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Simultaneous EEG/Functional Magnetic Resonance Imaging at 4 Tesla: Correlates of Brain Activity to Spontaneous Alpha Rhythm During Relaxation

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Summary

Simultaneous EEG and functional magnetic resonance imaging have been applied to the study of brain states associated with alpha waves using a magnetic field strength of 1.5 Tesla and has been shown in recent years to be feasible up to 3 Tesla for other applications. This study demonstrates this technique's continued viability at a field strength of 4 Tesla, affording a proportionally greater sensitivity to changes in Blood Oxygen Level Dependent (BOLD) signal. In addition, for the study of alpha correlations, the authors used a larger number of subjects and scanning sessions than in the previous work. Random effects group regression analysis of 35 EEG/functional magnetic resonance imaging sessions against occipital alpha magnitude in a relaxed state detected bilateral widespread activation of dorsal thalamus and portions of the anterior cingulate and cerebellum. In the same group analysis, deactivations arose predominantly in the fusiform and adjacent visual association areas with a small activation cluster also detected in dorsolateral prefrontal cortex. This pattern is consistent with a correspondence between alpha magnitude variations and resting state network dynamics ascertained by recent studies of low frequency spontaneous BOLD fluctuations. The central role of the thalamus in resting state networks correlated with alpha activity is highlighted. Demonstrating the applicability of simultaneous EEG/functional magnetic resonance imaging up to 4 Tesla is particularly important for clinically relevant research involving challenging spontaneous EEG abnormalities, such as those of epilepsy.

Keywords

Simultaneous EEG/fMRI; Alpha rhythm; Spontaneous brain activity; Thalamus; Windowed Fourier analysis; High-field MR imaging

The brain is known to enter states characterized by synchronous oscillatory behavior of large collections of neurons. Neuronal oscillations, spanning five orders of magnitude in frequency (Buzsaki and Draguhn, 2004), have been the subject of intense study since their first measurement in humans nearly eight decades ago (Berger, 1929). Although plausibly nothing

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more than a manifestation of neurophysiological noise, a growing body of work supports the view that rhythmic states play important roles in the workings of complex interacting neuronal networks by modulating input sensitivity, encoding information, and permitting plasticity (Buzsaki and Draguhn, 2004; Steriade et al., 1993). Thus, the understanding of the sources and function of neuronal oscillations is a key for framing the complexity of brain behavior.

Synchronous electrical activity of cortical neurons is detectable noninvasively by EEG or magnetoencephalography. Using these methods, a range of frequencies principally from 1 to 40 Hz is routinely measured for clinical and research applications. Frequency subranges are further delineated according to correspondence to functional state. The longest known and most prominent of these is the alpha rhythm occupying the frequency range of 8 to 13 Hz with greatest power detectable by EEG electrodes overlying the occipital lobes (Berger, 1929). Alpha oscillations are used as an electrophysiological indicator of vigilance and arousal, attenuating during targeted cognitive activity and strengthening while the brain is unoccupied by specific mental tasks and devoid of significant sensory input, particularly visual input. Current understanding of the origins of occipital alpha rhythm is incomplete, but a plausible scheme includes a complex interplay between the lateral geniculate and reticular nuclei of the thalamus and the primary visual cortex (Destexhe et al., 1998; Fuentealba and Steriade, 2005; Hughes et al., 2004; Jones, 2002). Cortical neurons and relay cells of the geniculate body form a closed loop circuit with axons that pass through and form side branches into the lateral reticular nucleus. GABA-ergic reticular cells subsequently feed back to inhibit the relay cells in the lateral geniculate sustaining a hyperpolarized state with oscillatory firing that is subsequently imposed on the visual cortex. Measurements of alpha-like rhythms in these structures in animal models lend support to this scenario (Buzsaki, 1991; da Silva et al., 1973). Rhythmic activity can be attenuated by excitatory inputs to the relay cells either from retinal afferents or from layer V neurons of other cortical areas. In this way, the thalamus is viewed not only as a relay of sensory input to the cortex, but also as an integrator of cortical networks through cortico-cortical communication.

