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Review

Recent advances in recording electrophysiological data simultaneously with magnetic resonance imaging

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Simultaneous recording of brain activity by different neurophysiological modalities can yield insights that reach beyond those obtained by each technique individually, even when compared to those from the post-hoc integration of results from each technique recorded sequentially. Success in the endeavour of real-time multimodal experiments requires special hardware and software as well as purpose-tailored experimental design and analysis strategies.

Here, we review the key methodological issues in recording electrophysiological data in humans simultaneously with magnetic resonance imaging (MRI), focusing on recent technical and analytical advances in the field. Examples are derived from simultaneous electroencephalography (EEG) and electromyography (EMG) during functional MRI in cognitive and systems neuroscience as well as in clinical neurology, in particular in epilepsy and movement disorders. We conclude with an outlook on current and future efforts to achieve true integration of electrical and haemodynamic measures of neuronal activity using data fusion models.

Keywords: EEG; EMG; fMRI; Fusion; Multimodal imaging; Electrophysiology; Human; Magnetic resonance imaging

Contents

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Introduction

The emergence of a young research domain

Recording electrophysiological data simultaneously with functional MRI (fMRI) has rapidly progressed technically as witnessed by the number of related publications with related reviews of this field already available (Salek-Haddadi et al., 2003; Gotman et al., 2006; Ritter and Villringer, 2006; Herrmann and Debener, 2007). The present review will limit itself to human studies using simultaneous EEG/EMG/fMRI and focus on selected methodological references that highlight the key principles in data acquisition and analysis.

The development of EEG recording during fMRI was motivated by the clinical interest in mapping changes in neural activity associated with epileptic discharges observed on surface EEG onto images of brain anatomy (Ives et al., 1993). At first glance, this may appear an indirect approach to this clinical question where procedures to localize electrical sources of EEG activity, possibly in conjunction with constraints derived from individual anatomy, should provide a more straightforward solution. Yet, while the temporal resolution of EEG is far more adequate for tracking dysfunctional activity embedded into physiological brain processes, the precision of fMRI in localizing

with confidence the spatial topography of neural processes is undeniably superior to that of EEG (Ives et al., 1993; Grova et al., 2008). This drove epilepsy researchers to "marry the blind (EEG) and the lame (fMRI)". EEG is "blind" with respect to the localization of sources, a handicap that arises from the so-called inverse problem and transpires into the accuracy of spatial localization and resolution. And fMRI is "lame" in that the hemodynamic signal changes that it captures are delayed and temporally dispersed with respect to the underlying neural events and hence compromised with respect to temporal resolution (Horwitz and Poeppel, 2002). While these two respective handicaps motivate the interest in combining the two modalities the actual success of this combination required methodological milestones including both MRI compatible EEG acquisition hardware and artifact reduction algorithms (Lemieux et al., 1997; Allen et al., 1998). Subsequent reports established the feasibility of related studies on physiological human brain function, mainly of event-related potentials (ERP), first interleaved with fMRI (Bonmassar et al., 1999; Kruggel et al., 2000) and later – once artifact correction algorithms ensured the necessary EEG quality (Warbrick and Bagshaw, 2007) – during image acquisition (Becker et al., 2005; Comi et al., 2005; Henning et al., 2005). From the perspective of experimental design, both spontaneous interictal EEG spikes and stimulus-driven evoked potentials fall into the previously already well-established framework of event-related fMRI studies. In another more recent field of application, however, this methodology was extended to the study of ongoing EEG activity, thus requiring continuous, high quality data across all frequency bands of interest reaching from theta (Mantini et al., 2007a,b; Sammer et al., 2007; Scheeringa et al., 2007), over alpha (Goldman et al., 2001; Laufs et al., 2003a,b; Moosmann et al., 2003; Feige et al., 2005; Laufs et al., 2006a,b,c; Mantini et al., 2007a,b) and beta (Laufs et al., 2003a,b; Mantini et al., 2007a,b) up to low gamma (in the cited publication about auditory activity this relates to up to 40 Hz) activity (Giraud et al., 2007) rather than good signal-to-noise ratio allowing the identification of individual discrete events like interictal discharges (Allen et al., 1998). Currently, brain activity patterns associated with ongoing electrical brain activity have been studied from the awake resting state, all the way to deep sleep (Goldman et al., 2001; Laufs et al., 2003a,b; Moosmann et al., 2003; Kaufmann et al., 2006; Laufs et al., 2006a,b,c; Horovitz et al., 2007; Laufs et al., 2007; Schabus et al., 2007). Also, the recent interest in resting state brain activity has stimulated multimodal EEG/fMRI studies since the additional information provided by the EEG allows further interpretation of the resting state fMRI data while not affecting the resting state in a way any experimental manipulation would do. For example, the so-called 'default mode' (Raichle et al., 2001) set of brain areas could be connected with EEG activity in different frequency bands (Laufs et al., 2003a,b; Mantini et al., 2007a,b), was shown do be dynamically active across different vigilance states (Laufs et al., 2003a,b; Horovitz et al., 2007; Laufs et al., 2007) and suspension of 'default mode' brain activity was suggested to occur during interictal focal and generalized epileptic activity (Gotman et al., 2005; Hamandi et al., 2006; Kobayashi et al., 2006; Laufs et al., 2006a,b,c; De Tiege et al., 2007; Hamandi et al., 2008).

Recording of other biologically informative signals in combination with fMRI (with or without EEG) has been of interest since the early days of fMRI (Ives et al., 1993; Hill et al., 1995; Kleinschmidt et al., 1996; Warach et al., 1996; Lemieux et al., 1997; Seeck et al., 1998) but seen wider progress more recently. Two examples of such signals are the measurement of skin conductance with respect to autonomous function (Patterson et al., 2002; Nagai et al., 2004a,b) and electromyography (EMG) with respect to motor behaviour (van Duinen et al., 2005; Richardson et al., 2006; Post et al., 2007). Due to this body of experience and published work, much of the required methodological development is broadly available and sufficiently advanced to facilitate a broad range of applications. Meanwhile, both hardware and software have become more reliable, widely used, and commercially available, thus reducing the time and expertise required to conduct these studies.

Applications for multimodal imaging

Mapping epileptic zones formed a clinically relevant early objective of EEG/fMRI (Gotman et al., 2006; Laufs and Duncan, 2007). Yet, this example contains two more generic aspects that have been a driving force in the further evolution of simultaneous multi-modal approaches joining electrophysiology and fMRI.

