the change is in the same range as found by Steinbrink et al. (2000).

The origin of the fast signal detected by our frequency-domain method cannot be instrumental because we did not find any such signal when measuring on a phantom block. The fast signal cannot be caused by random noise either, because it is significantly different from noise. Moreover, it cannot be generated by direct coupling to light from our checkerboard for a number of reasons: we carefully shielded the sensor against ambient light, the general brightness of the checkerboard did not change during the reversing periods, and light coupled from the checkerboard is not intensity modulated and would therefore only affect the DC and not the AC. Thus direct coupling between the checkerboard and the detectors at the sensor can be excluded.

Table 1. Description of the Fast Signals

Code	Subject	Signal	Periodicity	Amplitude	p	Latency		Wavelength
						Max	Min	
1	A	AC	1	0.072%	<.01	113 ms	38 ms	830 nm
2	A	AC	2	0.062%	<.025	-63 ms	63 ms	830 nm
3	A	AC	2	0.025%	<.025	213 ms	25 ms	830 nm
4	A	DC	2	0.021%	<.001	213 ms	25 ms	830 nm
5	B	AC	2	0.382%	<.05	250 ms	47 ms	670 nm

The code refers to the numbers used in Figures 4 and 5. AC is the amplitude and DC the mean intensity of the detected light intensity wave. For a periodicity of 1 each reversal of the checkerboard was treated as a stimulation, while for a periodicity of 2 two reversals (forth and back) were considered as one stimulation period. The amplitude is given in percent of the mean signal level. p is the significance of the fast signal. The latency is the period of time between stimulation and the closest maximum or minimum of the cross-correlation function.