Even while relaxing (no directed task) and awake with eyes closed, alpha power is observed to modulate spontaneously. The observed variation in alpha strength occurs at sub-Hertz frequencies aligned with those recently reported for spontaneous fluctuations in resting state Blood Oxygen Level Dependent (BOLD) signals (Damoiseaux et al., 2006; Gusnard et al., 2001). Spatiotemporal coherencies of BOLD signal variation suggest that the brain, unoccupied by external demands, alternates significant, ongoing mental activity between large introspective and extrospective functional networks (Fox et al., 2005; Fransson, 2005). Functional neuroimaging relies on contrast between task-directed brain activity and a more relaxed control condition. Spatiotemporal patterns of cognitive effort arising spontaneously during relaxation constitute a background with potential impact on the interpretation of functional data.

Although multichannel EEG or magnetoencephalography data can be inverted to solve for effective source distributions, the technique inherently lacks uniqueness and suffers from low spatial resolution (Koles, 1998). Moreover, these measurement modalities are sensitive only to neuronal activity in the superficial cortex and are unable to probe deep structures as reliably as other techniques, e.g., positron emission tomography or functional magnetic resonance imaging (fMRI).

To elucidate the functional underpinnings of the relaxed condition throughout the brain defined by the presence and absence of alpha activity, investigators have recently combined the fine temporal resolution of EEG (milliseconds) with the high spatial resolving power (millimeters) and spatial coverage of functional imaging (Goldman et al., 2002; Laufs et al., 2003a,b; Moosmann et al., 2003). Positron emission tomography studies conducted concurrently with

EEG have reported a mixture of conflicting results for the association of glucose metabolism or blood flow with alpha activity. An investigation of alpha power correlates to regional cerebral blood flow found positive correlation in the thalamus and negative correlation in the occipital cortex and frontal lobe (Sadato et al., 1998). In contrast, other work, measuring glucose metabolic rate determined that alpha correlated negatively with thalamic metabolism (Lindgren et al., 1999). These positron emission tomography studies, however, used an index of alpha power averaged over time periods of 90 seconds to 30 minutes, which would include numerous modulations of spontaneous alpha intensity according to present observations. The combination of fMRI with EEG to study spontaneous alpha sources, first by interleaving the functional imaging with EEG (Goldman et al., 2002), has implicated the thalamus as increasing in neuronal activity as alpha power increases at the cortex while the occipital and portions of the parietal lobes are more active when the alpha power decreases. The advent of methods to subtract the large EEG artifact caused by the rapidly switching gradient coil currents during fMRI has permitted the use of all time points of EEG data acquired concurrently with imaging (Allen et al., 2000). Acquisitions of this kind have had inconclusive results as well. The first study to use fully concurrent acquisitions reported, for random effects group analysis, negative alpha activation clusters exceeding 10 voxels in the visual association cortex and the cuneus (Moosmann et al., 2003). The same data generated positive activation in the thalamus, but only after dramatic relaxation of the significance threshold. Other investigators (Laufs et al., 2003a,b), using similar techniques, have found positive activation to be absent throughout the brain for both group and individual analyses. Instead of finding clusters of negative activation in the occipital regions, this research found broadly distributed negatively correlated clusters to alpha rhythm in the middle and inferior frontal gyri and the posterior and inferior parietal gyri. Follow-up analysis of these same data (Laufs et al., 2006a) and other recent work (Goncalves et al., 2006) have suggested that BOLD excursions related to alpha power follow cortical patterns that vary greatly between individuals and even over time for the same individual (Goncalves et al., 2006).

Performing concurrent EEG and fMRI, particularly for spontaneous source detection, presents a number of challenges. The efficiency of the paradigm is not controlled because it relies on the degree and frequency of EEG modulation during a particular scan. Sensitivity can thereby suffer. Although the random paradigm associated with spontaneous alpha may afford relatively high efficiency (Dale, 1999), other clinically relevant spontaneous activity such as epileptic spikes may result in paradigms of low efficiency. Bolstering sensitivity can be achieved by performing fMRI at higher field strengths. Direct comparisons of BOLD sensitivity between 1.5 Tesla and higher field strengths have found substantial increases in signal strength and extent of activation for tasks associated with visual and motor cortex (Gati et al., 1997; Kruger et al., 2001; Turner et al., 1993; Yang et al., 1999) as well as cognitive function (Krasnow et al., 2003). This advantage, however, is accompanied by the cost of exacerbating the challenge of gradient coil-induced artifacts. Simultaneous EEG/fMRI work focused on spontaneous alpha activity has, thus far, used field strength of up to 1.5 Tesla. Successful correction for ballistocardiographic and gradient artifacts has been demonstrated recently at fields up to 3 Tesla for other applications of concurrent EEG and fMRI (Debener et al., 2005; Vincent et al., 2007). This study extends the application to 4 Tesla to provide the greatest BOLD sensitivity, yet for this measurement technique.