The first aspect relates to the fact that interictal epileptiform activity (IEA) is in general unpredictable, not reliably induced by external stimuli, and relatively short-lived. Hence, fMRI can only localize the haemodynamic correlates of these spontaneous events if they are simultaneously recorded (and detectable) on surface EEG. In other words, the simultaneous electrophysiological recording is necessary to generate the information required to interrogate the fMRI data set in a sensitive, hypothesis-driven way. If the EEG information of interest corresponds to prolonged states and is relatively stable, EEG can be combined with functional imaging methods with slower sampling, e.g. positron emission tomography, as has been performed successfully in sleep studies (Maquet, 2000). Yet, in many instances beyond epilepsy, the electrophysiological effect of interest occurs only transiently at fairly unpredictable timepoints. This extends for example to pathologically enhanced EEG–EMG coherence in the study of movement disorders (Richardson et al., 2006). Such scenarios require the temporal resolution of fMRI in conjunction with a technique that can simultaneously record the biological effect of interest and thus inform the analysis of the fMRI time series data. Of note, the intrinsic sluggishness of haemodynamic signals recorded in fMRI means that the result of such an analysis will not isolate the 'pure' correlate of the electrophysiological signal but also its immediate origin and consequences. Again, this can be illustrated by an example from epilepsy research where EEG/fMRI can identify not only the (normally cortical) sources that generate discharges but also the brain regions affected by interictal EEG activity (Laufs and Duncan, 2007). This approach therefore allows the delineation of 'networks' of IEA as well as secondary functional consequences in brain regions which are not primarily expressing, but are modulated by, the occurrence of epileptic activity elsewhere (Gotman et al., 2005; Laufs et al., 2006a,b,c). Such findings are an accomplishment that neither surface EEG nor fMRI could reach individually.

Horovitz et al. introduced the idea of correlating the amplitude of event-related potentials with fMRI data acquired in a separate session (Horovitz et al., 2004). Simultaneous recordings allowed the extension of this idea to the study of event-related single-trial EEG signals (Debener et al., 2005; Eichele et al., 2005; Debener et al., 2006; Benar et al., 2007). One might think that when neural responses are elicited from carefully controlled external stimuli, fMRI and EEG results might as well be obtained in separate

sessions (Vitacco et al., 2002; Bledowski et al., 2004; Bledowski et al., 2006), in contrast to intrinsically generated epileptic discharges (Bledowski et al., 2004; Bledowski et al., 2006). The benefit from separate modality acquisition is that it avoids one modality's experimental set-up compromising the other. However, the down-side is that off-line multimodal correlation can only address that part of stimulus-related brain function which is reproducible. For example, identical sensory stimulation cannot be achieved and subjective experience and behaviour of the subject cannot be expected (Debener et al., 2006). Observations of trial-by-trial variability suggest a major contribution from non-reproducible effects to single trial responses. This variability can be related to state fluctuations (attention, mood, motivation) and can drift due to time-dependent effects for reasons ranging from fatigue to learning. If such effects are of interest, even if just to control their impact, it appears useful to accept the limitations and constraints from truly multimodal recording for the sake of obtaining super-additive information from real-time trial-by-trial correlation across the modalities applied.

Methodological aspects

While the previous section introduced some theoretical considerations, the following sections will cover practical aspects when planning and conducting a simultaneous multimodal experiment. Firstly, the choice of hardware to provide patient safety and comfort, while delivering high quality EEG and fMRI data is discussed. Secondly, we examine the choice of post-processing methods applied to the electrophysiological data for scanner- and subject-induced artifact reduction.

Hardware

The signal transduction chain of the electrophysiological signal of interest (e.g. EEG, EMG, skin impedance) starts at the subject's surface where electrodes make skin contact with the aid of a conductive gel or paste (Fig. 1). The currents generated by synchronously active and parallel oriented pyramidal neurons will cause a potential between EEG electrodes which then generate current flow detected by the amplifier which is digitised and recorded. The signal is relayed between the electrode and amplifier through wires. Either, these [metallic] wires reach from inside the scanner bore to the outside of the electro-magnetically shielded scanner room, in which case, conventional EEG amplification and digitization hardware can be used (provided a sufficient amplitude recording range and sampling rate can be obtained). Or, preferably, the signal is amplified and digitized within or near the scanner bore before leaving the scanner room through optical fibres (Allen et al., 2000). This has the advantages of both increased signal fidelity and patient safety. An interesting alternative is the use of the MR receiver hardware to transmit the EEG signals via the MR scanner receiver coil encoded alongside the MR signals (Van Audekerkea et al., 2000; Hanson et al., 2006).

The induced artifact in the EEG is due to a complex combination of factors including the field strength (and so frequency), orientation, positioning of the recording equipment relative to the RF coil, and the geometric relationship between the magnetic field gradients relative to the electrophysiological equipment. When measuring limb EMG, for example, increasing distance between the recording locations and the magnet isocentre does not necessarily translate into reduced artifact (despite decreasing field strength) because the field homogeneity decreases and hence motion will cause greater artifact than in the homogenous field. Generally, artifact will increase with the distance relative to the gradient direction and within the linear part of the gradients be determined significantly by the distance between measurement and reference electrode.

Subject safety issues pertain to current flow and heating within the body that is normally greatest close to the electrodes. The time-varying (switching) magnetic field gradients can induce voltages in electrodes and leads. Where the subject provides significant impedance within this circuit, current will flow within tissue which in turn could potentially cause stimulation, electric shock and tissue damage. Similarly, movement of an electric circuit (loop) in the static magnetic field will cause current flow and could cause injury via the same mechanisms (Lemieux et al., 1997).

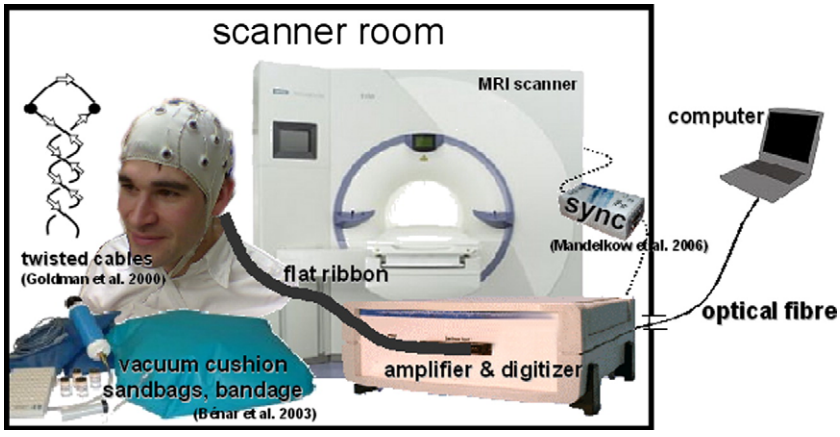

Fig. 1. Schematic of an EEG/fMRI experimental setup. EEG ring electrodes with current limiting safety resistors are woven into a cap. Their bundled wires converge into a flat ribbon cable which connects to the battery-driven amplifier and digitizer, which is usually positioned at the head end of the scanner bore (a second amplifier and digitizer, e.g. for EMG recordings, may be positioned near the subject's lower extremities). The digitizer is connected to the MRI scanner clock via a synchronization device (frequency divider). A fibre optical cable transmits the digitized electrophysiological signals through the wave guide to a recording computer outside the scanner room. Vacuum cushions serve subject comfort and can reduce subject motion. Twisting of wires has also been proposed and may be useful in bipolar recordings.

Especially at higher field strengths, the MR sequence (and coil) used in the presence of the multimodal recording equipment should not lead to excess energy deposition (e.g. specific absorption rate, Angelone et al., 2004; Angelone et al., 2006).