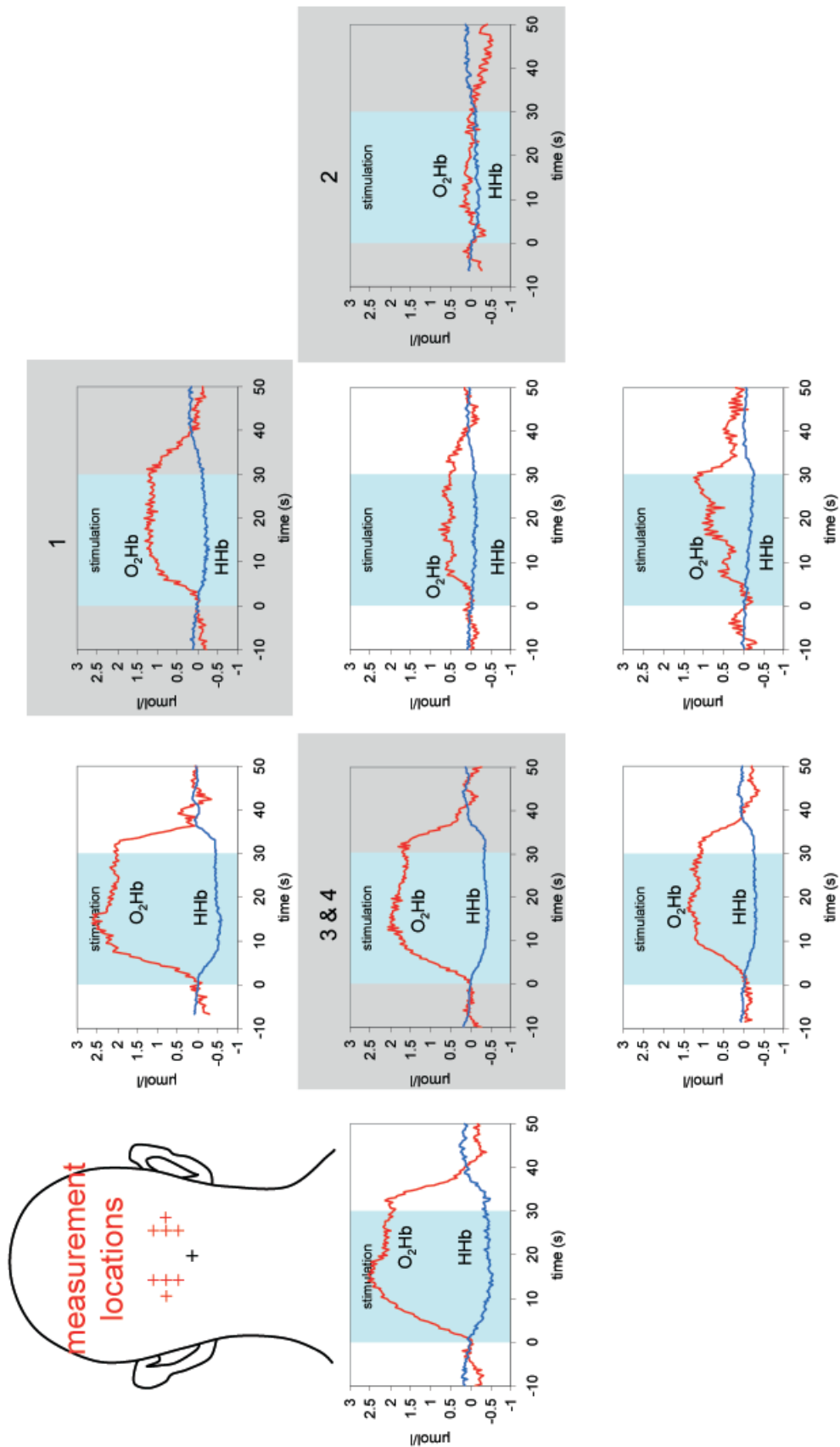


Figure 4. Locations of the functional signals in Subject A. The diagrams are arranged according to the location on the back of the head, where they were measured. The slow hemodynamic signal is clearly visible in most of the locations. The dark-shaded areas indicate locations where a fast neuronal signal was found. The locations of the fast signal are not completely congruent with the slow signal. The fast signals are numbered according to Table 1.