MATERIALS AND METHODS

Subjects and Paradigm

Twenty healthy adult subjects (8 women and 12 men) with mean $\pm$ SD age of 30.7 ± 9.0 years (range 18–46 years) took part in this study after providing informed consent for protocol approved by the institutional review board of the University of Cincinnati. Each subject was

instructed to lie still, relax, and keep eyes closed in the magnetic resonance scanner for 20-minute sessions during which functional imaging and EEG data were acquired simultaneously. Maintenance of a relaxed state was facilitated by playing soothing music of the subject's choice continuously during EEG/fMRI acquisition to reduce the preoccupation with fatigue associated with lying still for an extended period of time. Two separate 20-minute sessions were scheduled consecutively for each subject for a total of 40 scanning sessions.

Imaging

The fMRI was performed on a 61.5-cm bore Varian Unity INOVA system (Varian, Inc., Palo Alto, CA) equipped with a standard head coil. T2*-weighted, BOLD sensitive, echo-planar images were acquired with parameters: repetition time/echo time = 1800/25 milliseconds, field of view = 256 mm × 256 mm, matrix = 64 × 64, 5-mm slice thickness with axial orientation. For all but four of the scanning sessions, a total of 400 image volumes were acquired at the rate of one every 3 seconds for a total acquisition time of 20 minutes per session. The remaining four scanning sessions, each exceeding 15 minutes in duration, were cut short because of subject fatigue. Reconstruction of functional images included correction for ghosting artifact and geometric distortion by well-established methods (Schmithorst et al., 2001).

A three-dimensional structural image was acquired using a modified driven equilibrium Fourier transform method (Duewell et al., 1996; Ugurbil et al., 1993) with repetition time/echo time = 13.1/6.0 milliseconds, inversion delay, τ = 1,100 milliseconds, field of view = 256 mm × 196 mm × 196 mm, and matrix = 256 × 196 × 196. The three-dimensional structural image served as a T1-weighted anatomic reference onto which the functional results could be overlaid after coregistration.

EEG Acquisition and Processing

Concurrent with the fMRI, we recorded 64 channels of EEG with electrodes arranged according to the international standard 10/20 system. Eye movement and ECG data were also collected for subsequent ballistocardiographic artifact removal. During EEG acquisition, time marks generated by the scanner were inserted automatically in the data stream corresponding to the beginning of each functional image acquisition. As previously, the EEG data were collected continuously at 10 kHz using a MRI-compatible system (MagLink by Neuroscan, Division of Compumedics Ltd., El Paso, TX) with software that included an algorithm to subtract gradient and ballistocardiographic artifacts (scan 4.3.5) (Espay et al., 2008). After low-pass filtering with a cutoff frequency of 60 Hz, the echo-planar image gradient artifacts induced by imaging were averaged over the first three repetition time periods precisely aligned using the embedded time marks. Average gradient signals were then subtracted from each epoch of the raw data following a method described by Allen et al. (2000). In the Scan software, this subtraction procedure was further improved by optimizing temporal alignment between the average gradient waveform and the raw data based on the position of the peak of their cross-correlation. Ballistocardiographic artifact was removed in the same fashion using ECG events as time marks. The cleaned data were subsequently decimated to 200 Hz and derived in a standard, 16-channel bipolar montage. Panels a and b of Figure 1 show a segment of representative EEG data before and after processing.