The primary safety risk is due to heating arising from the interaction of the radio frequency (RF) fields used for MRI signal excitation with the electrophysiology recording equipment. It should be noted that no direct connections need to be present at RF frequencies for low impedance loops to be formed that will have current induced within them due to the RF fields. Maximum heating will occur when a conductor is resonant at the frequency of the RF field. It is important to realise that a single wire can be resonant (effectively acting as an RF antenna) and cause dangerous heating in nearby tissue, particularly at the ends of the wire where the electric field is normally concentrated (Achenbach et al., 1997; Dempsey et al., 2001; Pictet et al., 2002). Resonant lengths can vary between tens of centimetres and several metres depending on a number of factors including scanner frequency (i.e. field strength), wire environment, shape and position. From this it follows that careful choice and testing of leads and electrodes used within an MRI scanner is necessary and inductance should be reduced by minimising the length of wires and avoiding loops (Ives et al., 1993; Lemieux et al., 1997; Goldman et al., 2000; Dempsey et al., 2001; Lazeyras et al., 2001). Empirical evidence (Baumann and Noll, 1999) and theoretical considerations suggest that it is best to guide wires in close proximity to the axis around which the gradient switching occurs, i.e. the *z*-axis of the scanner. Such a geometry minimises the angle between the changing magnetic field and the electrical conductor — and at the same time avoids loop formation (Lazeyras et al., 2001). These advantages outweigh the effect of the electrical field parallel to the *z*-axis as long as the field decays quickly outside the (head) coil. In addition, current limiting resistance will be of protective benefit and can be implemented either by putting resistors close to the electrodes or distributed within the leads (Lemieux et al., 1997; Dempsey et al., 2001; Vasios et al., 2006).

Both reduced (non-optical) lead length and increased lead impedance limit the induced amplitude of the artifact in the recorded EEG. While these procedures reduce the required input range of the amplifier, they also correspondingly reduce the signal. Electrode caps help to keep wires in an optimized predefined position (Baumann and Noll, 1999), without loops and direct electrical contact yet bundled together. Twisting of all wires together has been proposed with the idea that induced fields cancel each other out (Goldman et al., 2000), but to work this assumes very similar resistances of the conductors. Even if achieved in practice, any remaining voltage difference would still be amplified. Generally, cables should be fixed to protect them against motion, such as gradient switching-generated vibrations (Thees et al., 2003), by means of sandbags (dampening effect), tape or bandage (Benar et al., 2003).

Materials should be non-ferrous (wires are mostly copper or carbon), and all equipment introduced into the shielded MRI room must not emit RF in the scanner frequency band (Ives et al., 1993) such that scanner functionality, image quality and subject safety are not compromised (Angelone et al., 2004; Angelone et al., 2006). Obviously, the electrophysiology recording equipment needs to remain operational within the MR scanner environment and during scanner operation (Ives et al., 1993). A balance must be struck between tolerable artifact on the images and practicality of the materials used. In that respect, for example, gold electrodes have been preferred over carbon electrodes (Krakow et al., 2000). Sintered Ag/AgCl ring "floating" electrodes are also widely used and include a surface mounted safety resistor. These electrodes i) do not directly touch the skin, ii) have good artifact characteristics, and iii) provide ease of use.

The amount of conductive agent used should be minimised, and it should be tested for related image artifacts, especially within the brain (Krakow et al., 2000; Bonmassar et al., 2001). Conversely, signal alterations confined to the electrode positions themselves may in fact be used for their localization. Finally, the entire ensemble should be tested together, as the MRI 'signal to noise ratio' (SNR) will be a function of 'radio frequency (RF) coil loading' that is increased with the amount of conductive material introduced into the RF scanner coil: in materials of high electrical conductivity RF (involved in excitation and detection of the MR signal) generates large surface current densities which act to screen the RF field from the interior of the material and hence compromise image quality. These currents also disturb the B1-field within regions in close proximity to the conductor, and finally, due to RF field-conductor interaction, the RF coil resistance increases further reducing SNR. Specifically, shielding-effects of multi electrode set-ups (Scarff et al., 2004) and altered B_0 and/or B_1 field homogeneity including that caused by EOG and ECG leads can manifest in the human head (via flip angle reduction) and thus may reduce the SNR of the images in areas of interest (Mullinger et al., 2007).

Directing special effort at subject comfort is warranted for increasing tolerance of the subject and thus also limiting head motion. Using a vacuum head cushion (Benar et al., 2003) has been found to minimise both motion-induced artifacts on the images as well as motion-induced currents contaminating the electrophysiological signal. This is especially important for patient studies in general and when recording EMG which is highly motion sensitive (Salek-Haddadi et al., 2003; Hamandi et al., 2004). The use of sedative agents to suppress motion needs careful consideration as 'neuroactive' substances can alter net synaptic activity in a region-specific manner and thus fMRI signal intensity (Bloom et al., 1999; Kleinschmidt et al., 1999; Iannetti and Wise, 2007). Depending on the study design, the administration of such substances may confound the results such that observations can be falsely attributed to the effect of interest while they may in fact be to major parts caused by the pharmacologic agent (Ricci et al., 2004; Iannetti and Wise, 2007). Under certain circumstances sedation cannot always be avoided, e.g. when studying very young children with fMRI (Jacobs et al., 2007), but valuable patient data sets acquired without sedation can often be recovered if motion effects are modelled sufficiently at the analysis stage (Lemieux et al., 2007).

EMG recordings during fMRI (Fig. 2) are particularly affected by artifact induced by motion in the static field because even during isometric contractions (i.e. muscle contraction without gross limb movement) some degree of electrode movement in the field is inevitable. Moreover, this artifact will tend to be grossly task-correlated while still irregular and thus difficult to model (van Duinen et al., 2005; Richardson et al., 2006; Post et al., 2007). In these cases, true bipolar recordings are advantageous as artifact common to closely positioned electrodes is already reduced prior to correction (Goldman et al., 2000; Richardson et al., 2006). If required for polygraphic measurements, other physiological data can be recorded such as respiration and pulse oximetry in addition to the various electrophysiological measurements (Laufs et al., 2007). Respective pneumatic and optic devices are provided by