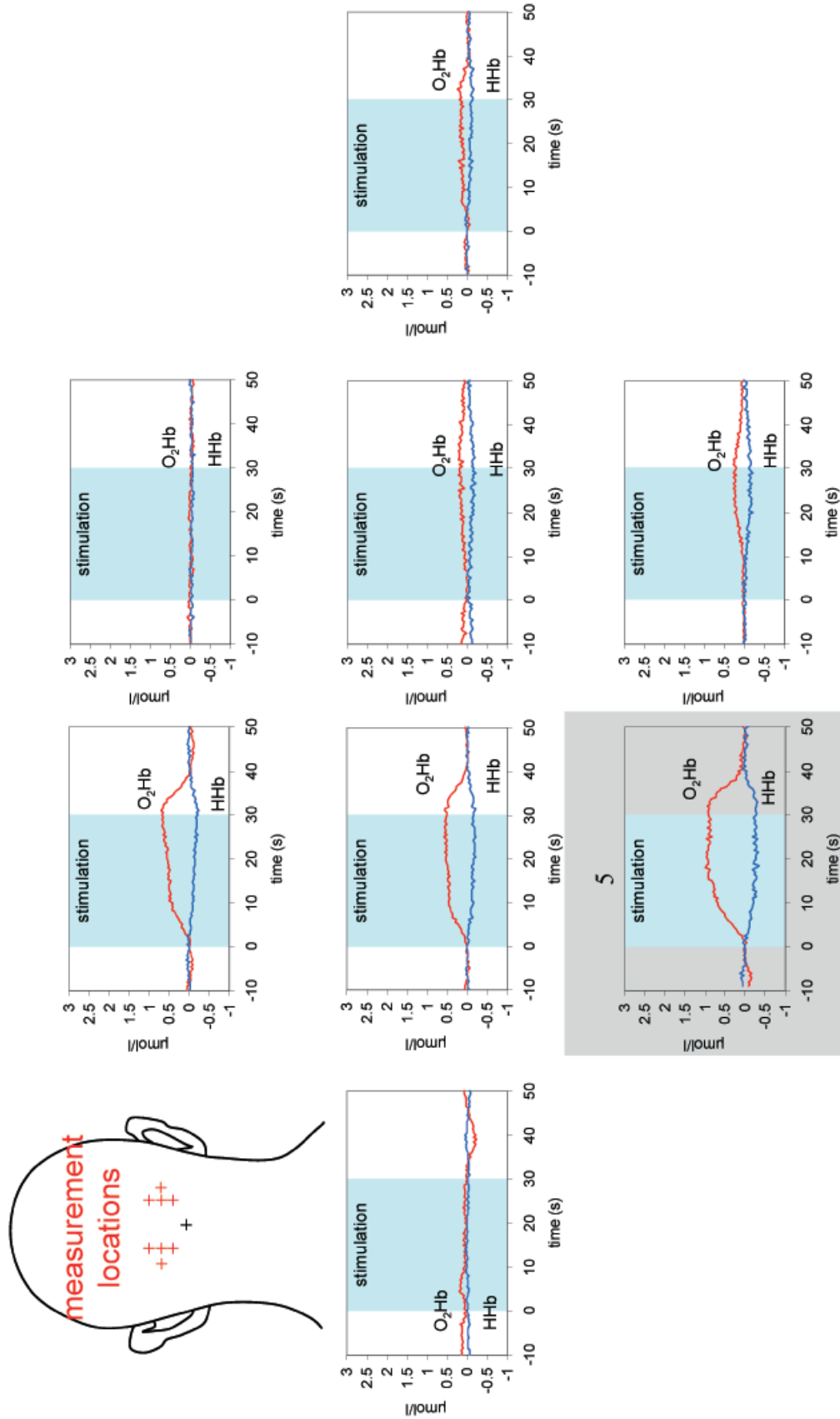


Figure 5. Locations of the functional signals in Subject B. The display is analogous to Figure 4. The fast neuronal signal was found in the same location as the largest slow signal.

The fact that the signal amplitude is higher during stimulation than during rest demonstrates that our signal is related to the stimulation. We can exclude systemic physiological effects as an origin of the fast signal because they should not appear within 100 ms. For all these reasons we conclude that the fast signal originates in the brain.

Several studies on the origin of the fast signal have been conducted in single nerve cells, animals, and humans. Extensive reviews can be found in Cohen (1973), Hochman (1997), and Tasaki (1999). In short, according to these studies, a likely origin of the fast signal is a change in birefringence of the cell membrane, which occurs simultaneously to the action potential and swelling of the neuronal cell.

Furthermore Salzberg et al. (1985) studied the intact neurohypophysis of mice and reported large and rapid decreases in light scattering, which accompanied the secretion by nerve terminals. These changes occurred in two phases and the fractional light intensity change was approximately 0.2%.

In humans, Gratton and Fabiani (2001), Gratton, Fabiani, and Corballis (1997), Gratton, Fabiani, Corballis, and Gratton (1997), Gratton, Fabiani, Corballis, Hood, et al. (1997), Gratton et al. (1995, 2000), and Rinne et al. (1999) report increases in the phase measured by a frequency-domain NIRS instrument. Although all authors of the cited literature assume a change in light scattering from the methodological point of view, it cannot be excluded that the reported fast signals in human subjects are related to a change in light absorption. We were not able to clarify this aspect in our current study because we found no significant fast signal in the ϕ .

In our study, the fast signal exhibits a higher localized spatial pattern than the slow signal (Figures 4 and 5). From data in the literature, we would expect that the region of neuronal activity is much smaller than the region of subsequent hemodynamic changes (Malonek & Grinvald, 1996). Also in our study, the fast signal is only detected in a few locations, which may indicate that the signal is highly localized. However, our signal-to-noise ratio may not have been sufficient to detect the fast signal in other locations.

It is interesting to note that we found more fast neuronal signals when we considered a double reversal of the checkerboard as one period of stimulation instead of a single reversal. This demonstrates that the brain is responding more to a double reversal than to a single reversal stimulus.

The mean latency of 40 ms of the minima of the CCF is in the lower range of a latency determined by EEG (Lesser, Luders, Klem, & Dinner, 1985). The latency depends to a large degree on the specific type of stimulation. The variation of our latencies between signals is small ($SD = 16$ ms), which further indicates that our signals are real.