Each EEG session, after processing, was assessed for quality by the same board certified electrophysiologist (J.P.S.); all reviewed EEGs were normal. A score was given to each interscan interval according to evidence of motion artifact or drowsiness/sleep. Five entire sessions were subsequently eliminated from the study because of the finding of excessive artifact throughout the EEG data. The remaining 35 sessions were analyzed in their entirety using an objective filter for alpha activity in each interscan interval as described below.

Image Processing and Statistical Analysis

Spatial preprocessing and statistical analysis under the general linear model were performed using Statistical Parametric Mapping software (SPM5, <http://www.fil.ion.ucl.ac.uk/spm/>). The initial five images of each session were first discarded to ensure attainment of T1 relaxation equilibrium. Each session was spatially processed following a recipe commonly used in SPM5 including motion correction by rigid body realignment of all functional images to the first image of the session, normalization to the Montreal Neurological Institute template using affine and nonlinear transformations, and spatial Gaussian filtering with an 8-mm kernel. Transformation parameters for normalization were first determined for the structural image using gray matter, white matter, and cerebral spinal fluid templates.

A regressor time course was built for each session by first applying a Hamming-windowed fast Fourier transform to each 3-second interscan interval of the four bipolar channels of the EEG data that included an occipital electrode (P3-O1, P4-O2, P7-O1, and P8-O2). Individual interscan intervals were scored for alpha activity according to the mean of the magnitudes of the Fourier transform frequency components between 8 and 13 Hz. The mean value of this score across the four occipital bipolar channels comprised the alpha time course as shown in the following equation:

$$A_{i,j} = \sum_{k=1}^{N_k} \left(\sum_{l=1}^{N_l} |F_{i,j,k}(\omega_l)| / N_l \right) / N_k$$

where $A_{i,j}$ is the alpha intensity, $|F_{i,j,k}(\omega_l)|$ is the magnitude of the Fourier component, N_l is the number of frequency components between 8 and 13 Hz, and N_k is the number of occipital bipolar EEG channels, for session i , interscan interval j , occipital EEG channel k , and Fourier frequency component l , between 8 and 13 Hz. Representative examples of the four occipital channels and their corresponding Fourier transforms are shown for a segment of data in Figure 1, panels c and d, respectively.

In rare, isolated cases, individual interscan intervals had outlying alpha scores resulting from subsecond, spurious EEG excursions presumed to arise from imperfect gradient artifact removal. Consequently, any alpha score deviating from the session mean by more than three standard deviations was reset to the session mean. The resulting alpha state regressor, after high-pass filtering and convolution with the canonical hemodynamic response function under SPM5, comprised the general linear model design matrix together with the three translational and the three rotational motion correction time courses introduced as nuisance variables. An example alpha time course is displayed in panel e of Figure 1. Given the uncontrolled nature of the alpha paradigm and the potential for developing inefficient time courses, summary statistics across sessions, including the mean percentage of time that alpha activity is present and the mean alpha duration were calculated. These quantities provide the average temporal character of the alpha regressors observed.

Global scaling was imposed on the fMRI data of each session resulting in all images being normalized to the same average intensity over the brain volume. Normalizing global intensity was motivated by observed fluctuations between complementary brain states during relaxation that seemingly maintain a balance of positive and negative BOLD signal changes over time (Fox et al., 2005; Fransson, 2005). Typically, functional data acquired under more traditional task-related paradigms are not scaled globally. This is because in such data the relative changes attributable to the task may have altered the proportion of activations and deactivations. Therefore, the effect of global scaling on activation patterns for the spontaneous alpha paradigm was further assessed by comparison with repeated analyses without scaling.

Regression, voxel by voxel, using this design matrix resulted in a T-score map for each session for correlation of the BOLD signal to alpha state, accounting for the variance explained by residual motion effects. A composite T-score map for all subjects was then constructed by random effects analysis. After applying a threshold for statistical significance, activated voxels in the composite were localized by overlay on an anatomic reference image generated by averaging the individual structural images of all subjects.