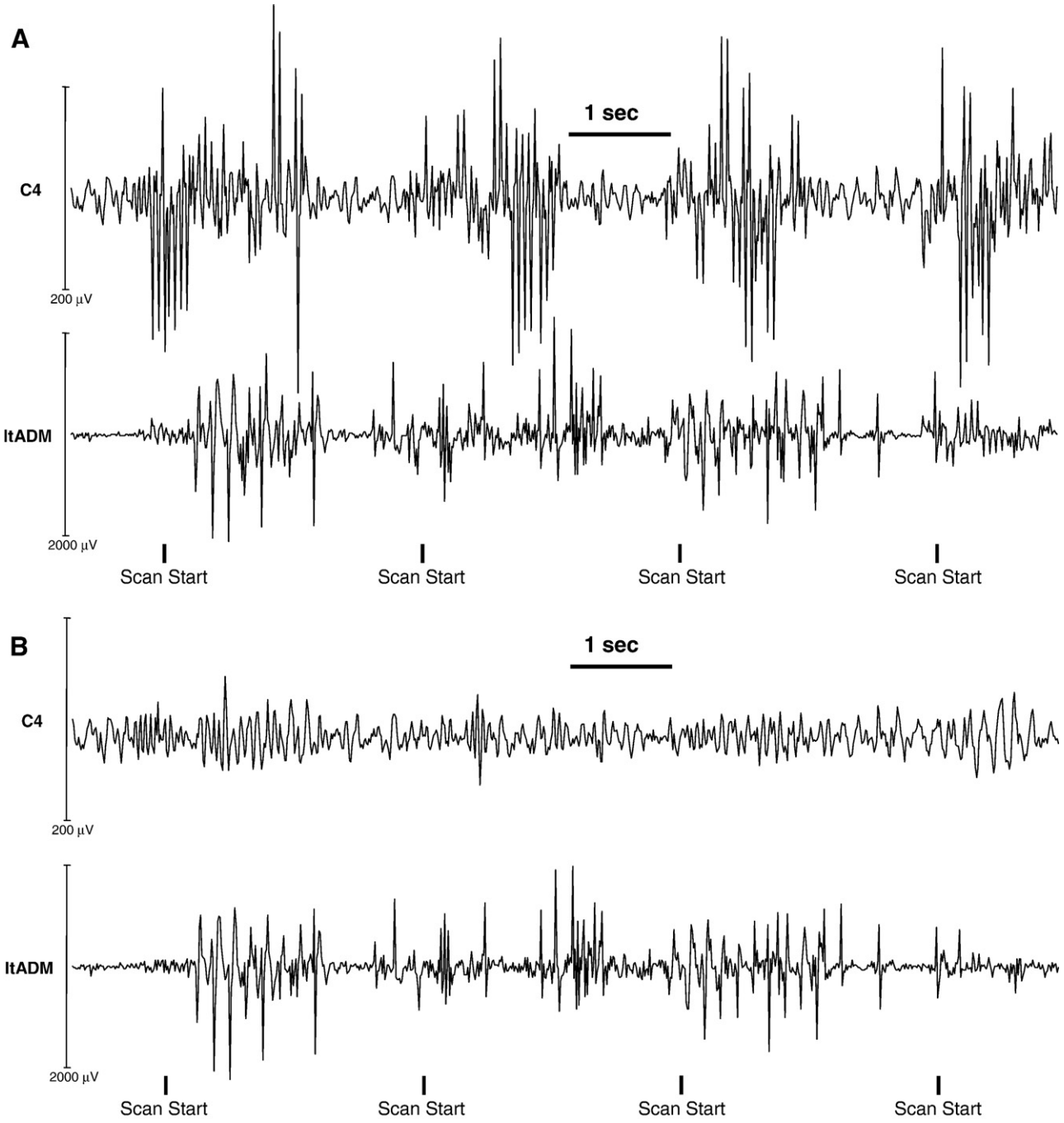

Fig. 2. Sample of EEG and EMG data acquired simultaneously with fMRI at 3T (A) before and (B) after MRI artifact subtraction. Both EEG and EMG data were recorded using surface ring electrodes including a 5 kΩ safety resistor. Data are shown from a right central electrode position (EEG, C4 referenced to FCz, 10–20 system) and the left abductor digiti minimi (ltADM, bipolar EMG, muscle belly vs. phalangeal epiphysis, twisted wires). Twenty-five coronal slices (acquisition start of the first slice indicated by the vertical Scan Start markers) were acquired covering the motor cortex, cerebellum and basal ganglia using an echo planar imaging sequence (TR = 1500 ms + 1100 ms gap, TE = 30 ms, 3.75 × 3.75 × 4 mm^3 voxels). A 50-year-old male with cortical myoclonus was studied; in this example data set, both EEG and EMG quality were sufficient for EEG–EMG and EMG–EMG coherence analyses and back-averaging to be carried out (not shown). Data were acquired by M. Richardson and H. Laufs at the National Society for Epilepsy in Chalfont, St. Peter, Buckinghamshire, UK in collaboration with Peter Brown, ION, University College London, UK. Note that the artifact on the bipolar EMG channel is small in comparison to the EMG signal even before (A) artifact subtraction, which yet needs improvement.

most scanner manufacturers and thus do not require special consideration of MR-compatibility.

Raw data quality remains essential despite sophisticated gradient and pulse artifact reduction algorithms. The generic set-up outlined above thus needs to be adapted to and optimized for every scanner, electrophysiological recording equipment and site. One should also consider switching off the scanner cooling pump and AC power sockets in the room to avoid these additional artifact

sources. Finally, synchronization of EEG sampling with the MR sequence vastly improves the effectiveness of MRI artifact reduction methods (Mandelkow et al., 2006). For their correction to work gradient artifacts must not exceed the amplitude range of the amplifier, the latter additionally requiring suitable signal-to-noise recording characteristics (see below). Special care should be taken during electrode preparation since relatively high skin-electrode impedances, which can still yield good data quality when the MR scanner is not running, will become detrimental to signal quality once scanning is underway.

MR-compatible EEG amplifiers should allow sampling of the electrophysiological signal including the gradient artifact at a high temporal rate and within a large amplitude range. The temporal resolution – unless perfect synchronization is warranted between the scanner and the recording equipment (Anami et al., 2003; Mandelkow et al., 2006) – is required because of the high slew rates of MRI sequences, and a large amplitude input range in order to avoid clipping of the signal and allow artifact reduction (see below). Widely used amplifiers permit MR-synchronized recording of 128 or more data channels at 5000 Hz with a dynamic amplitude range of ±3.2 mV to ±325 mV and respective resolution (16-bit sampling); noise characteristics <1 uVpp, 125 dB common mode rejection, switchable 10 MΩ/10 GΩ input impedance. With such an amplifier, conventional echo planar imaging sequences for blood oxygen level-dependent (BOLD) contrast and arterial spin labelling (ASL) have been successfully applied at up to 3 T (Hamandi et al., 2008). No human EEG/fMRI studies have yet been published for higher field strengths, but safety evaluation and experiments carried out in non-human primates suggest that respective studies in humans may follow in due course (Angelone et al., 2004; Angelone et al., 2006; Schmid et al., 2006; Vasios et al., 2006).