Why was the fast signal only found in 2 subjects? According to the organization of the visual cortex, where neurons with similar orientation preferences are grouped together into columns (Frostig et al., 1990), the fast signal is expected to be very localized (within the range of millimeters). For this reason the regions of the brain that are altered by visual stimulation can easily be missed, despite our efforts to cover the region of interest with a high spatial resolution of 0.5 cm. In the future, this resolution can be increased, for example, by using an instrument with a higher number of detectors and sources.

If it was not a coincidence that the signal was found in all our female subjects and not in the male subjects, the signal will be more difficult to detect in males. This may be explained by a higher thickness of the male skull, which reduces our signal.

Conclusion

We were able to detect small fast neuronal signals in the human visual cortex during visual stimulation with NIRS. These signals showed a higher degree of localization than the slow hemodynamic signals. Moreover, four out of five fast signals were collocated with a strong slow signal. The latencies of 40 ± 16 ms of the fast signals were similar between locations. Our results also indicate that the brain responds more to a double reversal than to a single reversal stimulus.

REFERENCES

- Chiappa, K. H. (1997). *Evoked potentials in clinical medicine*. Philadelphia: Lippincott-Raven.
- Cohen, L. B. (1973). Changes in neuron structure during action potential propagation and synaptic transmission. *Physiological Reviews*, 53, 373–418.
- Colier, W. N., Quaresima, V., Wenzel, R., van der Sluijs, M. C., Oeseburg, B., Ferrari, M., & Villringer, A. (1999). Cortical blood oxygenation changes in the left and right occipital area induced by selective visual stimuli in humans. *Advances in Experimental Medicine and Biology*, 471, 35–41.
- Colier, W. N., Quaresima, V., Wenzel, R., van der Sluijs, M. C., Oeseburg, B., Ferrari, M., & Villringer, A. (2001). Simultaneous near-infrared spectrophotometry monitoring of left and right occipital areas reveals contra-lateral hemodynamic changes upon hemi-field paradigm. *Vision Research*, 41, 97–102.
- Delpy, D. T., Cope, M., van der Zee, P., Arridge, S., Wray, S., & Wyatt, J. (1988). Estimation of optical pathlengths through tissue from direct time of flight measurement. *Physics in Medicine and Biology*, 33, 1433–1442.
- Filiaci, M. E. (2001). Spatial localization and temporal analysis of optical property fluctuations by multiplexed near-infrared photon density waves in turbid media: In vitro and in vivo studies. (Thesis, University of Illinois at Urbana-Champaign).
- Filiaci, M., Toronov, V., Fantini, S., & Gratton, E. (1998). Optical probe and frequency domain instrumentation to study spatial and temporal correlations and fluctuations in tissues. *Optical Society of America Biomedical Topical Meetings Technical Digest*, 22, 183–187.
- Frostig, R. D., Lieke, E. E., Ts'o, D. Y., & Grinvald, A. (1990). Cortical functional architecture and local coupling between neuronal activity and the microcirculation revealed by in vivo high-resolution optical imaging of intrinsic signals. *Proceedings of the National Academy of Sciences, USA*, 87, 6082–6086.
- Gratton, G., & Corballis, P. M. (1995). Removing the heart from the brain: Compensation for the pulse artifact in the photon migration signal. *Psychophysiology*, 32, 292–299.
- Gratton, G., Corballis, P. M., Cho, E., Fabiani, M., & Hood, D. C. (1995). Shades of gray matter: Noninvasive optical images of human brain responses during visual stimulation. *Psychophysiology*, 32, 505–509.
- Gratton, G., & Fabiani, M. (2001). Shedding light on brain function: The event-related optical signal. *Trends in Cognitive Sciences*, 5, 357–363.
- Gratton, G., Fabiani, M., & Corballis, P. M. (1997). Can we measure correlates of neuronal activity with non-invasive optical methods? *Advances in Experimental Medicine and Biology*, 413, 53–62.
- Gratton, G., Fabiani, M., Corballis, P. M., & Gratton, E. (1997). Noninvasive detection of fast signals from the cortex using frequency-