RESULTS

Both activation and deactivation was observed in the composite maps as shown in selected axial slices in Figure 2. Deactivation refers to negative correlation of BOLD signal changes to alpha activity. Activation is displayed in orange at a P value threshold of 0.001, uncorrected. Correcting for multiple comparisons, a false discovery rate (Benjamini and Hochberg, 1995) <0.17 applies for all suprathreshold voxels. A cluster size threshold of 20 voxels is also imposed. Dorsal thalamus, both right and left, exhibits activation extending from anterior to posterior borders. A cluster is also present in medial cerebellum. In the cortex, a prominent activation is seen in the anterior cingulate accompanied by a small portion of the somatosensory association region. The sizes and locations of clusters activating with alpha are summarized in Table 1.

Blue voxels in Figure 2 represent deactivations at a threshold of $P < 0.001$, uncorrected, with those satisfying this threshold having a false discovery rate <0.08 . Only clusters comprised of 20 voxels or more are displayed. Deactivations arose in bilateral posterior cortical regions including fusiform gyri extending into visual association cortex. The only frontal area activating was a small cluster in the left dorsolateral prefrontal cortex. Table 2 summarizes the locations and sizes of deactivated clusters for this study.

The same activation/deactivation patterns arose qualitatively if no global scaling was used on the fMRI data. Quantitatively, however, voxels in activation clusters in the composite maps corresponding to the thalamus increased in T value whereas the T value of voxels of the deactivation clusters in the fusiform gyrus decreased. This outcome is illustrated in Figure 3, where a side-by-side comparison of two representative composite map slices is shown for scaled and nonscaled functional data. Note that similar patterns arise in the unscaled case, but only after imposing a more stringent T threshold on the thalamic activation and a less stringent T threshold on the fusiform deactivation.

The quality assessment using visual inspection detected artifacts, mostly associated with motion, in each of the 35 EEGs used in this study. Accordingly, 60% of the sessions were found to have artifacts in less than 5% of their interscan intervals. The remaining sessions varied in the artifact rate up to a maximum of 55% with 86% of all sessions maintaining a rate below 30%.

Summary characteristics of the alpha regressors generated by this study help ascertain the performance of spontaneous alpha activity in relation to traditional block task paradigms. The mean percentage of time alpha is present and the mean alpha duration for all sessions are plotted in Figure 4 as a function of threshold value above which alpha is considered to be present. Each regressor was first normalized to fall within the range [0, 1] before making the calculations. These curves reveal that at a threshold that results in alpha presence 60% of the time, the mean duration is about 10 seconds.

A more detailed visual appreciation of session-to-session variation and temporal variation in alpha time courses is provided in Figure 5. A grayscale representation of alpha regressor values, again normalized to fall in the range [0, 1], is shown for each interscan interval of each of the 31 sessions that lasted a full 20 minutes. As observed in earlier studies of alpha (Goncalves et

al., 2006; Laufs et al., 2006a), some variety is seen in the temporal evolution of alpha intensity from session to session.

The issue of intersubject EEG/fMRI data variability was investigated further by considering the mean T value of voxels comprising select regions of interest (ROI) implicated in this work and in the work of others (Goncalves et al., 2006; Laufs et al., 2006a). Mean T values were calculated for each session of this study for six ROIs including two functional ROIs as follows: (1) the portion of thalamus activated by group analysis, and (2) the portion of the fusiform gyrus deactivated by group analysis, and four anatomic ROIs as follows: (1) the thalamus, (2) the fusiform gyrus, (3) the superior parietal lobule, and (4) the middle frontal gyrus. The four anatomic ROIs were defined using the Wake Forest University Pickatlas toolbox (Maldjian et al., 2003) in SPM5. Figure 6 shows a histogram of the mean T values for all sessions for each of the ROIs. As expected, the mean of the mean T values for activated thalamus and deactivated fusiform are significantly positive and negative, respectively. The broader thalamic and fusiform ROIs have the same outcome, but with less significance. The superior parietal and middle frontal ROIs, however, fail to have mean values significantly deviating from zero at the 5% level. More importantly, none of the ROIs have a mean T distribution for which the hypothesis of normality can be rejected at the 5% level according to the Lilliefors test (Conover, 1999; Lilliefors, 1967). Recognizing that this is a broad ROI assessment, individual voxels in the superior parietal and middle frontal ROIs were tested for normality using the Lilliefors test. Only 1% and 7% of voxels rejected the hypothesis of normality at the 5% significance level for the superior parietal and middle frontal ROIs, respectively.