Artifact reduction algorithms

Understanding how artifacts arise is the key to designing artifact reduction algorithms. Three types of artifacts in electrophysiological recordings originate specifically from the MR scanner. All these unavoidable artifacts manifest themselves as induced voltages that add linearly to the EEG signal and thus threaten to obscure the biological signal of interest (Fig. 3). The three artifact types arise from: 1) MRI scanning ('imaging artifact'): This is usually the largest in amplitude (in the order of mV) but the most stable over time (Allen et al., 2000). Its origin

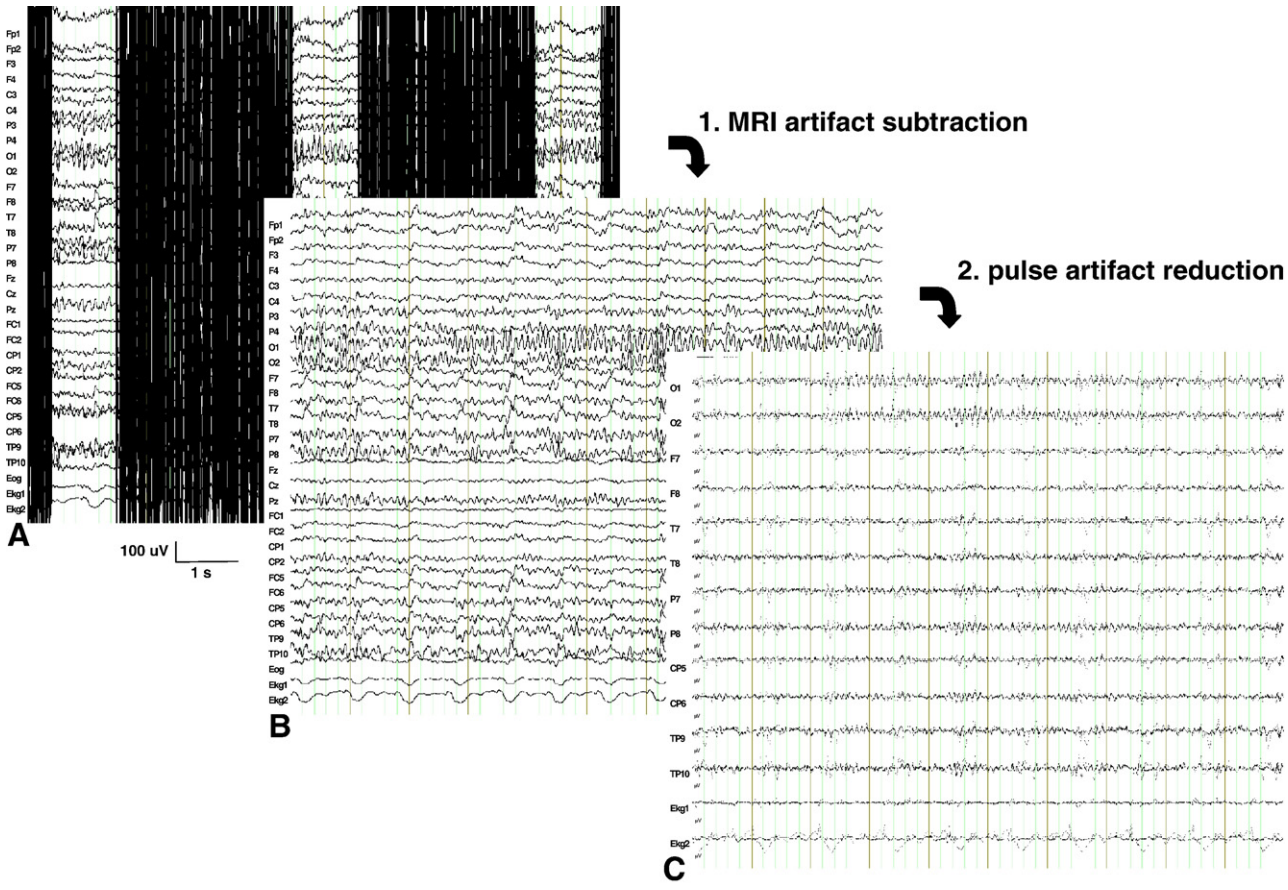

Fig. 3. Schematic of preprocessing stages of MRI and pulse artifact affected EEG. (A) Segment (10 s) of a 32 channel electrophysiological recording during "interleaved" fMRI acquisition at 1.5 T with about 3 s of imaging per acquired volume followed by a gap in scanning of about 1 s duration; (B) the same segment after channel-wise subtraction of a template MRI artifact obtained by averaging; (C) identical segment (dotted lines) after channel-wise pulse artifact reduction (solid line) via subtraction of an ECG-locked sliding average. Note "contribution" of the pulse artifact to the 12 Hz sinusoidal alpha oscillations in channels O1 and O2. These may be missed at the stage of panel B, i.e. before pulse artifact reduction. Electrode labels according to the international 10–10 system, reference at FCz; EKG = Electrokardiogramme.

has already been discussed above: the time varying electromagnetic fields induce currents resulting in artificial voltages in the recorded electrophysiological data; 2) cardiac pulsation ('pulse artifact') (Allen et al., 1998): This is thought to be due to heart beat-related movements (systolic pulsation) of the head or of electrodes adjacent to blood vessels, or of the blood itself caused by systolic acceleration and abrupt diastolic directional change of blood flow in large body vessels and – arguably (Nakamura et al., 2006) – due to fluctuations of the Hall-voltage due to the pulsatile arterial blood flow (Ellingson et al., 2004); 3) the amplitude and topography of the previous artifact types are affected, and the constant nature – which is the crucial basis for most artifact subtraction strategies – of 1) is compromised by subject motion, any change in position of the metallic recording components in the static field (Hill et al., 1995), drift of the electrode impedances and of the MR scanner magnetic field gradients that change by a small amount over time predominantly due to gradient heating.

The scanner-generated imaging artifact is theoretically the easiest one to remove owing to its periodicity. All currently available artifact subtraction methods exploit this regularity to varying degrees. However, since the regularity is not perfect, neither are the correction algorithms. Due to the scanner artifact's huge amplitude compared to the biological EEG signal (about a factor of 1000 for a standard set-up), even slight imperfections of the artifact correction leave EEG activity hard to visualise. In the absence of the perfect algorithm, depending on the purpose of the study, different approaches may be more or less suitable than others.

The principle of the first MRI scanner artifact reduction method was based on determining a template artifact waveform by time-locked averaging time-locked to the periodic MR-acquisition (Sijbers et al., 1999; Allen et al., 2000). This procedure is based on the rationale that those components of the recorded signal, which are not time-locked to image acquisition, should average to zero. Because of the additive property of the theoretically constant imaging artifact, averaging results in a template which can be subtracted from the data and thus recover the biological signal (and noise). Artifact drifts can be partly addressed by sliding average formation and subsequent linear filtering and, theoretically, adaptive noise cancellation (Allen et al., 2000; Wan et al., 2006).

These methods cannot entirely make up for asynchrony between the MR sequence and electrophysiology data sampling: despite EEG sampling rates of several kHz, MR slew rates at the order of several hundred T/m/s and gradient strengths of several dozen mT/m will result in very subtle temporal jitter and in turn compromise template accuracy. Digital up-sampling by interpolation of the recorded data and subsequent re-alignment of the segments before averaging (Allen et al., 2000), or grouping of segments to form several average 'families' based on correlation criteria (BrainVision Analyzer, Brainproducts, Munich, Germany) further improve correction quality — and can be performed online. But ideally EEG sampling should be *a priori* time-locked to the MR scanner and the TR an exact multiple of the sampling interval (Mandelkow et al., 2006).