- domain optical methods. *Annals of the New York Academy of Sciences*, 30, 286–298.
- Gratton, G., Fabiani, M., Corballis, P. M., Hood, D. C., Goodman-Wood, M. R., Hirsch, J., Kim, K., Friedman, D., & Gratton, E. (1997). Fast and localized event-related optical signals (EROS) in the human occipital cortex: Comparisons with the visual evoked potential and fMRI. *NeuroImage*, 6, 168–180.
- Gratton, G., Sarno, A., Maclin, E., Corballis, P. M., & Fabiani, M. (2000). Toward noninvasive 3-D imaging of the time course of cortical activity: Investigation of the depth of the event-related optical signal. *NeuroImage*, 11, 491–504.
- Heekeren, H. R., Obrig, H., Wenzel, R., Eberle, K., Ruben, J., Villringer, K., Kurth, R., & Villringer, A. (1997). Cerebral haemoglobin oxygenation during sustained visual stimulation—A near-infrared spectroscopy study. *Philosophical Transactions of the Royal Society of London B, Biological Sciences*, 29, 743–750.
- Hochman, D. (1997). Intrinsic optical changes in neuronal tissue. Basic mechanisms. *Neurosurgical Clinics of North America*, 8, 393–412.
- Hoshi, Y., & Tamura, M. (1993). Dynamic multichannel near-infrared optical imaging of human brain activity. *Journal of Applied Physiology*, 75, 1842–1846.
- Kato, T., Kamei, A., Takashima, S., & Ozaki, T. (1993). Human visual cortical function during photic stimulation monitoring by means of near-infrared spectroscopy. *Journal of Cerebral Blood Flow and Metabolism*, 13, 516–520.
- Lesser, R. P., Luders, H., Klem, G., & Dinner, D. S. (1985). Visual potentials evoked by light-emitting diodes mounted in goggles. *Cleveland Clinic Quarterly*, 52, 223–228.
- Malonek, D., & Grinvald, A. (1996). Interactions between electrical activity and cortical microcirculation revealed by imaging spectroscopy: Implications for functional brain mapping. *Science*, 272, 551–554.
- Meek, J. H., Elwell, C. E., Khan, M. J., Romaya, J., Wyatt, J. S., Delpy, D. T., & Zeki, S. (1995). Regional changes in cerebral haemodynamics as a result of a visual stimulus measured by near infrared spectroscopy. *Proceedings of the Royal Society of London B, Biological Sciences*, 261, 35–36.
- Rinne, T., Gratton, G., Fabiani, M., Cowan, N., Maclin, E., Stinard, A., Sinkkonen, J., Alho, K., & Näätänen, R. (1999). Scalp-recorded optical signals make sound processing in the auditory cortex visible? *NeuroImage*, 10, 620–624.
- Ruben, J., Wenzel, R., Obrig, H., Villringer, K., Bernarding, J., Hirth, C., Heekeren, H., Dirnagl, U., & Villringer, A. (1997). Haemoglobin oxygenation changes during visual stimulation in the occipital cortex. *Advances in Experimental Medicine and Biology*, 428, 181–187.
- Salzberg, B. M., Obaid, A. L., & Gainer, H. (1985). Large and rapid changes in light scattering accompany secretion by nerve terminals in the mammalian neurohypophysis. *Journal of General Physiology*, 86, 395–411.
- Steinbrink, J., Kohl, M., Obrig, H., Curio, G., Syre, F., Thomas, F., Wabnitz, H., Rinneberg, H., & Villringer, A. (2000). Somatosensory evoked fast optical intensity changes detected non-invasively in the adult human head. *Neuroscience Letters*, 291, 105–108.
- Takahashi, K., Ogata, S., Atsumi, Y., Yamamoto, R., Shiotsuka, S., Maki, A., Yamashita, Y., Yamamoto, T., Koizumi, H., Hirasawa, H., & Igawa, M. (2000). Activation of the visual cortex imaged by 24-channel near-infrared spectroscopy. *Journal of Biomedical Optics*, 5, 93–96.
- Tasaki, I. (1999). Rapid structural changes in nerve fibers and cells associated with their excitation processes. *Japanese Journal of Physiology*, 49, 125–138.
- Villringer, A., Planck, J., Hock, C., Schlinkofer, L., & Dirnagl, U. (1993). Near infrared spectroscopy (NIRS): A new tool to study hemodynamic changes during activation of brain function in human adults. *Neuroscience Letters*, 154, 101–104.

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