DISCUSSION AND CONCLUSIONS

This attempt to correlate brain function to the presence of alpha background rhythm in relaxed healthy adults builds on previous efforts by scanning at a higher magnetic field (4 Tesla) and by including a greater number of subjects. Increasing field strength improves sensitivity to the BOLD response but, at the same time, augments EEG artifacts including those induced by the gradient coils during image acquisition. This work demonstrates that artifact subtraction provides continuous EEG suitable for visual and digital analysis from a 4-Tesla environment. The attainable quality of the EEG is well represented by the data shown in Figure 1. EEG readings found evidence of artifacts in a small percentage of interscan intervals for a majority of sessions. Some sessions, however, contained a large number of intervals with artifacts by this subjective criterion. The nature and severity of these artifacts did not seem to preclude the detection of alpha activity by Fourier analysis. However, these results quantify the expected challenges of reading EEG time courses from a 4-Tesla environment clinically.

The alpha regressors resulting from windowed Fourier analysis were found to have temporal characteristics, illustrated in Figure 4, reminiscent of those of a typical block design paradigm if reasonable midrange threshold values are considered.

Consistency of alpha activity across sessions was assessed qualitatively. Inspection of Figure 5 suggests that alpha variations seem to be uniform over time and consistent between a large number of scanning sessions. Some sessions, however, display marked changes in temporal pattern. This behavior is not unlike that reported recently by other studies of spontaneous brain alpha activity (Goncalves et al., 2006; Laufs et al., 2003a).

The activation/deactivation patterns emerging from spontaneous fluctuations in occipital alpha rhythm during the relaxed state cannot be used alone to infer the functional role of alpha waves or the mechanisms of their generation. Nevertheless, the results can be discussed in the framework of current understanding of brain dynamics. The activation patterns observed support the view that the thalamus plays an active role in the generation of alpha waves and

the network dynamics of undirected thought. Increases in occipital alpha correlated with stronger synaptic activity in the thalamus whereas no BOLD change was detectable in primary visual cortex. In the thalamo-cortico-thalamic loop, thalamic relay axons project into cortical layer IV whereas projections in far greater numbers (Jones, 2002) return to the thalamus from pyramidal cells in layer VI. The BOLD effect is currently understood to derive principally from synaptic, or input, processes as opposed to spiking output (Logothetis et al., 2001). Cortico-thalamic synaptic action would therefore be expected to be the dominant source of BOLD increases during heightened alpha power. The anterior cingulate and somatosensory associative cortex increase input concomitantly with the thalamus. This is consistent with cortico-thalamo-cortical processes between layer V neurons of networked cortical areas through nuclei of the thalamus other than the lateral geniculate. Interestingly, the cerebellum also seems to increase synaptic activity, in line with the apparent modulatory role of this brain structure on cortico-thalamic function (Andre and Arrighi, 2003). Deactivations occur in areas receiving information processing inputs that increase BOLD signal while alpha decreases in the occipital region. As no visual sensory inputs are involved, the diminishing of occipital alpha implies a cortico-cortical interaction, passing through the lateral geniculate, to the visual association and fusiform gyri.

The typical periods of fluctuation of alpha observed in this study are similar to those recently reported for spontaneous fluctuations of BOLD intensity corresponding to the default state of the brain. There is a plausible parallel between the brain state variation defined by alpha waves and that defined by spontaneous correlated BOLD signals (Laufs et al., 2003b). With the waxing and waning of the alpha signal in the vicinity of the occipital lobes measured in this study, brain activity might be viewed as alternating between an introspective mode characterized by information processing in the anterior cingulate to a visual imagery mode suggesting a readiness for external stimuli represented by engagement of visual association cortex and fusiform gyri. The resting state networks emerging from this investigation have some elements in common with those identified by BOLD correlations. Particular parallels are seen with select networks, recently reported by researchers employing probabilistic independent component analysis of resting state fMRI data (De Luca et al., 2006) that included the thalamus.