A fixed temporal relation between EEG and MRI sampling is also a prerequisite for the 'stepping stone' technique, the idea of which is to avoid sampling EEG during periods of magnetic field gradient switching in the MRI pulse sequence but constrain sampling to periods without gradient switching where no related artifact is induced (Anami et al., 2003). However, this criterion imposes a constraint on the MRI sequences that can be used. Nonetheless further subsequent artifact correction is required, and continuous EEG is not obtained (Anami et al., 2003). Other approaches to imaging artifact correction have been suggested that also rely on the (*a priori* knowledge of the) specific sequence-related artifact shape (Hoffmann et al., 2000; Garreffa et al., 2003; Wan et al., 2006), its determination using principle component analysis (Negishi et al., 2004; Niazy et al., 2005) and subsequent respective artifact fitting and filtering steps. Combining different methods can prove very efficient (Niazy et al., 2005) however the correction of artifacts in EMG signals currently remains challenging (van Duinen et al., 2005; Richardson et al., 2006; Post et al., 2007), and algorithms will have to be developed accounting for artifact as a function of both electromagnetic field changes and simultaneous relative subject (electrode) movement therein.

The pulse artifact often requires more attention than the imaging artifact: it can be very subtle with an amplitude in the range of the biological signals (Allen et al., 1998). Non-invasive manipulation of this artifact for its exploration is difficult, but studying it at different field strengths demonstrated that the pulse artifact adds a spatio-temporally complex, non-stationary signal to the EEG (Debener et al., 2007a,b). Depending on the planned analysis, reducing the pulse artifact may not be required at all – despite its contribution to a broad frequency range –, for example, where discrete features such as IEA need to – and can readily – be identified on the EEG standing out clearly from the background (Benar et al., 2003). However, automated IEA detection algorithms may be compromised (Siniatchkin et al., 2007), and frequency analysis can be impaired by pulse artifact (Laufs et al., 2003a,b).

Methods for pulse artifact subtraction very much resemble those discussed above for the imaging artifact: due to its periodic nature, the average subtraction approach can be applied (Allen et al., 1998). However, the periodicity of this biological artifact is subject to heart rate variability and drift artifacts, leading to greater instability of the pulse artifact compared to the imaging artifact. This is the reason why a sliding average approach with or without additional weighting is beneficial (Allen et al., 1998; Sijbers et al., 1999; Goldman et al., 2000; Ellingson et al., 2004) or the use of several artifact templates per channel (Niazy et al., 2005). Again, similar approaches to the average artifact subtraction have been suggested, for example measuring pulsatile motion (and not the ECG itself) directly with a piezoelectric transducer before regressing it out (Bonmassar et al., 2002; Ellingson et al., 2004) or adding an additional, wavelet-based de-noising step after the average template subtraction (Kim et al., 2004).

All averaging methods critically rely on the exact detection of corresponding instances of the cardiac cycle, such that averaging results in an accurate template (Negishi et al., 2004). Here, sophisticated QRS or "R-peak" detection methods (Meyer et al., 2006) developed for automatic ECG processing are of use, but those relying on the typical morphology of the ECG waveform may fail because of the distorted ECG shape inside a strong magnetic field. Due to physiologically (Snisarenko, 1978) and psychologically induced (Michal et al., 2007) changes in heart rate, the length of the template to subtract will vary and should fit the shortest beat-to-beat period of the correction epoch. The amplitude of the artifact reflects the strength of the pulse wave, and the template should thus be centred such that it encompasses the part of the cardiac cycle most strongly affecting the electrophysiological data. The delay between the detected ECG feature and the amplitude 'centre of mass' obviously varies and can be determined empirically, e.g. ~0.2 s average delay when the QRS complex has

been marked in healthy young adults (Allen et al., 1998). However, if a different part of the ECG is marked, the delay needs to be defined individually and ideally automatically. One practical way to achieve this is to calculate the smoothed global field power (Skrandies, 1990) only for those channels containing above average artifact power and to determine the temporal midpoint between the local minima bordering the global field power maximum (documented within and implemented as "CBC Parameters" [2006] in BrainVision Analyzer, Brain Products, Munich, Germany).

Other methods use channel-wise ECG-locked temporal PCA (Negishi et al., 2004; Niazy et al., 2005), or PCA of representative epochs of data building a spatial filter to remove pulse-artifact related components (Benar et al., 2003). The question of the selection and refinement of the components to remove is a problem inherent to data driven approaches. Similarly for ICA, that can be used to further process the averaged pulse-contributed signal (Nakamura et al., 2006) or to determine and remove related components (Benar et al., 2003; Niazy et al., 2005; Srivastava et al., 2005; Nakamura et al., 2006; Mantini et al., 2007a,b). Again, combining different approaches can be advantageous (Debener et al., 2007a,b), but because of the non-stationarity of the signal, the use of statistical procedures such as ICA and PCA is limited (Debener et al., 2007a,b).

In principle, the quality of different pulse and imaging artifact correction methods can be evaluated by looking at signal to noise measures, or, in the context of a specific application, by seeing whether a signal property is preserved, such as the known topography of an event related potential component (Debener et al., 2007a,b). Nevertheless, it would be very difficult to prove superiority of one artifact subtraction method over another, as the outcome depends not only on the quality criteria applied but also on whether all methods in question have been optimally applied. For example, methods not requiring exact triggers (e.g. scan onset markers or ECG feature markers) may seemingly outperform those which are dependent on accurate timing signals, when reliable synchronization has not been achieved. It is thus reasonable to select or further develop one of the existing methods for artifact subtraction based on the individual needs and data (Grouiller et al., 2007). Because of the additive nature of all artifacts to the biologically generated electrical field, average artifact subtraction techniques are *a priori* valid; in addition, they are unbiased, computationally efficient and can be performed online (Allen et al., 2000). The latter feature is for instance relevant in the context of electrophysiological biofeedback fMRI investigations extending existing work in this area (Nagai et al., 2004a,b).

Analysis strategies

Importantly, fMRI measures the haemodynamic changes associated with synaptic activity (Logothetis et al., 2001; Heeger and Ress, 2002; Shmuel et al., 2006) while EEG measures the electrical field of a subset of (neuronal) cells on the scalp. The exact relationship between these measurements remains to be determined. Accordingly, so far, two main perspectives have been used to integrate fMRI and EEG data; firstly, using fMRI to better determine the source of the measured electrical EEG signal and secondly, trying to find the common neural "origin" of both the EEG and fMRI signals in a broader sense (Fig. 4). The first approach is usually based on averaged EEG event related responses used with fMRI-derived activations to constrain EEG source localization. The second relates more generally to the identification of a functional state of the brain associated with the EEG features that can be used to interrogate the simultaneously measured fMRI data, e.g. in the form of an EEG-based general linear model. It has been shown in generalised, and to some extent focal epilepsy (Gotman et al., 2006; Laufs et al., 2006a,b,c), as well as in ongoing background EEG activity, that fMRI signal changes can reflect associated changes in brain function characterised by EEG, e.g. attentional state (Laufs et al., 2003a,b). This is opposed to EEG-correlated fMRI activations representing the electrical sources of the measured EEG phenomena.

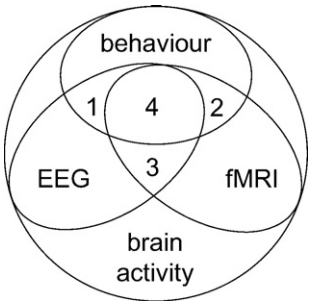

Fig. 4. Analytical perspective on the integration of electrophysiological and haemodynamic data. Across all neural processes, only a fraction is reflected by EEG, fMRI and behaviour. Some neural processes will manifest in EEG and behaviour (1) or fMRI and behaviour (2). Of the neural processes reflected in both EEG and fMRI there may also be measurable behavioural manifestations (4) or not (3). In both cases, 3 and 4, however, the correlation between EEG and fMRI is direct in that there is a common substrate of neural activity. If behaviour is related to neural processes that also manifest in EEG (1) and fMRI (2) independently, yet without being the identical processes at the source of EEG and fMRI effects, this situation can still result in an indirect but meaningful correlation between fMRI and EEG. Simultaneous multi-modal experiments benefit from situations where common neural processes are at the origin of EEG and fMRI signals but the most benefit is derived when these neural processes cannot be monitored by or recalibrated to behaviour (3). The difficulty in using either technique, EEG or fMRI, for predicting or constraining results in the other lies in the uncertainty as to whether one is recording data from situations 3 or 4 or from a joint situation of 1 and 2. In the latter case, prediction and constraint are not justified because the neural processes recorded in the two modalities are non-identical.

We chose to present three approaches to combining electrophysiology with fMRI based on previous work (Horwitz and Poeppel, 2002; Kilner et al., 2005): integration through i) prediction; ii) constraints; iii) fusion with forward models. i) Particularly under largely uncontrolled experimental conditions such as stimulus- and task-free relaxed wakefulness or sleep, spontaneous physiological as well as pathological brain activity can be studied with electrophysiologically-informed fMRI: The electrophysiological data is used to predict variance in the fMRI data (forward model from the electrophysiological data to fMRI). For example IEA or sleep spindles are identified on the EEG to define events or epochs in an fMRI analysis (Gotman et al., 2006; Laufs et al., 2007). Similarly, continuous EEG/EMG information, e.g. in the frequency domain, can be correlated with fluctuations in the ongoing fMRI activity (Salek-Haddadi et al., 2003; Menon and Crottaz-Herbette, 2005; Richardson et al., 2006; Ritter and Villringer, 2006). More sophisticated EEG pre-processing has allowed the generation of predictors for fMRI analysis from single

trial ERP data (Eichele et al., 2005; Debener et al., 2006; Bagshaw and Warbrick, 2007). This approach is an excellent example of a paradigm-controlled experiment in which EEG data adds information beyond that inherent in the paradigm, in distinction to combined EEG/fMRI studies not requiring simultaneous multimodal recordings (Bledowski et al., 2006). In addition, electrophysiological data can be used not only to model factors which confound the fMRI data such as cardiac activity and respiration (Glover et al., 2000; Liston et al., 2006; Laufs et al., 2007) but also "mental confounds", i.e. uncontrolled variability in brain activity during task performance, for example related to alertness (Henning et al., 2006; Laufs et al., 2006a,b,c; Scheeringa et al., 2007). An 'inverse mapping' approach has been applied where fMRI data from a region of interest was correlated with EEG frequency bands in order to identify the specificity of their contribution to an effect observed in the fMRI data (Laufs et al., 2003a,b). ii) Focal fMRI activations (e.g. in response to a known paradigm) can be used to constrain source estimates of the electrophysiological data (Dale et al., 2000; Phillips et al., 2002; Daunizeau et al., 2005; Im and Lee, 2006), mainly applied in ERP research. In clinical applications, BOLD signal changes could be used to inform EEG source modelling, but it has to be kept in mind that the two modalities do not measure exclusively identical phenomena: it has become clear that although a subset of IEA-related fMRI activations partly overlap with IEA sources (Benar et al., 2006), [whole brain] fMRI signal changes can in addition reflect network activity and global brain state changes that are likely a consequence and not the source of the IEA (Gotman et al., 2005; Laufs et al., 2006a,b,c). iii) The goal of a fusion approach is to utilise both electrical and haemodynamic measurements simultaneously and symmetrically in spatio-temporal assessment of brain function (Kilner et al., 2005; Daunizeau et al., 2007). Because the aforementioned EEG/fMRI combination strategies rely on the introduction of constraints derived from a preliminary analysis of fMRI into the EEG source reconstruction problem (Liu et al., 1998; Babiloni et al., 2003) these approaches are said to be *asymmetrical*. In other words, they