This study found the thalamus BOLD signal to correlate bilaterally to occipital alpha in random effects group analysis. Previous research, using lower field strengths and comprised of fewer subjects, either failed to detect thalamic activation (Laufs et al., 2003a; Laufs et al., 2003b) or reported it at severely reduced statistical threshold (Moosmann et al., 2003). Supporting the benefit of increased field strength, thalamic activation associated with generalized spike-and-wave discharges has recently been shown to be detectable at 3 Tesla but not at 1.5 Tesla in a single subject undergoing simultaneous EEG and fMRI (Laufs et al., 2006b). However, extensive parietal and frontal deactivations observed by these same investigators did not appear here, possibly because of the reported inconsistency of individual deactivation patterns leading to suppression by random effects analysis.

Past work by other investigators (Goncalves et al., 2006; Laufs et al., 2006a) has focused on the variability of functional patterns related to alpha waves. Their reports suggest that there may be a segregation of functional patterns among individuals into two or more distinct categories including one defined by occipital deactivation and one defined by parietal/frontal deactivation. The ROI analysis summarized in Figure 6 attempted to find evidence of multimodal behavior among sessions in this study. There are individual sessions for each ROI that fail to show activation or indeed activate in a manner opposite to that observed in the group analysis. However, in contrast to the data from the literature (Goncalves et al., 2006; Laufs et al., 2006a), normality tests fail to suggest a multimodal distribution of activation patterns in any of the ROIs considered. The activated thalamus histogram, however, suggests a bimodal

distribution and comes close to rejection of the hypothesis of normality. The deviations of individual session activations from composite activation are consistent with being part of a normal distribution. The difference in findings between this study and other studies is likely related to the higher number of subjects enrolled in this study and the substantially higher number of analyzed EEG/fMRI sessions.

Simultaneous EEG/fMRI studies to date have used similar methods of data acquisition and analysis to find neural correlates of alpha waves. Nevertheless, subtle differences in methodology exist that cannot be ruled out for explaining discrepancies in outcome. The present study used alpha band fast Fourier transform magnitude whereas the others used power, for instance. Although most extracted the alpha time course with a windowed fast Fourier transform, others used wavelets (Moosmann et al., 2003). Scanning parameters for fMRI also differed among investigations. For example, voxel volumes ranged from about 33 mm³ (Goncalves et al., 2006) to 80 mm³ for the work described here. Disparities of this sort may well lead to differences in global sensitivity, but fail to explain why more thalamic activity is observed in this study whereas greater parietal activity is seen in others (Laufs et al., 2003a).

Another difference among studies that is of note is in the choice of electrode derivations from which to extract the alpha regressor. The present work used spatially restricted bipolar derivations comprised of O1 and O2 and their closest parietal neighbors. In contrast, other studies have either used O1 and O2 referenced to the zenith (Laufs et al., 2003a; Moosmann et al., 2003) or bipolar derivations that extended into temporal and central regions (Goldman et al., 2002; Goncalves et al., 2006). These broader derivations are plausibly more influenced by alpha-like rhythms, such as the mu rhythm (Niedermeyer, 1997) that emanate predominantly from central regions. As mu alpha is distinct in origin from occipital alpha, another possible explanation for the observed difference in fronto-parietal deactivation is thus provided.

Listening to music during the scanning sessions of this study might have introduced unintended attentional modulations. For instance, attentional variations between subjects might have been amplified contributing to inconsistency of BOLD response in frontal/parietal regions and the subsequent lack of group deactivation there. It should be observed, however, that at least one study (Nakamura et al., 1999) found that listening to music does not modulate alpha power significantly. Music listening was employed to relax the subject and to relieve the annoyance of lying still for an extended time with periodic gradient noise. It should be noted that despite the fact that subjects were listening to music, no auditory activation was detected in association with alpha state. Music was played consistently for the entirety of each scanning session, and thus it could also be argued that it provided a constant background similar to the scanner noise to which the subject could be vigilant at will. From this viewpoint, alpha activity would still be expected to wax and wane spontaneously with each subject's relaxed mental state.