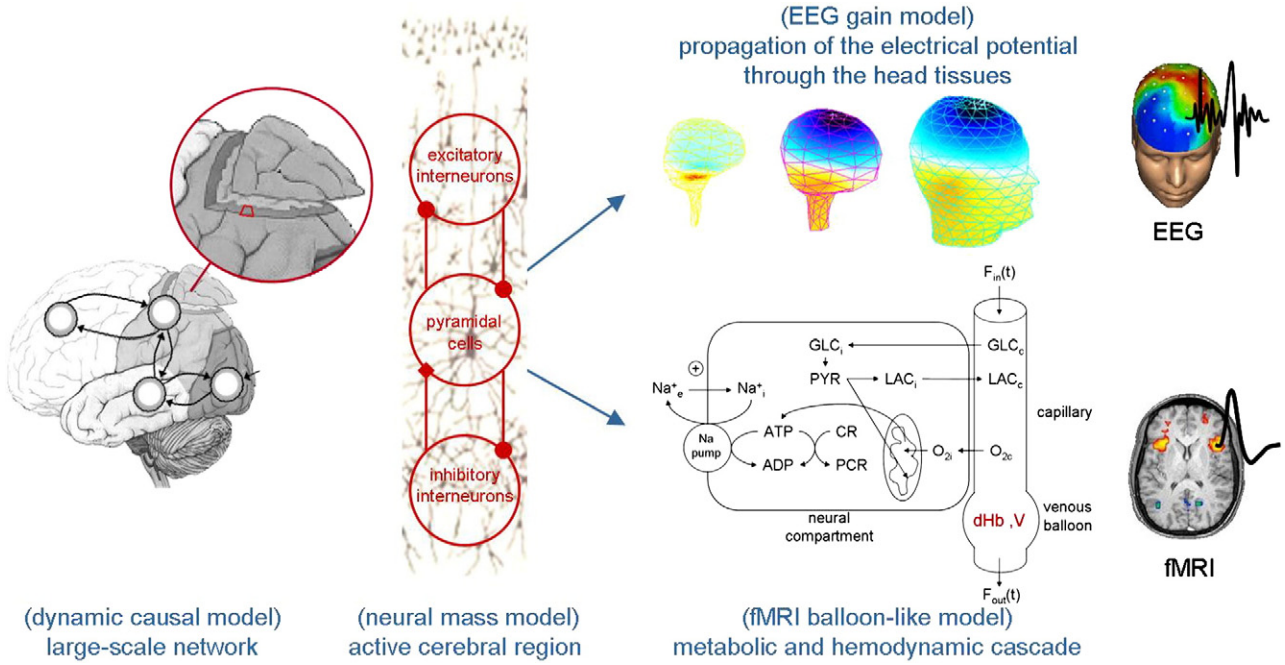

Fig. 5. Integrative EEG/fMRI symmetrical fusion model. Recent advances in understanding physiological mechanisms at different spatiotemporal scales have provided a framework within which to develop sophisticated biophysical models that permit an integration of different imaging modalities, each sharing a common aetiology. More precisely, evolution and observation equations encoding the relationship between bioelectric and hemodynamic mesostates can be motivated using both physiological and physical facts. Increasing experimental evidences have shown that the dynamical behaviour of an active region is the resultant of both its intrinsic and extrinsic connectivity. Dynamic causal models [*Friston, K.J., Harrison, L., Penny, W., 2003. Dynamic causal modelling. NeuroImage 19, 1273–1302*] relying on neural masses [*Jansen, B.H., Rit, V.G., 1995. Electroencephalogram and visual evoked potential generation in a mathematical model of coupled cortical columns. Biol. Cybern. 73, 357–366*] may then seem an appropriate framework for deriving models of bioelectric activity of the active neural populations [*Kiebel, S.J., Garrido, M.I., Friston, K.J., 2007. Dynamic causal modelling of evoked responses: The role of intrinsic connections. NeuroImage 36, 571–580*]. On this common basis, both the EEG/Meg and fMRI data can be predicted. On the one hand, the EEG scalp data is assumed to be an instantaneous measure of the electrical potential generated by the activity of a subpopulation of active neural masses (the pyramidal cells), which has been propagated (and spread) through the head tissues [*Baillet, S., Mosher, J.C., et Leahy, R.M. (2001b). Electromagnetic brain mapping. IEEE Sign. Proc. Mag., 18*: *14–30*]. On the other hand, the fMRI data seem to be related to a temporally over-smoothed response to mostly presynaptic neuronal activity [*Logothetis, N.K., Pauls, J., Augath, M., Trinath, T., Oeltermann, A., 2001. Neuro-physiological investigation of the basis of the fMRI signal. Nature 412, 150–157*]. This response is the final consequence of a slow cascade of both metabolic and hemodynamic events [*Aubert, A. et Costalat, R. (2002). A model of coupling between brain electrical activity, metabolism and hemodynamics*: *application to the interpretation of functional neuroimaging. Neuroimage, 17*: *1162–1181*], [*K.J. Friston, A. Mechelli, R. Turner, and C.J. Price. Nonlinear responses in fMRI: the Balloon model, Volterra kernels, and other hemodynamics. Neuroimage, 12:466–477, 2000*] and [J. *Riera, X. Wan, J.C. Jimenez, and R. Kawashima. Nonlinear local electrovascularcoupling. I: A theoretical model. Hum. Brain Mapp., 25, 2006*]. Inversion of such an integrative model for EEG and fMRI might then provide us with the key components of both neuronal activity and intrinsic/extrinsic connectivity structure.

do not consider EEG and fMRI data sets as equivalent and do not analyze them jointly (Trujillo-Barreto et al., 2001). Importantly, a potential discrepancy between bioelectric and haemodynamic activities will result in an estimation bias affecting the cortical current density (Ahlfors and Simpson, 2004). Therefore, several authors have proposed assessing the relevance of the fMRI-derived prior information regarding the electrophysiology data itself, so as to choose between the fMRI-constrained and non-constrained current density estimates (Daunizeau et al., 2005; Mattout et al., 2006).*Symmetrical* fusion approaches (Fig. 5) require the explicit definition of the common neuronal process that elicits both EEG and fMRI signals (Fig. 4). This entails building a forward model that encompasses our knowledge about the link between neural and surface bioelectric and haemodynamic signal changes (Logothetis et al., 2001; Shmuel et al., 2006; Daunizeau et al., 2007). The first steps towards true symmetrical electrophysiologic–haemodynamic data information fusion is currently grounded in compound neural masses and haemodynamics models (Riera et al., 2005; Babajani and Soltanian-Zadeh, 2006; Sotero et al., 2007).

Naturally, any EEG/fMRI fusion procedure which may be qualified as a 'model-based' approach (as is any symmetrical fusion approach) will suffer from the usual limitation of modelling: refutability. Whether the assumptions of the model are satisfied or not in a given experimental context is a question in itself. There is a subtle balance between the assumption's plausibility, and the interpretation power of any model. The tighter the prior belief regarding the underlying causes of the observations, the more stringent the interpretations of the data. However, the more exploratory the data analysis, the more flexible the posterior opinion regarding the unknown causes of the observations. A promising way of dealing with that issue is information-theoretic model comparison. Probabilistic inference methods already exist that furnish model accuracy measures (model marginal likelihood/evidence), which allows one to select models in a principled way. Along these lines, an increasing number of Bayesian model comparison methods have already been applied to the various modalities of neuroimaging data analysis (Penny et al., 2004; Trujillo-Barreto et al., 2004; Daunizeau et al., 2005; Penny et al., 2006; Behrens et al., 2007; Jbabdi et al., 2007; Kiebel et al., 2008; Stephan et al., 2007). In the context of EEG/fMRI symmetrical fusion, model comparison may be a way to both selecting the more plausible neuro-vascular coupling scenarios and quantifying the uncertainty about them.

Summary and outlook

The potential to study "spontaneous", i.e. task-unrelated neuronal events and their haemodynamic correlates is a unique feature of non-invasive multimodal electrophysiology and fMRI recordings in humans. Other advantages in contrast to recording different modalities separately include the avoidance of order effects and guaranteed identical sensory stimulation and subjective experience respectively as well as identical behaviour. The main technical challenges have been successfully addressed both in terms of acquisition and post-processing of the inevitably artifact-laden data. EMG recordings are feasible but await further improved artifact reduction methods dealing with motion in the field. Analytically, we expect the next milestone to be i) the refinement and *in vivo* validation of data fusion models, which should optimally utilise the information obtained by multi-modal imaging studies and ii) the identification of candidate neuronal and haemodynamic (un-)coupling processes and their disambiguation. Intracranial EEG and fMRI have been simultaneously acquired in animal studies, and this approach leads the way to empirically determining the bioelectric-haemodynamic coupling function (Logothetis et al., 2001; Shmuel et al., 2006). This next technical milestone for investigations of the human brain is already being addressed by related safety studies and simulations (Georgi et al., 2004; Boucousis et al., 2007; Carmichael et al., 2007a,b). For obvious ethical reasons the only comparable human data that could possibly be obtained would be acquired in patient populations. Although the prerequisite technical and safety studies remain to prove that this is feasible, some related studies with implanted deep brain stimulator electrodes appear promising (Georgi et al., 2004; Phillips et al., 2006). Even if obtained in patients, these data could provide information about physiological processes that would be at least as interesting as observations of the specific pathology and that would be invaluable for the validation of models required for symmetric EEG/fMRI fusion.

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