Application of global normalization of fMRI images before general linear model analysis has been identified as a potential cause of inferential artifact (Aguirre et al., 1998). It is used here, however, under the premise that in the relaxed state with no directed task a homeostasis of mental effort might be established. This is suggested qualitatively by the tendency of the resting state to be characterized by balanced, opposing, fluctuations in emphasis between two or more default networks (Fox et al., 2005; Fransson, 2005). This forms the basis of a conjecture that the average BOLD intensity across the brain unchallenged by a specific task remains constant. It should be noted that without global scaling the same activation/deactivations patterns arise, but with more emphasis concentrated on the thalamic activations (Fig. 3).

This report extends application of the modality of concurrent multichannel EEG/fMRI to the magnetic field strength of 4 Tesla to study correlates of brain activity to alpha waves. As research and clinical scanners continue to evolve to higher field strength and as EEG technology

geared to the magnetic resonance environment improves (Vasios et al., 2006), this technique promises to remain viable. The greater BOLD sensitivity afforded by higher field strength will have an important impact on clinically relevant uses of this method with more challenging spontaneous paradigms, such as epileptic spiking.

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FIGURE 1.

Processing of EEG data to extract an alpha activity regressor for general linear model analysis.

a, A segment comprising three interscan intervals of raw EEG data taken simultaneously with fMRI at a rate of 10 kHz. **b,** The same EEG segment after 60 Hz low-pass filter, elimination of ballistocardiographic and gradient artifacts, decimation to 200 Hz, and bipolar derivation. **c,** The four bipolar EEG channels that include an occipital channel. The beginning of each interscan interval is marked by an asterisk (*). **d,** Fourier transforms of the interscan intervals in panel (c) with the alpha band from 8 to 13 Hz indicated. **e,** The alpha activity regressor spanning the entire session calculated as the mean alpha band magnitude for each interscan

interval averaged across the four occipital bipolar channels. Convolution with the canonical hemodynamic function was also applied in SPM5.

FIGURE 2.

Composite random effects activation/deactivation patterns for the presence of occipital alpha waves. The orange and blue colors indicate activations and deactivations, respectively. For activations, $P < 0.001$, uncorrected, and for all activated voxels, false discovery rate < 0.17 . Deactivations shown correspond to $P < 0.001$, uncorrected, and false discovery rate < 0.08 . A cluster size threshold of 20 voxels is imposed. Images are in radiologic convention (viewing the head from below). Z coordinate in Montreal Neurological Institute space is indicated in the upper left hand corner of each slice.

FIGURE 3.

Comparison of activation and deactivation patterns in two representative slices with and without global scaling. Results with global scaling are shown on the left using positive/negative T thresholds of 3.35/−3.35. The images on the right show the outcome without global scaling using a positive T threshold of 3.78 and a negative T threshold of −2.44.

FIGURE 4.

Average, across sessions, of the mean alpha duration and the percent of time alpha is present versus threshold for alpha regressors normalized to the range [0, 1]. Error bars indicate the standard deviations.

FIGURE 5.

Grayscale encoded intensities of normalized alpha regressors for each interscan interval of each of the 31 scanning sessions with a duration of 20 minutes.

FIGURE 6.

Histograms of session mean T values in select functional and anatomic ROIs. Mean, standard deviation (SD), and *P* value for the rejection of the hypothesis of zero mean are shown for each ROI. The Lilliefors test failed to reject the hypothesis of normality at the 5% level for any of the ROI histograms shown. Functional ROIs: (a) activated thalamus, (b) deactivated fusiform gyrus. Anatomic ROIs: (c) thalamus, (d) fusiform gyrus, (e) superior parietal lobule, and (f) middle frontal gyrus.

TABLE 1

Clusters Activating With Occipital Alpha Activity

Anatomic Region	Brodmann Area(s)	Cluster Size (Voxels)
Thalamus, left	—	133
Thalamus, right	—	144
Cerebellum	—	148
Anterior cingulate	24, 32	279
Somatosensory association	7	73

TABLE 2

Clusters Deactivating With Occipital Alpha Activity

Anatomic Region(s)	Brodmann Area(s)	Cluster Size (Voxels)
Fusiform and visual association, left	19, 37	418
Fusiform, parahippocampal and visual association, right	19, 36, 37	514
Visual association, right	19	241
Fusiform and middle temporal, left	21, 37	144
Dorsolateral prefrontal, left	46	41